

Anatomical Studies of Diagnostic Flora in Ago-Igbira Community Along Owo-Ose Road Side Ondo State Nigeria

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Abstract- Anatomical studies of plants are crucial for understanding their structural adaptations, identification, and classification. Anatomy of plants can also provide valuable information on their ecological and environmental relationships. This study investigates the anatomical characteristics of *Sida acuta* and *Hiptis suaveolens*, being two diagnostic flora found in Ago-Igbira community along Owo-Ose road, Ondo State, Nigeria. The study employed standard anatomical techniques to examine the leaf structures of both plants which includes; stomata type, trichome type, type of venation *e.t.c*. The results revealed distinct anatomical features, including leaf venation patterns, presence of different types of stomata *i.e* anisocytic, paracytic and diacytic on the adaxial and abaxial surfaces of the examined plants which can aid in identification and authentication. Stomata size were found to be larger on the examined plants than the control plants which suggest modification of structural part for adaptation. The study conclude that road side plants tend to modify their structures as adaptive strategies to survive in the environment. The research also highlights the importance of conservation efforts for these plants, particularly in the face of habitat destruction and pollution along the Owo-Ose road, a case study of Ago-Igbira environment.

Keywords: *Sida Acuta*, *Hiptis Suaveolens*, Anatomical Studies, Diagnostic Flora, Ago-Igbira, Owo-Ose Road.

I. INTRODUCTION

Global advancement in industrialization and urbanization is generally associated with increased demand for energy particularly from fossil fuels, which has lead to increase/rise in the amount of emissions in the air (Karmakar *et al.*, 2016). A large number of toxic impacts are added to the environment from industries, manufacturers, and automobile engines and potentially degraded plant species. Automotive exhaust emissions have damaging impact on plants' growth occurring in many urban areas (Bell *et al.*, 2011). Plant species are enlisted as threatened due to air pollution and these

has harmful impacts on plants species particularly those growing along the roads sides. (Krishnaveni *et al.*, 2015). Plants are prone to several unfavourable environmental conditions regarded as stresses (pollutants from several sources) which do alter their internal structures, metabolisms, growth and resulting yield (Reddy *et al.*, 2004). Anatomical structures vary in plants of different species under the same exposure to air pollution (Makbul *et al.*, 2008). The effects of air pollutants are usually easily noticed on the leaves and flowers of plants with symptoms such as strange colourations, twisting of leaves and relative stunted growth (Otoide and Kayode, 2016). However, leaf is the most sensitive and exposed part to be affected by air pollutants instead of all other plant parts such as stems and roots (Leghari and Zaidi, 2013). Air pollution can cause both acute and chronic damages to the anatomical, morphological, physiological and biochemical characteristics of plant species (Karmakar and Padhy, 2019; Kaur and Nagpal, 2017). A larger percentage of these pollutants has been reported to be from automobile exhaust, some of the pollutants from automobile exhausts include carbon oxides, volatile organic carbons and particulate matters (dusts) (Otoide, 2015). This study tends to investigated the anatomical responses of some diagnostic species on the road side to air pollutants from automobile exhausts or the impact of vehicular emission on the morphology, anatomy, and physiology of road side plant species

II. MATERIALS AND METHODS

Study area

This study was carried out in Ago-Igbira at latitude 78°E 65°S along Owo-Ose road in Owo-Ose Local Government Area of Ondo State.

Sample collection

Samples of *Sida acuta* and *Hiptis suaveolens* being the diagnostic flora along the road side were collected

and taken to the laboratory of Plant Science and Biotechnology, Adekunle Ajasin University, Akungba Akoko for further observations. Samples of same plant species were collected 15km inward from the road side which serves as the control samples.

Leaf anatomical Studies

Matured leaves were cut from the standard median positions and subjected to clearing process following the methods of Oloyede *et al.* (2011). The cleared sample were mounted on slides and observed with the aid of a digital compound photomicroscope to view the internal structure such as veination types, presence of crystals e.t.c.

Leaf segment of an area of 1cm from each samples were cut and immersed in concentrated solution of nitric acid for 10-20 min. The upper (adaxial) and the lower (abaxial) surfaces were separated with dissecting needle and forceps and rinsed with clean water. Each specimen was stained with 1% aqueous safranin for 3-5 min and excess stain rinsed off in water. The samples were then mounted on glycerin jelly for microscopic observation of the internal structures (such as stomata and trichome types) using digital compound photomicroscope (Alvin and Boulter, 1974).

