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Identification of the chemical composition of extracts from the trunk bark of *Pseudocedrelakotschyi* and *Khaya senegalensis* by Gas Chromatography-Mass Spectrometry coupling

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ABSTRACT*:The use of plants for therapeutic purposes dates back to the appearance of man in the face of the various adversities of nature. The GC-MS technique was used to characterize metabolites from the stem barks of Pseudocedrelakotschyi and Khaya senegalensis. Through the results obtained, we note at the level of the bark of Khaya senegalensis ten major compounds such as: 4-hydroxyionone, 1,2,4,5-tetramethylbenzene, 11-hydroxyresibufogenine; 17-Hydroxy-3,20-dioxopregna-4,9,11-trien-21-yl acetate; pimara-7,15-dien-3-one; 9,19-cycloergost-24-en-3-ol, 4,14-dimethyl-acetate; cholesterol; lycopene; deacetylcinobufagine; 3-hydroxy-17-oxo-androsta-5,7,9-triene. On the other hand at the level of the trunk bark of Pseudocedrelakotschyi; astaxanthin; campesterol; stigmasterol; sitosterol and stigmast-4-en-3-one have been identified as major compounds. The richness in metabolites of these two plants could explain their use in traditional medicine.*
KEYWORDS: *Pseudocedrelakotschyi,Khaya senegalensis, Soxhlet, Extract, Chromatography, Metabolites.*

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I. INTRODUCTION

The global market for herbal medicines is continuously expanding, with an annual global value estimated at around US$800 million (OOAS, 2013). In recent years, human knowledge of medicinal plants has constantly expanded and deepened from one civilization to another (Kamari*et al*., 2009). Plants have a great importance onboth humans and animals well-being (Bako et al., 2005). They are used in medicine, food, cosmetics and energy. Plants have shown some therapeutic efficacy in the treatment of many pathologies against which modern medicine has remained powerless. *Pseudocedrelakotschyi* (syn: Cedrelakotschyi) and Khaya senegalensis (Swietenia senegalensis), of the family Meliaceae are widely distributed in Africa (Kerharo, 1974).The Meliaceae are large trees with bark that is often fragrant, greyish, dark and scaly and sometimes low in latex (kerharo and Adam, 1974; Shahina and Saranna, 1989). These leaves are alternate, usually pinnately compound, sometimes trifoliate, unifoliate or simple leaves without stipules (Shahina andSaranna, 1989). Their different organs are heavily exploited in traditional medicine. They are used to treat deworming, malaria, fever, jaundice, colic, scabies, leprosy, wound dermatitis, anemia diarrhea and migraine, gastrointestinal diseases, rheumatism, dysentery, epilepsy...(Adjanohoun*et al*., 1986; Olayinka et al., 1992; Traoré, 1999; Doumbia, 1994; Togola, 2002; Georgewill, 2008;Akande and Hayashi, 1998; Tapsoba and Deschamps, 2006). Previous work has proven that the different organs of *P. kotschyi* and *K. senegalensis* possess antidysenteric, antiulcer, analgesic, anti-inflammatory, antidiabetic, antimalarial, antibacterial and antidiarrheal activities (Asase*et al*., 2005; Kassim*et al*, 2009; Anuka*et al*., 1999; Musa *et al*., 2008; Georgewill, 2008; Sablassou, 1996).In the literature, very few works have addressed the characterization of extracts of *Pseudocedrelakotschyi* and *Khaya senegalensis*. In view of the use of these two plants in traditional medicine and their biological activities, it becomes necessary to deepen the characterization of their metabolites.

II. MATERIAL AND METHODS

- Plant collection

*Pseudocedrelakotschyi*and*Khaya senegalensis* plants materials were collected from Abomey-calavi and Dassa (Benin)

- Processing of plant samples

The samples of *Pseudocedrelakotschyi*and *Khaya senegalensis*, once harvested, were dried in the laboratory

until their plant mass stabilized before being made powder

- Extract preparation

One gram of powder from the bark of each plant (*Pseudocedrelakotschyi* and *Khaya senegalensis*) was introduced into the cartridge of the Soxhlet extractor. 50 mL of dichloromethane were poured into the flask topped with the Soxhlet extractor system. The whole was heated at 40°C for 12 hours and the dichloromethane extract was recovered.

Characterization of the compounds of the trunk barks *Pseudocedrelakotschyi* and*Khaya senegalensis*:-

The structural determination of molecules of dichloromethane extract of trunk bark of *Pseudocedrelakotschyi* and*Khaya senegalensis* was made by varian GC/MS type 1200 T, in positive electron impact mode (ionization energy: 70 eV), equipped with a single injector and a VF-5MS type fused silica capillary column (25 m long; 0.25 mm external diameter and 0.25 µm internal diameter). The carrier gas used is helium with a flow rate of 1mL/min which is constant throughout theanalysis. The temperature of the injector is 40°C that of the source is 281°C and 298.6°C at the level of the detector and the transfer line. The oven temperature program is defined by an isothermal temperature of 40°C for 5 min with a ramp of 5°C/min up to 310°C, followed by a second isotherm at 310°C for 1min. The quantity of injected extract is 1µL. The injection was made in splitless mode. The mass spectrometry data wereablished in scan mode whose m/z ratio varies between 50-800 amu. Theidentification ofthe compounds present in the extracts was made by comparing the spectra obtained with the information provided by the NIST database (Eswaran*et al*., 2012; Bojaxa*et al*., 2012; Apostolides*et al*., 2013).

