ISSN: 0975-833X

*Available online at http://www.journalcra.com*

*International Journal of Current Research*
*Vol. 11, Issue, 10, pp.7728-7733, October, 2019*

**DOI: https://doi.org/10.24941/ijcr.36903.10.2019**

**INTERNATIONAL JOURNAL OF CURRENT RESEARCH**

## RESEARCH ARTICLE

# BIOLOGICAL ACTIVITIES AND PHYTOCHEMICAL COMPOSITION OF THE METHANOL EXTRACT OF *PAULLINIA PINNATA LINN* STEMS FROM CÔTE D'IVOIRE

**Ouattara L.H.[1,2,*], Kabran G.R.M.[2], Konan K.M.[2,4], Koffi K.L.[3], Koffi K.E.[3] Mamyrbekova-Bekro J.A.[2] and Bekro Y.A.[2]**

[1]Université Péléforo Gon Coulibaly, BP 1328 Korhogo, Côte d'Ivoire
[2]Laboratoire de Chimie Bio-Organique et de Substances Naturelles (www.lablcbosn.com), UFR-SFA Université Nangui Abrogoua, 02 BP 801 Abidjan 02
[3]Unité de Gestion des Ressources Animales (UGRA), Institut Pasteur de Côte d'Ivoire, 01 BP 490 Abidjan 01, Côte d'Ivoire
[4]Université du Québec à Trois-Rivières, 3351 Boulevard des Forges, Trois-Rivières, QC G8Z 4M3, Canada

**ARTICLE INFO**

*Article History:*
Received 24th July, 2019
Received in revised form
19th August, 2019
Accepted 15th September, 2019
Published online 30th October, 2019

*Key Words:*

*Paullinia Pinnata*,
Methanol, Biology,
Phytochemical Composition, GC-MS.

**ABSTRACT**

The phytochemical composition of the methanol extract of *Paullinia pinnata* stems harvested in Dabakala (Côte d'Ivoire) was determined by GC-MS. The results revealed the presence of carboxylic acids with an abundance of gallic, vanillic, 2-methyl-2-hydroxypropanoic and palmitic acids. Biological tests have shown that *P. pinnata* stems not only have significant hemolytic and analgesic activities, but also have relatively low toxicity. Therefore, it can be concluded that the biological potential of *P. pinnata* stems is related to its phytochemical composition.



***Citation: Ouattara L.H., Kabran G.R.M., Konan K.M., Koffi K.L., Koffi K.E., Mamyrbekova-Bekro J.A. and Bekro Y.A.* 2019.** "Biological activities and phytochemical composition of the methanol extract of Paullinia pinnata Linn. stems from Côte d'Ivoire", *International Journal of Current Research*, 11, (10), 7728-7733.

## INTRODUCTION

*Paullinia pinnata* Linn. (Sapindaceae) is a liana 6 m long, widespread in secondary forest areas under reforestation, along streams and in savannas (Adjanohoun et Aké-Assi, 1979). In Côte d'Ivoire, *Paullinia pinnata* Linn. is known under various vernacular names: *ahè-biébun* in Akyé, *trondi* in Baoulé, *mlannomon* in Malinké and *gbessagbébroh* in Bété (Aké-Assi, 2011). In traditional African medicine, it is used to treat malaria, erectile dysfunctions, bacterial infections (Lunga *et al.*, 2014), epilepsy (Maiha et *al.*, 2009), hemorrhoids (Ouattara *et al.*, 2016) and facilitates deliveries (Aké-Assi, 2011). Several studies indicate that *P. pinnata* stems, leaves and roots exhibit antioxidant activities (Zamblé *et al.*, 2006), analgesic, anti-inflammatory (Ior *et al.*, 2011), antibacterial (Lunga *et al.*, 2014) and anti-cancer activities (N'guessan *et al.*, 2011), which are believed to be due to the co-presence of steroids, terpenoids, saponins, flavonoids, coumarins, tannins, phenolic acids and alkaloids (Ouattara *et al.*, 2016, Ior *et al.* 2011, Abourashed *et al.*, 1999). In addition, quantification of polyphenols, tannins and total flavonoids of *P. pinnata* stems showed rates of 5231.53 µgEAG/g SM, 52.25 µgECT/mg and 5.23 % respectively (Ouattara *et al.*, 2016).The purpose of this work was to evaluate the analgesic, hemolytic and toxicological activities of the methanol extract of *P. pinnata stems*, and to determine its chemical composition using the GC-MS (Gas Chromatography coupled with Mass Spectrometry).

## MATERIALS

**Plant material:** The stems of *Paullinia pinnata* were harvested in Dabakala (8° 21' 48" North, 4° 25' 43" West), a city located in central Côte d'Ivoire, in the region of Hambol (Department of Dabakala ). After authentication at the herbarium of the National Center of Floristic (Centre National de Floristique (CNF)) of Abidjan (5° 20' 11" North, 4° 01" 36" West), the stems were cleaned and dried first away from the sunlight for 2 days, then under air conditioning (18 °C) for

***Corresponding author:* Ouattara L.H.,**
Université Péléforo Gon Coulibaly, BP 1328 Korhogo, Côte d'Ivoire.

7 days and finally stored in an oven (45 °C) for 3 days. They were subsequently transformed into powder using an electric grinder (RETSCH, type SM 100).