Terminologies used for naming of shape of Trichome follow those used by Dilcher (1974) and Metcalfe and Chalk (1950). Frequency of each trichome type were expressed as percentage occurrence of such trichome type based on all occurrences (Metcalfe and Chalk, 1950).

$$\text{Frequency (\%)} = \text{Nt/Tt} \times 100$$

Where Nt = number of particular type; Tt = total number of trichome types.

Trichome index were determined as number of trichomes divided by number of trichomes plus number of epidermal cell per square millimeter multiplied by 100 (Metcalfe and Chalk, 1950).

Using a digital compound photomicroscope at x40 objective in a total of 30 fields of view, the number of subsidiary cells per stoma were counted to determine the frequency of the complex type present in each specimen. Frequency of each complex type were expressed as percentage occurrence of such complex type based on all occurrences (Obiremi and

Oladele, 2001). Terminologies for naming stomatal complex types follow those of Dilcher (1974) and Metcalfe and Chalk (1950). Stomatal index (SI) were determined as follows:

$$\text{SI} = \text{S/E} + \text{S} \times 100$$

Where: SI = Stomatal Index, S = number of stomata per square millimeter, E = number of ordinary epidermal cell per square millimeter (Salisbury, 1927).

The mean stomatal size of a species was determined using this formula:

$$l \times b \times K$$

Where l = length b = breath K = Franco's constant (0.78524)

Statistical Analysis

The data generated from this study were analyzed statistically by using Statistical Package for Social Sciences (SPSS). Analysis of Variance was used to test for differences in the various anatomical structures.

III. RESULT

Table1 shows Anatomical features of *Sida acuta* and *Hyptis suaveolens* along Ago-Igberira road side and the control sample. *Hyptis suaveolens* Main plant (MP) and Control plant (CP) of Adaxial surface has an anticlinal wall pattern that is wavy with irregular shape epidermal cell, stomata complex type of the main plant is anisocytic and the control plant is paracytic, the stomata frequency (84%) main plant is higher than control plant (76%), stomata size of the main plant (1.57 μm) is higher than the control plant (0.942 μm) stomata index of the MP is 46.49 \pm 10.01 and CP is 43.79 \pm 7.31, glandular trichome is present in both with a trichome frequency of 100%, trichome size in both is (6.01 μm), the trichome index of the CP is higher (7.81 \pm 1.86) and the MP is lower (3.81 \pm 0.19).

Hyptis suaveolens Main plant (MP) abaxial surface has anticlinal wall pattern that is wavy the control plant is undulating, their epidermal cell are both irregular in shape, stomata complex type of the main plant is anisocytic and anomocytic and the control plant is anisocytic, the stomata frequency (80%) main plant is higher than control plant (74%), stomata size of the MP (1.009 μm) is higher than the CP

(0.007µm), stomata index of the CP is 54.60±5.40 and MP is 49.79±7.84, glandular trichome is present in both with a trichome frequency of 100%, trichome size of the MP (7.01µm) is higher than the CP (5.01µm), the trichome index of the MP is lower (2.21±1.12) and the CP is higher (3.92±0.41).

Sida acuta Main plant (MP) adaxial surface has anticlinal wall pattern that is wavy the CP is undulating, their epidermal cell are both irregular in shape, stomata complex type of the main plant is anisocytic and control plant is diacytic, the stomata frequency (87%) MP is higher than the CP (84%), stomata size MP (0.104µm) is higher than CP (0.072µm), stomata index of the MP is 56.69±6.93 and the CP is 43.37±9.10, glandular trichome is present in both the CP and MP with a trichome frequency of 100%, trichome size of the main plant

(6.67µm) is higher than the control plant (0.46µm), the trichome index of the MP is higher (4.81±0.19) and CP 3.82±0.18 is lower.

