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Pollens of Melliferous Plants of Ecologic Zones IV and V of Togo

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Authors' contributions

This work was carried out in collaboration between all authors. Author CMK designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript and managed literature searches. Authors TE, KG and SC managed the analyses of the study and literature searches. All authors read and approved the final manuscript.

Original Research Article

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ABSTRACT

Pollinic analysis carried out on samples of honey harvested between 2009 and 2010 from natural beehives located in some parts of zones ecologic IV and V of the Guinean Zone of Togo: Danyi-Elavanyo, Danyi-Akayo, Igblélékoutsè-Béna and Azianfokopé has permitted to obtain 3625 grains of pollen which are grouped in 82 different types of pollen of melliferous plants; only 43 of them have been identified up to species level. *Elaeis guineensis*'s pollens are the most abundant and the only one found in all the samples of honey analysed. As a whole, the "Arboreal Pollens" are more represented than the "Non Arboreal Pollens" namely 87% against 23%. This study has been done to enrich the palynologic data bank of Togo which was very poor.
Besides, the photo of some determined pollens has been presented. The pictures have been taken with a light microscope, followed with their numerisation by a

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photomicroscope equipped with a camera, a computer and the softwares Quancoul and Motic Images Plus 2.0.

Keywords: Togo; Guinean Zone; Honey; Pollinic Analysis; Pollen; Melliferous Plants.

1. INTRODUCTION

Pollens are male reproduction cells of flowering plants. They are issued from stamens of flowers. They are obtained from the transformation of mother cells contained in cylindrical sacks pollinics of anthers.

A study among others contributes to the identification and classification of plants including melliferous ones. Melliferous plants are vegetable species from which bees extract their substances, particularly the nectar, the pollen and the resin to feed and for its diverse productions, one of which is honey [1].

These studies realized on the morphology of pollens allowed to bring out pollinic characteristics of each family, genre and species.

The morphology of the pollen grain of the plants met in Africa has been the object of study for a few specialists [2,3,4,5,6] Unfortunately, these studies do not cover the continent as a whole and concern less flowering plants especially those which produce pollens; even if nowadays the project « African Pollen Database » attempt to gather great number of photography of African pollen.

In Togo, only the works conducted by Lobreau-Callen and collaborators [7] have allowed to list certain melliferous threes from study of morphology of the pollen grain identified in the samples of the honey studied.

This study, carried out between 2009 and 2010, aimed specifically, on one hand, at determining the pollinic composition of the honeys harvested during this period in the areas of study, by the same time trying to draw up the list of melliferous plants presented in the areas, and on the other hand, at describing certain pollen of these plants.

2. MATERIAL AND METHODS

2.1 Material

Honey has been the base material used for this study. It has been harvested in four localities from the natural beehives put on trees in two ecologic zones:- tree localities at zone IV, two of them on plateau of Dayes (Danyi-Elavanyo et Danyi-Akayo) and one on plateau of Akposso at Bena (Igbélékoutsé); - one locality at zone V (Azianfokopé-Takpla).

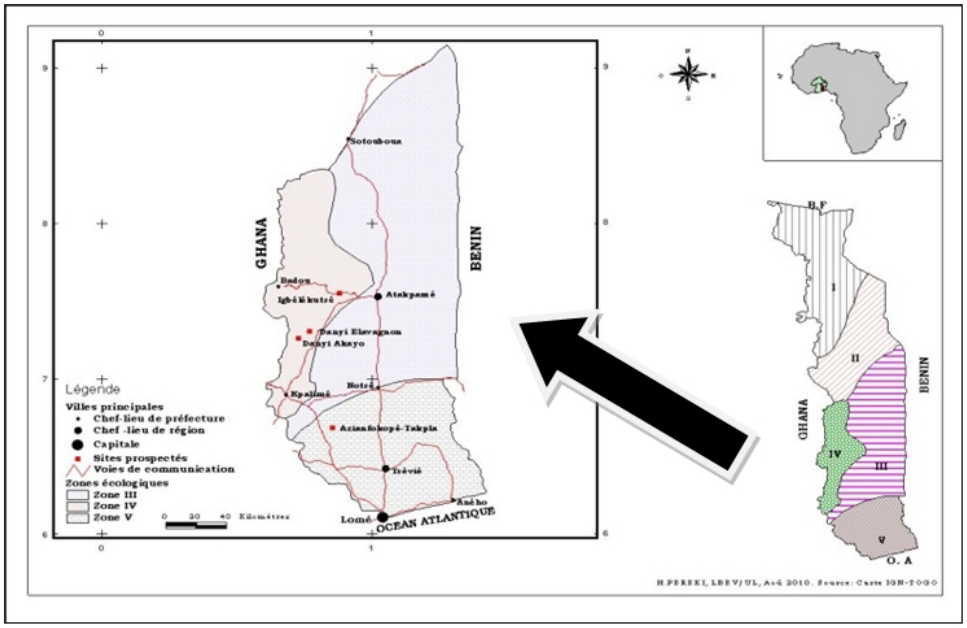

Fig. 1. Environment studied
Source: Koudégnan and al.; 2010

Fig. 2. Trunk of *Albizia adianthifolia* (Caesalpiniaceae) at Danyi-Akayo or Yésuvikopé
Source: Koudégnan and al.; 2010

2.2 Methods

2.2.1 Pollinic analysis of honey

Thirty millimeters of honey have been taken and treated according to the method of Erdtman [8] which consist in eliminating of the ingredients (sugars, proteins, wax and pectocellulosic membrane, etc) by a few acidify bathing. Then, the solution acidified has been centrifuged. For each sample of honey, roughly 20 µl of residue obtained after centrifugation are, first

time, put up between blades and strips, and then, observed under microscope for identify and count their palynologic population.

