

Common Road Tree Pollen Grain Contents and Their Cause on Allergy

Wafaa Kamal Taia¹ and Ahmed A. Zayed²

1-Alexandria University, Faculty of Science, Botany and Microbiology Department, Alexandria, Egypt.

2- Alexandria University, Faculty of Medicine, Alexandria, Egypt.

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ABSTRACT:

Background: Air pollution beside climatic conditions has severe effect on pollen grains collected from different plant species. Outdoor allergens are an important cause of allergic rhinitis, conjunctivitis and asthma, especially pollen grains and fungal spores being the major outdoor allergens that induce symptoms in atopic patients. Specific airborne pollen grain types, especially those collected from anemophilous plants, trigger respiratory allergy symptoms in sensitive individuals and cause immunity disorders. This work aims to investigate the effect of eight road trees commonly planted in Alexandria city streets by analyzing the protein contents and some element contents as an allergy inducing particles. Eight road tree species were chosen for this investigation. The chosen trees are *Bauhinia galpinii*, *B.variegata*, *Casia javanica*, *Parkinsonia aculeata*, *Peltophorum roxburghii*, *Delonix regii*, *Croton cotinifolia*, and *Jacaranda mimosifolia*. The pollen grains have been gathered during the period of July till November 2019, acetolyzed and described, meanwhile non-acetolyzed pollen grains have been sputtered on Aluminum stubs and coated with Gold for SEM examination and photographs. Pollen grains have been smeared onto glass slides, stained and photographed for protein contents evaluations. Mineral contents have been estimated using X-ray analyses. The results obtained revealed that allergic symptoms appeared in response to the density, dispersion and protrusions of the pollen grains in combination with the degree of pollution and climatic conditions. Meanwhile, the high protein contents, C, S and K can induce breath disorders.

Conclusion: From the present study *Delonix regia* and *Parkensonia aculeata* are the most responsible road trees which stimulate the human immune system, as they have small size pollen grain and high contents of C, S and K. As well as the results obtained indicated that many factors inducing allergic diseases and affect the immune system as environmental conditions include climate change, temperature, humidity, air pollution and loss of biodiversity.

KEY WORDS: Allergy- Immunology-Pollen grains- Road trees



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How to Cite

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INTRODUCTION:

Allergy became from the most globally important issues in this time, especially after the increase in air pollution and viral infections. Symptoms of allergy are due to the interaction between the body and any foreign substances. These interactions stimulate the immune system to a harmless materials carried in the air to the production of IgE by the mast cells and basophils. The degree of sensitivity to the inhaled substances depends on the people history, age, sex, as well as the type and quantity of these substances. Smaller particles can penetrate the bronchi very easily and cause allergenic effects and in severe cases mortality. Meanwhile,^{1,2,3,4} linked between air pollution and the increased number of Covid-19 death risk. Air pollution may be according to both biotic and abiotic factors. From the biotic factors are the pollen grains, which are the yellow or orange dusts found on the petals of opened flowers. They are male gametes in both Gymnosperms and Angiosperms, and considered from the most important outdoor sources of allergens especially those released from anemophilous plants i.e. flowers pollinated by air, including trees, grasses, and weeds. In Mediterranean countries, governments planted many types of road trees for ornamentation and sheltering from exposed sun rays besides decreasing the hot feelings in summer. Pollen grains released by these trees constitute one of the most important causes of pollinosis⁵. Exposure to these pollen grains led to a variety of allergic symptoms ranging from seasonal rhino conjunctivitis to severe asthma in susceptible individuals⁶.⁷ Reported that different types of pollen grains stimulate the production of IgE which is the allergens indicator.⁸ Noted to the annual periodicity of pollinosis as an important feature with symptoms usually occurring at the same time of the year, during pollination time.

