

Original research papers

POLLENS OF MELLIFEROUS PLANTS OF ECOLOGIC ZONES IV AND V OF TOGO

Comlan M. KOUDEGNAN¹, Thérèse EDORH², Kudzo GUELLY³, Siendou COULIBALY⁴.

¹Palynology, Algology and Paleoecology Laboratory; Botanic and Ecology Laboratory; Faculty of sciences; University of Lomé

²Palynology, Algology and Paleoecology Laboratory; Faculty of sciences; University of Lomé

³Botanic and Ecology Laboratory; Faculty of sciences; University of Lomé

⁴Botanic Laboratory, UFR Biosciences; University of Félix Houphouët Boigny (Ivory Coast)

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, Igbélékoutsè-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 of these pollens determined has been presented. The photographs have been carried out with light microscope, followed of their numerisation by a 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 contributes among things to the identification and classification of plants including melliferous plants. 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 conducted on the morphology of pollens allowed bringing out pollinic characteristics of each family, genus 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, alone the works conducted by Lobreau-Callen and collaborators ([7]) have allowed to listing certain melliferous trees from study of morphology of the pollen grain identified in the samples of the honey studies.

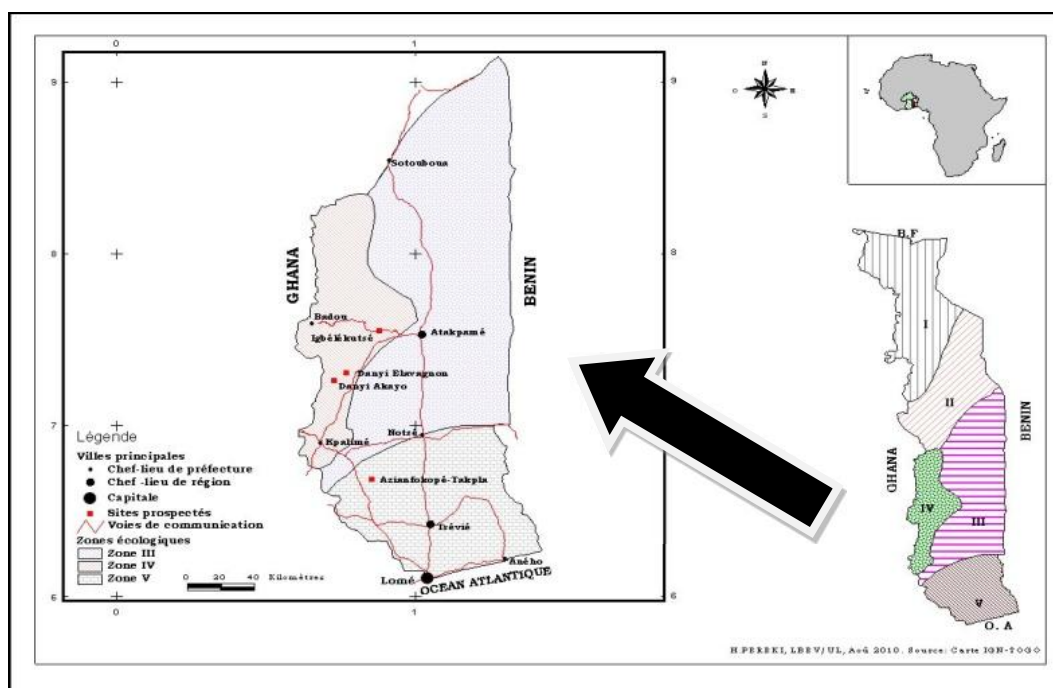
This study, carried out between 2009 and 2010, aimed specifically, on the one hand, at determining the pollinic composition of the honeys harvested during this period in the areas of study, by the same time trying to draw up the list of melliferous plants presented in the areas, on the other hand, at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



Source: Koudegnan and al.; 2010

Figure 1. Environment studied



Hive opening

Figure 2. Trunk of *Albizia adianthifolia* (Caesalpiniaceae) at Danyi-Akayo or Yésuvikopé

Source: Koudegnan 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 μ l 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 genus level (*Ficus*), or again family level (Poaceae). In most of these cases the pollinic types are determined and represented by the genus (*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 to 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 this used 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
DICOTYLEDONES				
AMARANTHACEAE				
<i>Amaranthus</i> -type	+	-	-	-
<i>Celosia argentea</i> -type	+	-	-	-
<i>Cyathula</i> -type <i>prostrata</i>	++	-	-	-
<i>Amaranthaceae undiff.</i>	-	-	+	++
ANACARDIACEAE				
<i>Mangifera indica</i>	+	-	++	-
<i>Lannea</i> -type	+	-	+	-
<i>Lannea</i> -type <i>kerstingii</i>	-	-	-	++
ANNONACEAE				
<i>Annonaceae undiff.</i>	-	-	-	+
APOCYNACEAE				
<i>Tabernaemontana crassa</i> -type	-	-	-	+
ASTERACEAE				
<i>Ageratum</i> -type <i>conyzoides</i>	++	+	-	-