**Animals:** Healthy Swiss albino (*Mus musculus*) rats of both sex weighing 22-32 g were used in this study. All animals were obtained from Animal house of Institute Pasteur, Abidjan, Côte d'Ivoire. The mice were acclimated to the experimental environment in well-ventilated clean cages for one week prior to experimentation. They had free access to water and standard pelletized laboratory animal diet ad libitum. This study was approved by an ethics committee which gave its consent in absolute accordance with the recommendations of the International Association for the study of Pain.

## METHODS

### Biological analysis

**Preparation of the extract:** The plant extract to be tested was prepared according to Zirihi *et al.*, 2003. 50 g of powder were macerated in 375 ml of 80 % MeOH (3 x 24 h), at room temperature with constant stirring. After filtration under vacuum macerated were collected and then concentrated with a rotary evaporator (BÜCHI Waterbath B-480). The concentrate was dried in an oven at 50 °C for 48 h, then stored in a refrigerator (4 °C) in a hermetically sealed glass jar.

**Analgesic test:** The analgesic test of the methanol extract of *Paullinia pinnata* stems was performed according to Koster *et al.*, 1959 ; Chatter *et al.*, 2011. Three (3) groups of four (4) mice were constituted. Control group was treated with 0.1 ml of physiological saline. Positive control was treated with 0.1 ml of paracetamol (reference analgesic) at 200 mg/kg body weight (BW) and last group by 0.1 ml plant extract. Half an hour after administration of the extract, the mice were given 0.1 ml of acetic acid ($CH_3COOH$) at 1 %by intraperitoneal injection. The number of abdominal contortions was counted in each mouse for 30 min. The analgesic effect of the extract was evaluated by determining the percentage inhibition of contortions according to the following formula:

$$\% \text{ inhibition } = \frac{\text{Mtb} - \text{Mext}}{\text{Mtb}} \times 100$$

**Mtb**: average number of contortions of the mice in control group;

**Mext**: average number of contortions of the mice treated with the plant extract and paracetamol.

**Hemolysis test:** The study of the hemolysis activity was conducted according to the method described by De Freitas *et al.*, (2008). Different concentrations (5, 4, 3, 2 and 1 mg/ml) of the methanol extract of *P. pinnata* and escin (triterpene saponin) from Sigma Aldrich (France) were prepared in a NaCl physiological solution at 0.85 % (w/v). The NaCl solution was the negative control while the escin was used as a positive control. These different solutions were subsequently mixed in EDTA tubes with 25 µl of fresh sheep blood, kept away from food throughout the night. The mixture was homogenized, incubated for 30 min at 37 °C and centrifuged at 3,500 rpm for 10 min. During centrifugation, the erythrocyte lysis may or may not be observed. The absorbance of the supernatants was measured at 540 nm.

The absorbance values in relation to the NaCl concentration made it possible to plot a sigmoidal regression curve given by Boltzmann formula, by means of which the percentage of hemolysis was calculated according to the following formula:

$$\text{Hemolysis (\%)} = \frac{\text{A1} - \text{A2}}{1 + e^{\frac{(\text{S}-\text{H50})}{\text{ds}}}} + \text{A2}$$

**A1:** maximum average of the absorption values;
**A2:** minimum average of the absorption values;
**S:** NaCl concentration;
**H50:** NaCl concentration which causes 50 % of hemolysis;
**ds:** amplitude of sigmoid curve of transition between A1 and A2.

**Toxicity test:** The toxicity study was conducted according to the Test Guidelines 423 of the OECD Guidelines for the Testing of Chemicals **(OECD, 2001)**. 4 batches of mice previously kept away from food for 16 h, were gavageed 1 ml of plant extract by doses varying between 300 and 2,000 mg/kg BW. After administration of the extracts, the animals were observed for 14 days. The lethal dose 50 % ($LD_{50}$) was determined from the mortality rate.

### GC-MS analysis

**Preparation of the extract:** 20 g of powder of *Paullinia pinnata* stems were macerated in 125 ml of 80 % MeOH (3 × 24 h), with constant stirring. After filtration under vacuum, macerated were collected and concentrated using a rotary evaporator. The concentrate was refrigerated (24 h) to rid it of lipophilic compounds. 30 ml of the concentrate were treated with 3 × 20 ml of ethyl acetate. After removing the solvent extract was dried in an oven at 50 °C (48 h) for the preparation of the sample to be analyzed. 100 mg of extract was heated in 50 ml of HCl (2N) for 2 h. After cooling, the hydrolyzate was treated with ethyl acetate (3 × 50 ml).

