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POLLENS-A TOOL FOR PHARMACOGNOSY

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ABSTRACT

Medicinal plants are the natural sources of drugs to cure various ailments. There are number of ancient literatures about the natural drugs, these literatures are usually found in Sanskrit and in vernacular language. Names of these botanicals are based on the morphology of officinal part. Now a day most of the phyto-pharmaceutical companies are depends on wanders for raw material supply in bulk. The wanders were collecting herbs in the wild without knowing the difference among the herbs, sometimes intentionally. In case of closely related species, it is very difficult to identify and differentiates the species. Our approach is to investigating the pollens of medicinal plants. Pollens play a very important role to identify the species due to their uniqueness in the shape, apertures, polarity, ornamentations and symmetry. Present investigation was conducted on pollens of 25 medicinal plants of varied habits belonging to 12 families of angiosperms. Medicinal plants *Cardiospermum canescens*. Wall and *Cardiospermum halicacabum*. L., belongs to Sapindaceae have triangular shaped pollens but they have difference in the apex of their angular structure. Whereas in *Malvastrum coromandelianum.* (L) Garcke, *Hibiscus sabdariffa.* L and *Abelmoschus moschatus.* Medik, belonging to Malvaceae, all three medicinal plants have almost spheroidal pollen grains and there is difference in spine's shape, arrangement and density. Investigated pollens of 25 medicinal plant species are unique in any one or more features of pollen parameters. Pollens of botanicals belonging to the same genus or different genus of the same family may have a similar shape and structure but there is a minor difference in the shape, aperture, polarity and other parameters of the pollens. Each botanical has its own unique pollen as a finger print. So pollen can be used as a one of the parameter in pharmacognostic studies of natural drugs in addition to organoleptic, microscopic, physico-chemical and other parameters. It can be include to The Ayurvedic Pharmacopeia of India.

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INTRODUCTION

In Indian system of medicine, numbers of medicinal plants are used in fresh and dry form as raw materials for preparation of medicine. Sometimes herbal drugs are misunderstood due to morphological similarity and adulteration due to unscientific naming in the ancient literature. In such case the organoleptic characters, stomatal numbers, stomatal index, vein islets numbers, histology of plant parts and phytochemical studies play an important role for identification and authentication of herbal drugs as a pharmacognostic parameters. These parameters are important and are time consuming and expensive. Sometimes it is very difficult to identify the botanicals of closely related genus or species through these parameters. In such cases pollen parameter play a very important role in judging the botanical identity. Pollen morphology has increased its application in plant taxonomy, paleo-botany and allergy in respiratory system & on skin, has been recognised [1]. Pollens are extremely tiny particles which cannot be seen by the naked eye. Pollen grains come in infinite variety of shape with complex surface in nature [2]. A pollen study is useful to identify various species and taxa in their respective families [3]. Pollens are highly resist to decaying and are ubiquitous in nature. On account of unique characters of pollen are often referred as finger print of plants [4, 5]. Pollen study is less expensive and it requires short time and very scientific. It can be conduct on all angiosperm medicinal plants. Each botanical has its own pollen features. Pollen features studies definitely helpful in pharmacognostical studies and can be resolve the problems of identity and authenticity of botanicals. Present research was conducted on 25 botanicals of different habits belongs to 12 families of angiosperm plants. These are *Wrightia tinctoria.* DC., *Holarrhena antidysenterica*. Wall, *Catharanthus roseus.* (L) G. Don, *Thevetia neriifolia*. Juss, belonging to Apocynaceae; *Malvastrum coromandelianum*. (L) Garcke, *Hibiscus sabdariffa*. L, and *Abelmoschus moschatus.* Medik, belong to Malvaceae; *Callicarpa macrophylla*. Vahl, *Leucas aspera*. (Willd) Link, and *Clerodendron serratum, var. dentatum* H. J. Lam, belonging to Lamiaceae; *Solanum torvum.* SW, *Solanum indicum*. L., and *Solanum nigrum.* L, belonging to Solanaceae; *Cardiospermum canescens*. Wall, and *Cardiospermum halicacabum*. L., belonging to sapindaceae; *Convolvulus pluricaulis.* Choisy, and *Evolvulus alsinoides*. L., belonging Convolvulaceae; *Clitoria ternatea*. L, and *Bauhinia variegata.* (L.) belonging to Fabaceae; *Adhatoda vasica*. Nees and *Barleria lupulina.* Lindl, belonging to Acanthaceae; *Euphorbia fusiformis.* Buch.-Ham.ex.D.Don of Euphorbiaceae; *Lannea coromandelica.* (Houtt.) Merr of Anacardiaceae; *Woodfordia fruticosa.* (L.) Kurz, of Lythraceae and *Plumbago zeylanica*. L, of Plumbaginaceae. Since pollens also play an important role in identification and authentication of drugs of plant origin. Pollens of the above botanical were studied under compound light microscope and were documented.