<i>Aspilia africana</i>	-	+	-	-
<i>Bidens-type</i>	++++	-	-	+
<i>Pluchea ovalis-type</i>	+	-	-	-
<i>Vernonia-type</i>	+	-	-	+
BOMBACACEAE				
<i>Ceiba pentandra</i>	+	+	-	-
BORAGINACEAE				
<i>Cordia</i>	-	-	++	-
CAESALPINIACEAE				
<i>Berlinia-type</i>	-	-	-	++
<i>Cassia -type</i>	-	-	-	+
<i>Daniellia oliveri-type</i>	-	-	-	+
<i>Delonix regia-type</i>	+	-	-	-
<i>Dialium guineense-type</i>	-	-	+	-
CELASTRACEAE				
<i>Maytenus</i>	-	-	-	+
CHRYSOBALANACEAE				
<i>Parinari-type</i>	-	-	+	-
CLUSIACEAE				
<i>Harungana madagascariensis-type</i>	-	-	-	++
COMBRETACEAE				
<i>Combretum-type racemosum</i>	-	-	+	-
<i>Terminalia-type glaucescens</i>	-	++	+	-
<i>Combretaceae undiff.</i>	+	-	-	-

CONVOLVULACEAE				
<i>Ipomoea</i> -type	+	-	-	-
DILLENIACEAE				
<i>Tetracera alnifolia</i> -type	-	++	-	+
EUPHORBIACEAE				
<i>Alchornea cordifolia</i>	-	++	-	-
<i>Bridelia ferruginea</i> -type	-	++	-	-
<i>Hymenocardia acida</i> -type	-	-	-	++
<i>Macaranga</i> -type	-	+	+	+
<i>Mallotus</i> -type	+	-	-	+
<i>Securinega virosa</i>	+	+	-	++
<i>Tetrorchidium didymostemon</i> -type	-	++	+	+++
<i>Euphorbiaceae undiff.</i>	-	-	-	++
FABACEAE				
<i>Canavalia</i> -type	+	-	-	-
<i>Centrosema pubescens</i> -type	+	-	-	-
<i>Indigofera</i> -type <i>microcarpa</i>	-	+	-	-
<i>Millettia</i> -type	-	-	-	++
<i>Pterocarpus</i> -type	-	+	-	-
<i>Fabaceae undiff.</i>	-	++	++	+++
MALVACEAE				
<i>Abutilon</i> -type	+	-	-	-
<i>Sida</i>	+	-	-	-
MIMOSACEAE				
<i>Acacia</i> -type <i>mellifera</i>	-	-	-	+

* Tel.: (+228) 90081341. – B.P. 1515 (University of Lomé - Togo)
E-mail address: kcomlanmawussi@yahoo.fr; comawuk.77@gmail.com

<i>Albizia-type adianthifolia</i>	-	++	-	-
<i>Dichrostachys cinerea</i>	-	-	-	+
<i>Entada-type abyssinica</i>	-	++	++	+++
<i>Pentaclethra macrophylla</i>	-	++	-	-
<i>Schrankia leptocarpa</i>	-	++	-	+
type <i>Xylia schliebenii</i>	-	+	-	-
type <i>Xylia torreana</i>	-	++	-	-
MORACEAE				
<i>Antiaris-type africana</i>	-	+++	-	-
<i>Ficus</i>	-	-	++++	++
MYRTACEAE				
<i>Psidium guajava-type</i>	+	+	+	-
<i>Syzygium-type</i>	-	-	+	+
OCHNACEAE				
<i>Lophira alata-type</i>	-	-	-	+
OLACACEAE				
<i>Olax</i>	-	+	-	-
PORTULACACEAE				
<i>Talinum-type</i>	+	-	-	-
RHAMNACEAE				
<i>Rhamnaceae undiff.</i>	-	-	-	++
RUBIACEAE				
<i>Canthium-type subcordatum</i>	+	-	-	-
<i>Crossopteryx febrifuga</i>	-	+	-	-

<i>Mitragyna inermis</i> -type	-	-	-	+
<i>Nauclea</i> -type	-	+	+	+
<i>Rubiaceae undiff.</i>	-	+	+	++
RUTACEAE				
<i>Citrus</i>	+	++	-	+
<i>Zanthoxylum</i> -type <i>xanthoxylloides</i>	-	+	+	+
<i>Rutaceae undiff.</i>	-	-	+	-
SAPINDACEAE				
<i>Cardiospermum halicacabum</i>	+	-	-	-
<i>Lecaniodiscus cupanioides</i>	-	-	+	-
<i>Paullinia pinnata</i>	-	-	+	+
<i>Sapindaceae undiff.</i>	-	-	+	-
STERCULIACEAE				
<i>Cola</i> -type	-	-	+	-
TILIACEAE				
<i>Grewia</i> -type	+	-	-	+
ULMACEAE				
<i>Celtis</i>	-	+++	-	-
<i>Holoptelea grandis</i>	-	++	-	-
VITACEAE				
<i>Cissus</i>	-	-	-	++
MONOCOTYLEDONES				
AGAVACEAE				
<i>Agavaceae undiff.</i>	-	-	-	+

