DOI: 10.21608/TAEC.2024.277137.1054 Taeckholmia 44 (2024):48-72

Comparative study on the effect of pollution on leaf morphology of common ornamental species between polluted and peri-urban regions in Egypt

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Abstract

Environmental pollution is one of the major problems facing the megacities. This study was carried out to investigate the effect of air pollution on the leaf morphological characters of 12 species commonly planted in the streets of Giza district, Cairo a highly polluted site and Smouha district, Alexandria as peri-urban non polluted site of Egypt. Both the Light (LM) and Scanning Electron Microscopes (SEM) were used to investigate the leaf characters. The thirty-one macro-and micro-morphological leaf characters were subjected to T-TEST to assess the influence of pollution on the leaf characters and to find out the significant variations among the species in the two sites (polluted and non-polluted), using SPSS software, in addition to, Bivariate correlation using Pearson correlation. The study results proved that leaf characters of the studied species exhibited significant reduction at polluted sites especially in leaf length, width, area, and number of lateral veins compared with non-polluted sites, and identified the most adapted species to the polluted sites.

Keywords: Air pollutants; leaf characters; metropolitan city; phyllo remediation; stomata

Introduction

By the beginning of this century earth faces a great change in climate which affects all the living creatures as well as human health responding to the rise in air temperature and air pollutants (Semenza, 2014). This change in air temperature is significantly sensitive in urban areas with crowded populations (Roth, 2013). Planting of the ornamental plants in megacities like Giza clean up the environment is called phylloremediation, this is a process to accelerate the uptake of

Received on 5/3/2024 and accepted on 23/5/2024

contaminants in the surrounding environment by the plant leaves, as well as capturing the dust from the air (Rai et al., 2010, Corada et al., 2021, Shao and Kim, 2022). Capuana (2020) reported several tree species with high capacity to absorb, tolerate, and translocate contaminants in urban cities. Gardens and green open areas are prominent in any city to absorb the pollutant, shading and reflect heat waves (Al-Obaidy et al., 2019).

Several studies have recorded great changes in morphology, anatomy, and even physiological pathways of plants as a result of environmental pollutants (Ekpemerechi, et al. 2014, Abu Zaida et al., 2015, Al Obaidy et al., 2019, Niklas et al., 2023, Shakeel et al., 2023). Many studies revealed changes in leaf mass, area, shape, as well as nutrient contents, and even both photosynthesis and respiration, beside stomatal characters, also leaf venation and vein characters in response to air pollution (Shi et al., 2021, Shi et al., 2022). The size and wall thickening of the guard cells as well as the epicuticular wax deposition beside trichome densities on the lamina are noticeably changed in polluted areas (Uka et al., 2019).

Song et al. (2019) and Andersona et al. (2020) warned against using unsuitable ornamental plants which can disturb the surrounding environment or compete with neighboring plants. Although Wang et al. (2018), Zhao et al. (2019) and Zhu et al. (2020) reported that trees planted in megacities and garden trees differ than those grow in the wild. Li and Wang (2021) focused to the necessity of studying leaf morphological characters which include leaf size, type, shape, margin type beside number of teeth and all the leaf morphological features to understand their adaptive ways in obtaining their different resources and enable them to overcome the surrounding disorders.

Although a lot of studies have been done to investigate the relationship between urban vegetation and air pollution, it remains unclear to which degree air pollutants can be captured by trees (Prigioniero et al., 2023). Canopies of the urban trees and shrubs can stabilize ecosystem services in two ways; keeping pollutants on their leaf surfaces or absorb them into their tissues (Livesley et al., 2016, Łukowski et al., 2020). Wei et al. (2017) gave a review in the role of plant leaves in stabilizing the air pollutants.

Ottelé et al. (2010) used SEM to investigate the quantity of PM (Particulate Matter; the term for a mixture of solid particles and liquid droplets found in the air) pollution on leaf surfaces. Their observations switched the light to many scientists to investigate which are the most important micro-morphological leaf characters for capturing PM (Chen et al., 2017, Weerakkody et al., 2018a, Wang et al., 2019, Zhang et al., 2019, He et al., 2020). Previous studies reported that the most effective micro-morphological features for PM retention are leaf surface roughness, presence of trichomes, cuticular wax, and stomatal density (Weerakkody et al., 2017, Zhang et al., 2018).

This work has been done to investigate the leaf morphological feature in twelve plant species of the common ornamental plants found in the streets of one of the metropolitan cities namely Giza city within the great Cairo and compared with the same species grown in Smouha district, Alexandria city. The main objectives of this work are: 1- to investigate the leaf macro- and micro-morphological characters of these species. 2- to investigate their response to air pollutants, 3- to recommend which taxa are more suitable for planting in urban polluted areas to improve the air quality to achieve the goal of Egypt Vision 2030 to reduce 50% of air pollution by 2050.

Materials and Methods

A- Study sites

Giza district west of Cairo (polluted site) is a very important part of the Great Cairo, the metropolitan city in Egypt. The air quality in Giza is full of industrial emissions besides the dust storms from El-Mokatam Plateau and CO_2 gas from agricultural waste burning in the Nile Delta (Hassan and Khoder, 2017). The percentage of air pollution in Great Cairo exceeded 11 times the limit approved by the World Health Organization (https://www.iqair.com/egypt). Giza climate is very hot in summer and considered the hot, arid place (https://en-climate-data.org). Smouha district, Alexandria (non-polluted site) is a peri-urban region with less polluted air referring to its position close to Al-Nozha Great Park and Smouha Club with plenty of green areas and trees.

B- Plant material

Reconnaissance of roads in Giza and Smouha sites have been carried out from March 2023 to September 2023, to identify the most common planted species in both sites. This survey revealed the selection of twelve common ornamental species (Table 1). Young branches were collected from ten individuals/species at five roads/sites, were subjected for investigation to make the necessary measurements. Description and terminology of the macro-morphological characters was according to Stearn (1986).

C- Data collection

Measurements of leaf macro-characters (leaf length, width and petiole length) were carried out using ordinary ruler in ten mature leaves/species. Young leaf was chosen to make the peels from both surfaces by gentle heating in water with few drops of Tepol and Nitric acid. The peels mounted onto clean microscopic slides enriched with few drops of water and glycerol, then examined carefully, and photographed using 40x10x magnification lens in OPTICA (B-150D) light microscope fitted with USB digital-Video Camera and Computer Software.