The determination of the pollens has been done up to species level (*Elaeis guineensis*), or genre level (*Ficus*), or again family level (Poaceae). In most of these cases the pollinic types are determined and represented by the genre (*Bidens*-type) or the species (*Bridelia ferruginea*-type). For the determination at the family level, it adds the suffix "undiff", which means undifferentiated to designate this pollinic type (Euphorbiaceae undiff).

Also the terms "Arboreal Pollens" (AP) and "Non Arboreal Pollens" (NAP), have been used to designate respectively pollen from ligneous species (trees, small shrubs, ligneous creeper) and these from grassy species.

Generally it has been admitted that the total number of pollen grains to count must reach at least 300 and the number of pollinic taxons must be equal or superior to 20 per sample [9]. The sum pollinic calculation doesn't take into account the indeterminable pollen grains. It helps calculate the percentage of different pollinic taxons and to obtain the pollinic spectre of each sample. A pollinic spectre is made of the identified pollinic taxons together with their numbers and representative rate.

2.2.2 Terminology and photography of pollen grains

The terminology of pollen grains used in this study is the one by certain authors [3],[4] and other palynologic collections. The pictures of the pollen grains have been taken with a light microscope at lens 100x followed by their digitalization by a camera incorporated light microscope, a computer and the softwares Quancould and Motic Images Plus 2.0.

3. RESULTS AND DISCUSSION

3.1 Results

3.1.1 Pollinic spectres

All in all, 3625 pollen grains have been numbered in the samples of honey analysed and grouped into 82 pollinic taxons belonging to 35 melliferous families. Only 43 pollinic taxons have been identified up to level species, 28 identified up to level gender and 04 identified up to level family. The indeterminable pollens have not been taken into account.

The table hereafter presents the honey's pollinic spectres of different localities considered.

Table 1. Pollinic spectres of honey harvested in the localities studies

Pollinics Taxons	Locality 1	Locality 2	Locality 3	Locality 4
DICOTYLEDONAE				
Amaranthaceae				
Amaranthus-type	+	-	-	-
Celosia argentea-type	+	-	-	-
Cyathula-type *prostrata*	++	-	-	-
Amaranthaceae undiff.	-	-	**+**	++
Anacardiaceae				
Mangifera indica	+	-	++	-
Lannea-type	+	-	+	-
Lannea-type *kerstingii*	-	-	-	++
Annonaceae				
Annonaceae undiff.	-	-	-	+
Apocynaceae				
Tabernaemontana crassa-type	-	-	-	+
Asteraceae				
Ageratum-type *conyzoides*	++	+	-	-
Aspilia africana	-	+	-	-
Bidens-type	++++	-	-	+
Pluchea ovalis-type	+	-	-	-
Vernonia-type	+	-	-	+
Bombacaceae				
Ceiba pentandra	+	+	-	-
Boraginaceae				
Cordia	-	-	++	-
Caesalpiniaceae				
Berlinia-type	-	-	-	++
Cassia -type	-	-	-	+
Daniellia oliveri-type	-	-	-	+
Delonix regia-type	+	-	-	-
Dialium guineense-type	-	-	+	-
Celastraceae				
Maytenus	-	-	-	**+**
Chrysobalanaceae				
Parinari-type	-	-	+	-
Clusiaceae				
Harungana madagascariensis-type	-	-	-	++
Combretaceae				
Combretum-type *racemosum*	-	-	+	-
Terminalia-type *glaucescens*	-	++	+	-
Combretaceae undiff.	+	-	-	-
Convolvulaceae				
Ipomoea-type	+	-	-	-
Dilleniaceae				
Tetracera alnifolia-type	-	++	-	+
Euphorbiaceae				

Table 1 Continues.....

Alchornea cordifolia	-	++	-	-
Bridelia ferruginea-type	-	++	-	-
Hymenocardia acida-type	-	-	-	++
Macaranga-type	-	+	+	+
Mallotus-type	+	-	-	+
Securinega virosa	+	+	-	++
Tetrorchidium didymostemon-type	-	++	+	+++
Euphorbiaceae undiff.	-	-	-	++
Fabaceae				
Canavalia-type	+	-	-	-
Centrosema pubescens-type	+	-	-	-
Indigofera-type *microcarpa*	-	+	-	-
Millettia-type	-	-	-	++
Pterocarpus-type	-	+	-	-
Fabaceae undiff.	-	++	++	+++
Malvaceae				
Abutilon-type	+	-	-	-
Sida	+	-	-	-
Mimosaceae				
Acacia-type *mellifera*	-	-	-	+
Albizia-type *adianthifolia*	-	++	-	-
Dichrostachys cinerea	-	-	-	+
Entada-type *abyssinica*	-	++	++	+++
Pentaclethra macrophylla	-	++	-	-
Schrankia leptocarpa	-	++	-	+
type *Xylia schliebenii*	-	+	-	-
type *Xylia torreana*	-	++	-	-
Moraceae				
Antiaris-type *africana*	-	+++	-	-
Ficus	-	-	++++	++
Myrtaceae				
Psidium guajava-type	+	+	+	-
Syzygium-type	-	-	+	+
Ochnaceae				
Lophira alata-type	-	-	-	+
Olacaceae				
Olax	-	+	-	-
Portulacaceae				
Talinum-type	+	-	-	-
Rhamnaceae				
Rhamnaceae undiff.	-	-	-	++
Rubiaceae				
Canthium-type *subcordatum*	+	-	-	-
Crossopteryx febrifuga	-	+	-	-
Mitragyna inermis-type	-	-	-	+
Nauclea-type	-	+	+	+

Table 1 Continues.....