The organic fractions obtained were dried over anhydrous $MgSO_4$ concentrated under reduced pressure with a rotary evaporator, and then dried under nitrogen. 5 mg of the organic fraction were collected and subjected to silylation by adding 0.5 ml of acetone and 0.2 ml of N-methyl-N-trimethylsilyltrifluoroacetamide (MSTFA). After 12 h incubation at room temperature, the reaction mass was concentrated under reduced pressure and treated with 2 ml of MeOH in a pillbox. 0.1 µl extract obtained was injected into the gas chromatograph coupled to a mass spectrometer (Agilent Technologies trademark 6890 N). The injections (1 µl) were performed in split mode in 45 min with an ionization energy of 70 eV, and helium was used as a carrier gas with a linear velocity of 1 ml/min. The following temperature program has been applied: 50 °C for 5 min, then a gradient at 3 °C/min was achieved from 50 to 250 °C and finally maintained at 250° C at 15 min. The injector temperature was 230 °C, and that of the transfer line 250 °C. The constituents of the samples were identified by comparing their retention times ($t_r$) and their mass spectra with the spectral data of the reference compounds of the apparatus (NIST libraries 05). Each analyze was made in duplicate.

## RESULTS AND DISCUSSION

### Biological analysis

**Analgesic effect:** Figure 1 shows the effect of the methanol extract of *Paullinia pinnata* stems on abdominal contractions. Figure 1 shows that the methanol extract of *P. pinnata* stems significantly inhibited pain (86.57±0.67 %), compared by paracetamol (100±0.00 %). This pharmacological ability of *P. pinnata* stems may be justified by its phytochemical composition (Ouattara *et al.*, 2016). Indeed, several studies have shown the analgesic effects of some secondary metabolites. This is the case of terpenes (Bruneton, 1999), flavonoids (Bent *et* Havsteen, 2002), saponins (Amzal, 2010), alkaloids (Zenk *et* Jueng, 2007) and coumarins (Reddy *et al.*, 2005). These bioactive phytocompounds inhibit the release of mediators (serotonin, acetylcholine, histamine, bradykinin, substance P and prostaglandins ($PGE_{2\alpha}$, $PGF_{2\alpha}$)) involved in the pain process.

**Hemolytic activity:** Figure 2 shows the hemolytic effect of the methanol extract of *P. pinnata* stems. Percentages of hemolysis of *P. pinnata* stems (14.58±0.67 to 24.73±0.35 %) vary at different concentrations. However, they are relatively less significant than those of escin (21.6±0'84 to 41.13±0.97 %) at the same concentrations. However, this observed activity seems to be explained by the presence of certain bioactive phytocomposites, such as saponins and coumarins **(Ouattara** *et al*., 2016) whose hemolysing actions are widely described in various studies (Ladyguina *et al.*, 1983, Park *et al.*, 2005, Amzal, 2010). The manifestation of the hemolytic activity of methanol extract of *P. pinnata* stems could explain its use in traditional treatment of hemorrhoidal pathologies (Ouattara *et al.*, 2016), thus opening a path for the design of a phytoremedia improved.

**Toxicity:** The toxicity test was performed by treating 4 batches of mice with different doses of *P. pinnata* stem extract. The mortality rates were recorded and presented in Table 1. The treated mice showed some clinical signs such as: fast heart rate, breathing difficulties, convulsions. However, the surviving animals return to normal appearance the following days. In addition, only the second dose of 2,000 mg/kg BW caused the death of a single mouse on the second day of observation. As a result, according to the OECD guidelines (OECD, 2001), *P. pinnata* stems would fall into Category 5, with an $LD_{50}$ > 2,000 mg/kg BW. The toxicity scale according to Hodge *et* Sterner (1980) indicates that the *P. pinnata* stems are slightly toxic with regard to toxicity which is comprised between 500 and 5000 mg/kg BW; hence the recurrent use of this plant in popular traditional medicinal practice.

**Phytochemical composition:** The GC-SM spectrum (Figure 3) of methanol extract of *P. pinnata* stems shows presence of at least 20 molecular peaks, corresponding to coexistence of phytocompounds, recorded in 4 chemical families: phenolic (11,76 %) and carboxylic acids (35.29 %), carbohydrates (35.29 %), and alcohols (17.65 %) (Table 2). The fragmentation analysis revealed the existence of a large peak m/z 73 in almost all mass spectra, reflecting loss of a group of TMS (trimethylsilyl). In addition, the (M-89) fragment has been observed in most compounds containing a carboxylic acid function. The latter shows departure of a TMSO group. These observations were also reported by Charalampos and Michael (Charalampos *et* Michael, 2013).