MATERIALS AND METHODS

In the present investigation, available polliniferous sample was collected from herb garden, Sri Sri College of Ayurvedic Science and Research, Bangalore and field survey was conducted to collect the rest of the sample from different places of Karnataka like Savanadurga of Ramanagara district, M M hills and B R hills of Chamarajanagar district during flowering season between April, 2016 to August, 2019. Plant specimens were identified by referring floras [6-8]. The anthers were collected from flower buds directly from the field and preserved in 70% ethyl alcohol for further investigation. The preserved matured pollen sample was prepared by acetolysis method according to Erdtman technique [9] for light microscope which involves the introduction of acetic anhydride mixed with concentrated sulphuric acid in the ratio 9:1. The tubes were immersed in boiling water bath for 3-5 min. Centrifuge the mixture and the supernatant was decanted, few drops of glycerine was added and mounted on glass slide. The prepared slides were studied under light microscope for morphological feature and photomicrograph of pollen grain was taken.

RESULTS AND DISCUSSION

The Pollen microphotographs and micro-morphology with respect to aperture, shape and exine stratification are represented in figure: I & II and in table: I & II, respectively. Polar view of pollens of *W. tinctora* [Fig.I.1] is non angular isopolar & radial symmetry [Tab.I.1]. *H. antidysenterica* [Fig.I.2] is non angular isopolar, bilateral symmetry [Tab.I.2], In *T. neriifolia*a [Fig.I.3] pollen is triangular straight obtuse, bilateral, heteropolar symmetry [Tab.I.3]. Pollen of C. *roseus* [Fig.I.4] in equatorial view is rectangular convex and obtuse having isopolar bilateral symmetry [Tab.I.4]. *M. Coromandelianum* [Fig.I.5], *A. moschatus* [Fig.I.6] and *H. sabdariffa* [Fig.I.7] pollens are in polar view are non-angular and have radial symmetry [Tab.I.5-7]. *S. torvum* [Fig.I.8] has non angular elliptical, truncate-isopolar, bilateral symmetry [Tab.I.8], *S. indicum* [Fig.I.9] pollen is elliptic, isopolar radial symmetry [Tab.I.9] and *S.nigrum* [Fig.I.10] pollen is non angular, heteropolar radial symmetry [Tab.I.10]. *C.canescens* [Fig.I .11] pollen is triangular, concave and obtuse; it has heteropolar, radial symmetry [Tab.I.11]. *C. halicacabum* [Fig.I.12] is triangular, convex, obtuse and heteropolar, radial symmetric pollen [Tab.I.12]. *C. pluricaulis* [Fig.I.13] and *Evolvulus alsinoides* [Fig.I.14] have non angular circular isopolar, radially symmetric pollens [Tab.I.13, 14]. *A. vasica* [Fig.I.15] pollen in equatorial view is rectangular, obtuse convex and isopolar, radial symmetry [Tab.I.15]. *B.lupulina* [Fig.I.16] has isopolar radial symmetric pollen grain [Tab.I.16]. *C. ternatea* [Fig.I.17] pollen, in polar view is triangular, concave and obtuse and has heteropolar bilateral symmetry [Tab.I.17]. *B.variegata* [Fig.I.18] pollen is isopolar and radial symmetric [Tab.I.18]. *C. macrophylla* [Fig.I.19] pollen grain in equatorial view is non angular-elliptical, truncate and has isopolar, bilateral symmetry [Tab.I.19]. *C.serratum* [Fig.II.20] pollen is circular, isopolar and radial symmetry [Tab.II.20]. *E. fusiformis* [Fig.II.21] pollen is non angular, heteropolar and radial symmetry in nature [Tab.II.21]. *W. fruticosa* [Fig.II.22] pollen is circular, isopolar and radial symmetry [Tab.II.22]. *L. coromandelica* [Fig.II.23] Pollen is circular, heteropolar and radial symmetry [Tab.II.23]. *L.aspera* [Fig.II.24] pollen is elliptical, isopolar and radial symmetry [Tab.II.24]. In *P.zeylanica* [Fig.II.25] pollen grain in polar view is elliptical, with isopolar and radial symmetry, aperture with long furrows [Tab.II.25].