Table 1. Studied species and their taxonomic position according to Engler and Prantl system

No	Order	Family	Genus	Species	Citation	Origin
1	Malvales	Malvaceae	*Hibiscus* L.	*H. rosa-sinensis* L.	Sp. Pl.: 694 (1753)	Tropical Asia
2	Rosales	Moraceae	*Ficus* L.	*F. carica* L.	Sp. Pl.: 1059 (1753)	Mediterranean region
3				*F. elastica* Roxb. ex Hornem	Hort. Bot. Hafn., Suppl.: 7 (1819)	Asia
4		Rosaceae	*Rosa* L.	*R. hybrida* Vill.	Hist. Pl. Dauphiné (Villars) 3(1): 554. 1788	Egypt
5		Fabaceae	*Cassia* L.	*C. fistula* L.	Sp. Pl. 377. 1753	India, Malaysia and S.E. Asia
6				*C. javanica* L.	Sp. Pl.: 379 (1753)	South East Asia
7			*Delonix* Raf.	*D. regia* (Bojer ex Hook.) Raf.	Sp. Pl.: 379 (1753)	Madagascar
8			*Senna* Mill.	*S. surattensis* (Burm. f.) Irwin & Barenby	Mem. New York Bot. Gard.35(1): 81(1982)	Tropical Asia to Australia.
9	Myrtales	Lythraceae	*Punica* L.	*P. granatum* L.	Sp. Pl.: 472 (1753)	Afghanistan and Iran.
10	Lamiales	Bignoniaceae	*Jacaranda* Juss.	*J. mimosifolia* D. Don	Bot. Reg. 8: t.631 (1822)	Argentina and Bolivia
11			*Tecoma* Juss.	*T. stans* (L.) Juss. ex Kunth	Nov. Gen. Sp. Pl. 3:144 (1819)	S. America
12		Verbenaceae	*Lantana* L.	*L. camara* L.	Sp. Pl.: 627 (1753)	Mexico

Density of trichomes and epidermal cells outline were observed from the careful examination of the peels under the light microscope. Stomatal type, guard cell size, stomatal density and index were examined under light microscope. The terminology used in stomata description was according to Dilcher (1974). The stomatal density (D) and stomatal index (I) were calculated according to Kadiri and Olowokudejo (2008).

D (stomata in mm^2) = SN/A, I (stomatal index) =SN/(SN+EC) X 100 Where SN=Stomata Number, EC=Epidermal Cell & A=Area (mm^2).

Leaf area was measured using the "millimeter graph paper method" (Pandey and Singh, 2011), for all the species investigated leaves. The terminology used in stomata description was according to Dilcher (1974).

The detailed leaf epidermal structures (leaf surface, stomata, periclinal wall of the epidermal cell and trichomes) were investigated in dry leaves using SEM. Fragments of the leaves were stuck onto the Aluminum stubs using double cello tape and coated with 30 nm gold in a polaron JFC-1100E coating unit then examined and photographed under 15 Kev with JEOL JSM-IT200 SEM in the electron microscopes unit of faculty of science in Alexandria University.

D- Data analysis

The normality of the collected data was checked according to Kol-mogorov-Smirnov test; and then subjected to T-TEST (Independent samples-T test) to assess the influence of pollution on the leaf characters and to find out the significant variations of the species in both sites (polluted in Giza and non-polluted in Smouha) using SPSS software (version 20 for Windows). The similarity between the studied species were studied based on thirty-one macro-and micro-morphological leaf characters using the SPSS program and bivariate correlation using Pearson correlation. Finally, Histograms were developed using the Excel program (Microsoft 365).

Results

A- Macro-morphological characters:

The macro-morphological characters of the studied leaves from 12 species from the polluted and non-polluted sites are summarized in (Table 2). All the studied leaves were petiolate with different petiole length varied from 0.1 cm in *Delonix regia* to 10.5 cm in *Ficus carica*. Leaves were mostly exstipulate, except in *Hibiscus rosa-sinensis*, and *Senna surattensis* two filamentous hairy stipules were distinguished, while leafy stipules were found in *Cassia javanica*. In *Rosa hybrida* stipules were adnate to the petiole. Most of the studied species had simple leaves, except *Rosa hybrida* and *Tecoma stans* had imparipinnate compound leaves. While the paripinnate compound leaves were found in *Cassia javanica* and *Senna surattensis,* while bipinnate leaves were recorded in *Delonix regia* and *Jacaranda mimosifolia*. The leaf length and width varied greatly between the taxa growing in the two sites (Figs 1, 2).

A.1. Leaf area

The estimated leaf area was greatly varied between the polluted and non-polluted sites also with the leaf blade type (Fig. 3). Notable variation was observed in the leaf area of *Ficus carica* (20384 mm^2 in polluted site versus 30912 mm^2 within the non-polluted sites), while in *Ficus elastica* it was 11070 mm^2 in the polluted site and 19292 mm^2 in the non-polluted site. (Table 4).

A.2. Leaf blade texture and base

Leaf blade texture was the same in both polluted and non-polluted sites. It was mostly papery in most of the studied taxa, while it was leathery in *Hibiscus rosa-sinensis, Ficus carica* and *Ficus elastica,* and scarious in *Rosa hybrida*. The leaf blade base was anguste in most of the studied taxa; cordate in *Ficus carica* while it was truncate in *Ficus elastica.* On the other hand, asymmetric base was found in *Rosa hybrida, Delonix regia* and *Senna surattensis* (Table 2).

A.3. Leaf apex

The apex was apiculate in *Hibiscus rosa-sinensis, Rosa hybrida* and *Cassia fistula,* while obtuse apex was recorded in *Delonix regia* and *Jacaranda mimosifolia*. *Senna surattensis* had the retuse apex. *Tecoma stans* had the acuminate apex, mucronate apex was recorded in *Ficus elasica* and *Punica granaum*. Pungent apex recorded only in *Lantana camara* (Table 2).

A.4. Leaf margin and blade venation

The leaf margin of the studied species was mostly entire. Serrate margin was recorded in *Rosa hybrida* and *Tecoma stans*; crenate margin was observed in *Lantana camara*. The number of dentae, serrae or crenae/cm varied in the studied species between polluted (P) and non-polluted (NP) sites (Table 2). The type of leaf blade venation was unchanged between both sites. It varied between the studied species from the actinodromous in *Hibiscus rosa-sinensis* to the semicraspedodromous in *Tecoma stans,* while *Lantana camara* had mixed crasspedodromous. Brochidodromous type was recorded in four species: *Ficus elastica*, *Delonix regia, Senna surattensis* and *Punica granatum*; while the rest of the studied taxa have reticulodromous venation. The number of lateral veins greatly varied in the studied species between the two locations (P and NP, Fig. 4). There were ranged between 5 to 88 in *Ficus carica* and *Cassia fistula* at Giza site (P), while from 6 to 102 in the same species respectively at Smouha site (NP).

A.5. Trichomes

The leaves of the studied species appeared either hairy or glabrous under light microscope with variation in the trichome density between the two sites. The type and position of the trichomes were the same in the two sites. In the hairy leaves, the trichomes are distributed all over the leaf blade with different densities. While, in *Delonix regia* the trichomes were allocated at the base of the leaf blade only. In *Senna surattensis* the trichomes were sparse on the upper surface and dense on the lower surface in the polluted site whereas it was sparse on the upper surface and moderate on the lower surface in the non-polluted site (Table 2 cont.).

A.6. Stomata

Results of the stomatal characters were summarized in (Table 2 cont., Figs. 5 & 6). The leaves were amphistomatic i.e. both surfaces had stomata nearly of the same type in the two sites. All the observed stomata were of the anomocytic type. Guard cells were kidney- shaped, crescent, or linear with different lengths (Plate I, Figs. 7-18). From the kidney-shaped guard cells were *Hibiscus rosa-sinensis, Senna surattensis, Punica granatum* (Plate I, Figs. 7, 14 &15). While from the linear guard cells were *Tecoma stans* (Table 2 cont.). One of the linear- shaped stomata appeared with wavy margins as in *Ficus elastica* only (Plate I, Fig. 9). In both *Ficus carica* and *Jacaranda mimosifolia* (Plate I, Figs. 8 & 16) the guard cells were crescent.