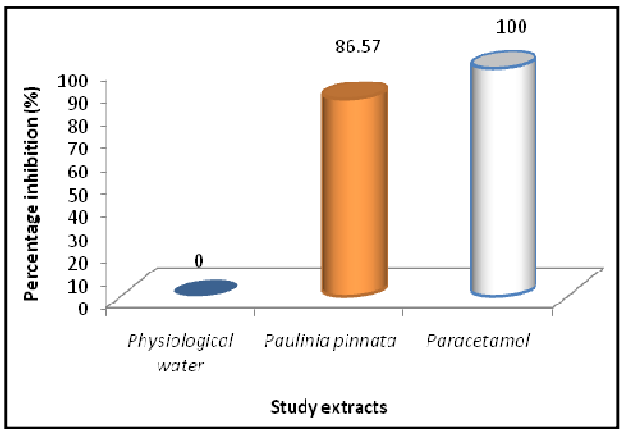


**Figure 1. Percentage inhibition of abdominal contortions indued by methanol extract of *P. pinnata* stems**

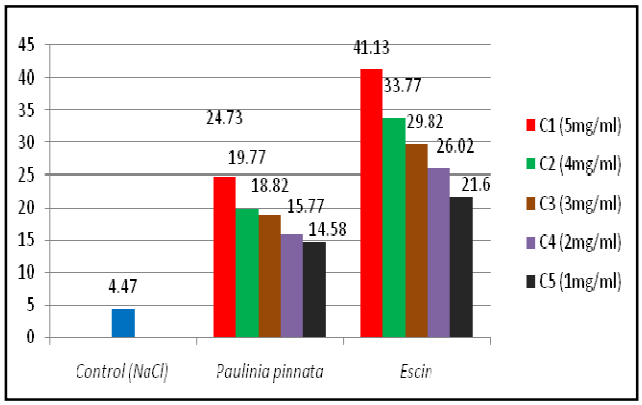


**Figure 2: Hemolysis activity of *P. pinnata* extract at different concentrations**

Thus, presence of phenolic acids was materialized by peaks 13 and 17. Indeed, peak 13 (tr: 15.455 min) with a m/z 312 molecular peak and the following fragments: m/z 297 (M-15) (loss of a methyl); 282 (M-30) (loss of a formaldehyde); 267 (M-45) (loss of $CO_2$ after rearrangement); 223(M-89) ; 193 and 73 indicates presence of vanillic acid. As for peak 17, with a retention time of 17.453 min, it was identified as gallic acid. It was found with a molecular peak m/z 458 and predominant peak at m/z 281 (M-177). In addition, loss of certain compounds such as $CH_3$, TMSO and TMS were observed. The carboxylic acids were visualized by peaks 3, 4, 6, 12, 19 and 20. With concentrations of 8,18 %, 2-methyl-2-hydroxy-propanoic (Peak 3) and palmitic (Peak 19) acids are the most abundant.2-methyl-2-hydroxypropanoic acid revealed itself with m/z 248 molecular peak and following fragments: m/z 173 (M-7CH3); 115 (M-TMS-4CH3) and 73.As for peak 4, with a molecular peak m/z 260 and the following fragments: (m/z 245 (M-$CH_3$); m/z 171 (M-TMSO); m/z 98 (M-TMSO-TMS)) corresponds to fumaric acid. For palmitic acid, the recorded molecular peak is m/z 328 and resulting fragments are: m/z 313 (M-$CH_3$); 299 (M-$C_2H_5$); 285 (M-$C_3H_7$); 269 (M-$C_2H_5$ -2$CH_3$); 234 (M-TMSO-$CH_3$); 201 (M-$C_{10}H_{21}$); 73 and 43 (M-$C_{10}H_{24}O_2$-TMS). Peak 6, with m/z 213 (M-2$CH_3$) fragment and peak 12 with the following fragments: m/z 247 (M-$CH_3$); 233 (M-$C_2H_5$); 217 (M-3$CH_3$) and 189 (M-TMS) correspond respectively to octanedioic and 3-hydroxy-valeric acids whose molecular peaks are respectively 318 and 262. Then, carbohydrates are represented by peaks 11, 14, 15, 16, 18 and 21. D-ribopyranose with a molecular peak of m/z 438 and following fragments:

