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ANATOMICAL STRUCTURE STUDY OF *Solanum torvum* Sw.

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ABSTRACT

This study, conducted with meticulous attention to detail, aims to complete the anatomical survey of Solanum torvum and to identify characteristic microscopic features that can serve as specific markers, including the type of trachea in the stem, tracheids and fibers in the stem, and the stem cross-section at the branching part. The research data were analyzed using a qualitative descriptive method by comparing the characteristics of the samples with those reported in the references. The results of this study include image and qualitative descriptive data on the anatomy of the roots, stems, and leaves of Solanum torvum. The results showed that the leaf and root exhibited a similar pattern of anatomical structures, as in the references. The stem part is composed of an epidermis with star-shaped non-glandular trichomes, lamellar collenchyma, cortex, vascular tissue comprising xylem and phloem, and pith. The anatomical difference between unbranched and branched stems lies in the arrangement of the vascular bundles, the thickness of the supportive tissue, and the complexity of the vascular tissue, indicating structural adaptations to the position and function of the stem. On the stem found scalariform trachea, annular trachea, reticulate trachea, thickened reticulate trachea, tracheid, fiber tracheid, and libriform fiber.

INTRODUCTION

The study of plant anatomy, a crucial field in botany, aims to unravel the intricate internal structure of plants, including cells, tissues, and organs at the microscopic level (Evert & Eichhorn, 2020). This study is not just an academic pursuit, but a key to understanding how plants adapt to environmental conditions (Dewi et al., 2025) and identifying the characteristics that connect ancient plants in the fossil record with those that still exist today (Sokoloff et al., 2021). Our research on the anatomy of plants in the Solanaceae family, a group with high diversity and significant morphological and anatomical variation, including Solanum torvum, is of paramount importance. Previous anatomical studies of Solanaceae showed significant variations in internode thickness, vascular tissue patterns, and trichome differentiation among Solanaceae genera, including *Solanum tuberosum*, *Capsicum annuum*, and *Datura stramonium* (Tribble et al., 2021). Some species of *Hyoscyamus* exhibit anatomical differences in stems, stomatal types, and mesophyll tissue (Satil et al., 2015). Several key characters are important for distinguishing species within the genus (Ainiah et al., 2020) and for understanding the similarity relationships among species within a family (Jannah et al., 2023).

According to Satil (2015), mesophilic types of several Solanaceae species can be divided into bifacial (*Hyoscyamus niger* L., *Hyoscyamus albus* L.) and equifacial (*Hyoscyamus aureus* L., *Hyoscyamus pusillus* L., *Hyoscyamus reticulatus* L.). Nevertheless, specific data regarding the anatomy of the *Solanum torvum* species in Indonesia is still limited. In Indonesia, *Solanum torvum* grows wild in the forest, and its fruit is used as a side dish. Based on previous research, *Solanum torvum* is a plant with many benefits, including antibacterial properties (Wiryani et al., 2023), and its leaves contain high levels of nutrients and antioxidant compounds (Helilusiatiningsih, 2021).

Our research on the anatomy of Solanum torvum, a plant with many benefits, including antibacterial properties (Wiryani et al., 2023), and high levels of nutrients and antioxidant compounds in its leaves (Helilusiatiningsih, 2021), not only contributes to scientific knowledge but also serves as a valuable educational resource. According to Nurit-silva et al. (2011), in their research on the anatomy of *Solanum torvum*, its leaves are amphistomatic, with anisocytic and anomocytic stomata present simultaneously, and the epidermal cells, in frontal view, have undulating anticlinal walls on the upper surface and wavy anticlinal walls on the lower surface. The mesophyll is dorsiventral; the petiole has a central bicollateral vascular bundle; and the leaf stalk has three or four. The stem shows external phloem, xylem, internal phloem, and parenchyma pith. The roots are axial and elongated in shape, and the secondary xylem is a massive cylinder. The macro- and microscopic vegetative characters constitute a set of diagnostic parameters for the studied species (Nurit-silva et al., 2011; Amantayeva et al., 2023). However, there is still no recent, detailed research on the anatomy of *Solanum torvum*, especially regarding several other parts, such as the type of trachea in the stem, tracheids and fibers in the stem, and the cross-section of the stem at the branching point. Therefore, this research aims to complement the anatomical study of *Solanum torvum* and to identify characteristic microscopic features that can serve as specific anatomical markers. This research is important for contributing to the scientific documentation of local plant taxonomy, supporting the utilization of plants in the fields of pharmacy and conservation, and serving as a reference for botanical education in Biology. Microscopic information can serve as an important foundation for characterizing the morpho-anatomy of this species.