Stomatal density and index differed between the two sites as it was bigger in the polluted area (Giza) than the non-polluted area (Smouha) as shown in (Fig. 6). The stomatal density in the polluted site varied from 6.1 in *Jacaranda mimosifolia* to 38.1 in *Punica granatum*. While the stomatal density varied in the non-polluted site from 5.8 to 32.6 in the same two species. The stomatal index in the polluted site varied from 14.2 in *Jacaranda mimosifolia* to 52.0 in *Punica granatum*. While the stomatal index varied in the non-polluted site from 13.2 in *Cassia javanica* and *Jacaranda mimosifolia* to 46.2 in *Punica granatum*.

B- Micro-morphological characters (SEM)

B.1. Epidermal cells

The leaf micro-morphological characters observed using the SEM were summarized in (Table 3). The epidermal system showed that the epidermal cells were mostly symmetric in all the studied species (Plate II, Figs. 19-41). The outline of the epidermal cell shape was isodiametric with either straight (Plate I, Figs. 8, 9 & Plate II, Fig. 32) or wavy walls (Plate I, Figs. 7, 14, & Plate II, Figs. 37, 39). Epidermal cells varied in shape between the studied species, either pentagonal (Plate I, Figs. 12, 13, 16 & Plate II, Fig. 32) or hexagonal (Plate I, Figs. 11, 17). The cell arrangements were areolate, alveolate, reticulate areolate, or reticulate foveolate (Plate II, Figs. 19, 27, 29). Ruminate arrangement was recorded only in *Tecoma stans* (Plate II, Figs. 37, 38), undulate arrangement was observed in *Punica granatum* (Plate II, Fig. 34). The thickness of the periclinal epidermal cells wall varied from thin, moderate, or even thick within the different species. The relief of the periclinal epidermal cell were either channeled, superficial, or raised, while the curvature of the anticlinal walls was concave, convex, or straight (Table 3). The anticlinal wall appearance was mostly granulated or striated. Wrinkled appearance was found in *Punica granatum* (Plate II, Fig. 34), while smooth appearance was recorded in *Rosa hybrida* (Plate II, Figs. 25, 26) and *Delonix regia* (Plate II, Figs. 30, 31). Wax depositions were of different densities, starting from absence in *Jacaranda mimosifolia* (Plate II, Fig. 35). Wax deposits were recorded as rosette wax platelets in *Delonix regia* and *Senna surattensis* (Plate II, Figs. 30-33).

B.2. Stomata and trichomes

Stomatal appearance and trichome characters were undifferentiated between the two sites (Giza and Smouha). The stomatal appearance was either sunken or superficial, where raised stomata were seen in *Hibiscus rosa-sinensis*, *Ficus carica* and *Ficus elastica*, (Plate II, Figs. 19, 22, 24). The stomata were mostly with a wide lenticular opening, except in *Ficus elastica* the opening was wide circular (Plate II, Fig. 24). The length of guard cells was varied greatly from 7.2 µm in *Punica granatum* to 18.7 µm in *Hibiscus rosa-sinensis* collected from polluted site, and from 8.0 µm in *Punica granatum* to 18.1 µm in *Hibiscus rosa-sinensis* collected from non-polluted site (Fig. 5, Plate I, Figs. 15, 7).

The studied species were either glabrous or hairy with different densities and types. Hairy species are either monomorphic, dimorphic, or polymorphic (Table 3). Dimorphic trichomes were observed in *Hibiscus rosa-sinensis* while polymorphic were recorded *Cassia fistula* and *Lantana camara* only (Table 3, Plate II, Figs. 19, 27, 39, 40). Several types of hairs had been recorded; biforked, stellate, uni-or/and multi-cellular non glandular pointed or hooked and papillary; on both the abaxial and adaxial surfaces. Capitate glandular trichomes were recorded in *Lantana* *camara* (Table 3, Plate II, Figs. 40, 41). The rest of the hairy species have multicellular uniserriate trichomes only. The hairs have rosette like base in both *Ficus carica* and *Lantana camara* (Plate II, Figs. 22, 41) or with undistinguished bases in the rest of the taxa. The trichome characters were the same in both sites (P & NP), except their density.

Table 2. Macro-morphological characters of the studied species
Abbreviations: - =absent, Ac=Actinodromous, Act=Acuminate, Ad=Adnate, Ag=Anguste, Al=Allover, An=Anomocytic, Ap=Apiculate, At=Acute Bi=Bi-pinnate, Br=Brochidodromous, C=Crescent, Cl=Cladodromous, Co=Cordate, Cre=Crenate, D=Dense, Dn=Dentate, El=Elliptical, En=Entire, H=Hairy, Im=Im-paripinnate, K=Kidney, L=Linear, Lc=Lanceolate, Lf=Leafy, Ls=Lower surface, Lth= Leathery, LW=Linear wavy, M=Moderate, Mu=Mucronate, Mx=Mixed craspedodromous, NLV=Number of lateral veins, Np=Non-polluted, Nt=Number of teeth/cm, Obl=Oblong, Ol=Oblanceolate, Obt=Obtuse, Ov=Ovate, P=Polluted, Pa=Papery, Par=Paripinnate, Pet.L=Petiole length, Pn=pungent, Rt=Reticulodromous, Sc=Scariosus, Se=Semicraspedodromous, Sp=Sparse, Sim=Simple, Sr=Serrate, Tl=Trilobate, U=Upper surface.

No.	Species	Pet. L (cm)	Stipule	Blade								
				Type	NT/(cm) (Mean)		Margin	Venation	Shape	Texture	Base	Apex
					P	Np						
1	***Hibiscus rosa-sinensis***	2.3±0.6	H	Sim	2	3	Dn	Ac	El	Lth	Ag	Ap
2	***Ficus carica***	10.5±2.0	-	Sim	3	3	Dn	Rt	Tl	Lth	Co	At
3	***Ficus elastica***	4.2±1.6	-	Sim	0	0	En	Br	Ov	Lth	Tr	Mu
4	***Rosa hybrida***	1.0±0.4	Ad	Im	4	6	Sr	Rt	El	Sc	In	Ap
5	***Cassia fistula***	0.9±0.1	-	Sim	0	0	En	Rt	El	Pa	Ag	Ap
6	***Cassia javanica***	0.3±0.1	Lf	Par	0	0	En	Cl	Ol	Pa	Ag	At
7	***Delonix regia***	0.15±0.1	-	Bi	0	0	En	Br	Ol	Pa	In	Obt
8	***Senna surattensis***	0.2±0.1	H	Par	0	0	En	Br	Ol	Pa	In	Ret
9	***Punica granatum***	0.5±0.1	-	Sim	0	0	En	Br	Lc	Pa	Ag	Mu
10	***Jacaranda mimosifolia***	0.45±0.1	-	Bi	0	0	En	Rt	Lc	Pa	Ag	Obt
11	***Tecoma st ans***	0.2±0.1	-	Im	3	5	Sr	Se	El	Pa	Ag	Act
12	***Lantana camara***	0.8±0.3	-	Sim	5	5	Cre	Mx	Ov	Pa	Ag	Pn