Actually Pollen allergens are water-soluble proteins, or glycol-proteins, sometimes starch and fats, which make them capable of evoking an IgE antibody-mediated allergic reaction in seconds to sensitive peoples.⁹ Recognized 15 distinct groups of proteins with diverse biochemical properties as allergens in taxa of subfamily Pooideae, family Poaceae. While¹⁰ clarified the allergens by being proteins capable of citing powerful T helper lymphocyte type 2 (Th2) responses, resulting in immunoglobulin (Ig)E antibody production. Specific selection of aeroallergens for the skin prick test by allergists is not always evidence-based.^{6,7} Recognized that trees belonging to orders Fabales, Fagales, Lamiales, Proteales, and Pinales are recognized as the most potent allergen sources. They recognized both *Prosopis juliflora* and *Peltophorum pterocarpum* trees as the source of the important allergen. Whereas¹¹ found that date palms produce clinically relevant pollen allergens.¹² Found that allergy is not restricted to certain trees, shrubs, or herbs, but it depends on the number of pollen grains released in the air and several environmental and climatic factors

This work provides the pollen characteristics and their quantitative protein contents and some important mineral contents of eight common tree species growing in Alexandria streets, Egypt. Alexandria city, which lies in the Mediterranean coastal region of Egypt, has its own weather with high humidity in summer and rainfall in winter. The chosen trees are commonly planted in the streets everywhere in Alexandria, Egypt and their flowers are flourished in the summer from July till November. This weather hydrates the pollen grain easily to excrete their allergens substances stimulating the human immune system causing severe symptoms to allergic people. This work will help in aeropalynological, immunological, and

horticultural studies and identification of allergenically significant road trees pollen grains. The present research has been done to investigate if the protein and element contents of the cultivated road trees stimulate the human allergic response and the immune system.

Mechanisms underlying allergy response

Allergy is defined as an abnormal adaptive immune response against noninfectious environmental compounds, i.e., allergens, including noninfectious components of some infectious organisms. When allergens crosslink the performed immunoglobulin E (IgE) bound to the high-affinity receptor FcεRI on mast cells, the allergic reaction occurs. Not only are mast cells crucial for the development of allergic reactions, but also function as sensors of psychological and environmental stress¹³. The most important role of IgE, however, lies in its potential to promote the release of biologically active mediators in an antigen-specific manner by sensitizing mast cells. More precisely, upon activation, mast cells synthesize and release chemokines, lipid mediators, and additional cytokines such as IL-4 and IL-13 to perpetuate the TH2 response. The differentiation of naïve allergen-specific T cells into TH2 cells is favored by an early burst of IL-4 presence. IL-4 seems to be derived from a specialized subset of T cells. The allergen-specific TH2 cells produce IL-4 and IL-13 that act on allergen-specific B-cells to produce IgE. The IgE, produced in response to the specific allergen, binds

to the high-affinity receptor for IgE on the abovementioned mast cells, activated eosinophils, and basophils. These cells can amplify the production of IgE since they produce IL-4 and CD40 ligands upon their activation.

In the case of pollen allergy, the TH2-biased immune system reacts to pollen-derived allergens. The allergic reaction to pollen grains develops in the same manner as other allergic responses, and it includes two phases. The first phase, early response, depends on IgE receptors binding, which leads to activation of mast cells and basophils. The second phase, the late response, involves the engagement of adhesive molecules, eosinophils, lymphocytes and their products, and neuropeptides^{14, 15}. Uncovered a mechanism crucial for developing an allergy to pollen grain and forming allergic asthma or seasonal nasal allergies. Exposure of airways to pollens rapidly induces recruitment of neutrophils, a type of white blood cells that move quickly to an affected site and induce inflammation. A potential mechanism of action through which this happens is an induction of a state of sustained oxidative stress in the airways. Oxidative stress occurs due to repeated recruitment of activated ROS-generating neutrophils. Chronic oxidative stress can alter the function of dendritic cells, worsen allergic asthma, and modify the balance of TH1 and TH2.¹⁶ Pointed to the role of oxidative stress and allergy to pollen. The impact of oxidative stress induced

by pollen grain on dendritic cells has a dual-action. Besides activating the production of pro-inflammatory cytokine from dendritic cells linked to local innate immunity, oxidative stress is also an adjuvant factor in the adaptive immunity initiation against pollen allergens.