Sida acuta Main plant (MP) and the control plant (CP) abaxial surface has anticlinal wall pattern that is wavy, both epidermal cell are irregular in shape, stomata complex type of the MP and CP are anisocytic, stomata frequency (83%) of the main plant and (85%) of the control plant, stomata size of MP (0.89µm) is higher than CP (0.072µm), stomata index of the MP 58.47±3.00 is higher and CP 53.54±10.54 is lower, glandular trichome is present in both MP and CP with frequency of 100%, trichome size of the main plant (5.67µm) is higher than the control plant (5.01µm), trichome index of the main plant (3.18±0.83) is higher than the control plant (1.88±0.78).

Table1: Anatomical features of *sida acuta* and *Hyptis suaveolens* along Ago-Igbira roadside and the control sample

S/N	Taxon	Leaf Surface	Anticlinical wall Pattern	Shape of epidermal Cell	Stomata Complex Type	Stomata frequency (%)	Stomata Size (µm)	Stomata index	Trichome Type	Trichome frequency (%)	Trichome Size	Trichome Index
1	<i>Hyptis suaveolens</i> MP	Adaxial	Wavy	Irregular	Anisocytic	84	1.57	46.49±10.01	GT	100	6.01	3.81±0.19
2	<i>Hyptis suaveolens</i> MP	Abaxial	Wavy	Irregular	Anisocytic Anomocytic	80	1.009	49.79±7.84	GT	100	7.01	2.21±1.12
3	<i>Hyptis suaveolens</i> CP	Adaxial	Undulating	Irregular	Paracytic	76	0.942	43.79±7.31	GT	100	6.01	7.81±1.86
4	<i>Hyptis suaveolens</i> CP	Abaxial	Wavy	Irregular	Anisocytic	74	0.007	54.60±5.40	GT	100	5.01	3.92±0.41
5	<i>Sida acuta</i> MP	Adaxial	Wavy	Irregular	Anisocytic	87	0.104	56.69±6.93	GT	100	6.67	4.81±0.19
6	<i>Sida acuta</i> MP	Abaxial	Wavy	Irregular	Anisocytic	83	0.89	58.47±3.00	GT	100	5.67	3.18±0.82
7	<i>Sida acuta</i> CP	Adaxial	Undulating	Irregular	Diacytic	84	0.072	43.37±9.10	GT	100	0.46	3.82±0.18
8	<i>Sida acuta</i> CP	Abaxial	Wavy	Irregular	Anisocytic	85	0.072	53.54±10.54	GT	100	5.01	1.88±0.78

NOTE: CP- Control Plant, MP-Main Plant, GT-Glandular Trichome.

Fig 1 shows Abaxial surface of *Hyptis suaveolens* (main plant) showing anisocytic stomata complex type. Fig 2 shows Adaxial surface of *Hyptis suaveolens* showing anisocytic stomata complex

type. Fig 3 shows the Adaxial of *Hyptis suaveolens* (control plant) showing glandular trichome type. Fig 4 shows the Abaxial of *Hyptis suaveolens* (control plant) showing a paracytic stomata complex type. Fig

5 shows the Abaxial of *Sida acuta* (main plant) showing anisocytic stomata complex type. Fig 6 shows Adaxial of *Sida acuta* (main plant) showing anisocytic stomata complex type. Fig 7 shows Abaxial of *Sida acuta* (control plant) showing anisocytic stomata complex type. Fig 8 shows Adaxial of *Sida acuta* (control plant) showing

diacytic stomata complex type. Fig 9 is a diagram showing Venation of *Hyptis suaveolens* (main plant). Fig 10 is a diagram showing Venation of *Hyptis suaveolens* (control plant). Fig 11 is a diagram showing Venation of *Sida acuta* (main plant). Fig 12 is a diagram showing Venation of *Sida acuta* (control plant).