Rubiaceae undiff.	-	+	+	++
Rutaceae				
Citrus	+	++	-	+
Zanthoxylum-type *xanthoxylloides*	-	+	+	+
Rutaceae undiff.	-	-	+	-
Sapindaceae				
Cardiospermum halicacabum	+	-	-	-
Lecaniodiscus cupanioides	-	-	+	-
Paullinia pinnata	-	-	+	+
Sapindaceae undiff.	-	-	+	-
Sterculiaceae				
Cola-type	-	-	+	-
Tiliaceae				
Grewia-type	+	-	-	+
Ulmaceae				
Celtis	-	+++	-	-
Holoptelea grandis	-	++	-	-
Vitaceae				
Cissus	-	-	-	++
MONOCOTYLEDONAE				
Agavaceae				
Agavaceae undiff.	-	-	-	+
Arecaceae				
Elaeis guineensis	+++++	+++	++++	+
Poaceae				
Poaceae undiff.	+	+	+	-
Typhaceae				
Typha	+	-	-	-
indeterminate	-	+	+	++
TOTAL OF POLLEN NUMBERED	**2182**	**311**	**810**	**322**

Locality 1 : Azianfokopé-Takpla ; locality 2 : Danyi-Elavanyo ; locality 3 : Danyi-Akayo ; locality 4 : Igbélékoutsè (Béna) ; undiff : undifferentiated ; - : absent ; + : % between 0 and 1 ; ++ : % between 1 and 10 ; +++ : % between 10 and 30 ; ++++ : % between 30 and 50 ; +++++ : % > 50.

Source: Koudégnan and al.; 2010

3.1.2 Distribution of the pollinic taxons according to pollen categories

Following the precision made in material and methods, two categories of pollens have been identified: “Arboreal Pollens” (AP) and “Non Arboreal Pollens”. The graphic below presents for each locality the distribution of pollinic taxons according to the two categories of pollen identified.

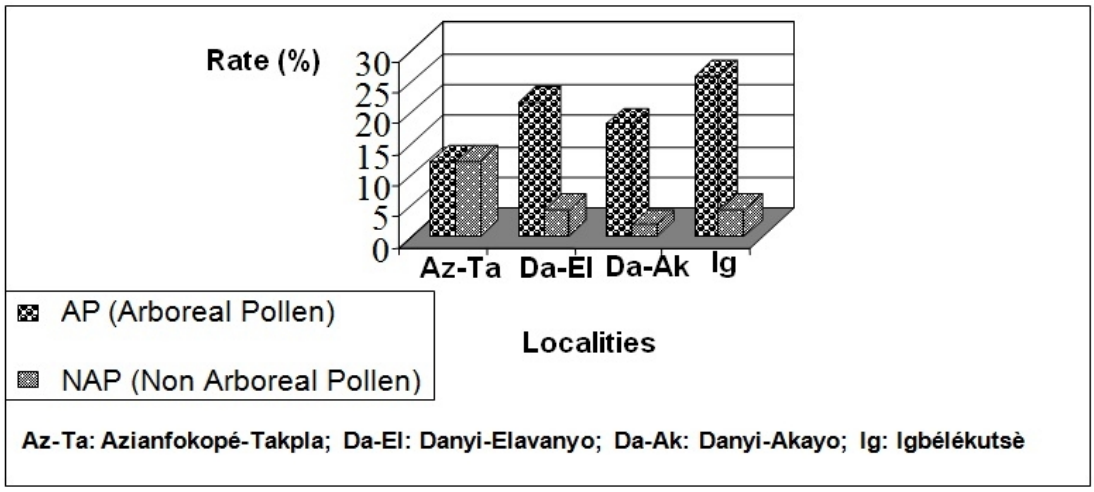

Fig. 3. Frequency of pollinic taxons of localities according to pollen categories
Source: Koudégnan and al.; 2010

In Azianfokopé-Takpla the rate of Non Arboreal Pollens (NAP) is near the rate of Arboreal Pollens (AP). But in the other tree localities, the rate of the NAP remains largely lower than the rate of AP (16.67 % against 83.33 % at Danyi-Elavanyo; 8.7 % against 91.3 % at Danyi-Akayo; 14.29 % against 85.71 % at Igbélékoutsè-Béna).

3.2 Discussion

The pollinic spectre obtained from analysis reveal the presence of some of these pollens in both honey of the two ecologic zones IV and V. The Fig.4. stands for these pollens according to the total of the numbered pollens.

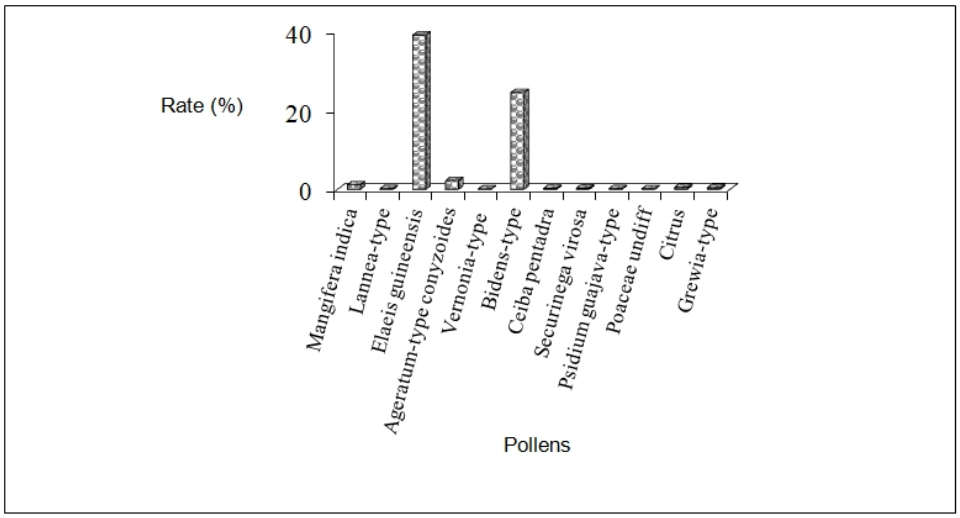

Fig. 4. Pollens present in the two ecologic zones
Source: Koudégnan and al.; 2010

Furthermore, it is important to note that only pollen of *Elaeis guineensis* have been found in all of the samples of honey harvested. This is due to the fact that plants which produce them are abundantly available in the localities and also constitute the provisions source for bees [1,10,11].