Materials and Methods:

Collected flowers and flower buds of eight road trees, four deciduous, and four evergreens have been subjected in this study during July-October 2019. The confirmation of the identifications and data of collections are listed in table 1. This collection is from perennial trees, commonly planted in Alexandria streets, *Bauhinia galpinii*, *B.variegata*, *Casia javanica*, *Parkinsonia aculeate*, *Peltophorum roxburghii*, *Delonix regii*, *Croton cotinifolia*, and *Jacaranda mimosifolia*. The plants are identified with the aid of ^{17,18} The anthers are carefully removed from the flowers immediately after gathering by the aid of stereomicroscope and the pollen grains were sputtered on glass slides with a thin film of egg albumin. The slides were directly stained with bromophenol blue for 2 min., washed in tap water, cleared in xylol, mounted in Canada balsam, then covered for examination by Olympus light microscope and selected species with the different protein concentrations photographed¹⁹. For SEM examinations, non-acetolyzed pollens were sputtered onto Aluminum stubs, coated with gold, and examined and photographed using JEOL JSL IT 200 SEM

allocated at Faculty of Science, Alexandria University at 15 Kev. For mineral contents, pollen grain pellets have been made of the studied species and subjected to X-ray analysis under 20 kv, using JEOL JSL IT 200 SEM. Acetolyzed pollen grains were examined by Olympus light microscope for measurements; at least 30 pollen/ taxa were subjected in this study. The terminology used here is according to²⁰.

Results:

Description of the pollen grains and their protein contents are photographed and summarized in table 2, while the mineral contents are listed in table 3. Full pollen morphology descriptions are given below.

A-Pollen morphology

The studied taxa have been grouped according to the pollen size into three categories; taxa have small pollen grains, others with moderate size pollen grains and the third with large size pollen grains. The taxa with small pollen grains are *P.aculeata* and *D.regia*, the second group has four taxa; *B.galpinii*, *C.javanica*, *Cr.cotinifolia* and *J.mimosifolia*; while the third group with large pollen grains has two species; *B.variegata* and *P.roxburghiana*. The size of the pollen grains has no relation with the state of the tree; evergreen or deciduous; or the protein contents (Fig.2).

1-Bauhinia galpinii N.E.Br. Family Caesalpinaceae, evergreen tree (Photos. 1-2)

Symmetric, isopolar P/E ratio:1.5, polar axis P 37.2 (43.15) 48.5 μ m and equatorial diameter 25.3 (29.12) 35.2 μ m. Prolate, sometimes subprolate, tricolporate, colpi, length 27.2 (29.4) 31.54 μ m and width 2.9 (3.4) 4.5 μ m, with granulate colpus membrane. Ora protruding lalongate. Mesocolpium 12.0 (13.34) 14.7 μ m. Apocolpium 7.0 (9.23) 11.5 μ m. Exine 1.00 (1.9) 2.5 μ m thick. Sexine as thick as nexine. Tectum foveolate rugate . The protein contents are moderate as indicated by the degree of stain (photo. 2).

2-Bauhinia variegata L. Family Caesalpinaceae, evergreen tree (Photos. 3-4)

Symmetric, isopolar P/E ratio:0.9, polar axis P 54.2 (56.15) 62.5 μ m and equatorial diameter 55.3 (59.12) 64.2 μ m. Oblate spheroidal, sometimes subprolate, tricolporate, colpi, length 38.2 (39.4) 41.54 μ m and width 3.9 (5.4) 6.45 μ m, with granulate colpus membrane. Ora protruding lalongate. Mesocolpium 12.6 (13.44) 17.7 μ m. Apocolpium 10.0 (12..47) 14.5 μ m. Exine 1.00 (1.9) 2.5 μ m thick. Sexine as thick as nexine. Tectum striate. The protein contents are moderate as indicated by the degree of stain (photo. 4).

3-Cassia javanica L. Family Caesalpinaceae, deciduous tree (Photo 5-6)

Symmetric, isopolar P/E ratio 1.2, polar axis P 27.8 (32.15) 37.2 μ m and equatorial diameter E 21.7

(24.9) 28.2 μ m. Subprolate to prolate, tricolporate. Colpi length 16.2 (15.2) 18.3 μ m with lalongate ora. Mesocolpium 9.8 (10.44) 14.7 μ m. Apocolpium 10.19 (11.75) 14.8 μ m. Exine 1.0 (1.8) 2.0 μ m thick. Sexine as thick as nexine. Exine widely reticulate with granulate endexine. The protein contents are very high as indicated by the degree of stain (photo. 6).