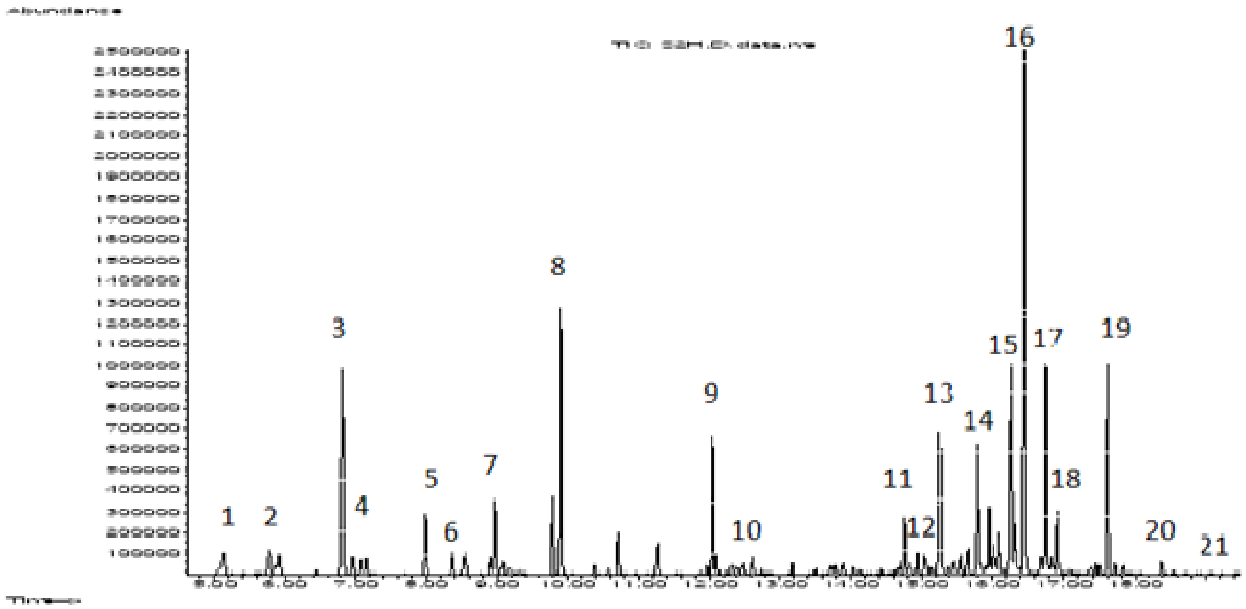


**Figure 3. GC-SM profile of methanol extract of Paullinia pinnata stems**

**Table 1. Mortality rate of mice by *P. pinnata* methanol extract**

| Batch of mice | | Batch 1 | Batch 2 | Batch 3 | Batch 4 |
|---|---|---|---|---|---|
| Dose administered (mg/kg BW) | | 300 | 300 | 2,000 | 2,000 |
| Number of deaths per day | Day 1 | 0 | 0 | 0 | 0 |
| | Day 2 | 0 | 0 | 0 | 1 |
| | Day 3 | 0 | 0 | 0 | 0 |
| | Day 4 | 0 | 0 | 0 | 0 |
| | Day 5 | 0 | 0 | 0 | 0 |
| | Day 6 | 0 | 0 | 0 | 0 |
| | Day 7 | 0 | 0 | 0 | 0 |
| | Day 8 | 0 | 0 | 0 | 0 |
| | Day 9 | 0 | 0 | 0 | 0 |
| | Day 10 | 0 | 0 | 0 | 0 |
| | Day 11 | 0 | 0 | 0 | 0 |
| | Day 12 | 0 | 0 | 0 | 0 |
| | Day 13 | 0 | 0 | 0 | 0 |
| | Day 14 | 0 | 0 | 0 | 0 |
| Mortality | | 0 | 0 | 0 | 1 |
| Mortality rate | | 0 | 0 | 0 | 33.33 |

**Table 2. Silyl phytocompounds identified by GC-SM in *Paullinia pinnata* extract of stems**

| Peak | $T_r$ (min) | Molar mass (g/mol) | Empirical formula | Percentage (%) | Fragmentation | Identified compound |
|---|---|---|---|---|---|---|
| 1 | 5.130 | / | / | 0.91 | 256; 228; 200; 184; 169; 156 | NI |
| 2 | 6.463 | / | / | 1.06 | 221; 208; 187; 169; 159; 146 | NI |
| 3 | 6.811 | 248 | $C_{10}H_{24}O_3Si_2$ | 8.18 | 173; 115; 73 | 2-methyl-2-hydroxypropanoic acid |
| 4 | 7.148 | 260 | $C_{10}H_{20}O_4Si_2$ | 0.61 | 245;171;98; 73 | Fumaric acid |
| 5 | 8.367 | / | / | 2.42 | 251;173;145; 73 | NI |
| 6 | 8.549 | 318 | $C_{14}H_{30}O_4Si_2$ | 0.91 | 245; 229 | Octanedioic acid |
| 7 | 8.959 | 228 | $C_{13}H_{28}OSi$ | 3.03 | 213;170;155; 73 | 1-cyclohexyl-2-méthylpropan-1-ol |
| 8 | 9.893 | 202 | $C_{11}H_{26}OSi$ | 11.82 | 173;131;99; 73 | Octan-1-ol |
| 9 | 12.046 | 260 | $C_{12}H_{28}O_2Si_2$ | 5.45 | 245;171; 73 | 4-methylpent-1-ene-2,4-diol |
| 10 | 12.331 | / | / | 0.61 | 287;185; 73 | NI |
| 11 | 14.713 | 438 | $C_{17}H_{42}O_5Si_4$ | 2.42 | 304; 189;73 | D-ribopyranose |
| 12 | 15.175 | 262 | $C_{11}H_{26}O_3Si_2$ | 0.91 | 247;233;217;189; 73 | 3-hydroxy-valeric acid |
| 13 | 15.455 | 312 | $C_{14}H_{24}O_4Si_2$ | 6.82 | 297;282;267;223;193; 73 | Vanillic acid |
| 14 | 15.938 | 554 | $C_{21}H_{50}O_7Si_5$ | 5.15 | 450;360;305;217; 73 | D-glucuronic acid |
| 15 | 16.264 | 540 | $C_{21}H_{52}O_6Si_5$ | 9.70 | 435;363;291;169; 73 | β-D-galactofuranose |
| 16 | 16.327 | 452 | $C_{18}H_{44}O_5Si_4$ | 20.45 | 392;233;273;243; 73 | 6-desoxy-L-mannose |
| 17 | 17.453 | 458 | $C_{19}H_{38}O_5Si_4$ | 8.18 | 281; 73 | Gallic acid |
| 18 | 17.613 | 540 | $C_{21}H_{52}O_6Si_5$ | 2.42 | 435;361;331; 73 | D-glucose |
| 19 | 18.371 | 328 | $C_{19}H_{40}O_2Si$ | 8.18 | 313;299;285;269;234;201; 73 | Palmitic acid |
| 20 | 19.948 | 354 | $C_{21}H_{42}O_2Si$ | 0.61 | 339;311;295;222; 73 | Oleic acid |
| 21 | 20.680 | 540 | $C_{21}H_{52}O_6Si_5$ | 0.15 | 361;169; 73 | α-D-galactopyranoside |