MATERIALS AND METHODS

This research was a collaborative effort, conducted from January to May 2025. Samples of the *Solanum torvum* plant were taken from the roots, stems, and leaves. Specimen preparation was performed using manual, paraffin, and maceration methods, each method contributing to a comprehensive understanding of the plant's anatomy. The manual process was used for cross-sections of the stem, leaves, and abaxial and adaxial leaf sections. Meanwhile, the paraffin method was applied to cross-sections of the roots and the stem branches. The maceration method was used for the stem part. The preparation of anatomical specimens of the plant (*Solanum torvum*) was conducted in the Botanical Laboratory and in the Biology Education Laboratory of the Faculty of Teacher Training and Education at Sriwijaya University, Indralaya, a testament to the synergy between these institutions in advancing botanical research. The research data were analyzed using descriptive qualitative methods, comparing the characteristics of the samples with references from the books by Gembong Tjitrosoepomo (2010) and Fritz Hans Schweingruber & Annett Börner (2018). This aims to facilitate the observation of plant anatomy. This is in accordance with the research conducted by Ermayanti (2017), which states that representation is needed in the form of 2- or 3-dimensional images or verbal representation to more easily understand the anatomical characteristics of each plant organ (Ermayanti et al., 2018; Mukti et al., 2022).

Manual Method (Free-Hands Section)

The manual method, also known as the Free-Hands Section, is a precise process that involves careful handling of the plant (*Solanum torvum*). The intact plant or the one transferred into a polybag is used for this method. The stems and leaves are cut into small pieces about 3 cm long. Cross-sections of the stem and leaves, as well as abaxial-adaxial cuts of each plant sample being observed, are manually made using a razor blade as thin as possible. The cuts are immediately placed on a clean, sterile glass slide, and then droplets of distilled water and safranin dye are added. A cover slip is then carefully placed over the sample, using a technique that avoids excess pressure to prevent air bubbles from forming. The prepared specimens are then observed using a binocular microscope at varying magnifications, from low to high. Documentation is then carried out for all thin, clear samples (Ermayanti, 2023).

Paraffin Method

The paraffin method, a meticulous process, was used for the plant (*Solanum torvum*). Sections of its stem, leaf, root, and fruit measuring 1-2 cm were fixed by immersing them into a FAA solution (70% alcohol, glacial acetic acid, and formalin) for 24 hours. The aspiration phase used a desiccator until the bubbles that appeared had disappeared. The dehydration phase involved replacing FAA with 70% alcohol every 2 hours, then maintaining the 70% alcohol with sapranin for 48 hours for staining, followed by a series of 96% and 100% alcohol, each for 2 times and 2 hours. The de-alcoholization process used xylene as the alcohol in the following ratio.

Table 1. Comparison table of alcohol series and xylene

Comparison (Alcohol : Xylene)	Time
3:1	1.5 hours
1:1	1.5 hours
1:3	1.5 hours
Xylene I	1.5 hours
Xylene II	1.5 hours

The infiltration stage involves inserting paraffin into vials, replacing it every 2-3 days, and heating the vials in an oven for 1-4 hours until xylene completely evaporates. The embedding and trimming stages use paper molds sized to the microtome block holder. The block is allowed to freeze, then removed from the box and cut to a thickness of 15-30 µm using a microtome. The paraffin ribbons are taken with a mounted needle and then submerged in water until they float on the surface. The paraffin ribbons are placed on a glass slide and heated on a hot plate at 50°C until the remaining wax melts. Position the glass slide at a 30° angle so that the liquid paraffin flows downward, and gently wipe it clean with a tissue until the paraffin is gone. Your role in this stage is crucial as you observe the sections with a binocular microscope and document them (Ermayanti, 2023).