4-Parkinsonia aculeate L. Family Caesalpinaceae, evergreen tree (Photos 7-8)

Symmetric, isopolar P/E ratio:1.3, polar axis P 19.2 (20.15) 22.5 μ m and equatorial diameter E 14.05 (15.9) 18.2 μ m. Prolate, tricolporate, colpi, length 15.9 (19.2) 20.54 μ m and breadth 6.4 (7.45) 9.25 μ m, ora lalongate. Mesocolpium 12.6 (13.44) 14.7 μ m. Apocolpium 14.59 (16.47) 19.5 μ m. Exine 1.00 (1.9) 2.5 μ m thick. Sexine thicker than nexine. Tectum reticulate. The protein contents are very high as indicated by the degree of stain (photo. 8).

5-Peltophorum roxburghii (G. Don) Degener. Family Caesalpinaceae, deciduous tree (Photos 9-10)

Symmetric, isopolar P/E ratio 1.3, polar axis P 48.8 (57.79) 65.2 μ m and equatorial diameter E. 39.5 (47.2) 59.7 μ m. Prolate to prolate, tricolporate, colpi length 7.6 (12.85) 15.8 μ m and breadth 5.8 (7.65) 11.75 μ m. Mesocolpium 24.2 (33.36) 44.2 μ m. Apocolpium 37.9 (52.9) 56.2 μ m. Exine 2.0 (2.5) 3.0 μ m thick. Sexine thicker than nexine.

Tectum coarsely reticulate. The protein contents are very high as indicated by the degree of stain (photo. 10).

6-*Delonix regia* (Bojer) Rafin. Family Caesalpinaceae, evergreen tree (Photos. 11-12)

Symmetric, isopolar P/E ratio 1.25, Polar axis P 16.8(19.95) 21µm and equatorial diameter 12.6(15.75) 16.8 µm. Sub-prolate, tricolporate, triangular, colpi length 6.3(9.45) 10.5µm and breadth 2.1(4.935) 6.3 µm, oral alongate. Mesocolpium 12.6 (16.38) 18.9 µm. Apocolpium 14.7(17.85) 18.9 µm. Exine 1.8 (2.0) 2.3 µm, sexine thicker than nexine. Tectum reticulate-rugulate. The protein contents are low as indicated by the degree of stain (photo. 12).

7- *Croton cotinifolia* L. Family Euphorbiaceae, deciduous tree (Photos 13-14)

Symmetric, isopolar P/E ratio 1.3, Polar axis P 38.4 (41.95) 46.8 µm and equatorial diameter 22.6(27.75) 31.8 µm. Prolate, tricolporate, colpi length 19.3(27.45) 31.5µm and breadth 1.8(2.9) 3.3 µm, ora very small, lalongate. Mesocolpium 14.3 (16.38) 18.5µm. Apocolpium 15.7(17.85) 19.2µm. Exine 1.8 (2.0) 2.3 µm, sexine as thick as nexine. Tectum reticulate. The protein contents are very high as indicated by the degree of stain (photo. 14).

8-*Jaccaranda mimosifolia* D.Don Family Bignoniaceae deciduous tree (Photos 15-16)

Symmetric, heteropolar P/E ratio 1.3, polar axis P 38.2 (41.8) 46.7µm and equatorial diameter 25.8

(32.3) 39.75 µm. Prolate, tricolporate, the colpi syncolpate at one pole, colpi length 25.8 (28.3) 32.5 µm and breadth 3.2 (4.5) 6.7 µm, ora small and plugged lalongate. Exine 2,0 (3.5) 3.8 µm, sexine thicker than nexine, tectatepsilate, or faintly punctuate. The protein contents are moderate, as indicated by the degree of stain (photo.16).