The pollens of four species belong to Apocynaceae family differ in the shape, aperture but three are isopolar and one is heteropolar. Two are radial symmetry other two are bilateral symmetry in nature. In Malvaceae all three species are similar in terms of shape and symmetry and polarity but the difference is in the arrangement of spines, in *M. coromandelianum*, echines are densely arranged and have sharp pointed tip compared to other two, echine density is very less and the space between the echine is also larger in *A. moschatus* and base of the spines are broad and tip is rounded. In *H. sabdariffa* echines are not straight compare to other two species; middle part of the echine is curved. Pollens of *Cardiospermum* genus of Sapindaceae are triangular in shape in polar view. The difference is concave margin and apex was notched in case of *C.canescens*, Where as margin was convex and apex was obtuse in C. *halicacabum*. The pollen morphology varies among plant species; occur in varying shapes, forms, symmetry, polarity and exine structure.

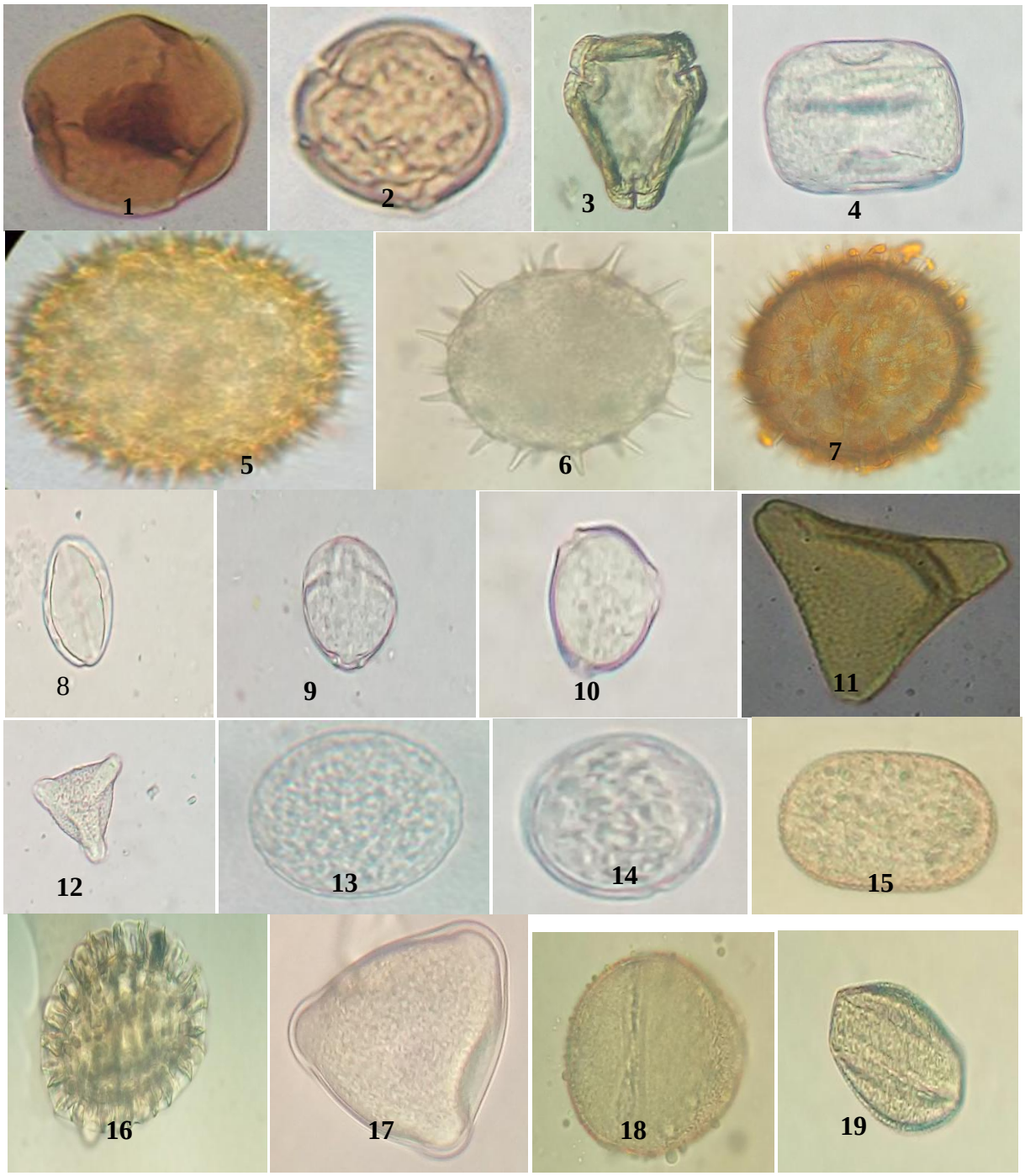

Figure: I. Pollen's photo micrographs.

1. *Wrightia tinctoria, 2. Holarrhena antidysenterica, 3. Thevetia neriifolia, 4. Catharanthus roseus, 5. Malvastrum coromandelianum, 6. Abelmoschus moschatus, 7. Hibiscus sabdariffa, 8. Solanum torvum, 9. Solanum indicum, 10. Solanum nigrum, 11. Cardiospermum canescens, 12. Cardiospermum halicacabum. 13. Convolvulus pluricaulis, 14. Evolvulus alsinoides, 15. Adhatoda vasica, 16. Barleria lupulina, 17. Clitoria ternatea, 18. Bauhinia variegata, 19. Callicarpa macrophylla.*

Table: I. Botanicals, family, habit and pollen features.