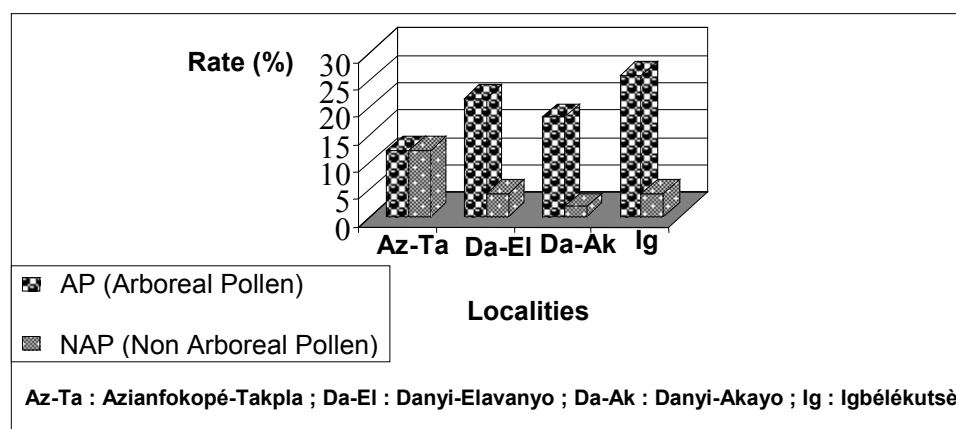
ARECACEAE				
<i>Elaeis guineensis</i>	+++++	+++	++++	+
POACEAE				
<i>Poaceae undiff.</i>	+	+	+	-
TYPHACEAE				
<i>Typha</i>	+	-	-	-
indeterminate	-	+	+	++
TOTAL OF POLLEN NUMBERED	2182	311	810	322

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: Koudegnan 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.



Source: Koudegnan and *al.*; 2010

Figure 3. Frequency of pollinic taxons of localities according to pollen categories

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

171

172

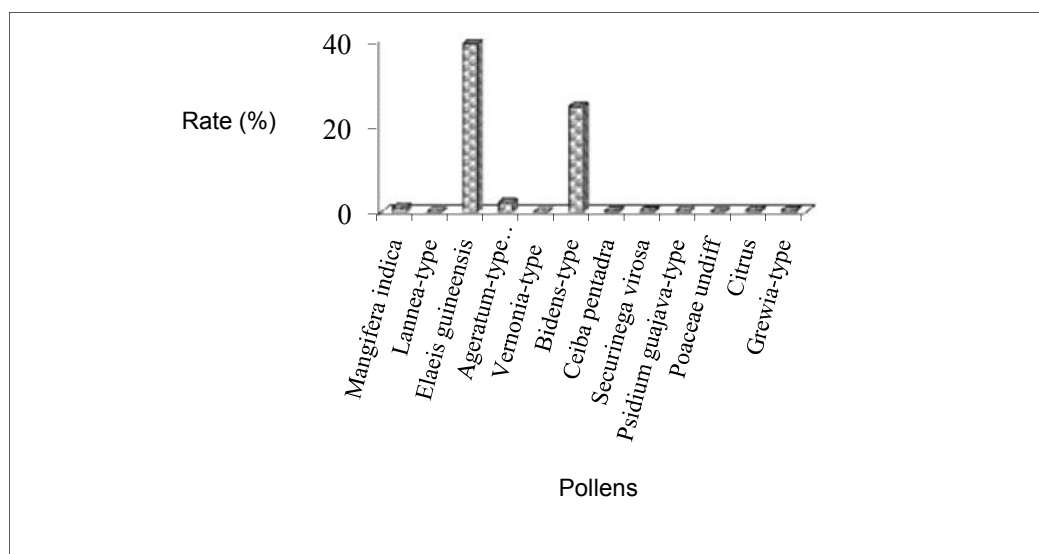
173 3.2 Discussion

174

175 The pollinic spectre obtained from analysis reveal the presence of some of these pollens in
 176 both honey of the two ecologic zones IV and V. The Figure 4 present the representation of
 177 these pollens according to total of numbered pollens.

178

179



180

181

Source: Koudegnan and al.; 2010

182

183

Figure 4. Pollens present in the two ecologic zones

184

185

186

187 Furthermore, it is important to note that only pollen of *Elaeis guineensis* have been found in
 188 all of the samples of honey harvested. This is due to fact that plants which produce them are
 189 abundantly available in the localities and also constitute the provisions source for bees ([1],
 190 [10], [11]).

191

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

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-flower honey depends on the relative proportion of different types of pollen which make it. A honey is calling one-flower if the number of only type of pollen is superior at 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 the quality of the pollen [10].