B-Element contents

Mineral contents are listed in table 3, in which all the minerals investigated are considerably low in all the pollen taxa, except the C and O which are in moderate quantities. The N content is slightly fair in *P.roxburghii* and Na content is very low in all the pollen taxa, except *J.mimosifolia*. The rest of the investigated elements; P, Mg, Al, Si, Cl, Ca, Fe, Cu and Zn; are considerably low in all the pollen grains under study, except the Zn is higher in *C.cotinifolia*. The two investigated elements; S & K; are very low in all the pollen taxa, except in both *D.regia* and *P.aculeata*.

Discussion:

The study of Pollen allergens becomes from the recent points of research now a day. These allergens arise from the process of pollination happened in wind pollinated trees, shrubs and herbs, especially grasses. The amount of these allergens depends on pollen-specific characteristics such as their density, dispersion, and profusion. ² Announced that the exposure to both pollen grains and fungal spores, especially in the spring and in early morning, induce severe allergic symptoms.

This observation may be due to the great amount of pollen grains and spores dispersed at that time. ⁶Found that the proteins found on the surface of the pollen grains of *Prosopis juliflora* trees can induce allergy. At the same time, ⁷found that the use of the pollen grains of one of the wide trees in India, *Peltophorum pterocarpum*, can be of Clinical and immune-biochemical characterization. Accordingly, the street trees can be one of the reasons causing pollinosis and meanwhile can be used in immunotherapy. ²¹Indicated that many factors inducing allergic diseases, such as climate change, temperature, humidity, and loss of biodiversity. These stimulants, besides air pollution and exposure to submicronic particles, may be causes of allergy (Fig.1). Now a day ^{4,5} linked air pollution and the increased number of Covid-19 death risk. This isn't just bad news for those with hay fever history, it seems, it indicated that, even in people who don't have the allergy, pollen can suppress the way the body responds to viruses by reducing the immune response in the airways with the risk of catching Covid-19 linked to the amount of airborne pollen circulating. The problem that the pollen season is lengthening; with great change in temperature and humidity; resulting to the increase in patients. In the same time most people are allergens to some pollen trees, grasses or even herbs. ¹²Found that the type of road trees is not the cause of pollinosis alone, but there are many factors in combination with the dispersed pollen grains that are all together reasons

for allergy. In this work, eight widely planted road trees are chosen to investigate their pollen grains allergic stimulations. The external morphology, protein contents as well as mineral contents have been estimated.

The results showed that both *Parkensonia aculeate* and *Delonix regia* have small pollen grains than the other trees and high contents in both S. and K as shown in tables 2 & 3 and Fig.2. The size of the pollen grains is considered an important character in inducing allergy. Small pollen grains are easily dispersed by air and inhaled²². The most effective reasons for pollinosis is the exposure to submicronic particles and the role of the pollen grains is to stimulate the allergens present in their cytoplasm, such as starch, proteins, ubish bodies and polysaccharides. These organic substances are specific to each plant species to recognize the stigma specific to this species. The results obtained revealed that all the studied trees have high Carbon and Oxygen contents, which is due to the high contents of Carbohydrates. Meanwhile, the Nitrogen content is low due to the decrease in Proteins. The rest of the elements are considerably low, and this is can be due to the difference in their physiological activity. ²³ Found that intralymphatic immunotherapy (ILIT) for allergic patients require only a few intralymphatic injections of the allergen. Worth mentioning is the fact pollens contain aqueous pollen extract proteins, proteases, NADPH oxidases, and lipids that stimulate innate immunity responses. These responses recruit the

abovementioned neutrophils and modulate functions of the dendritic cells, but also induce TH2 polarization, and promote allergic inflammation²⁴.²³ Found pollen allergens do not act on their own. Pollen-associated lipid mediators (PALMs) may propagate the overall TH2 favoring micro milieu in tissue exposed to pollen, independently of pollen allergens. PALMs may activate eosinophils and provide signals for dendritic cells to mature and acquire migratory phenotypes. In other words, PALMs could have a role in the immune system's response to pollen grains, besides pollen allergens only.

From this study, we can conclude that the most effective trees causing allergy are those having smaller pollen grains with the excess of starch, polysaccharides (as indicated by Carbon and Oxygen contents mentioned in table 2), and release more proteins on their surfaces. From the studied trees, *Delonix regia* and *Parkensonia aculeata* are the most responsible road trees which can stimulate the human immune system in this study, as they have small size pollen grain (table 1, Fig.1) and high contents of C, S and K (table 2) .