The melliferous flora constituted by taxons identified is dominated by ligneous plants (trees, small shrubs, ligneous creeper). This shows that the pollens taken from woody species are largely superior to those which are taken from grassy plants by bees. The high rate of these taxons in spectre implies its availability in the nearby vegetation. This result confirms the reality on the site studied. On the whole, the zone studied is dominated by the forest. The ligneous plants, being the most available and visible, attract worker bees in search of flowers nutritious elements (pollen nectar, resin, water, miellat) which are mainly transformed into honey and propolis. This predominance of ligneous taxons rhymes with the results of certain authors [12,13,14,15]. But it's opposed to the works of others [11,16].

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Nevertheless, it is necessary to notice that the results obtained in Azianfokopé-Takpla reveal that there is as much Arboreal Pollen as Non Arboreal Pollen. The high rate of pollen withdrawn from herb flowers comparatively to these of other localities is due to fact that this locality is a savannah and limited by agricultural domains. So, the bees would be influenced by these ecological conditions.

The pollinic spectre analysis obtained from this study shows that the samples of honey harvested have been made from several nectars a plant. The status of poly-flower honey or one-flowery honey depends on the relative proportion of different types of pollen which make it. Honey is called one-flower if only one type of pollen is superior to 45 % of all pollens identified [17]. From this standpoint, we can affirm that the honeys of Azianfokopé-Takpla and Danyi-Akayo are one-flower honeys. It is respectively, honey of *Elaeis guineensis* (51.15 % of all pollens) and that of *Ficus* (45.56% of all pollens). The flatly abundance of species pollens mentioned above would be linked not only to the quantity but also to the quality of the pollen [10].

At last, the pollinic spectre shows that the pollinic wealth (total of pollen grains numbered) of different honey analysed varies from one locality to other: Azianfokopé-Takpla (2182), Danyi-Elavanyo (311), Danyi-Akayo (810) and Igbélékutsè-Béna (322). This gap is not due to the honey analysed, nor to the type of bees which made the honey. The poverty in pollens of honey harvested in Danyi-Elavanyo and Igbélékutsè-Béna could be due to the fact that these samples of honey are essentially made nectar-based [7].

3.3 Photos of a Few Pollens (x 1000)

Plate 1 – Dicotyledonae

Amaranthaceae: *Amaranthus*-type (fig.3) ; **Anacardiaceae** : *Mangifera indica* (figs. 1a, 1b, 1c) ; *Lannea*-type (figs. 2a, 2b, 2c) ; **Asteraceae** : *Bidens*-type (figs. 4a, 4b)

Plate 2 – Dicotyledonae

Asteraceae: *Ageratum*-type *conyzoides* (figs. 5a, 5b, 5c); *Aspilia africana* (figs. 6a, 6b, 6c); *Vernonia*-type (figs. 7a, 7b); **Bombacaceae** : *Ceiba pentandra* (figs. 8a, 8b)

Plate 3 – Dicotyledonae

Combretaceae : *Terminalia*-type *glaucescens* (figs. 9a, 9b); **Euphorbiaceae** : *Hymenocardia acida*-type (figs. 10) ; **Malvaceae** : *Sida* (figs. 11a, 11b); **Moraceae** : *Ficus* (figs. 12a, 12b); **Mimosaceae** : *Acacia*-type *mellifera* (figs. 13)

Plate 4 – Dicotyledonae

Mimosaceae : *Shrankia leptocarpa* (figs. 14); **Portulacaceae** : *Talinum*-type (fig. 15); **Sapindaceae** : *Cardiospermum halicacabum* (figs. 16a, 16b) ; **Tiliaceae** : *Grewia*-type (figs. 17a, 17b)

Plate 5 – Dicotyledonae

Mimosaceae : *Dichrostachys cinerea* (figs. 18a, 18b)

Plate 6 – Monocotyledonae

Arecaceae : *Elaeis guineensis* (fig. 19a, 19b, 19c, 19d) ; **Poaceae** : Poaceae undiff (fig. 20a ; 20b, 20c)