Lastly, the pollinic spectre show 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 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 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)

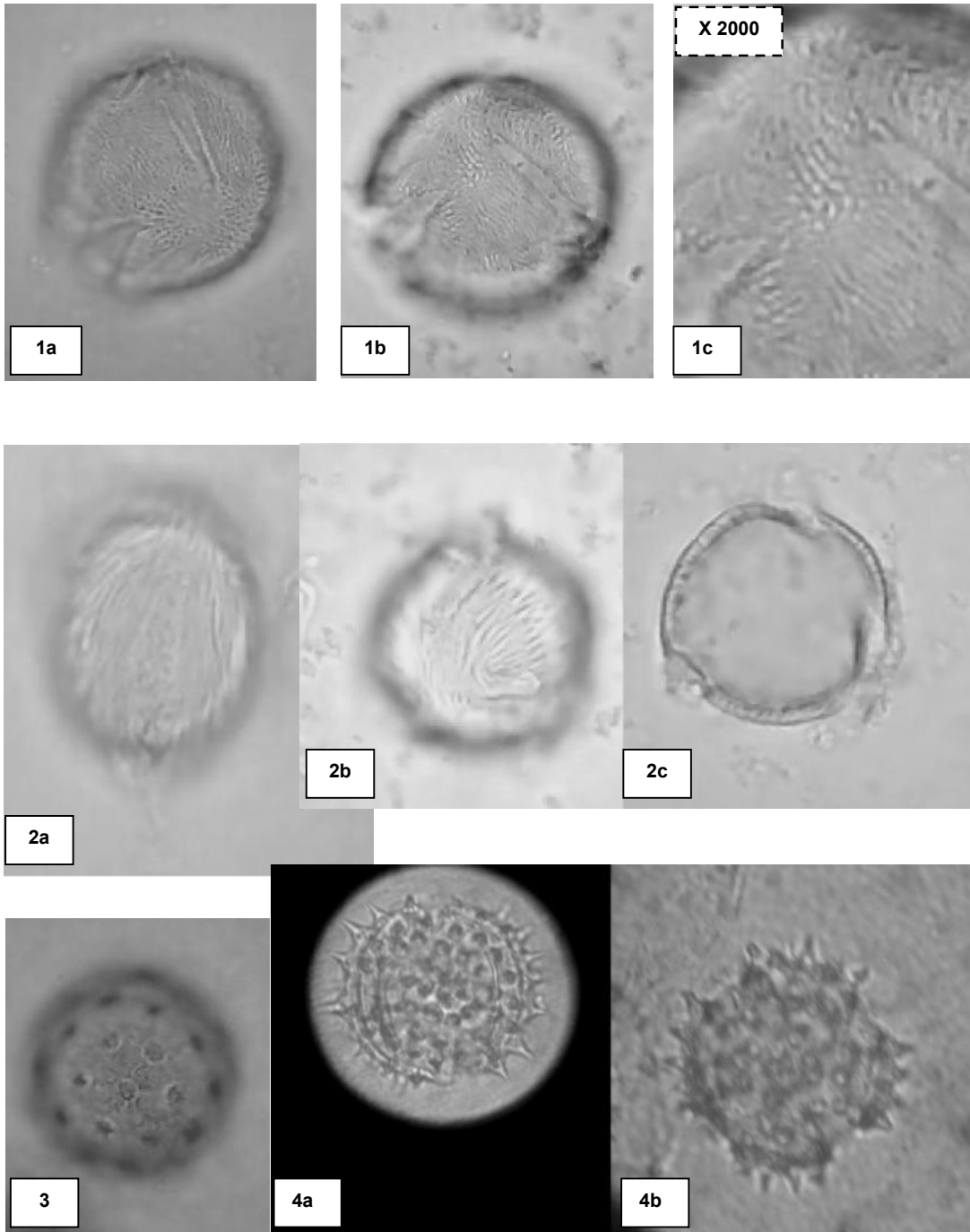