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Table1 Studied species, date of collection, confirmation of nomenclature and synonyms. Table1 Studied species, date of collection, confirmation of nomenclature and synonyms.

Species	Date of collection	Source of confirmation	Synonyms
<i>Bauhinia galpinii</i> N.E. Br. (Photos1 & 2)	5/7/2019	"ILDIS LegumeWeb entry for Bauhinia"	<i>B.galpinii</i> var. <i>galpinii</i> <i>Perlebia galpinii</i> (N.E.Br.) A.Schmitz
<i>B. variegata</i> (L.)Benth. (Photos 3 &4)	12/7/2019	"ILDIS LegumeWeb entry for Bauhinia"	<i>Phanera varigata</i> (L.) Bent
<i>Cassia javanica</i> L. (Photos 5 &6)	10/8/2019	IPNI (2022)	<i>Bactrylobium javanica</i> (L.) Hornem.,
<i>Parkinsonia aculeata</i> L. (Photos 7&8)	8/9/2019	IPNI (2022)	-
<i>Peltophorum. Roxburghii</i> (G.Don) Degener (Photos 9 & 10)	7/7/2019	IPNI (2022)	-
<i>Delonix regia</i> (Bojer ex Hook.) Raf. (Photos11 & 12)	19/9/2019	IPNI (2022)	<i>Aprevalia</i> Baill.
<i>Croton cotinifolia</i> L. (Photos13 & 14)	8/8/2019	IPNI (2022)	<i>Codiaeum variegatum</i>
<i>J. mimosifolia</i> D.Don (Photos 15& 16)	12/9/2019	IPNI (2022)	-

Table 2 Summary of the different features within the studied taxa

Abbreviations: Eg.=Ever green, D.=Deciduous; L.=Large $\geq 50\mu\text{m}$, M.=Moderate from 25-48 μm , S.=Small $\leq 25\mu\text{m}$; Ob-Sp=Oblate Spheroidal, S.Pr.=Subprolate; Orn=Ornamentation; Fov.=Foveolate

Taxa	Family	Status	Pollen characters					
			Polarity	Size	P/E	Shape	Exine Orn	Protein
<i>B. galpinii</i>	Caesalpinaceae	Eg.	Isopolar	M.	1.5	Prolate	Fov.rugate	Medium
<i>B. variegata</i>		Eg.	Isopolar	L.	0.9	Ob-Sp	Striate	Medium
<i>C. javanica</i>		D.	Isopolar	M.	1.2	S.Prolate	Reticulate	High
<i>P. aculeata</i>		Eg.	Isopolar	S.	1.3	Prolate	Reticulate	High ¹
<i>P. roxburghii</i>		D.	Isopolar	L.	1.3	Prolate	Reticulate	High
<i>D. regia</i>		Eg	Isopolar	S.	1.25	S.Prolate	Reticulate Rugulate	Low
<i>C. cotinifolia</i>	Euphorbiaceae	D.	Isopolar	M.	1.3	Prolate	Reticulate	High
<i>J. mimosifolia</i>	Bignoniaceae	D.	Heteropolar	M.	1.3	Prolate	Psilate, Faintly Punctate	Medium

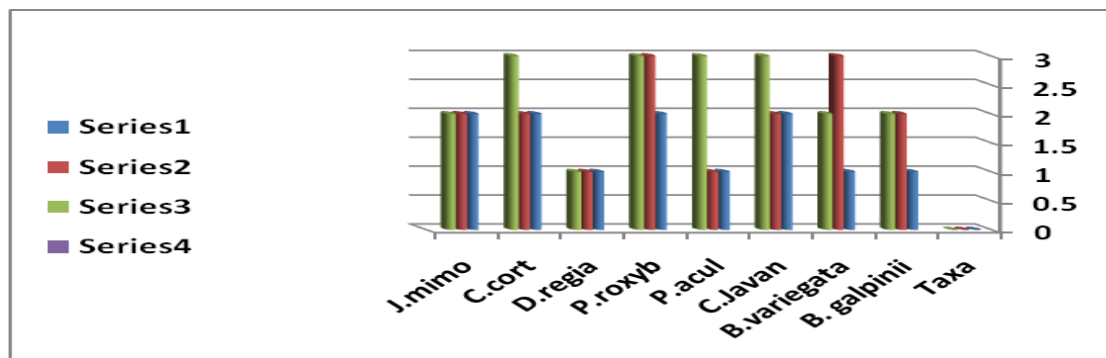


Fig.2 Series 1= State of the tree, Series2=Pollen size, Series 3= Protein contents, Series 4=different taxa
Table 3 Summary of the investigated minerals within the studied taxa as shown by X-ray analyses