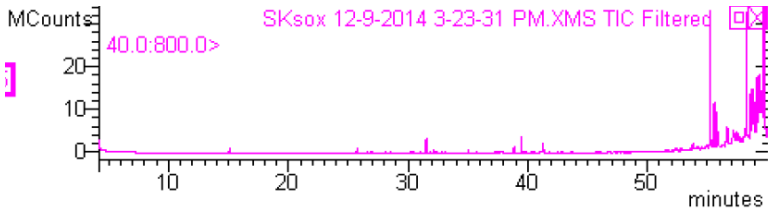
III. RESULTS AND DISCUSSION

Gas chromatography-mass spectrometry analysis of dichloromethane constituents of trunk barks of *K. senegalensis* and *P. kotschyi* led to the identification of several metabolites.

Metabolites from stem bark of*K. senegalensis* and *P. kotschyi*

- **Metabolites from the trunk bark of *K. senegalensis***

The compounds characterized in the dichloromethane extract of *Khaya senegalensis* are indicated by the chromatogram in figure 1.



FigureNo.1: Chromatographic profile of dichloromethane extract of trunk bark of*K. senegalensis*

In the dichloromethane extract of the trunk bark of *Khaya senegalensis*, 10 major compounds such as 4-hydroxyionone, 1,2,4,5-tetramethylbenzene, 11-hydroxyresibufogenine; 17-Hydroxy-3,20-dioxopregna-4,9,11-trien-21-yl acetate; pimara-7,15-dien-3-one; 9,19-cycloergost-24-en-3-ol, 4,14-dimethyl-acetate; cholesterol; lycopene; deacetylcinobufagine; 3-Hydroxy-17-oxo-androsta-5,7,9-triene have been identified with forty-six minority compounds. The richness and diversity in secondary metabolites of the trunk bark of *K. senegalensis* could justify its use in traditional medicine.An extract of the bark of *Khaya senegalensis* is commonly used in African traditional medicine for pain and inflammation. *Khaya senegalensis* bark extract was hypothesized to contain inhibitors of the cyclooxygenase-2 (COX-2) gene and to be useful in the prevention and treatment of colorectal cancer.

TableNo.1: Metabolites from the trunk bark of*K. senegalensis*

RT (min)	Metabolites
31.524	4-hydroxyionone
39.461	1,2, 4, 5- tetramethylbenzene
55.229	11-hydroxyresibufogenine
55.571	17-Hydroxy-3,20-dioxopregna-4, 9,11-trien-21-yl acetate
55.776	Pimara-7,15-dien-3-one
58.261	9,19-cycloergost-24-en-3-ol, 4,14-dimethyl-acetate
58.641	Cholesterone
58.823	Lycopene
59.178	Deacetylcinobufagine

59.347	**3-hydroxy-17-oxo-androsta-5, 7,9-triene**
15.076	2,7,7-trimethylbicyclo[2,2,1]hept-2-ene
25.793	Caryophyllene
26.953	4-methyl-1, 2, 3, 4, 4, 5, 6, 7-octahydro-2-naphthalenol
28.129	Cadinene
28.560	1,4-dimethyl-3-tetrahydroacetophenone
29.508	3-methylene-1, 5, 5-trimethylcyclohexene
30.270	1,4-dimethyl-3-tetrahydroacetophenone
32.186	1, 2, 3, 4-tetrahydronaphthalen-1,5-diol
32.397	5-isopropylidene-4,6-dimethylnona-3,6,8-trien-2-ol
33.074	2-octenoic acid, 4-isopropylidene-7-methyl-6-methylene-methyl ester
35.062	Endothaldimethyl ester
37.236	3-methyl-5-phenylhex-1-en-4-ol
37.381	Ambrettolide
37.711	Guaiene
37.846	1, 2, 3,4-tetrahydroisoquinoline
38.848	Tricyclo[4,2,2,0]deca-7-ene
40.310	Sericealactone
41.080	7-methyl-1,4,5,6,7,7hexahydro hind-2-en-2-one
41.182	2, 2-dimethyl-3-vinylbicyclo [2, 2,1] heptane
41.285	Cuminal
41.845	2-[2-methylpropenyl] cyclohexanone
42.513	P-menth-4-en-3-one
43.654	Norfenefrin
45.673	Diphenyl-3,3'-dimethyl-2,2'-ditertiobutyl-1,1'-dihydroxymethane
45.878	Neonicotine
46.081	(Z)-9-tetradecenal
46.749	Methylretinoate
46.842	2-Acetyl-5,8-dihydroxy-3-methoxynaphthoquinone
52.042	retinal
52.397	Ursodiole
52.786	3-ethyl-3-hydroxyandrostan-17-one
53.623	3-hydroxy-17-oxo-androsta-5,7,9-triene
53.784	Methylretinoate
54.887	Ergosterylacetate
55.057	4,6-cholestadien-3-ol
55.913	3-ethyl-3-hydroxyandrostan-17-one
56.626	2-(7-hydroxymethyl-3,11-dimethyldodeca-2, 6,10-trienyl)-[1,4] benzoquinone
56.737	Resibufogenin
57.194	11-hydroxyresibufogenin
57.583	17-hydroxy-3,20-dioxopregna-1, 4, 9-trien-21-yl acetate
57.761	Carotene
58.057	lupeylacetate
58.184	Sitosterol
58.924	Stigmasta-4, 22-dien-3-one