Plate 5 – Dicotyledonae

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

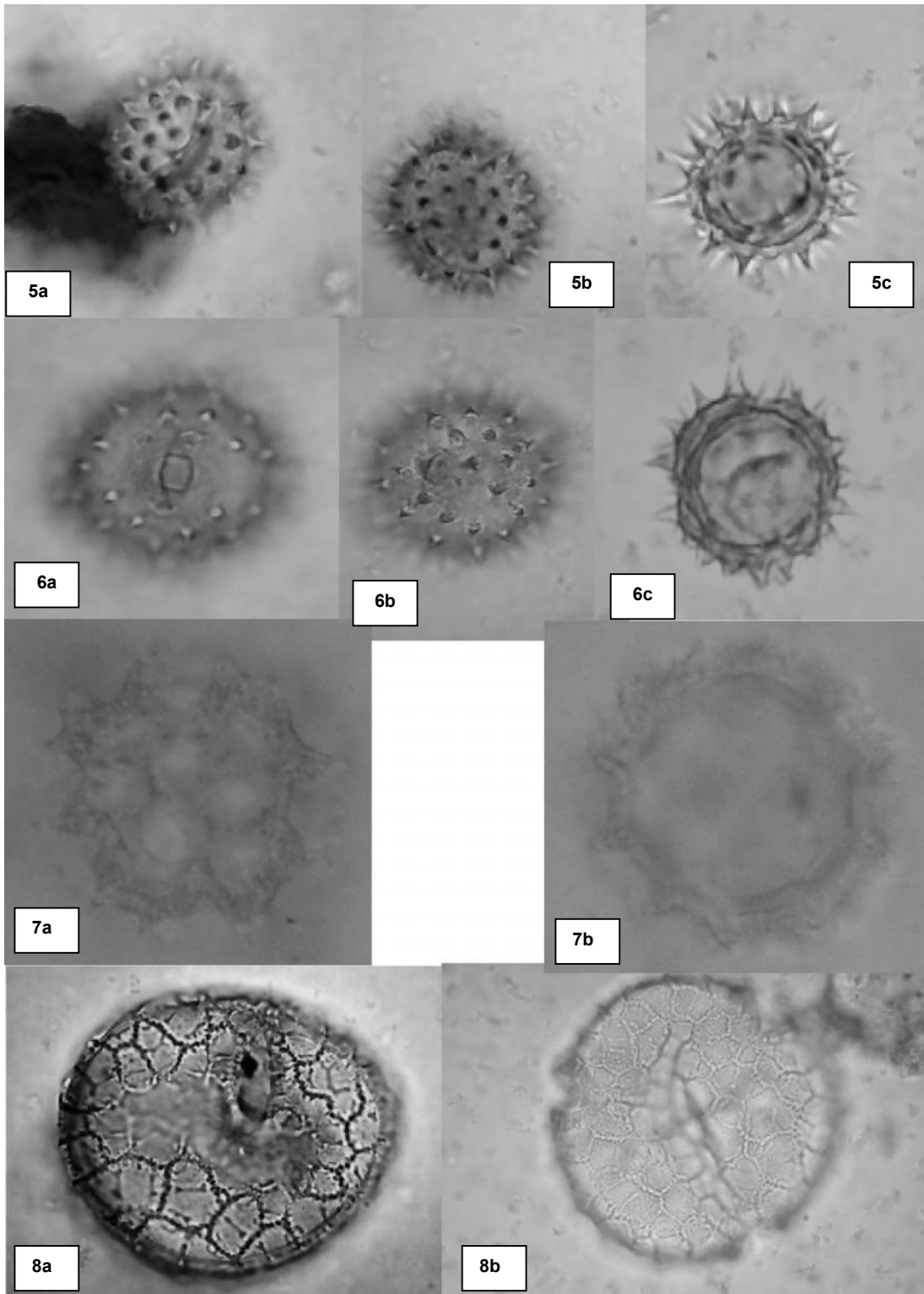
Plate 6 – Monocotyledonae

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