NI: non identified

(m/z 304 (M-TMS-3$CH_3$); 189(M-TMS-2$CH_3$)) and 6-desoxy-L-mannose (m/z 452, fragments: m/z 392(M-6$CH_3$); 333(M-TMSO-2$CH_3$); m/z 273(M-TMSO-6$CH_3$); 243(M-TMSO-8$CH_3$)) were visualized by peaks 11 and 16, respectively. 6-desoxy-L-mannose or rhamnose is the most prominent among carbohydrates. As for peak 14, with a molecular peak m/z 554 and the following fragments: m/z 450(M-TMS-$CH_3$); 360(M-TMSO-7$CH_3$) and 217 (M-4TMS-3$CH_3$) was identified as D-glucuronic acid. Finally, peaks 15; 18 and 21 of identical molecular peak m/z 540 are respectively β-D-galactofuranose, D-glucose and α-D-galactopyranoside. These three compounds generally have the fragments 361(M-TMSO-6$CH_3$) and 169 (M-4TMSO-$CH_3$) in common. β-D-galactofuranose and α-D-galactopyranoside are two isomers of D-galactose. Finally, peaks 7, 8 and 9 highlight the alcohols.Peak 7, with m/z 228 molecular weight and the followingfragments: m/z 213(M-$CH_3$); 170(M-$CH_3$-$C_3H_7$) and 155(M-TMS) corresponds to 1-cyclohexyl-2-methylpropan-1-ol.As for peak 8, it indicates presence of the major compound, octan-1-ol (11.82 %) with a molecular peak m/z 202 and following fragments: m/z 173(M-$C_2H_5$); 131(M-$C_5H_{11}$) and 99(M-$CH_2$OTMS). Finally, 4-methylpent-1-ene-2,4-diol (peak 9) was visualized with a m/z 260 molecular peak and the following fragments: m/z 245(M-$CH_3$); 171(M-TMSO) and 131: (C($CH_3$)$_2$ OTMS radical). The presence of these phytocompounds in *Paullinia pinnata* stems justifies its numerous pharmacological properties. Indeed, certain compounds such as phenolic acids have beneficial effects on health. This is the case of vanillic acid, which is known to be anti-inflammatory, and anti-tumor (Corder *et al.*, 2001). Similarly, for gallic acid, which is an excellent antioxidant (Gow-Chin *et al.*, 2002). In addition, it has antitumor (Bo *et* Woo, 2010), antibacterial and antiviral properties (Kratz *et al.*, 2008). However, an increased presence of carbohydrate was observed in the study extract, which could cause deleterious effects for the body (Dargent–Pare *et* Levy, 2001).

The existence of these effects would be caused by the glycosidic compounds. Indeed, several heterosidic compounds have been identified and isolated from *P. pinnata* organs. This is the case of diosmetin-7-O-(2''-O-β-D-apiofuranosyl-6''-β-D-glucopyranoside in leaves (Ehab *et al.*, 1999), from 3-O-β-D-glucopyranosyloxy-4-methyl-2 (5H)-furanone, 3-O-β-D-glucopyranosyl(1 → 6) -β-D-glucopyranosylolefinic acid (Lunga *et al.*, 2015) and (3β)-3-O-(β-D-glucopyranosyl-(1''-3')-2'-acetamido-2'-deoxy-β-D-galactopyranosyl)oleanolic acid (Lunga *et al.*, 2014) in the stem. The acid hydrolysis of the latter has caused lysis of the ester bonds, thus releasing aglycones and sugar units such as β-D-galactopyranoside, D-glucose, etc. In addition, CG-SM analysis of the roots of Ghanaian species revealed co-presence of a dozen fatty acids, majority of which are palmitic (30 %) and oleic (30.8 %) acids (Kofi *et al.*, 2009). These results are in accord with those of the Ivorian species, with regard to palmitic acid. On that basis, we can argue that *P. pinnata* is a source of palmitic acid. Thus, its presence could confer on the latter many biological properties including hypercholesterolemic effects in addition to energy intake (Guesnet *et al.*, 2005). Finally, the alcohol molecules recorded in *P. pinnata* stems extract are aresing from the various fragmentations and rearrangements observed during the analysis.