Sl.No	Species	Family	Habit	Pollen features
01	*Wrightia tinctoria*. DC	Apocynaceae	tree	Tricolporate, prolate-spheroidal,reticulate.
02	*Holarrhena antidysenterica.* (L) R.Br	Apocynaceae	tree	Tricolporate, prolate-spheroidal, regulate.
03	*Thevetia neriifplia.*(Pers)K.Schum	Apocynacaeae	tree	Tricolporate, sub-oblate, verrucate.
04	*Catharanthus roseus.* (L)G.Don	Apocynacaeae	herb	Tricolporate, prolate-subprolate, scabrate.
05	*Malvastrum coromandelianum.* (L)Garcke	Malvaceae	herb	Pantaporate, spheroidal, echinate.
06	*Abelmoschus moschatus.*Medik	Malvaceae	herb	Pantaporate, spheroidal, echinate.
07	*Hibiscus sabdariffa.*L	Malvaceae	herb	Pantoporate, spheroidal, echinate
08	*Solanum torvum*.SW	Solanaceae	herb	Tricolporate, prolate, psilate.
09	*Solanum indicum.* L	Solanaceae	herb	Tricolporate, prolate, psilate.
10	*Solanum nigrum*. L	Solanaceae	herb	Tricolporate, prolate-spheroidal, psilate.
11	*Cardiospermum canescens*.Wall.	Sapindaceae	climber	Tricolporate, triangular, reticulate.
12	*Cardiospermum halicacabum*.L.	Sapindaceae	climber	Tricolporate, triangular, reticulate.
13	*Convolvulus pluricaulis,*Choisy	Convolvulaceae	herb	Triporate, prolate-spheroidal, reticulate.
14	*Evolvulus alsinoides.*L.	Convolvulaceae	herb	Pentacolporate, prolate-spheroidal, reticulate.
15	*Adhatoda vasica.*Nees	Acanthaceae	shrub	Pantaporate, perprolate, reticulate.
16	*Barleria lupulina*.Lindl	Acanthaceae	herb	Trizonocolporate, oblate, reticulate.
17	*Clitoria ternatea.*L	Fabaceae	twinner	Triporate, triangular, psilate.
18	*Bauhinia variegata*. (L.)	Fabaceae	tree	Tricolporate, prolate-spheroidal, striate.
19	*Callicarpa macrophylla.*Vahl.	Verbenaceae	tree	Tricolporate, prolate, fossulate.