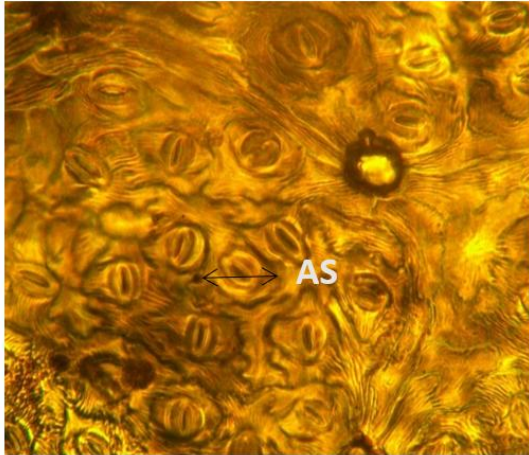


Fig. 1

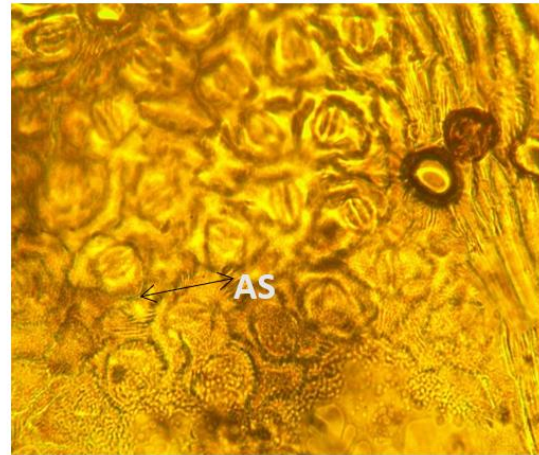


Fig. 2

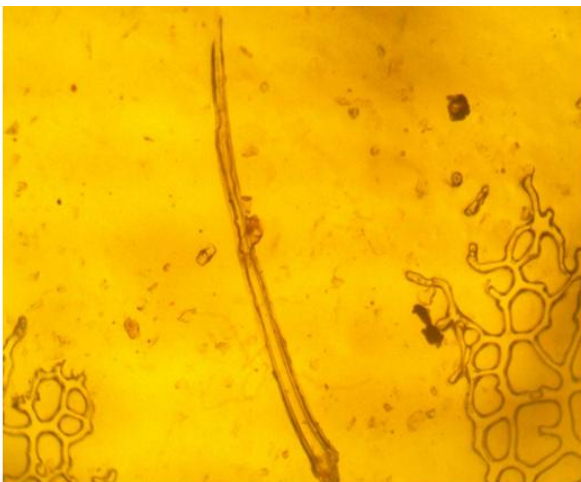


Fig. 3

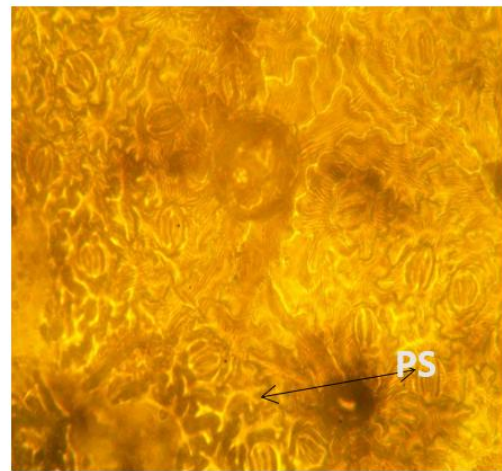


Fig. 4

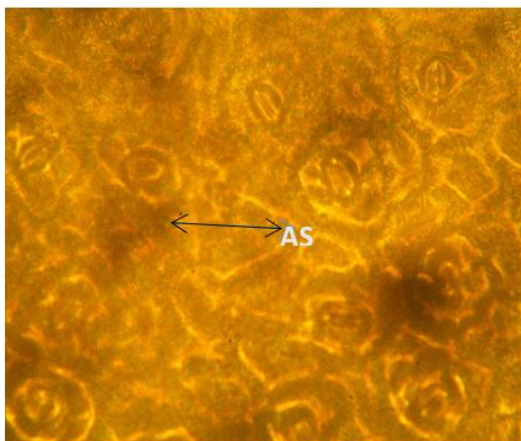


Fig. 5

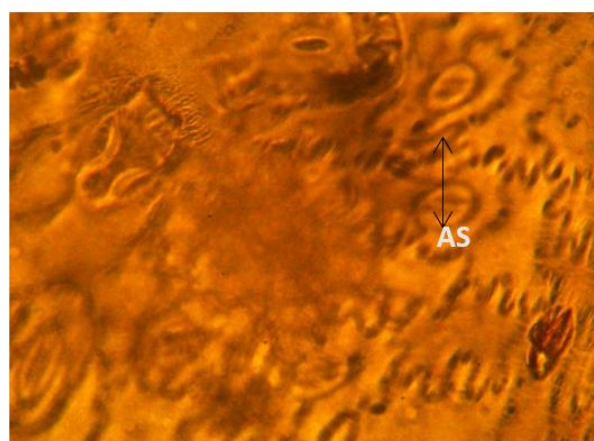


Fig. 6

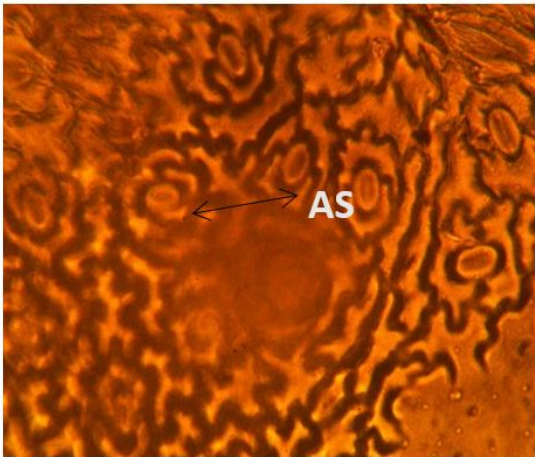


Fig. 7

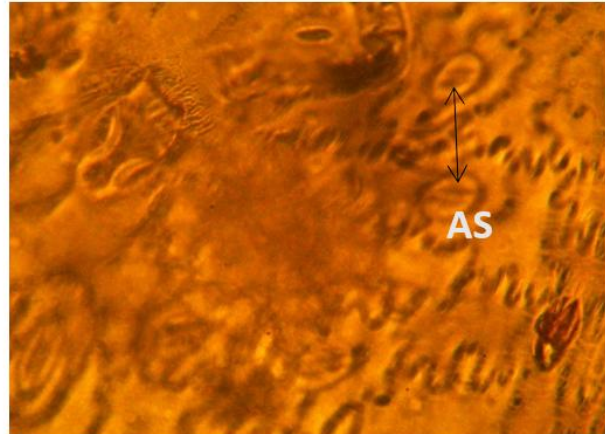


Fig. 8

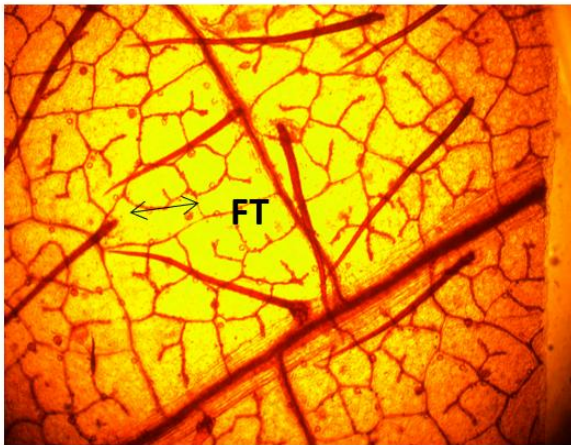


Fig. 9

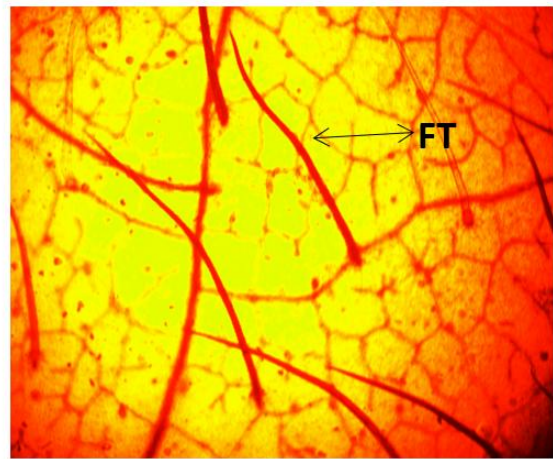


Fig. 10

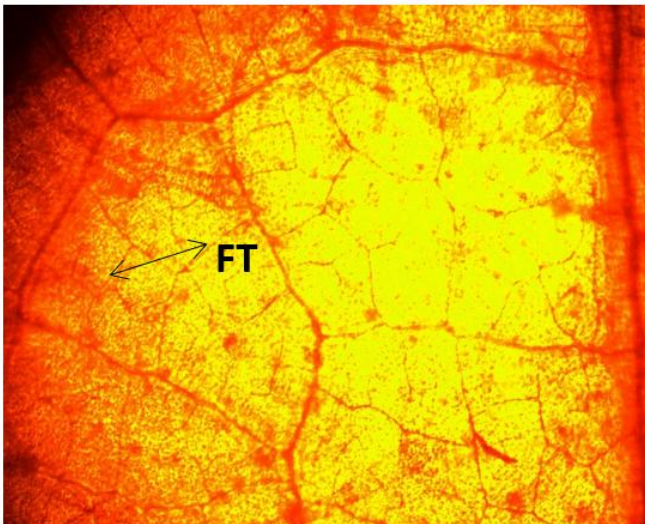


Fig. 11

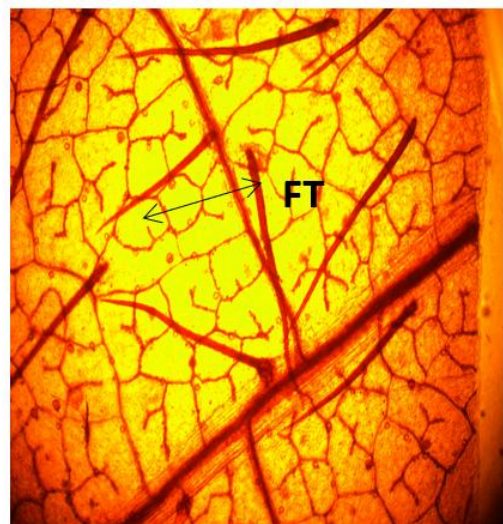


Fig. 12

IV. DISCUSSION

Sida acuta and *Hyptis suaveolens* are two tropical plant species widely distributed in Nigeria, particularly in the southwestern region. These plants

have been used in traditional medicine for various purposes, including antimicrobial, anti-inflammatory, and antioxidant activities (Kokate *et al.*, 2022). Despite their medicinal importance, there is a dearth of information on their anatomical

characteristics, particularly in specific regions like Ago-Igbira along Owo-Ose road in Ondo State, Nigeria.