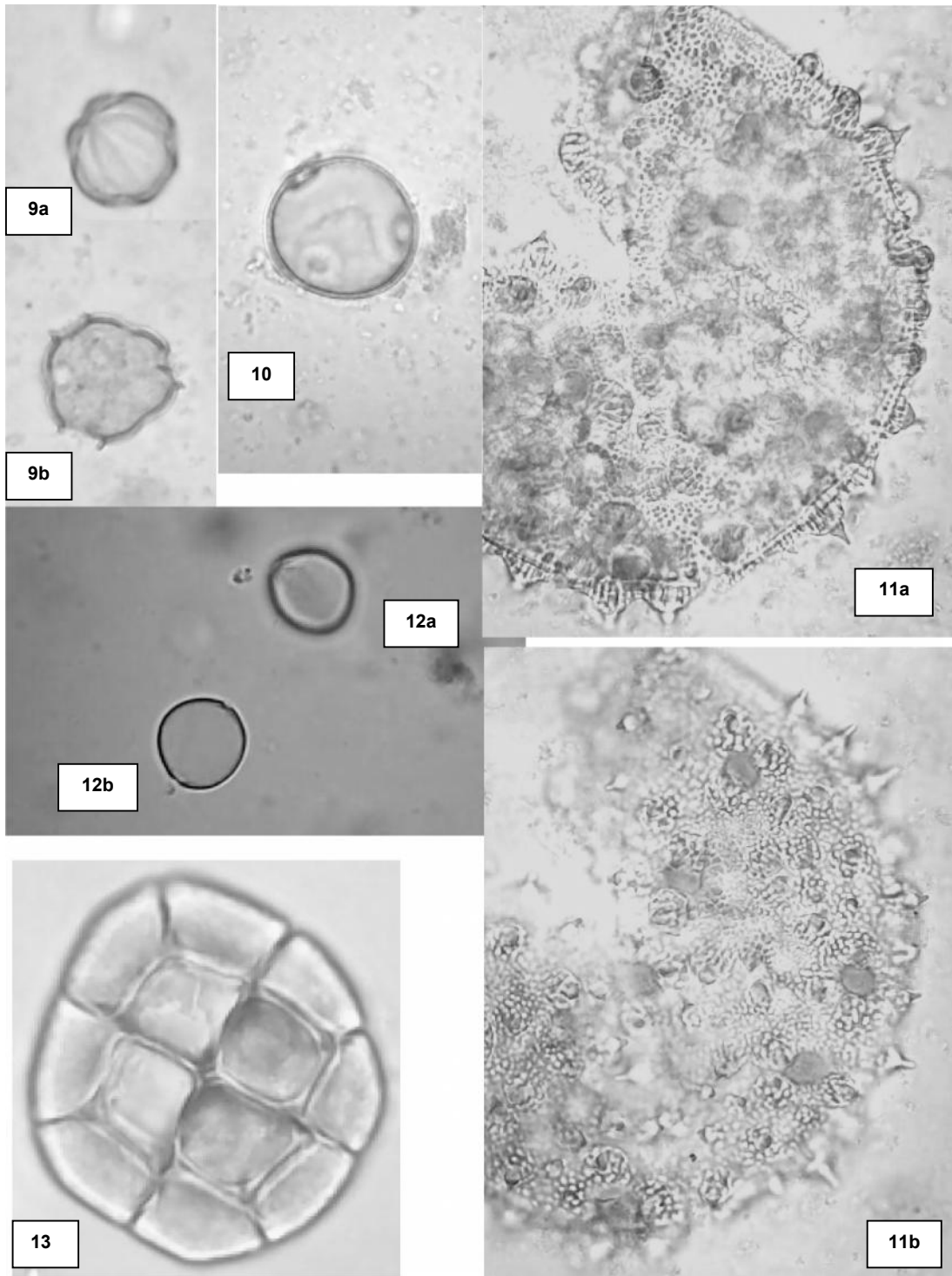
Plate 1 – Dicotyledonae



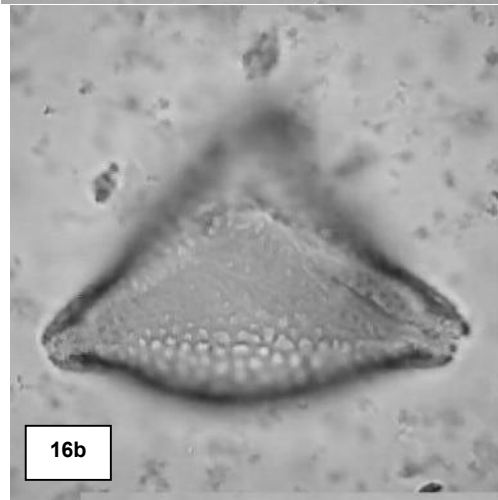
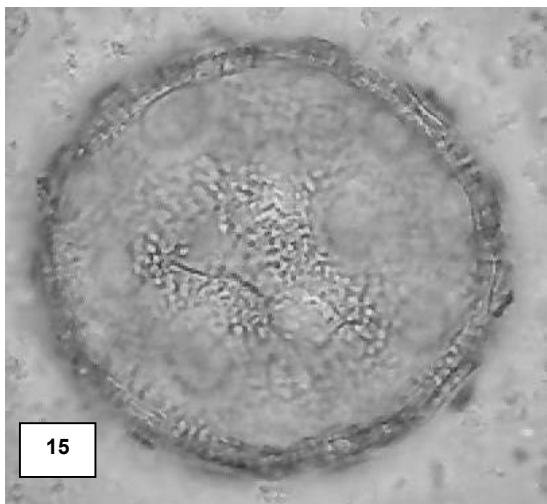
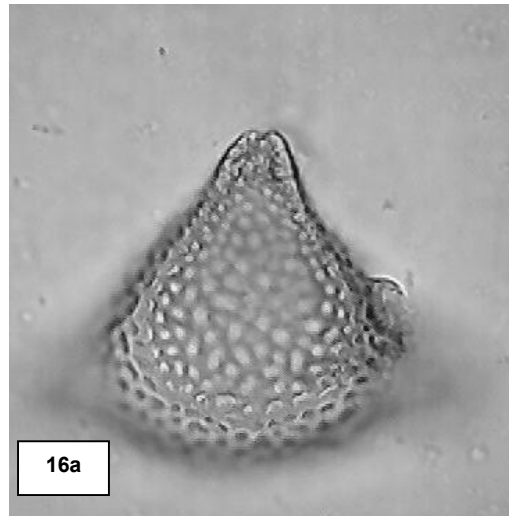
Source: Koudegnan and *al.*; 2010