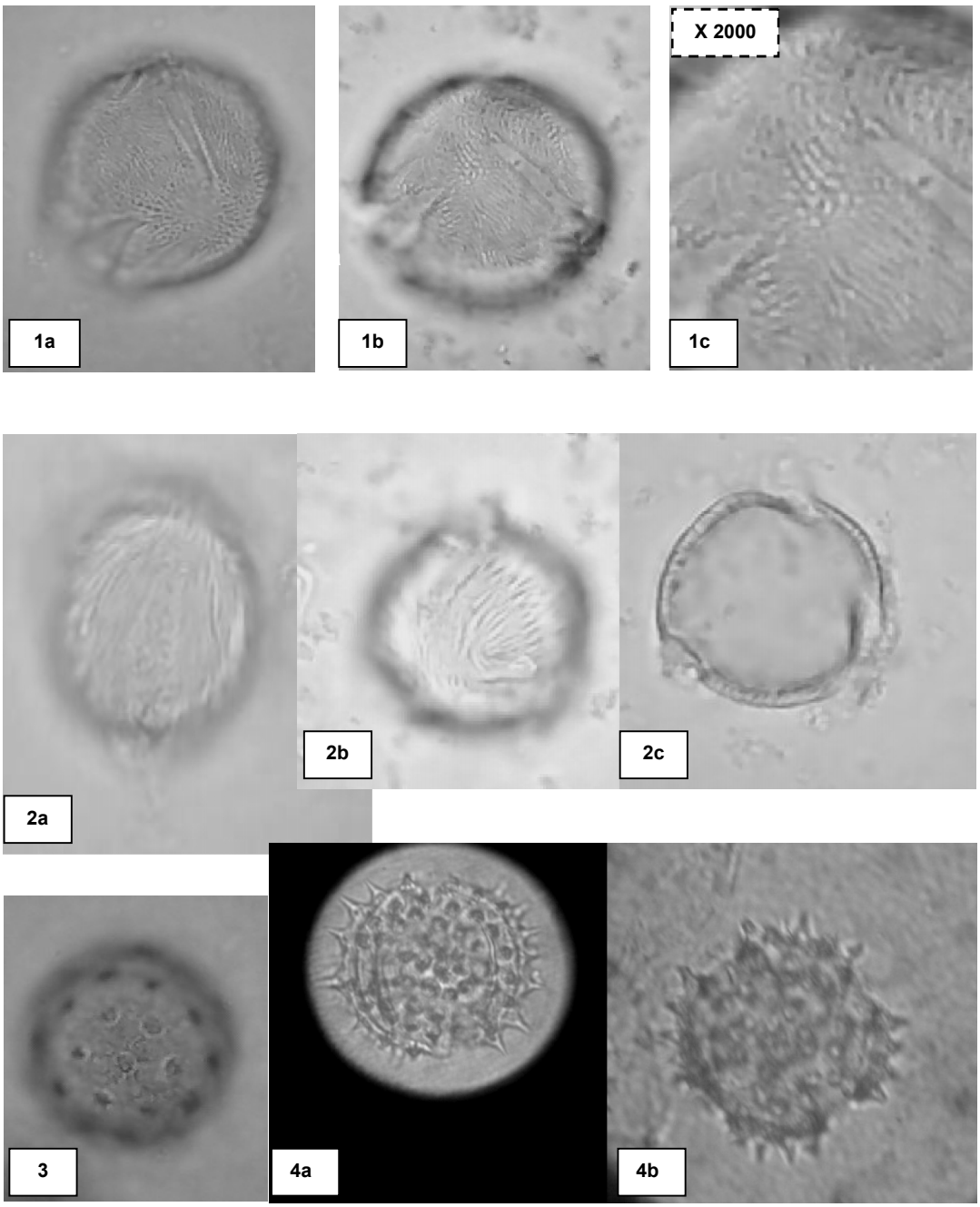

Plate 1 – Dicotyledonae
***Source**: Koudegnan and al.; 2010*

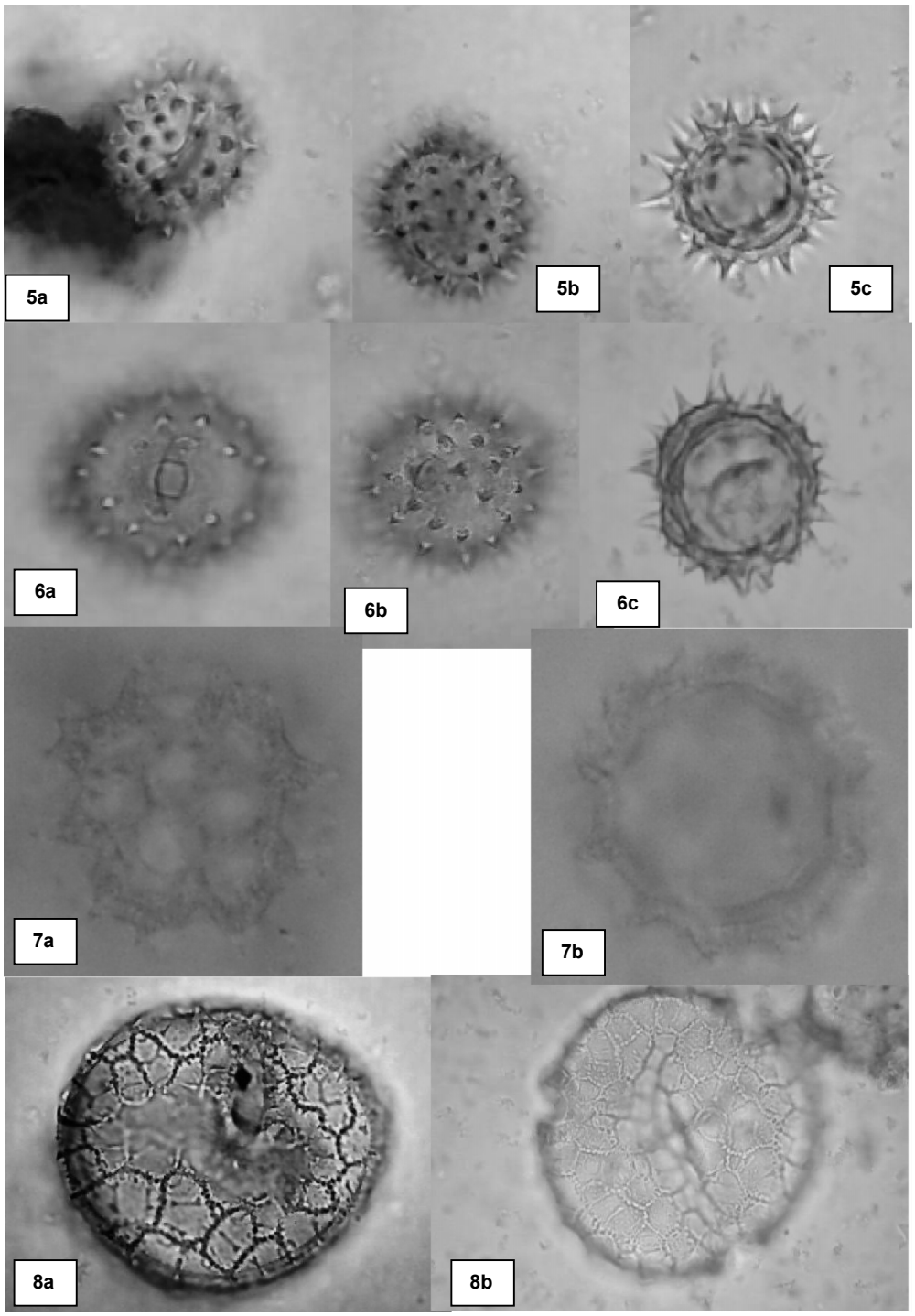

Plate 2 – Dicotyledonae
***Source**: Koudegnan and al.; 2010*

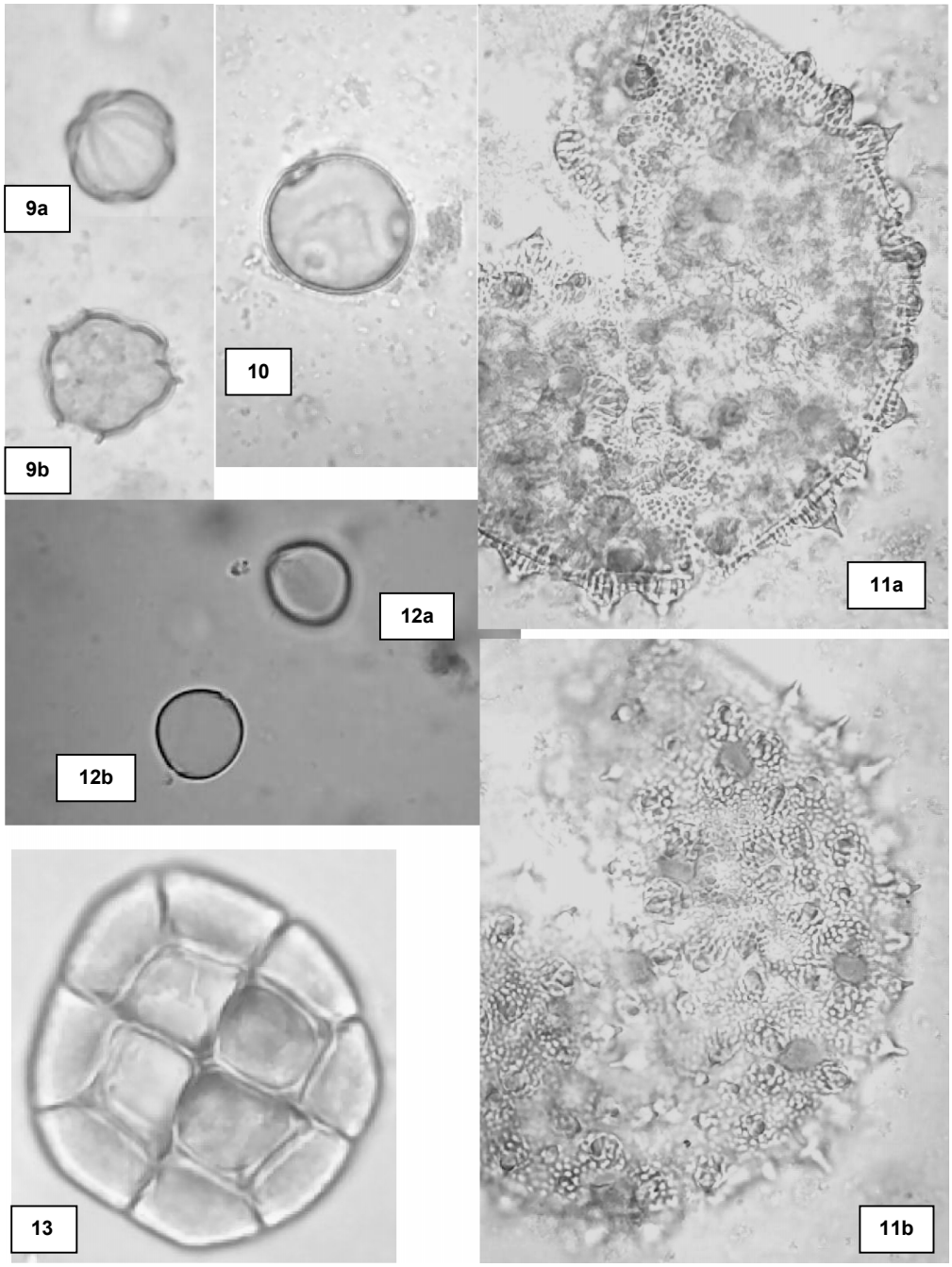

Plate 3 – Dicotyledonae
***Source**: Koudegnan and al.; 2010*

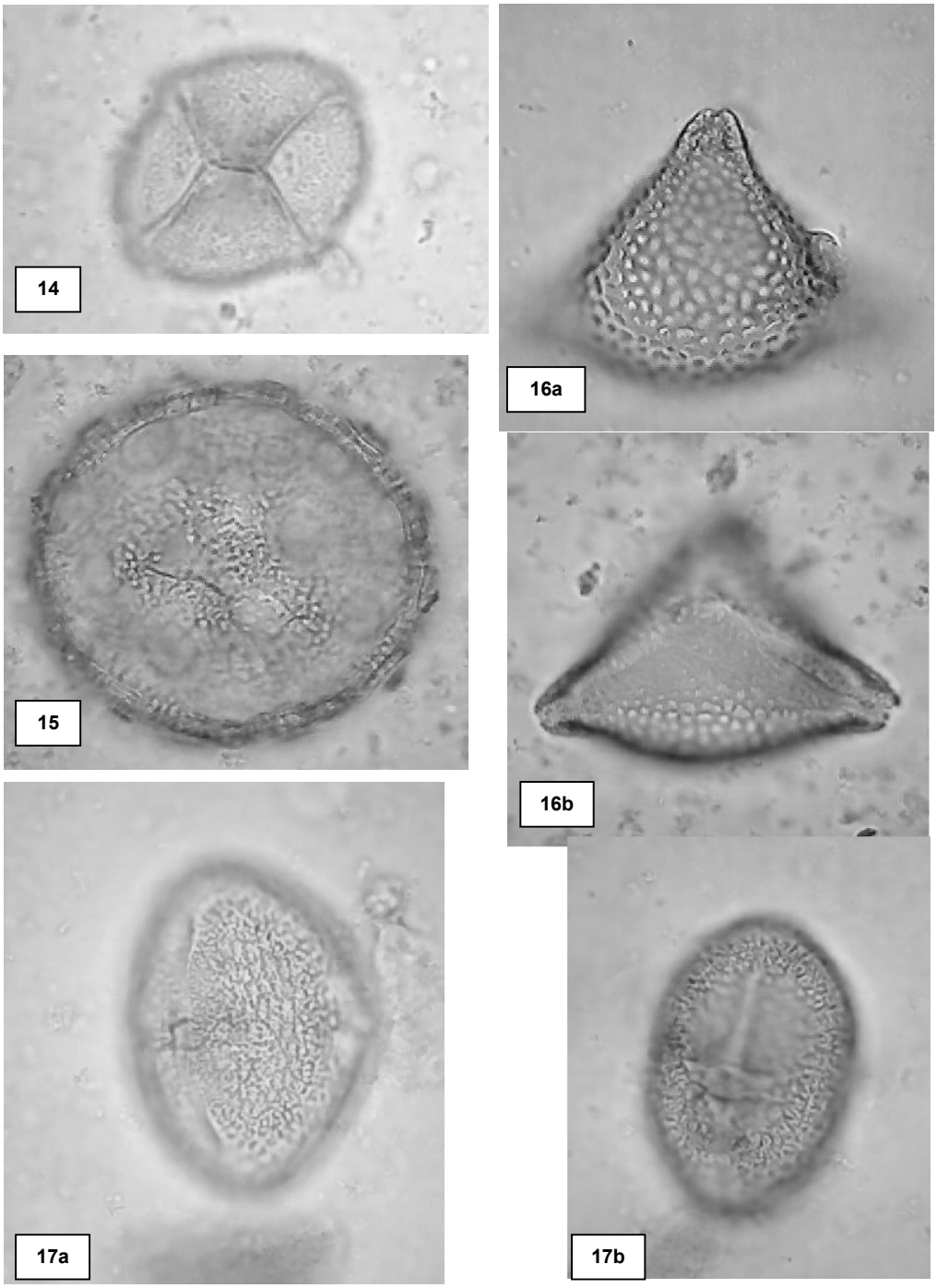

Plate 4 – Dicotyledonae
Source: Koudegnan and al.; 2010