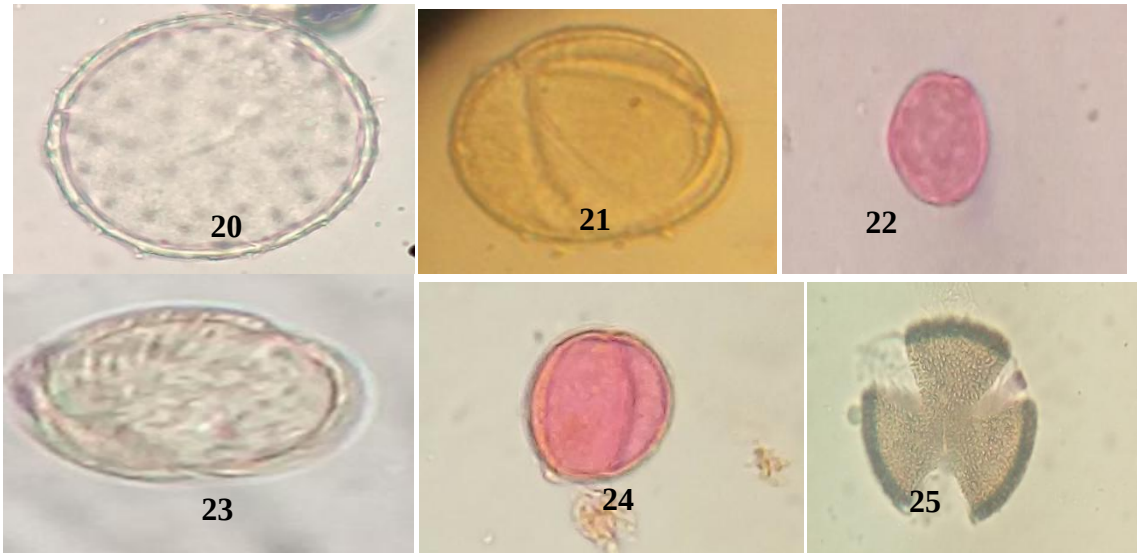

Figure: II. Pollen's photo micrographs.

20. *Clerodendron serratum var. dentatum,* 21. *Euphorbia fusiformis,* 22. *Woodfordia fruticosa,* 23. *Lannea coromandelica,* 24. *Leucas aspera,* 25. *Plumbago zeylanica*

Table: II. Botanicals, family, habit and pollen features.

Sl.No	Species	Family	Habit	Pollen features
20	*Clerodendron serratum.*var. *dentatum.* H.J.Lam	Verbenaceae	shrub	Tricolporate, prolate-spheroidal, scabrate.
21	*Euphorbia fusiformis.*Buch.-Ham.ex.D.Don	Euphorbiaceae	herb	Tricolporate, sub-spheroidal, psilate.
22	*Woodfordia fruticosa*. (L.) Kurz.	Lythraceae	shrub	Tricolporate, Prolate-spheroidal, psilate.
23	*Lannea coromandelica*. (Houtt.)Merr	Anacardiaceae	tree	Tricolporate, oblate-spheroidal, striate.
24	*Leucas aspera.*(Willd) Link	Lamiaceae	herb	Tricolpate, subprolate, psilate.
25	*Plumbago zeylanica*.L.	Plumbaginaceae	herb	Tricolpate, subprolate, reticulate.

CONCLUSION

Pollen morphology is beneficial in identification and authentication of plant species in their respective genus and families. Pollens are the finger prints of plant. It can be adopted as a one of the pharmacognostic parameter in identification of herbs in additions to other pharmacognostic parameters. Study showed a significant result that pollen is a tool for identification of botanicals. It can be include to The Ayurvedic Pharmacopeia of India. SEM pollen studies are recommended for future research

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Conflict of interest

The authors declared no conflict of interest.

ABBREVIATIONS

SEM: Scanning electron microscope

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