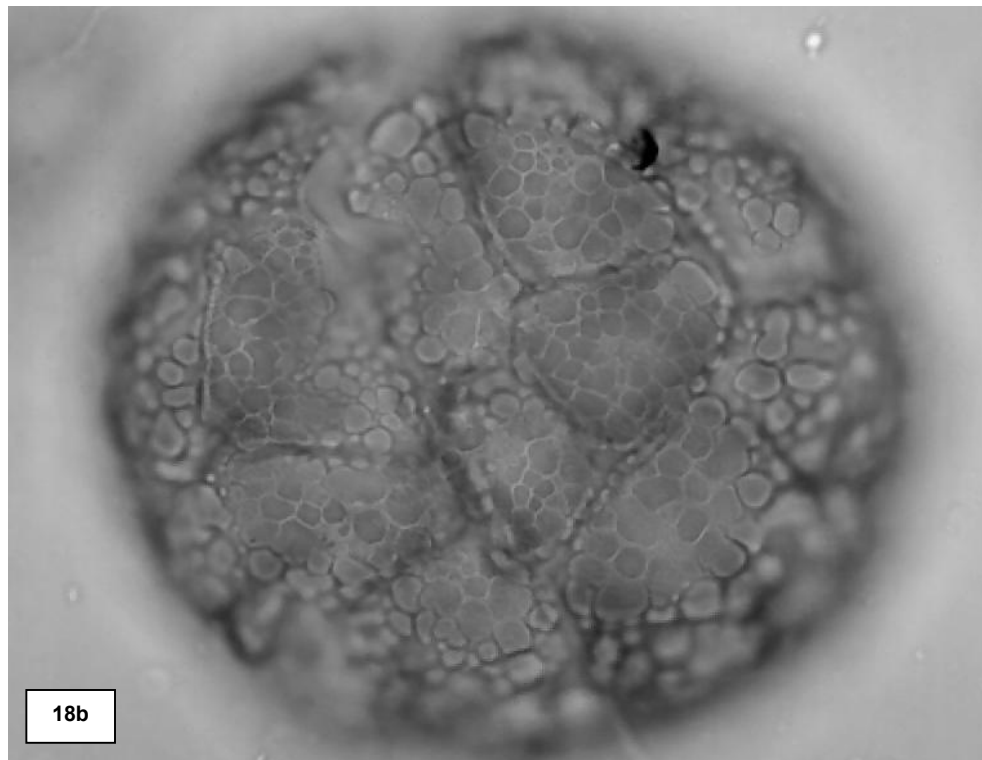
Source: Koudegnan and *al.*; 2010



Source: Koudegnan and *al.*; 2010

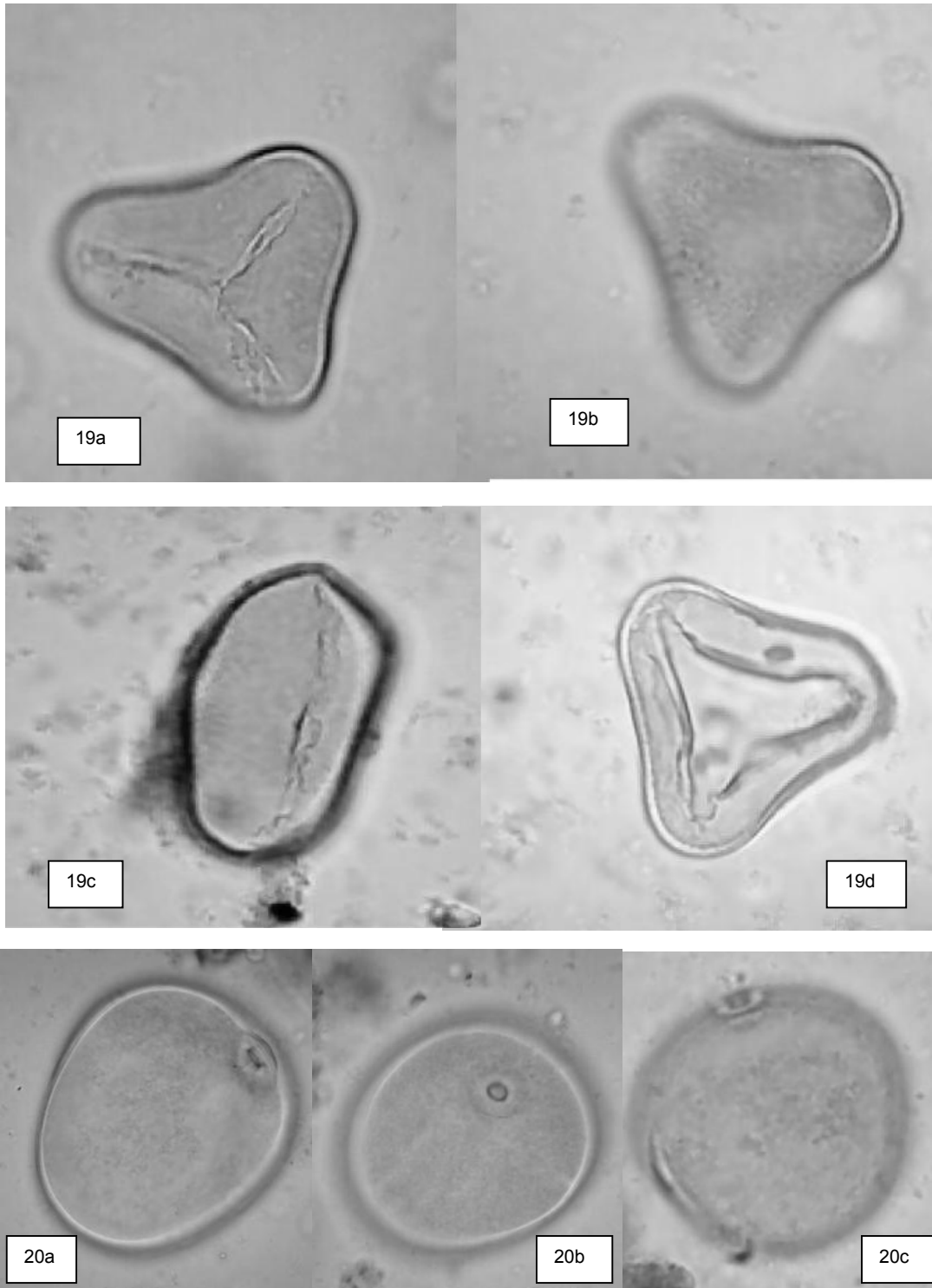


Source: Koudegnan and *al.*; 2010



Source: Koudegnan and *al.*; 2010

Plate 6 – Monocotyledonae



Source: Koudegnan and *al.*; 2010

4. CONCLUSION

The pollinic analysis of honey's samples harvested has help 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 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.