Maceration Method

The maceration method is a systematic process that, when followed correctly, yields reliable results. Cut the branches of the *Solanum torvum* plant into four pieces, each approximately 1-2 cm long. Boil in plain water for 5 minutes. Place the wood slices into a small test tube. Add 20% KOH to the test tube. Boil water in a 100 mL beaker on a hot plate until it reaches a boil. When the water is boiling, place the test tube containing the wood slices in it and boil for 30 minutes. Transfer the KOH solution containing the wood into another 100 mL beaker, then discard the KOH solution and wash with running water. Add a 20% nitric acid:1:1 solution to the beaker. Soak the wood slices in a 20% nitric acid solution for 2 hours. Wash with running water until the acid solution is gone using gauze and rubber assistance. Dehydrate with 30%, 50%, and 70% alcohol for 3 minutes each, then add safranin during the 70% alcohol dehydration. Leave it overnight in safranin for the color to adhere better to the cells. Dehydrate the wood again using 90% and 100% alcohol. Observe the macerated sample using a microscope (Ermayanti, 2023).

RESULTS AND DISCUSSION

The results of this study include image and qualitative descriptive data on the anatomy of the roots, stems, and leaves of *Solanum torvum*. The data were obtained by comparing the characteristics found with various references and previous studies. The transverse section of *Solanum torvum* root (Figure 1) showed the characteristic structural features of young dicot root tissue. The outermost part consisted of the epidermis (Ep), a thin layer of cells lacking a thick cuticle. The cortex layer (Ko) contained cortex parenchyma, which occupied most of the cortex. These cells had thin walls, with sufficiently wide intercellular spaces, and functioned as storage and lateral transport pathways for water and nutrients. The characteristic endodermis (En) consisted of a single layer of tightly packed cells. It featured a Casparian strip, a distinctive suberin- and lignin-containing structure that regulates the entry of dissolved substances into the central cylinder (stele) (Calvo-Polanco et al., 2021). Pericycle (Pe) or pericambium was a layer of parenchyma cells located between the endodermis and vascular tissues, such as xylem and phloem, in the roots of plants. Although it consisted of non-vascular cells, the pericycle was considered part of the vascular cylinder because it originated from the procambium, similar to the surrounding vascular tissues. The pericycle played an essential role in the formation of lateral roots. This process began with the division of pericycle cells adjacent to the protoxylem, forming lateral root primordia. These primordia then grew through the cortex and epidermis, eventually becoming branch roots. In dicotyledonous plants undergoing secondary growth, the pericycle contributes to the formation of the vascular cambium. Pericycle cells divided and differentiated into vascular cambium, which then produced secondary xylem and phloem, thereby increasing the diameter of the roots.

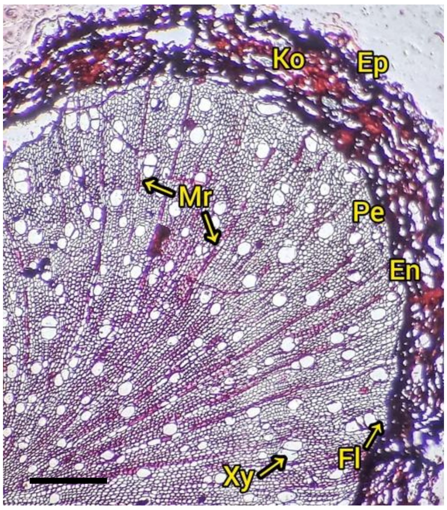


Figure 1 Cross section of *Solanum torvum* root. Ep: epidermis, Ko: cortex, En: endodermis, Pe: pericycle, Xy: xylem, Fl: phloem, Mr: medullary ray (scale bar = 100 μm).