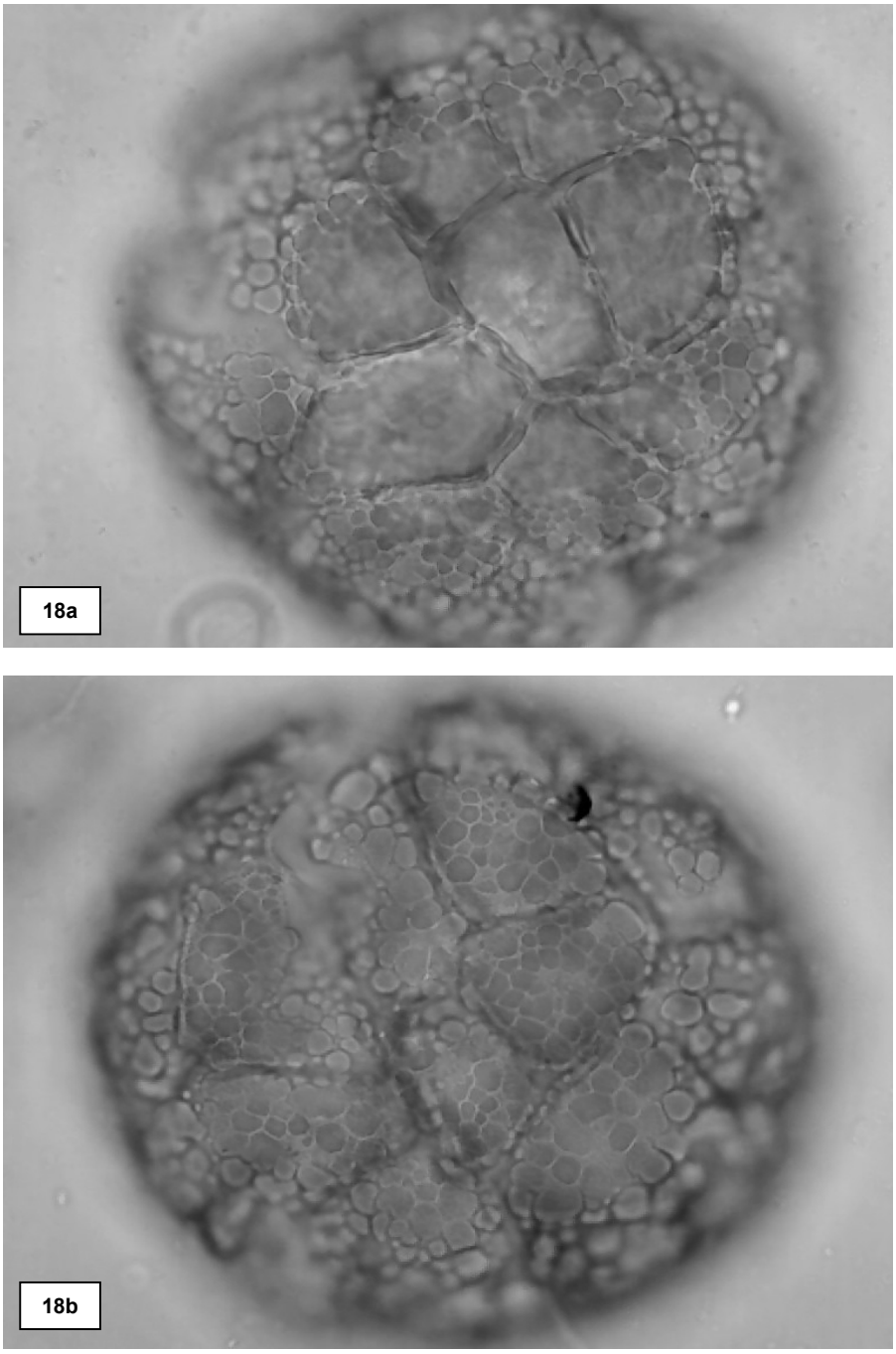

Plate 5 – Dicotyledonae
Source: Koudegnan and al.; 2010

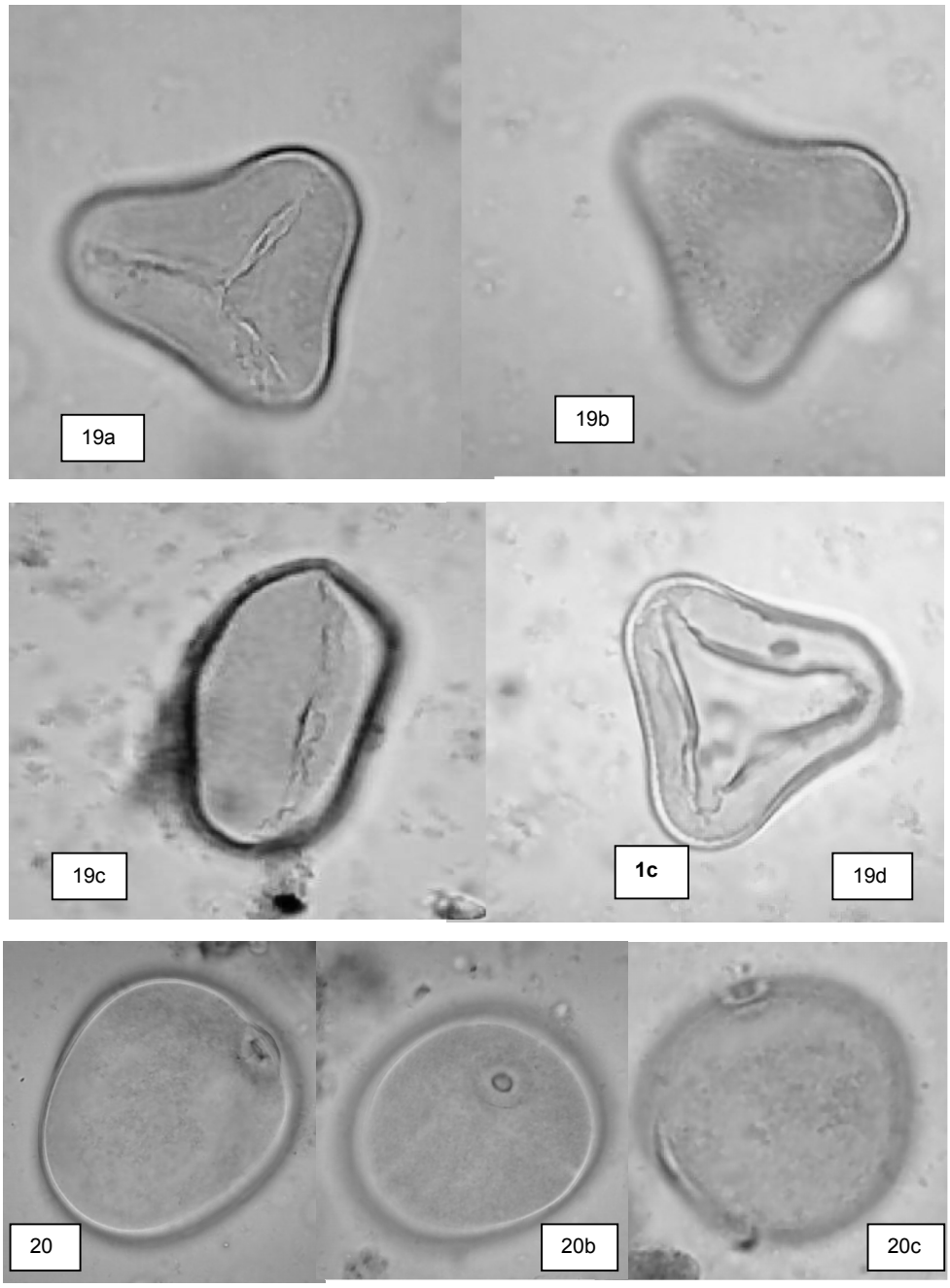

Plate 6 – Monocotyledonae
Source: Koudegnan and al.; 2010

4. CONCLUSION

The pollinic analysis of honey's samples harvested has helped us to count 3625 pollen grains which are grouped into 82 pollinic taxons belonging to 35 melliferous families. The analysis has proved that:

- the pollen of *Elaeis guineensis* (Arecaceae) are more collected than the others because it is very preferred by the bees;

- in the localities of harvest of the ecological zone IV studied, the Arboreal Pollens outnumbers the Non Arboreal Pollens (nearly 87 % against 23 %) compared to the locality of ecological zone V where the rate of both categories of pollens are identical;

- the honeys of Azianfokopé-Takpla and Danyi-Akayo are qualified as one-floral honeys;
- the pollinic wealth varies from one locality to other with a high numbered in honey of Azianfokopé-Takpla (2182) and Danyi-Akayo (810).

In the end, the photos of a few pollen grains taken with light microscope at lens 100x have been presented.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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