**Conclusion**

In order to provide a rational response to the traditional use of *Paullinia pinnata*, biological and chemical investigations have been carried out on the stems of this species, often used in traditional therapy. The investigations set out to evaluate analgesic, hemolytic and toxic potentials of methanol extract of *P. pinnata* stems. It turns out that *P. pinnata* stems showed a significant analgesic effect (86.57±0.67 %) compared to paracetamol (100±0.00 %).The notable hemolytic power of the stems has been noted with rates ranging from 14.58±0.67 to 24.73±0.35%. The $LD_{50}$> 2,000 mg/kg BW is evidence that the *P. pinnata* stems show low toxicity. Phytochemical analysis by CG-SM of methanol extract of *P. pinnata* stems revealed presence of 20 phytocompounds including phenolic and carboxylic acids, carbohydrates and alcohols. Phenolic and carboxylic acids are the most abundant, with an overall content of 47.05 %. A preponderance of gallic (8,18 %), vanillic (6,82 %), 2-methyl-2-hydroxy-propanoic (8,18%) and palmitic (8,18 %) acids is noticed. Biological and phytochemical investigations of methanol extract of *P. pinnata* stems that we carried out are a contribution to valorization of the species of Côte d'Ivoire; and also attest to its use in endogenous and popular traditional medicinal practice against certain pathologies.

## REFERENCES


Abourashed E.A., Toyang N.J., Choinski J., Khan I.A. 1999. Two new flavone glycosides from *Paulliniapinnata*. *Journal of Natural Products*, 62: 1179-1181.

Adjanohoun E., Aké-Assi L. 1979. Contribution au recensement des plantes médicinales de Côte d'Ivoire. Ministère de la recherche scientifique. Centre National de Floristique, Abidjan, Côte d'Ivoire, 359 p.

Amzal H. (2010). Étude de l'activité antioxydante des saponines du tourteau de l'arganier. Rabat : Université Mohammed V Agdal Faculté des Sciences Rabat ; 149 p.

Ake-Assi L. 2011. Abrégé de médecine et de pharmacopée africaine. Quelques plantes employées traditionnellement dans les couvertures des soins de santé primaire. NEI/CEDA, 157 p.

Bent H., Havsteen. 2002. The biochemistry and medical significance of the flavonoids. *Pharmacology & Therapeutics*, 96: 67-202.

Bo R.Y., Woo H.P. 2010. Gallic acid-induced lung cancer cell death is related to glutathiane depletion as well as reactive oxygen is species increase. *Toxicology in vitro,* 24: 1356-1362.

Bruneton J. 1999. Pharmacognosie, Phytochimie, Plantes médicinales. 3ème Edition, Lavoisier, EM Internationales, Tec. & Doc, Paris (France), 1120 p.

Charalampos P., Michael K. 2013. Analysis of naturally occurring phenolic compounds in aromatic plants by RP-HPLC coupled to Diode Array Detector (DAD) and GC/MS after silylation. *Foods*, 2: 90-99.

Chatter R.R., Tarhouni S., Kharrat R. 2011. Criblage de l'effet anti-inflammatoire et analgésique des algues marines de la mer méditerranée.

Corder R., Douthwaite J.A., Lees D.M., Khan N.Q., Viseu Dos Santos A.C., Wood E.G., Carrier M.J. 2001. Endothelin-1 synthesis meduced by red wine. *Nature*, 414: 863-864.

Dargent–Pare C. Levy G., Basdevant A., Laville M., Lerebours E. 2001. Traite de nutrition clinique de l'adulte, *Médecine-Sciences, Flammarion* 723 : 609-12.

De Freitas M.V., Netto R.C.M., Huss J.C.C., Souza T.M.T., Costa J.O., Firmino C.B., Penha-Silva N. 2008. Influence of aqueous crude extracts of medicinal plants on the osmotic

stability of human erythrocytes. *Toxicology in Vitro*, 22(1): 219-224.

Ehab A.A., Ngeh J.T., John C.J., Ikhlas A.K., (1999). Two new flavone glycosides from Paulliniapinnata.*J Nat Prod*. 62: 1179-1181.

Gow-Chin Yin., Pin-Der Duh., Hui-Ling Tsai. 2002. Antioxidant and pro-oxidant properties of ascorbic acid and gallicacide.*Food Chemistry*,79: 307-313.