The special structure of the medullary ray (Mr) consisted of parenchyma tissue organized radially from the centre of the root towards the outside. This structure played a role in the lateral movement of water, nutrients, and photosynthesis products, and participated in food storage. Medullary rays were also present in the anatomy of the *Acacia hybrid* stem, resulting from the crossbreeding of *Acacia mangium* and *Acacia auriculiformis* (Sunarti et al., 2018). The vascular tissue of the phloem (Fl) and xylem (Xy) was organized radially (arachnoid type). The entire central cylinder was surrounded by the pericycle (which was not clearly visible in the image). This structure played a role in the formation of lateral roots in dicot roots as the outer layer of the stele (surrounding the xylem-phloem).

The stems of *Solanum torvum* showed an apparent variation in anatomical structure between the unbranched and branched parts. In the unbranched stems (Figures 2A–2C), the constituent tissues appeared organized and symmetrical. The epidermis consisted of a single layer of cells with thick walls and star-shaped trichomes that served as protection (Hidayat, E., 1995; Wang et al., 2021). Beneath the epidermis layer, there was a reasonably wide cortex that consisted of parenchyma and lamellar collenchyma. The cambium was located between the xylem and phloem, forming a continuous ring that actively divided to produce secondary xylem toward the inside and secondary phloem toward the outside. This meristematic layer contributed to secondary growth and stem thickening. The vascular bundles were organized in a circular pattern, forming concentric layers. The xylem was large, while the smaller phloem outside the xylem was of the open collateral type and thus exhibited eustele type.

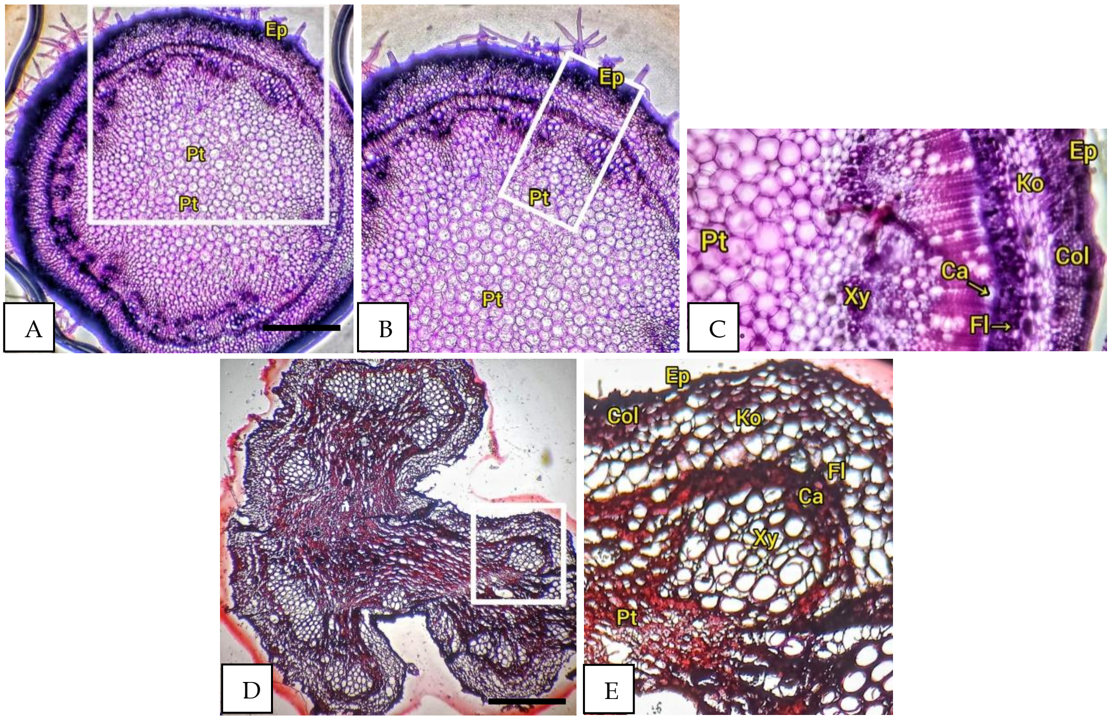
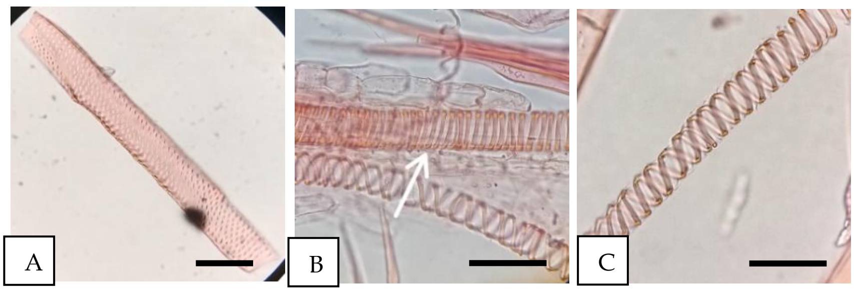