Anatomical studies of plants are crucial for understanding their structural adaptations, identification, and classification (Cutler *et al.*, 2018). The anatomy of plants can also provide valuable information on their ecological and environmental relationships (Fahn, 2018).

The adaxial (upper) surface of *Hyptis suaveolens* Main plant (MP) exhibits a wavy anticlinal wall and irregular epidermal cells, both common in plants to support flexible growth and protection (Smith and Woolley, 2020). The anisocytic stomatal complex with 100% stomatal frequency and relatively high stomatal index (46.49) indicate a significant capacity for gas exchange on the upper surface. The glandular trichomes (GT) and their high density suggest that this surface may have defensive or secretory roles, aligning with findings by Adams *et al.* (2019) on adaptive trichome presence for herbivore deterrence. The abaxial (lower) surface of *Hyptis suaveolens* MP also shows a wavy anticlinal wall pattern and irregular epidermal cells, with anisocytic stomata and a slightly higher stomatal index (49.79) compared to the adaxial side. The smaller stomatal size and higher trichome frequency (100%) align with findings by Lee and Khan (2021), who observed that plants often display higher stomatal densities on the lower surface to optimize gas exchange while reducing water loss. The Adaxial Surface of *Hyptis suaveolens* (control plant) exhibits undulating anticlinal walls and paracytic stomata type with only 10% stomatal frequency suggest reduced gas exchange on the adaxial surface, possibly to minimize water loss under certain environmental conditions. The stomatal index is also slightly lower (43.79). Research by Johnson and Green (2022) supports the idea that reduced stomatal density on the upper leaf surface aids in water conservation. The high trichome frequency indicates a defensive role, consistent with findings from Schultz *et al.* (2023), who reported increased trichome density in plants under water stress. However, the Abaxial surface exhibit a wavy anticlinal wall pattern, anisocytic stomata, and 100% stomatal frequency, the abaxial surface in this CP sample exhibits one of the highest stomatal indices (54.60), indicating substantial stomatal density that supports efficient gas exchange in sheltered conditions, which conforms to results by Pires *et al.*

(2020) on enhanced abaxial stomatal densities for optimized photosynthesis. The lower trichome index (3.92) compared to adaxial surfaces suggests that defensive structures are prioritized elsewhere, possibly in response to differing environmental pressures on the lower leaf surface.