Guesnet P., Alessandri J.-H., Astorg P., Lavialle M., 2005. Les rôles physiologiques majeurs exercés par les acides gras polyinsaturés (AGPI). *OCL vol 12 N° 5-6 : 333-343.*

Hodge A.C., Sterner J.H., (1980). In études de toxicité : quelques données fondamentales (A.K DONE). *Tempo Medical Afrique*, 7 :18.

Ior L., Uguru M., Olotu P., Ohenu T., Ukpe A. 2011. Evaluation of analgesic and antiinflammatory activities and phytochemical screening of the leaves extract of *Paulliniapinnata*(sapindaceae).*Journal of Chemical and Pharmaceutical Reseach*,3(4): 351-360.

Kofi A., Stephen G., Francis A. 2009. Antibacterial and radical scavenging activity of fatty acids from Paulliniapinnata L. *PharmacognMag*: 5: 119-23.

Koster R., Anderson M., Beer E.J. 1959. Acetic acid for analgesic screening.*Federation Proceedings*, 18: 412-417.

Kratz J.M., Andrighetti-Frohner C.R., Leal P.C., Nunes R.J., Yunes R.A., Trybala E., Bergstrom T., Baradi C.R., Simoes C.M. 2008. Evaluation of anti-HSV-2 activity on gallic acid pentylgallate.*Biological and pharmaceutical*, 31 (5): 7-903.

Ladyguina E., Safronitch L., Otriachenkova V., Bolandina I., Grinkevitch N., Sorokina A., Glizin V.I., Molodojnikova L.M., Mitin Y.S., Samilina I.A. &Ermakova V.A. 1983. Analyse chimique des plantes médicinales. Edition Moska ; VischayaChkola (traduit du russe) : 46-347.

Lunga P.K., Qin Xu-Jie., Yang Xing W., Kuiate J.-R., Du Z.Z., Gatsing D. 2014. Antimicrobial steroidal saponin and oleanane-type triterpenoidsaponins from *Paulliniapinnata*. *Bio Med Central Complementary and Alternative Medicine*, 14: 369.

Lunga P.K., Qin Xu-Jie., Yang Xing W., Kuiate J.-R., Du Z.Z., Gatsing D. 2015. A new antimicrobial and radical-scavenging glycoside from*Paulliniapinnata* var. cameroonensis, Natural Product Research: *Formerly Natural Product Letters*: 1-7.

Maiha B.B., Magaji M.G., Yaro A.H., Hamza A.H., Ahmed S.T., Magaji R.A. 2009. Anticonvulsant studies on *cochlospermumtinctorium* and *Paulliniapinnata* extracts in laboratory animals. Nigeria. *Journal of Pharmaceutical Sciences*, 8 (1): 102-108.

N'guessan A.H.O., Dago D.C.E., Mamyrbekova-Bekro J.A., Bekro Y.A. 2011. Teneurs en composés phénoliques de 10 plantes médicinales Employées dans la tradithérapie de l'hypertension artérielle, une pathologie émergente en Côte d'Ivoire. *Revue de génie industriel,* 6 : 55-61.

OCDE. 2001. Ligne directrice de l'OCDE pour les essais de produits chimiques. 14 p

Ouattara L.H., Kabran M.G.R., Kadja A.B., Tano B.M., Mamyrbekova-Bekro J.A., Bekro Y.A. 2016. Etude phytochmique et activiteanti-oxydante d'extraits de plantes de Côte d'Ivoire utilisées dans le traitement traditionnel des hémorroïdes. *Internationnal Journal of Innovation and Applied Studies*, 15(4): 881-893.

Park J.D., Rhee D.K., Lee Y.H. 2005. Biological activities and chemistry of saponins from *Panax ginseng* C. A. Meyer.*Phytochemistry Reviews*, 4: 159-175.

Reddy N.S., Gumireddy K., Mallireddigari M.R. 2005.Novel coumarin-3-(N-aryl)carboxamides arrest breast cancer cell growth by inhibiting ErbB-2 and ERK1. *Bioorganic and Medicinal Chemestry Letters*, 13: 3141-3147.

Zamblé A., Carpentier M., Kandoussi A., Sahpaz S., Petrault O., Ouk T., Martin-Nizard F. 2006. *Paullinia*pinnata extracts rich in polyphenols promote vascular relaxation via endothelium dependent mechanisms. *Journal Cardiovascular Pharmacology,* 47(4): 599-608.

Zenk M., Jueng M. 2007. Evolution and current status of the phytochemistry of nitrogenous compounds. *Phytochemistry*, 68: 2757-2772.

Zirihi G., Kra A.K.M., Guédé-Guina F. 2003. Évaluation de l'activité antifongique de *Microglossapyrifolia* (Lamarck O. KuntzeAsteraceae) « PYMI » sur la croissance *in-vitro* de *Candida albicans*. *Medecin& Pharmacology African* ; s17. 9/88 : 11-18.

*******