Legend.RT: Retention Time

- **Metabolites from the trunk bark of*P. kotschyi***

Figure 2 shows the chromatographic profile of the dichloromethane extract from the trunk bark of *P. kotschyi*.

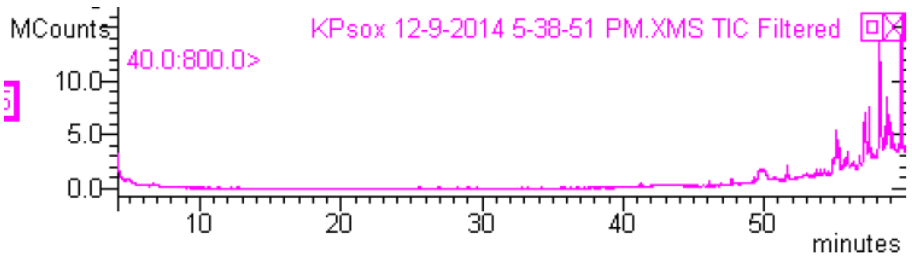


Figure No.2: Chromatographic profile of dichloromethane extract of stem bark of*P. kotschyi*

The compounds identified in dichloromethane extract of trunk bark of*P. kotschyi* are listed in Table 2.Sixteen (16) compounds were identified in dichloromethane extract of trunk bark of*P. kotschyi* with astaxanthin, campesterol,stigmasterol, sitosterol and stigmast-4-yn-3-one the majority compounds. The richness and diversity in secondary metabolites of trunk bark of *P. kotschyi* could justify its use the traditional treatment of various diseases. For example, cholesta-4, 6-dien-3-ol has interesting antimicrobial activity (Yong-Zhe*et al.*, 2018).

Table No.2: Chemical composition of dichloromethane extract from trunkbark of*P. kotschyi*

RT (min)	Metabolites
55.094	**Astaxanthin**
57.109	**Campesterol**
57.430	**Stigmasterol**
58.184	**Sitosterol**
59.732	**Stigmast-4-in-3-one**
51.639	3-[3-Methoxy-4-hydroxyphenyl]-2-oxopropanoic acid
53.656	Ursodiol
53.943	Cholesta-4,6-dien-3-ol
54.222	Ergost-5-en-3-ol acetate
54.578	3-ethyl-3-hydroxyandrostan-17-one
55.890	Vitamin E
58.285	Cyclolanost-24-en-3-ol acetate
58.641	Cholest-5-en-3-one
58.945	Cholesta-4,22-dien-3-one
59.114	Ursodeoxycholicacid

Legend. RT: Retention Time

IV. CONCLUSION

The use ofherbal medicine is common inAfricato treat various ailments and diseases. This research work has made it possible to identify the metabolites of the bark of *Khaya senegalensis* and *Pseudocedrelakotschyi*, plants that are in high demand in traditional medicine in Benin to treat several diseases. From the results obtained, the trunk barksof *Khaya senegalensis* and *Pseudocedrelakotschyi* are rich and diversified in secondary metabolites. The main metabolites of the bark of*Khaya senegalensis* are 4-hydroxyionone, 1,2,4,5-tetramethylbenzene, 11-hydroxyresibufogenin, 17-hydroxy-3,20-dioxopregna-4, 9,11-trièn-21- yl acetate, pimara-7,15-dien-3-one; 9,19-cycloergost-24-en-3-ol, 4,14-dimethyl-acetate; cholesterol, lycopene, deacetylcinobufagine, 3-hydroxy-17-oxo-androsta-5,7,9-triene. At the level of the trunk bark of *Pseudocedrelakotschyi*, astaxanthin, campesterol, stigmasterol, sitosterol and stigmast-4-yn-3-one are the main metabolites. The strong use of Khaya *senegalensis* and *Pseudocedrelakotschyi* in traditional medicine could be explained by their richness and diversification in metabolites.

- **Conflicts of Interest**

The authors declare that they have no conflicts of interest

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