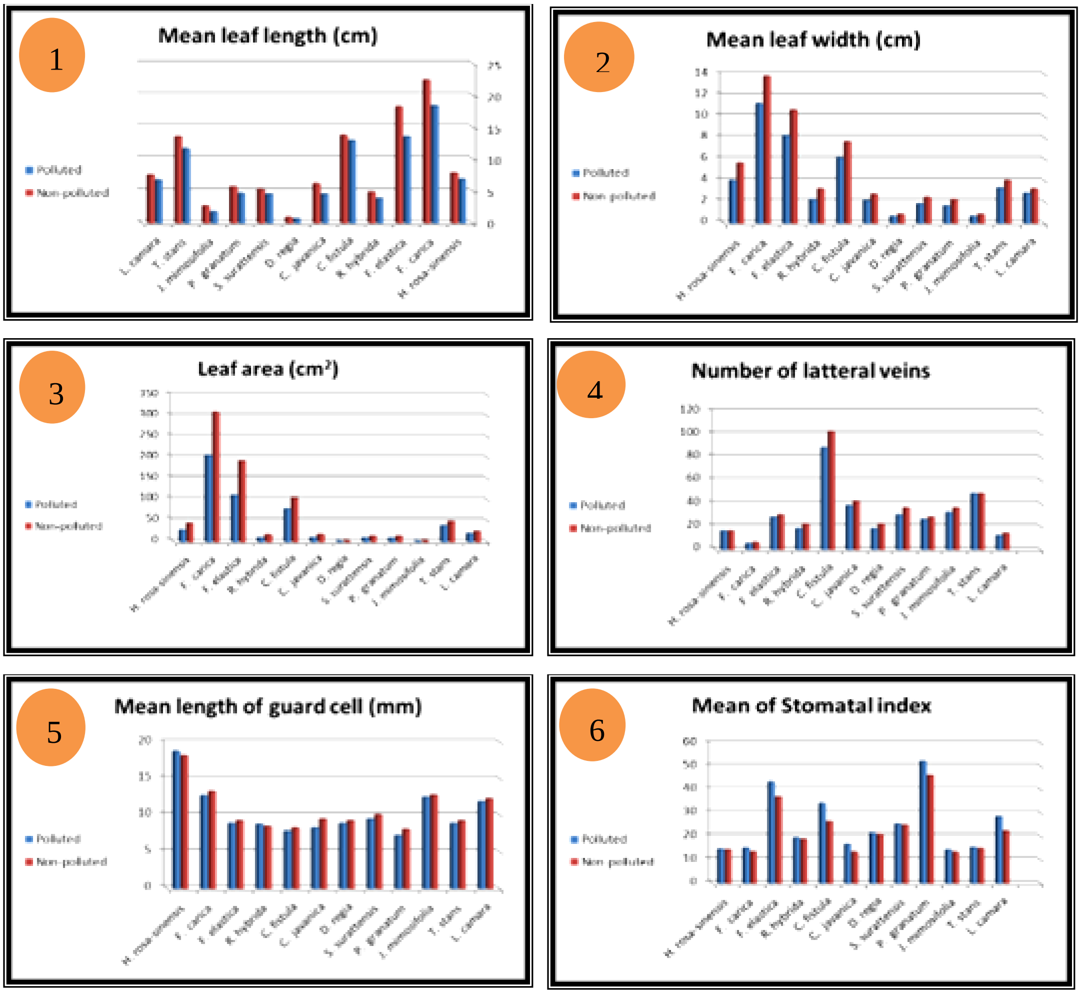
Table 2. Cont.

No.	Species	Venation		Hairs				Guard Cell Shape	
		NLV (Mean)		Distribution		Density			
		P	Np	P	Np	P	Np	P	Np
1	***Hibiscus rosa-sinensis***	16	16	Al	Al	Sp	Sp	K	K
2	***Ficus carica***	5	6	Al	Al	D	Sp	C	C
3	***Ficus elastica***	28	30	Al	Al	Sp	Sp	LW	K
4	***Rosa hybrida***	18	22	-	-	-	-	LW	K
5	***Cassia fistula***	88	102	Al	Al	D	M	L	K
6	***Cassia javanica***	38	42	-	-	-	-	L	L
7	***Delonix regia***	18	22	B	B	Sp	Sp	L	L
8	***Senna surattensis***	30	36	Al	Al	Sp /U D /Ls	Sp /U M/Ls	K	K
9	***Punica granatum***	26	28	-	-	-	-	K	K
10	***Jacaranda mimosifolia***	32	36	Al	Al	D /U Sp/Ls	D/U Sp/Ls	C	K
11	***Tecoma stans***	48	48	Al	Al	D	M	L	K
12	***Lantana camara***	12	14	Al	Al	D	M	L	L

Moreover, different hair wall ornamentation patterns were shown great variations between the studied species (Table 3). Smooth hair wall was found in *Ficus carica*, *Cassia fistula* and *Delonix regia*. While the warty walls were recorded in *Hibiscus rosa-sinensis*, *Senna surattensis*, *Jacaranda mimosifolia*, *Tecoma stans* and *Lantana camara* (Plate II, Figs. 20, 21, 33). Wrinkled hair wall surface was recorded in *Ficus elastica* only.

Table 3. Lamina micro-morphological characters of the studied species (SEM)
Abbreviations: A=Areolate Ab=Absent, Al=Alveolate, Ap=Apex, BC=Basal cell, Bf=Biforked, CAW=Curvature of anticlinal wall, C=Channeled, CA=Cell arrangement, CD=Cuticular deposition, CO=Cell outline, Cp=Capitate, CS=Cell shape, Cv=Concave, Cx=Convex, D=Dense, Di=Dimorphic, Es=Elongated straight, F=Flat, Fl=Flakes, G=Granulate, Gr=Granules, H=Hooked, Hx=Hexagonal, Is=Isodiametric straight, Iw=Isodiametric wavy, Ls=Lower surface, M=moderate, Mo=Monomorphic, Mu=Multicellular uniseriate, N=Normal, P=Pentagonal, Pa=Papillary, Pat=Patelliform, Pl=Polymorphic, Po=Pointed, Pt=Platelets, R=Raised, RA=Reticulate-areolate, RCB=Relief of cell boundary, Ref= Reticulate-foveate, Ro=Rosette, Rpt=Rosette of platelets, R=Rounded, Ru=Ruminate, Sc=Scalariform, Sf=Superficial, Sl=Stellate, Sm=Smooth, Sn=Sunken, Sp=Sparse, St=Striate, Sy=Symmetry, T=Type, Tk=Thick, Tn=Thin, Tt=Tetragonal, U=Unicellular, Ud=Undulate, Us=Upper surface, W=Wrinkled, Wk=Wrinkled, Wl= Wall, Wt=Warty, Wth=Wall thickness.