Figure 2. A-C cross sections of the stem of *Solanum torvum* without branching parts. D-E cross sections of the stem of *Solanum torvum* at the branching parts. Ep: epidermis, Col: collenchyma, Ko: cortex, Fl: phloem, Ca: cambium, Xy: xylem, Pt: pith (scale bar = 100 µm)

Branching stems (Figures 2D–2E) had a more complex and asymmetrical anatomical structure. The arrangement of vascular bundles was no longer neatly circular but more dispersed, with a tendency to be layered, with the same type of dispersal —open collateral —to accommodate the more intense transport needs towards the branches. The lamellar collenchyma and cortex tissues showed more significant thickening, providing additional strength to support branch growth. The stem cross-section was also not fully circular, reflecting functional and structural changes in response to branching. The striking differences between these two sections lie in the arrangement of the vascular bundles, the thickness of the supporting tissue, and the complexity of the vascular tissue, indicating structural adaptations to the stem's position and function.

The stem of *Solanum torvum* (Figure 3) showed the presence of scalariform tracheae (Figures 3A, 3D, 3E) that had elongated holes resembling a ladder, along with spiral tracheae (Figure 3C) and an annulus (Figure 3B) that demonstrated thick spiral and ring-shaped secondary walls, indicating that the tracheae remained flexible as the tissue elongated. Annulus tracheae were stronger than ring tracheae because their spirals were continuous. In some areas, reticulate tracheae (Figure 3F) were observed, consisting of thickened tissue with holes between, as well as simple perforated tracheae with a single large hole that allowed water flow. The trachea showed thickening in a reticulate, net-like pattern (Figure 3G). This thickening was recognized by the net-like tissue pattern that resulted from irregularly distributed secondary wall thickening, creating a weave that strengthened the tracheal cell structure. This pattern provided flexibility and sufficient mechanical strength to support the transportation of water and minerals in the plant. On the other hand, tracheids (Figure 3H) were characterized by long, tapered cells lacking perforations, with pits (minor points) commonly found in gymnosperms. Additionally, some illustrations showed xylem fibers, such as fiber tracheids and libriform fibers, characterized by elongated, slender shapes and thick secondary walls, serving as mechanical support.