ACKNOWLEDGEMENTS

This study had been possible favour in support of a few persons as:

- Pr Isabelle Adolé GLITHO, Dean of the Faculty of sciences (University of Lomé), who had accepted to register me in his department;
- Pr Thérèse EDORH, Responsible for Palynology, Algology and Paleoecology Laboratory (University of Lomé), First Supervisor of memory, who had help me to identify the pollens present in the different samples of honey harvested for this study;
- Pr Kudzo GUELLY, Second Supervisor of memory, who had helped me to identify the different plants in the localities of harvest;
- Pr Adzo Dzifa KOKUTSE who had accepted to give up me her microscope equipped for photography of pollens.

REFERENCES

1. Dongock N. D., Foko J., Pinta J. Y., Ngouo I. V., Tchoumboue J. & Zango P. Inventaire et identification des plantes melliferes de la zone soudano-guineenne d'altitude de l'ouest cameroun. *Tropicultura*. 2004 ; 22 (3) : 139-145.
2. Maley J. Atlas des pollens du Tchad. *Bull. Jard. Bot. Nat. Belgique*. 1970 ; 40 (1): 29-48.
3. Ybert J-P. Atlas de pollens de Côte d'Ivoire. Orstom, Initiations-Documentations Techniques, 40. Orstom. Paris. 1979 ; 40 p.
4. Bonnefille R. Riollot G. Pollens des savanes d'Afrique orientale. 1980 ; 254 p.
5. Ambougou A. V, Atlas des pollens des zones forestières humides d'Afrique. Thèse de doctorat de l'université paris 6. 1991 ; 34 p.
6. Sowunmi M. A. Pollen of Nigerian plants. II *Woody species*. *Grana*. 1995; 34, 120-141
7. Lobreau-Callen D., Darchen R., & Le Thomas A. Apport de la palynologie à la connaissance des relations abeilles/plantes en savanes arborées du Togo et du Bénin. *Apidologie*. 1986; 17 (4): 279-306.
8. Erdtman G. The acetolysis method. *A revised description*. *Svesk. Bot. Tidskv.* 1960; 54 (4), 561-564.
9. Lezine A. Le Pollen. Collection "Interactions". 2008; 122p.
10. Yédomonhan H. Plantes mellifères et miel du Bénin: cas de la forêt classée de la Lama. Mém. DEA, Univ. Lomé (Togo). 2004 ; 65p.

- 578 11. Yedomonhan H., Tossou M. G., Akoegninou A., Déménou B. B., Traore D. – Diversité
579 des plantes mellifères de la zone soudano-guinéenne : cas de l'arrondissement de Manigri
580 (centre-ouest du Bénin). *Int. J. Biol. Chem. Sci.* 2009 ; 3(2) 355-366.
- 581 12. Sawadogo M. Contribution a l'étude du cycle des miellées et du cycle biologique annuel
582 des colonies d'abeilles *Apis mellifica adansonii* lat. à l'ouest du Burkina Faso. Thèse de
583 doctorat, Université de Ouagadougou. 1993; 152p.
- 584 13. Aloma S. Etude des facteurs de production de miel dans deux zones écologiques
585 différentes au sud du Togo: les sites d'Aképé et d'Agotimé-Nyitoé. Mém. Dea, Univ. Bénin
586 (Togo). 2000 ; 36 p.
- 587 14. Nombé I. Etude des potentialités mellifères de deux zones du Burkina Faso : Garango
588 (province du Boulgou) et Nazinga (province du Nahouri). Thèse de Doctorat Unique, Univ.
589 de Ouagadougou, Ouagadougou (Burkina Faso). 2003; 156 p.
- 590 15. Dongock N. D., Tchoumboue J., Pinta J. Y., Zango P. Caractéristiques polliniques des
591 plantes mellifères de la zone soudano-guinéenne d'altitude de l'ouest Cameroun.
592 *Tropicultura*. 2008; 26 (3) : 150-154.
- 593 16. Fohounfo H. T. Plantes mellifères et composition pollinique des miels de la petite saison
594 des pluies et de la grande saison sèche au sud Bénin. Mémoire du Diplôme d'Ingénieur des
595 Travaux. CPU/UAC/ Cotonou – Bénin. 2002 ; 56 p. + annexes.
- 596 17. Louveau J., Maurizio A., Vorwohl G. Methods of melissopalynology. *Bee World*. 1970; 51
597 (3), 125-138.