No.	**Species**	**CO**	**CA**	**CS**	**Wth**	**RCB**	**CAW**	**CD**	**Epicuticular wax**				**Trichomes**				
									Type		**Density**						
									P	**Np**	**P**	**Np**	**Sy**	**T**	**Ap**	**BC**	**Wl**
1	***Hibiscus rosa-sinensis***	Is	Al	P	Tk	R	F	G	Gr	--	Sp	--	Di	Bf Sl	Po	Ab	Wt
2	***Ficus carica***	Is	RA	P	Tk	R	F	St	Gr	--	Sp	--	Mo	MU	Po	Ro	Sm
3	***Ficus elastica***	Iw	Ref	P	M	C	Cx	G	Absent				Mo	MU	Po	N Ab	Wk
4	***Rosa hybrida***	Is	Al	Hx	Tk	C	Cx	S	Pt	Gr	M	Sp	Absent				
5	***Cassia fistula***	Is	Ref	P	M	C	Cx	G	Rpt	--	M	--	Pl	MU MU U	Po H Po	Ab	Sm
6	***Cassia javanica***	Is	Ref	P	Tk	R	Cv	G	Gr	--	Sp	--	Absent				
7	***Delonix regia***	Iw	RA	Hx	M	C	Cx	S	Rpt	Pt	D	Sp	Mo	MU	Po	Ab	Sm
8	***Senna surattensis***	Is	A	P	M	C	Cx	G	Rpt	Pt	D	M	Mo	MU	Po	Ab	Wt
9	***Punica granatum***	Iw	Ud	Hx	Tk	C	Cx	W	Gr	--	Sp	--	Absent				
10	***Jacaranda mimosifolia***	Iw	RA	Hx	M	C	Cx	W	Absent				Mo	MU	Po	N	Wt
11	***Tecoma stans***	Iw	Ru	Hx	M	C	Cx	St	Pt	--	Sp	--	Mo	MU	Po	N	Wt
12	***Lantana camara***	Iw	Ru	Hx	M	C	Cx	St	Gr	--	Sp	--	Pl	Pa MU Cp	Po Po R	Ro Ro Ab	Wt Wt Sm



Figs 1-6. Variation in leaf macro-and micro-morphological characters between polluted and non-polluted sites, **1.** variations in the mean leaf length, **2.** variations in the mean leaf width, **3.** variations in the mean leaf area, **4.** variations in the mean of lateral vein numbers, **5.** variations in the of guard cell length, **6.** variations in the mean of stomatal indices.

Data analysis

The result obtained from the T-Test indicated that *Ficus carica*, *Ficus elastica*, *Cassia fistula*, *Cassia javanica*, *Punica granatum* and *Lantana camara* showed a significant difference at $P \leq 0.05$ for leaf length, leaf width, leaf area, length of guard cell, stomatal density, and stomatal index among polluted and non-polluted sites. On the other hand, *Tecoma stans* and *Delonix regia* showed non-significant differences at $P \leq 0.05$ between the polluted and non-polluted sites (Table 4). All the studied species showed significant difference at $P \leq 0.05$ for leaf area among polluted and non-polluted sites.

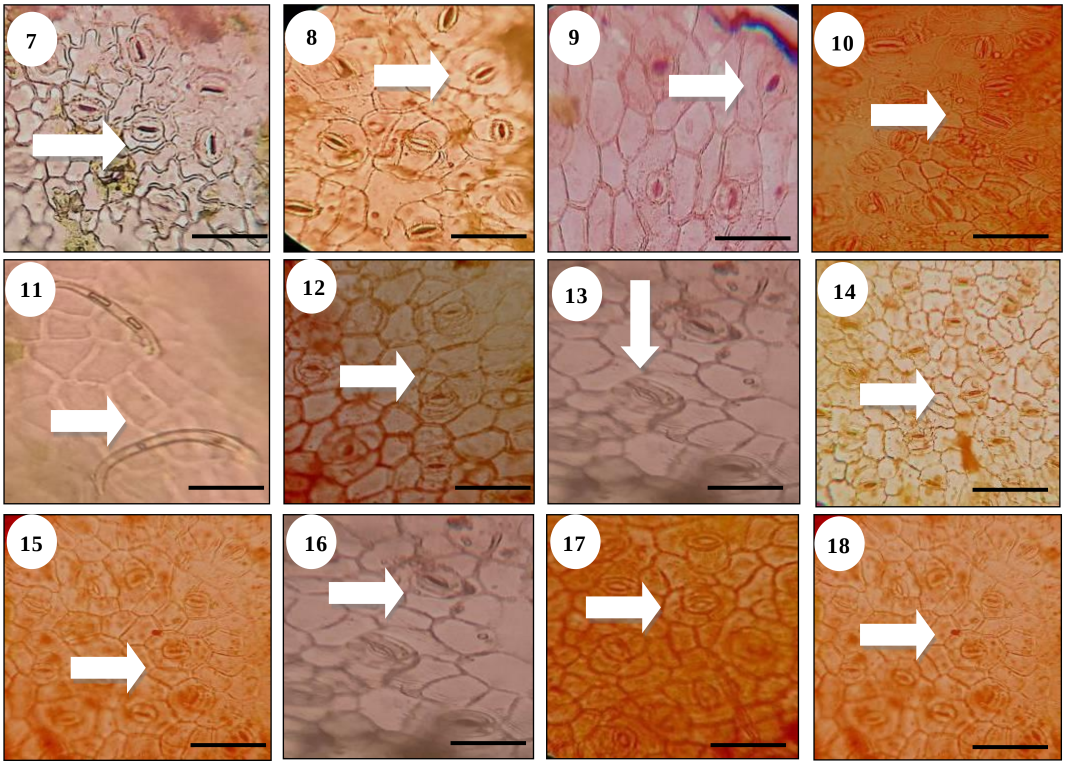


Plate I. Epidermal micrographs of the studied species under Light microscope **7.** *Hibiscus rosa sinensis*, **8.** *Ficus carica*, **9.** *Ficus elastica*, **10.** *Rosa hybrida*, **11.** *Cassia fistula*, **12.** *Cassia javanica*, **13.** *Delonix regia*, **14.** *Senna surattensis*, **15.** *Punica granatum*, **16.** *Jacaranda mimosifolia*, **17.** *Tecoma stans*, **18**. *Lantana camara*. Arrows indicate stomatal opening, guard cells and anticlinal walls or trichomes. Scale bar = 0.13 mm.

While Pearson correlation between the thirty-one macro and micro-morphological characters investigated between the studied species in both sites showed that *Hibiscus rosa-sinensis* and *Ficus carica* were significantly correlated (r = 0.417* and P = 0.020), high correlation between *Jacaranda mimosifolia* and *Tecoma stans* (r = 0.565**and P = 0.001) and between *Delonix regia* and *Senna surattensis* (r = 0.565**and P = 0.001) as shown in (Table 5). On the other hand, a negative significant correlation between *Hibiscus rosa-sinensis* and *Delonix regia* (r = -0.474-** and P = 0.007) and between *Ficus elastica* and *Cassia javanica* (r = -0.407*and P = 0.023).