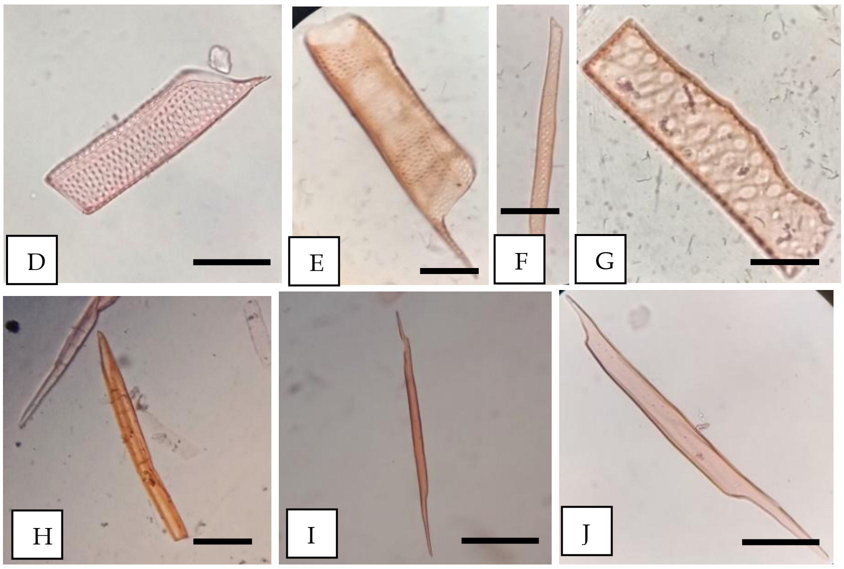


Figure 3 A-H trachea of *Solanum torvum* and I-J fibers in the stem of *Solanum torvum* A, D, E: scalariform trachea, B: annular trachea, C: spiral trachea, F: reticulate trachea, G: thickened reticulate trachea, H: tracheid, I: fiber tracheid, J: libriform fiber (scale bar = 20 μm)

The leaf petiole of *Solanum torvum* (Figures 4A-4D) had an anatomy that consisted of various tissues with specific functions, starting from the outer layer to the innermost. The epidermis (Ep) consisted of flat and tightly packed cells. Beneath the epidermis, there was lamellar collenchyma tissue (Col) with unevenly thick cell walls, particularly in young leaves and the petiole. Parenchyma tissue (Pa) dominated the petiole's structure, contributing to nutrient storage and photosynthesis, with cells having thin walls to facilitate efficient nutrient exchange. The vascular bundle consists of phloem (Fl) on the outside and xylem (Xy) on the inside with an open collateral arrangement, in accordance with the research results of Nugroho & Pratiwi (2023). In addition, there were idioblasts (Id), individual cells with specialized functions, particularly in protection and storage. The characteristic pattern of the leaf skeleton of *Solanum torvum* was marked by a straightforward arrangement of vascular bundles, dominance of parenchyma, and the presence of supporting tissues such as collenchyma, all of which contribute to the leaf's physiological and structural functions. This supported the research conducted by Filartiga et al. (2022) regarding the anatomy of the petiole of *Solanum torvum.*