Taxa	%of the Mass of Investigated Elements														
	C	N	O	Na	Mg	Al	Si	P	S	Cl	K	Ca	Fe	Cu	Zn
<i>B. galpinii</i>	59.24± 0.19	2.12± 0.21	33.52± 0.39	0.07± 0.02	0.05± 0.02	0.20± 0.02	0.00	0.03± 0.02	0.22± 0.02	0.0	0.65± 0.04	0.22± 0.03	0.06± 0.02	0.48± 0.07	0.24± 0.07
<i>B. variegata</i>	58.84± 0.19	2.22± 0.21	36.62± 0.39	0.06± 0.02	0.14± 0.02	0.05± 0.01	0.05± 0.0	0.28± ±0.02	0.17± 0.01	0.0	0.80± 0.03	0.33± 0.02	0.04± 0.02	0.28± 0.04	0.13± 0.04
<i>C. javanica</i>	62.85± 0.23	0.0	34.24± 0.45	0.11± 0.03	0.0	0.08± 0.02	0.00	0.48± 0.03	0.22± 0.02	0.05± 0.01	0.70± 0.0	0.38± 0.03	0.12± 0.03	0.39± 0.06	0.39± 0.06
<i>P. aculeata</i>	60.16± 0.20	2.11± 0.22	35.26± 0.39	0.08± 0.02	0.05± 0.02	0.20± 0.02	0.19± 0.02	0.22± 0.02	14± 0.01	0.13± 0.01	65± 0.03	0.28± 0.02	0.04± 0.02	0.26± 0.05	0.25± 0.05
<i>P. roxburghii</i>	59.59± 0.28	11.97± 0.51	24.77± 0.49	0.07± 0.03	0.03± 0.02	0.11± 0.02	0.42± 0.03	0.03± 0.02	0.0	1.99± 0.06	0.02± 0.02	0.22± 0.03	0.06± 0.02	0.48± 0.07	0.24± 0.07
<i>D. regia</i>	60.16± 0.20	2.11± 0.22	35.26± 0.39	0.08± 0.02	0.05± 0.02	0.20± 0.02	0.19± 0.02	0.22± 0.02	14± 0.01	0.13± 0.01	65± 0.03	0.28± 0.02	0.04± 0.02	0.26± 0.05	0.25± 0.05
<i>C. cotinifolia</i>	60.86± 0.24	2.75± 0.28	33.03± 0.46	0.03± 0.03	0.37± 0.03	0.02± 0.02	0.06± 0.02	0.66± 0.03	0.25± 0.02	0.36± 0.03	0.65± 0.04	0.41± 0.03	0.03± 0.02	0.27± 0.05	26± 0.06
<i>J. mimosifolia</i>	57.86± 0.30	2.28± 0.33	35.97± 0.60	35.97± 0.60	0.20± 0.03	0.06± 0.03	0.11± 0.03	0.61± 0.04	0.38± 0.03	0.02± 0.02	1.29± 0.07	0.45± 0.05	0.0	0.49± 0.09	0.06± 0.07

External shape: 1-2 *Bauhinia galpinii*, 3-4 *B. variegata*, 5-6 *Casia javanica*, 7-8 *Parkinsonia aculeate*, 9-10 *Peltophorumroxburghii*, 11-12 *Delonix regii*, 13-14 *Croton cotinifolia*, 15-16 *Jacaranda mimosifolia*

Pollen SEM photographs: 1,3,5,7,9,11,13 & 15, X=5000

Protein intensity 2,4,6,8,10,12,14&16 (X=40)