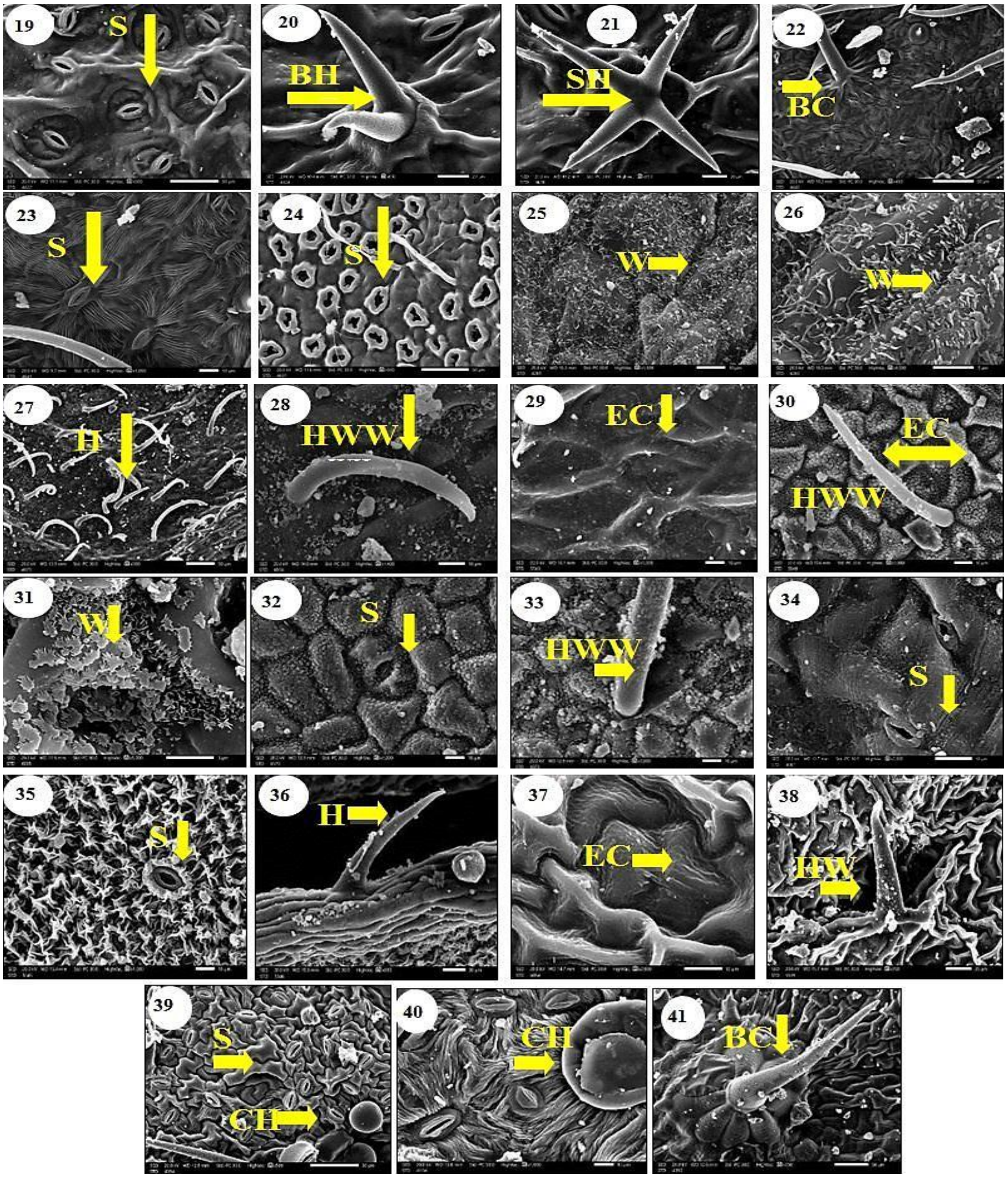


Plate II. SEM photographs showing the epidermal system of the studied species **19-21.** *Hibiscus rosa-sinensis*, **22-23.** *Ficus carica*, **24.** *Ficus elastica*, **25-26.** *Rosa hybrida*, **27-28.** *Cassia fistula*, **29.** *Cassia javanica*, **30-31.** *Delonix regia*, **32-33.** *Senna surattensis*, **34.** *Punica granatum*, **35-36.** *Jacaranda mimosifolia*, **37-38.** *Tecoma stans*, **39-41.** *Lantana camara*. Arrows indicate stomata, trichomes and epicuticular wax depositions. BC=Basal cell, BH=Biforked hairs, CH=Capitate hairs, EC=Epidermal cells (periclinal & anticlinal walls), H=Hair, HW=Hair wall, S=Stomata, SH=Stellate hair. Scale bar=5 µm in (26, 31). =10 µm in (22, 25, 28, 29, 30, 32, 33, 34, 35, 37, 40). =20 µm in (20, 21, 36, 38). =50 in (19, 23, 24, 27, 39, 41).

Table 4. Independent samples T-test between the studied species with Mean values, standard deviation, t-values and Sig. (P) values; $P \leq 0.05$. P= polluted and NP =non-polluted.

Species	**Blade length**		**Blade width**		**Leaf area**		**Guard Cell Length**		**Density**		**Index**	
	P	**NP**	**P**	**NP**	**P**	**NP**	**P**	**NP**	**P**	**NP**	**P**	**NP**
Hibiscus rosa-sinensis	6.8±0.40	7.8±0.58	4.0±0.04	5.6±0.03	2960±05	4816±17	18.7±0.24	18.1±0.38	10.2±0.57	9.7±0.27	14.6±0.01	14.3±0.30
t-value	1.414		26.417		1973.083		1.185		1.673		2.263	
Sig (2-tailed) p	0.195		0.000		0.000		0.270		0.147		0.08	
Ficus carica	18.3±0.08	22.3±0.08	11.2±0.01	13.8±0.03	20384±03	30912±18	12.6± 0.01	13.2±0.03	12.6±0.02	10.6±0.06	15±0.01	13.4±0.01
t-value	33.68		125.501		5339.618		1.744		393.409		253.289	
Sig (2-tailed) p	0.000		0.000		0.000		0.000		0.000		0.000	
Ficus elastica	13.4±0.06	18.1±0.03	8.2±0.02	10.6±0.01	11070±02	19292±09	8.8±0.02	9.2±0.04	18.6±0.01	14.1±0.04	43.2±0.02	36.8±0.01
t-value	64.151		160.325		1993.991		101.000		710.812		871.202	
Sig (2-tailed) p	0.000		0.000		0.000		0.000		0.000		0.000	
Rosa hybrida	3.8±0.40	4.8±0.31	2.2±0.32	3.2±0.40	704±05	1824±03	8.7±0.18	8.4±0.89	17.0±0.64	16.5±0.54	19.4±0.19	18.8±0.57
t-value	1.960		1.957		493.222		0.686		1.387		2.286	
Sig (2-tailed) p	0.081		0.088		0.000		0.528		0.204		0.072	
Cassia fistula	12.8±0.01	13.6±0.41	6.1±0.02	7.6±0.02	7808±14	10792±13	7.8±0.06	8.2±0.03	17.8±0.01	14.2±0.04	34.0±0.13	26.2±0.04
t-value	1.812		65.429		3010.581		70.711		569.842		404.259	
Sig (2-tailed) p	0.075		0.000		0.000		0.000		0.000		0.000	
Cassia javanica	4.5±0.03	6.1±0.34	2.1±0.04	2.7±0.08	900±14	1596±14	8.21±0.04	9.4±0.05	14.6±0.02	13.8±0.01	16.61±0.02	13.2±0.01
t-value	4.590		6.967		116.034		44.327		125.859		355.309	
Sig (2-tailed) p	0.002		0.000		0.000		0.000		0.000		0.000	
Delonix regia	1.9±0.20	2.4±0.01	0.6±0.01	0.7±0.01	90±01	192±03	8.8±0.48	9.2±0.33	13.6±0.89	11.4±0.89	21.4±1.09	20.9±1.09
t-value	1.045		1.869		51.892		0.390		1.768		0.577	
Sig (2-tailed) p	0.332		0.734		0.000		0.708		0.115		0.580	

Table 4. Cont.