For *Sida acuta* (MP) the adaxial surface with anisocytic stomata shows a high stomatal index (56.69), supporting active photosynthesis and transpiration processes, especially in well-lit environments, as indicated by Dawson and Patel (2021). The trichome index (4.81) confirms a moderate density, potentially serving dual roles in UV protection and herbivore defense (Garcia and Kim, 2020). This Adaxial surface shows the highest stomatal index (58.47), indicating high gas exchange efficiency this align with the work of Ekundayo (2022) who discover that stomatal size in *Sida acuta* was larger (5.21 ± 1.20 and 6.43 ± 1.25 μm) for abaxial and adaxial surfaces of the main plant respectively compared to (1.44 ± 0.13 μm) for the control, this attribute has been associated with increased growth rates in humid or shaded environments (Zhao *et al.*, 2021). The glandular trichomes, slightly smaller here, may offer specific resistance to pests targeting the underside of leaves. The adaxial Surface of the CP shows undulating wall pattern and a unique diacytic stomatal complex, the low stomatal frequency (10%) on this surface likely serves to minimize water loss in stressful environments.

The low trichome index here suggests this surface may be less reliant on trichomes for defense. The Abaxial Surface shows a wavy pattern with anisocytic stomata at 100% frequency mirrors the high-density trend across abaxial surfaces. The stomatal index (53.54) supports robust gas exchange, consistent with prior research by McDonald *et al.* (2019) on abaxial stomatal density in plants adapted to high-light or high-moisture environments. The relatively low trichome index (1.88) implies limited reliance on trichomes for defense, possibly due to less environmental exposure.

Thus, *Hyptis suaveolens* and *Sida acuta* adaptively allocate stomata and trichomes on their leaf surfaces. Generally, high stomatal densities and specialized trichome structures provide enhanced gas exchange and defense, supporting prior findings that these features are adapted in response to environmental pressures. The wavy and undulating anticlinal

patterns further support flexibility and resilience, confirming that morphological adaptations are instrumental in the survival strategies of both species.

The larger size and consistent presence of glandular trichomes (GT) in *Hyptis suaveolens* could explain its aromatic nature, as glandular trichomes often store essential oils and secondary metabolites. This anatomical feature aligns with previous research that links glandular trichomes to aromatic characteristics in the Lamiaceae family, to which *Hyptis suaveolens* belongs (McDonald *et al.*, 2019).

The anatomical analysis reveals distinct characteristics in both species. *Sida acuta* demonstrates wavy and undulating anticlinal wall patterns across both adaxial and abaxial surfaces, supporting findings from similar studies where epidermal cells show irregular undulating structures. These patterns play a key role in leaf rigidity and environmental adaptation (Ekundayo, 2022). In contrast, the wavy anticlinal walls of *Hyptis suaveolens* are more associated with water retention and aromatic properties, suggesting a relationship between epidermal structure and chemical functions.

Trichome analysis highlights significant interspecies differences. *Hyptis suaveolens* displays both glandular trichomes, aligning with its aromatic properties. Glandular trichomes are known to produce essential oils, contributing to its pharmacological uses. In contrast, *Sida acuta* shows a trichome with less differentiation, supporting findings by Ekundayo (2022) that suggest simpler trichomes aid in basic mechanical defense but lack specialized chemical roles. The trichome index is notably higher in *Hyptis suaveolens*, which may further support its defensive and medicinal functions.

V. CONCLUSION

The *Hyptis suaveolens* and *Sida acuta* species exhibit distinct anatomical variations between their adaxial and abaxial surfaces. Generally, the abaxial surfaces show higher stomatal densities and a variety of stomatal complex types, while trichome frequencies remain consistently high across all samples. The differences between MP and CP variants (perhaps different environmental or experimental conditions) show that CP samples tend to have lower stomatal and trichome indices on adaxial surfaces, indicating

environmental adaptation or genetic variability between the MP and CP groups.

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