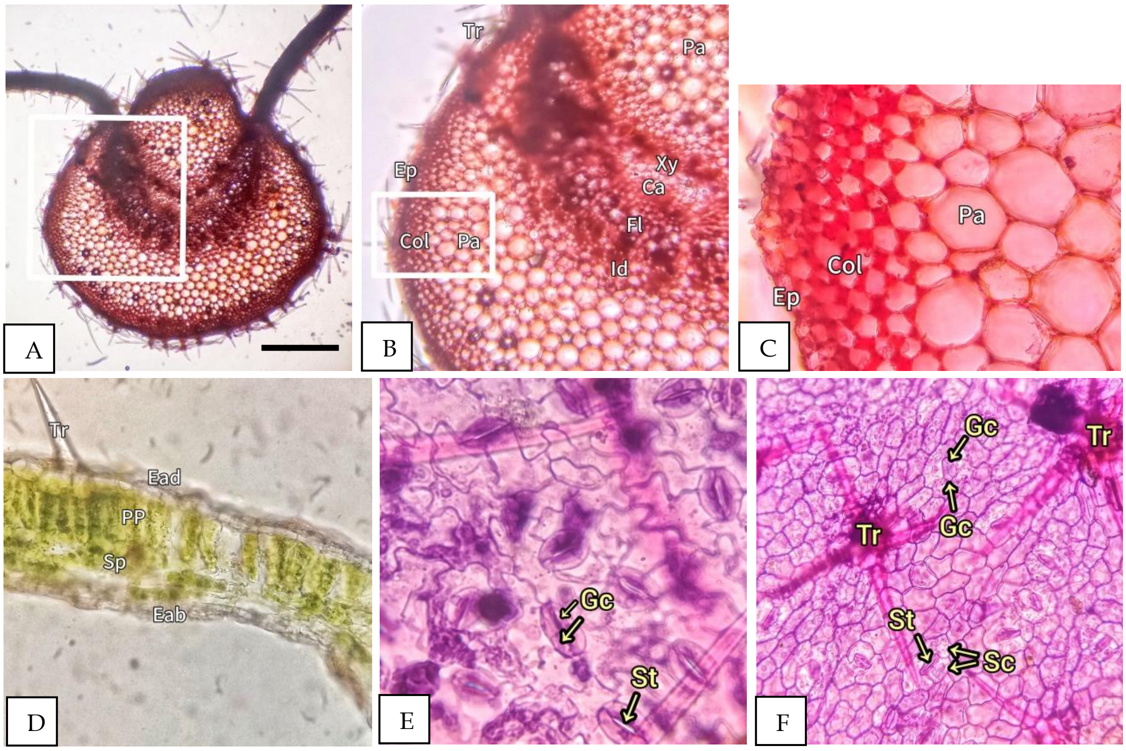


Figure 4 A-C cross-section of the leaf bone. Tr: trichome, Ep: epidermis, Col: collenchyma-lamellar type, Pa: parenchyma, Id: idioblast, Fl: phloem, Ca: cambium, Xy: xylem. D: cross-section of the leaf blade. Tr: star-shaped non-glandular trichome, Ead: adaxial epidermis, PP: palisade parenchyma, Sp: spongy parenchyma, Eab: abaxial epidermis. E: abaxial surface of the leaf, anomocytic stomata type. F: adaxial surface of the leaf, anisocytic stomata type. St: stoma/stomata, Gc: guard cell, Sc: subsidiary cell (scale bar = 100 µm)

The cross-section of the *Solanum torvum* leaf (Figure 4D) showed that the outer layer consisted of star-shaped trichomes (Tr) protruding from the epidermis, serving as a protective adaptation against herbivores and abiotic stress (Johnson, H.B., 2021). The adaxial epidermis (Ead) at the top of the leaf was composed of cuboidal-shaped cells with thickened outer walls and stomata surrounded by three neighboring cells of unequal size, an anisocytic type. This was similar to what Wahyuni, S. et al. (2021) stated in their research on SEM micrograph analysis of anomocytic stomata and their relation to water efficiency. Below the epidermis, the palisade parenchyma (PP) was tightly arranged, with elongated cells containing numerous chloroplasts, optimizing light absorption for photosynthesis. The deeper, spongy parenchyma (Sp) contained irregular cells with large intercellular spaces, facilitating gas exchange and the temporary storage of metabolic products. The abaxial epidermis (Eab) on the underside of the leaf contained scattered stomata, lacked specialized neighboring cells, and was surrounded by 3-4 ordinary epidermal cells that played a role in transpiration and gas exchange, of the anomocytic stomata type. The characteristic pattern of *S. torvum* included multiseriate palisade parenchyma (composed of several layers of cells) as an adaptation to high light intensity and an asymmetrical distribution of stomata with higher density on the abaxial surface (Gomez-Nuñez, J.C. et al., 2021).

CONCLUSION

The anatomical observation of *Solanum torvum* showed apparent variations between the unbranched and branched stem parts. The unbranched stem had a regular, symmetrical structure, whereas the branched stem displayed a more complex, asymmetrical arrangement of vascular tissues, reflecting adaptation to branching growth. Several types of tracheae, — scalariform, spiral, and reticulate, — were identified, along with tracheids and xylem fibers that strengthened the stem and supported efficient water transport. These findings indicated specific anatomical adaptations of the stem related to vascular differentiation and mechanical reinforcement in *Solanum torvum*. The leaf epidermis had a higher stomatal density on the abaxial surface. The adaxial epidermis of *Solanum torvum* had anisocytic-type stomata, while the abaxial epidermis possessed anomocytic-type stomata.

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