Species	Blade length		Blade width		Leaf area		Guard Cell Length		Density		Index	
	P	**NP**	**P**	**NP**	**P**	**NP**	**P**	**NP**	**P**	**NP**	**P**	**NP**
Senna surattensis	4.5±0.02	5.2±0.19	1.8±0.11	2.4±0.13	810±03	1284±07	9.4±0.43	10.0±0.54	15.6±0.89	14.8±0.87	25.1±1.41	24.7±1.64
t-value	35.530		42.144		984.764		1.65		1.414		0.494	
Sig (2-tailed) p	0.000		0.000		0.000		0.139		0.195		0.635	
Punica granatum	4.6±0.03	5.6±0.05	1.6±0.15	2.2±0.12	736±11	1232±19	7.2±0.02	8.0±0.05	38.1±0.03	32.6±0.03	52.0±0.04	46.0±0.11
t-value	48.24		43.643		101.798		35.029		870.259		107.792	
Sig (2-tailed) p	0.000		0.000		0.000		0.000		0.000		0.000	
Jacaranda mimosifolia	1.7±0.17	2.6±0.49	0.6±0.20	0.8±0.17	102±07	192±04	12.4±0.44	12.7±0.46	6.1±0.89	5.8±0.70	14.2±0.86	13.2±0.82
t-value	1.670		13.649		24.566		0.975		0.588		1.869	
Sig (2-tailed) p	0.156		0.000		0.000		0.358		0.573		0.099	
Tecoma stans	11.3±0.78	12.2±0.57	3.3±0.28	4.0±0.39	3465±11	6532±26	8.8±0.45	9.2±0.51	10.0±0.81	9.5±0.70	15.3±0.85	14.8±0.72
t-value	1.996		1.518		91.717		1.414		0.588		1.043	
Sig (2-tailed) p	0.158		0.167		0.000		0.195		0.575		0.328	
Lantana camara	6.7±0.08	7.5±0.07	2.8±0.09	3.2±0.04	1904±03	2400±11	11.8±0.01	12.2±0.04	21.2±0.03	18.3±0.01	28.3±0.04	22.2±0.01
t-value	29.004		25.12		299.877		19.288		457.898		323.255	
Sig (2-tailed) p	0.000		0.000		0.000		0.000		0.000		0.000	

Table 5. Pearson correlation between thirty-one (31) macro and micromorphological characters between the studied species. *. Correlation is significant at the 0.05 level (2-tailed). **. Correlation is significant at the 0.01 level (2-tailed)

		Hibiscus rosa-sinensis	*Ficus carica*	*Ficus elastica*	*Rosa hybrida*	*Cassia fistula*	*Cassia javanica*	*Delonix regia*	*Senna surattensis*	*Punica granatum*	*Jacaranda mimosifolia*	*Tecoma stans*	*Lantana camara*
Hibiscus rosa-sinensis	Pearson Correlation	1	.417*	-.223	.268	.103	.103	-.474-**	-.268	-.268	-.268	-.326	-.127
	Sig. (2-tailed)		.020	.227	.144	.582	.582	.007	.145	.145	.145	.074	.495
	N	31	31	31	31	31	31	31	31	31	31	31	31
Ficus carica	Pearson Correlation	.417*	1	.319	-.096	-.071	-.216	-.253	-.177	-.326	-.177	-.096	-.177
	Sig. (2-tailed)	.020		.080	.608	.706	.242	.171	.340	.074	.340	.608	.340
	N	31	31	31	31	31	31	31	31	31	31	31	31
Ficus elastica	Pearson Correlation	-.223	.319	1	-.137	.018	-.407-*	.167	.209	.209	-.079	-.137	-.079
	Sig. (2-tailed)	.227	.080		.461	.922	.023	.370	.258	.258	.672	.461	.672
	N	31	31	31	31	31	31	31	31	31	31	31	31
Rosa hybrida	Pearson Correlation	.268	-.096	-.137	1	-.216	.221	-.253	-.029	.120	-.326	-.253	-.177
	Sig. (2-tailed)	.144	.608	.461		.242	.232	.171	.878	.521	.074	.171	.340
	N	31	31	31	31	31	31	31	31	31	31	31	31
Cassia fistula	Pearson Correlation	.103	-.071	.018	-.216	1	.048	.075	.241	.103	.103	.221	.241
	Sig. (2-tailed)	.582	.706	.922	.242		.797	.687	.191	.582	.582	.232	.191
	N	31	31	31	31	31	31	31	31	31	31	31	31
Cassia javanica	Pearson Correlation	.103	-.216	-.407-*	.221	.048	1	.075	.241	.103	-.036	-.071	-.174
	Sig. (2-tailed)	.582	.242	.023	.232	.797		.687	.191	.582	.849	.706	.349
	N	31	31	31	31	31	31	31	31	31	31	31	31

Table 5. Cont.

		Hibiscus rosa-sinensis	*Ficus carica*	*Ficus elastica*	*Rosa hybrida*	*Cassia fistula*	*Cassia javanica*	*Delonix regia*	*Senna surattensis*	*Punica granatum*	*Jacaranda mimosifolia*	*Tecoma stans*	*Lantana camara*
Delonix regia	Pearson Correlation	**-.474-****	-.253	.167	-.253	.075	.075	1	**.565****	.268	.268	.217	.268
	Sig. (2-tailed)	.007	.171	.370	.171	.687	.687		.001	.144	.144	.241	.144
	N	31	31	31	31	31	31	31	31	31	31	31	31
Senna surattensis	Pearson Correlation	-.268	-.177	.209	-.029	.241	.241	**.565****	1	.155	.155	.120	.014
	Sig. (2-tailed)	.145	.340	.258	.878	.191	.191	.001		.406	.406	.521	.942
	N	31	31	31	31	31	31	31	31	31	31	31	31
Punica granatum	Pearson Correlation	-.268	-.326	.209	.120	.103	.103	.268	.155	1	.155	-.029	.295
	Sig. (2-tailed)	.145	.074	.258	.521	.582	.582	.144	.406		.406	.878	.107
	N	31	31	31	31	31	31	31	31	31	31	31	31
Jacaranda mimosifolia	Pearson Correlation	-.268	-.177	-.079	-.326	.103	-.036	.268	.155	.155	1	**.565****	.155
	Sig. (2-tailed)	.145	.340	.672	.074	.582	.849	.144	.406	.406		.001	.406
	N	31	31	31	31	31	31	31	31	31	31	31	31
Tecoma stans	Pearson Correlation	-.326	-.096	-.137	-.253	.221	-.071	.217	.120	-.029	**.565****	1	.268
	Sig. (2-tailed)	.074	.608	.461	.171	.232	.706	.241	.521	.878	.001		.144
	N	31	31	31	31	31	31	31	31	31	31	31	31
Lantana camara	Pearson Correlation	-.127	-.177	-.079	-.177	.241	-.174	.268	.014	.295	.155	.268	1
	Sig. (2-tailed)	.495	.340	.672	.340	.191	.349	.144	.942	.107	.406	.144	
	N	31	31	31	31	31	31	31	31	31	31	31	31

Discussion

Scientists and agronomists were seeking the most capable ornamental species to revive under the polluted environment. NiKolik and Stevovik (2015) found that plants belonging to some families like Asteraceae can be used as bio-removal of a wide range of pollutants in polluted areas. The leaf morphological characters are not only a tool for classification, but also, they could reflect the degree of plant adaptation to the surrounding environment (Ordoñez et al., 2009). This study was carried out to investigate the leaf morphological characters of twelve species commonly planted in the streets of Giza district, Cairo a highly polluted site and Smouha district, Alexandria a peri-urban non polluted site of Egypt. The leaves of these plants varied in their leaf type and characters. The results of measuring the macro-morphological leaf characters were significantly different between the two sites. This observation agrees with Abu Ziada et al. (2015), who found that air pollution greatly affected both the morphology and physiology of the plant. The same observation mentioned by Xu et al. (2023), who found that plants planted in the cities, exhibit great variations in their morphology due to air pollution.

The leaf area is an important character in the comparison between the two sites. The greater leaf area recorded in *Ficus carica* indicates its efficiency in capturing the dust and PM. Meanwhile the smallest leaf area can dissipate the high heat and reduce the transpiration losses, which is important for the survival of plants under heat stress. Moreover, all the studied species showed variation in the leaf area among polluted and non-polluted sites. This agrees with Allahnouri et al. (2018) who pointed to the importance of the dimensions of the leaf blades and leaf areas as an adaptation strategy to the surrounding environment and air pollution. The achieved results were allied with that of Uka and Ebenezer (2020), who found that trees in polluted areas exhibit disorders in their leaf macro-characters expressed in lowering the leaf areas.

The leaf area in some of the studied species showed significant variations, in polluted and the non-polluted sites. As in *Cassia fistula* where its area measured 7808 mm^2 in polluted site compared to 10792 mm^2 in non-polluted site (Smouha). *Delonix regia* showed the smallest leaf area 90 mm^2 in polluted site versus 192 mm^2 in non-polluted site. This agrees with Shi et al., (2021) and Shi et al., (2022) who revealed changes in leaf area, shape beside stomatal characters, also leaf venation and vein characters, in response to air pollution.

Leaf texture is among the important macro-morphological character in plant adaptation to the surrounding pollution. Within the studied taxa, scarious leaf texture in *Rosa hybrida* and leathery texture in *Hibiscus rosa-sinensis*, *Ficus carica*, and *Ficus elastica* could reflect the ability of this species in capturing the dust when compared with species with papery leaf texture. Wang et al. (2022) mentioned that tough leaf surfaces help to protect the plant from both damage, herbivores and provide resistance to environmental overheating. According to Wang et al. (2022) observations, the species have lathery and scariosus leaf texture are accommodate with the heat waves in Giza district much more than the species with papery leaves.

The studied species showed variation in number of lateral veins between the polluted and non-polluted sites, this come in line with Carlson et al. (2016) who reported that number of lateral veins could be a defense action toward herbivores and modulate their physiological process. Also, Diaz et al. (2016) reported that leaf characters such as specific leaf area, leaf thickness, stomatal density, and leaf vein density in addition to other physiological characters can reflect the adaptive strategies of plants to climate and surrounding environment. It can play role as defense action toward herbivores and modulate their physiological process as reported by Carlson et al. (2016).

The result of the micro-morphological leaf characters showed that stomata size, number and index were greatly variable between the studied species. There was an increase in epidermal cell number, stomatal density and indices as well as in hair length within the hairy species in the polluted site (Giza) compared to the non-polluted site (Smouha). These variations can be considered as pointers of environmental stress and ways of adaptation in capturing the air pollutants. These observations agree with Verma and Singh (2022) and Rabelo et al. (2012), they found that stomatal density increased in polluted areas.

The stomatal density, guard cell length and shape are influenced by many ecological factors (Woodruff et al., 2008) and by the degree of air pollution (Riikonen et al., 2010), while stomatal density, indices, elevation, and guard cell size besides stomatal opening reflect physiological processes (Wang et al., 2017a, b). From the results obtained the guard cell length was small in *Punica granatum* (7.2 µm) and *Cassia fistula* while *Hibiscus rosa-sinensis* has the longest guard cells within the studied taxa in both sites (polluted and non-polluted).

All the species have anomocytic stomata with variable stomatal densities and indices varied from very low stomatal density in *Cassia javanica* to very high in *Punica granatum* in both sites. Wide lenticular stomatal opening were observed in all the studied taxa except in *Ficus elastica*; the opening was wide circular. Larger stomatal area may be related to greater availability of CO_2 (James and Bell, 2001). This result reflects the variation of the studied species in terms of their suitability to pollution and other surrounding environmental stress.

Perini et al. (2017), Weerakkody et al. (2017), Muhammad et al. (2019) and others announced to the importance of choosing the planted species with certain traits in urban cities. They were preferred to plant those plants with dense wax epicuticular depositions on their leaves, higher wet ability (Muhammad et al., 2019) or with a higher stomatal density (Weerakkody et al., 2018b) as they were more suitable in capturing the PM and purify the ambient air. Trichomes as well considered an important character in purifying the air. In contrast to Perini et al. (2017) opinion who mentioned that hairy leaves were less effective in PM capture. This disagrees with other studies on more than hundred tree, shrub and climber species (Muhammad et al., 2019), which report important differences between hairy and non-hairy leaves and significant positive relationships between PM accumulation and trichome density.

Leaf micro-morphological investigations especially, the epicuticular secretions and trichomes are adaptation characters for environmental stresses. In addition, hairy leaves showed a lower rate of transpiration than glabrous leaves, and plants respond to the air pollutants by appearance of long trichomes (Pérez-Estrada et al., 2000). Among the species with trichomes were *Hibiscus rosa-sinensis* which has biforked and stellate pointed trichomes with warty surface pattern, *Cassia fistula* has multicellular uniseriate hooked trichomes with smooth surface pattern, and *Lantana camara* which has capitate trichomes with rounded apex and smooth surface pattern. Rosette basal cells can be noticed in *Ficus carica* and *Lantana camara*. The trichome density in the two sites differed as it was denser in the taxa collected from polluted site. Most of the studied species have epicuticular secretions with different densities and shapes; the achieved result agrees with Das and Prasad (2012).

Pearson correlation reflects the significant macro and micro-morphological leaf characters between the studied species; namely: leaf area, presence and density of trichomes, texture of leaf, epidermal cell outline, epidermal cell shape, thickness of the wall of trichomes, and density of epicuticular waxes. The achieved results agreed with Rabelo et al. (2012), Verma and Singh (2022) and Wang et al. (2017a, b).

Conclusion

From this investigation, we can conclude that the most affected morphological characters under polluted environment were leaf length, width, area, surface texture, epicuticular secretions and trichome length and density. The most adapted species which are recommended to be planted as purifier to air pollutants are *Hibiscus rosa- sinensis, Ficus carica*, *Ficus elastica*, *Cassia fistula, Tecoma stans* and *Lantana camara*.

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