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## Qualitative and quantitative analyses of three antimalarial plants of Ivorian floral

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### Abstract

In order to identify new antimalarial plants, phytochemical study was carried out on three plant matrices belonging to Euphorbiaceae family. Phytochemical screening mentioned the co-presence of several phytochemicals, which are involved in observed antioxidant potential. These results were confirmed by different dosages (polyphenols, flavonoids and total condensed tannins). In view of the relative similarity of the molecular fingerprints of *Alchornea cordifolia* (control plant, recognized as an antimalarial) and the stem bark of *Ricinodendron heudelotii*, we can be affirmed that the latter could be classified in restricted list of antimalarial plants. Therefore, it would be more recommended in malaria access management.

**Keywords:** Antimalarial plants, phytochemical, antioxidant activity, Côte d'Ivoire, *Ricinodendron heudelotii*

### 1. Introduction

Malaria is the world's leading parasitic endemic. It mainly affects countries in the intertropical zone [1]. This pathology is most often transmitted to humans by the bite of a female mosquito of the genus *Anopheles*. It is responsible for the death of a good number of people, especially children under 5 [2]. In Côte d'Ivoire, according to the National Malaria Control Program (PNLP-CI), the number of deaths linked to this disease in 2017 was estimated at 3,222 [3]. Despite the existence of several means of control (impregnated mosquito nets, insecticides, antimalarial drugs, etc.), we are still witnessing a resurgence of this disease. Several factors are at the origin of this situation. One of them is the high cost of antimalarial remedies, and the adverse effects caused by some of them. In addition, we have recently recorded a resistance of parasites to these drugs [4-6]. In this situation, most patients use medicinal plants, particularly those in rural areas [7]. In this framework, investigations carried out with traditional therapists from Agboville Department (Côte d'Ivoire) have led to the inventory a multitude of plants reputed to be antimalarial [8]. Within the present study, we were interested in three plants of Euphorbiaceae family for their accessibility and recurrent use by populations. These are *Alchornea cordifolia*, *Jatropha gossypifolia* and *Ricinodendron heudelotii*. Furthermore, research has revealed the antimalarial properties of *Alchornea cordifolia* leaves from Côte d'Ivoire. Indeed, the results indicated significant activity on the Fc B1 strain from Colombia (chloroquine resistant) of *Plasmodium falciparum* with an IC<sub>50</sub> of around 4.56 µg / ml [9, 10]. Thus, this work aims to provide a rational response to the use of these plants against malaria. For this purpose, a comparative study of antioxidant potential was carried out between *A. cordifolia* (control plant) and the other vegetal matrices in order to identify new antimalarial plants.

### 2. Material and methods

#### 2.1. Selection of plant species

The plant support consists of *Alchornea cordifolia* (leaves), *Jatropha gossypifolia* (leaves) and *Ricinodendron heudelotii* (leaves and stem bark), three plants traditionally used for the care of people who have malaria by the Abbey and Krobou people of Agboville (5°55'41" North, 4°13'01" West) (Côte d'Ivoire) [8]. They were harvested in August 2015 in Grand-Morié (5°59'00" North, 4°08'00" West) (Department of Agboville, Agneby-tiassa Region) and certified at the National Floristic Centre of Félix Houphouët-Boigny University (Abidjan / Côte d'Ivoire), in accordance with existing specimens. The different organs were cleaned, dried in an air-conditioned room at 16°C for 10 days, and then reduced to powder.

## 2.2. Preparation of extracts

15 g of powder were macerated in 20 ml of methanol (MeOH, 80%, 3 × 24 h) at room temperature (25°C), with constant stirring. After filtration under vacuum, the filtrates were combined and then concentrated using a rotary evaporator (Büchi) at 40°C. The aqueous extracts obtained were kept in the refrigerator (4°C) for 24 h to precipitate the lipophilic compounds. After decantation and filtration, the aqueous extracts were separated into two parts. The first was used on the one hand for quantification of total polyphenols and condensed tannins and on the other hand for evaluation of antioxidant power by spectrophotometry. The second part was exhausted successively with different solvents of increasing polarity: hexane (n-C<sub>6</sub>H<sub>14</sub>), chloroform (CHCl<sub>3</sub>), ethyl acetate (AcOEt) and n-butanol (n-BuOH). These were used for phytochemical screening and detection of antioxidant activity by TLC.

### 2.2.1. Qualitative study

#### 2.2.1.1. Phytochemical screening by TLC

Phytochemical screening was carried out by means of TLC, according to the methodologies described in literature [11-13]. Chromatographic plates used are in silica gel 60 F<sub>254</sub>, with a rigid aluminium support. The developers used composed of a mixture of solvent: n-C<sub>6</sub>H<sub>14</sub> / AcOEt in proportions 5: 2 (v / v) for hexanic extracts; CHCl<sub>3</sub> / AcOEt / n-C<sub>6</sub>H<sub>14</sub> 4: 4: 1 (v / v / v) for chloroformic extracts; the combination of AcOEt / MeOH / H<sub>2</sub>O / n-C<sub>6</sub>H<sub>14</sub> in the ratio 4: 1.5: 0.5: 1.5 (v / v / v / v) and 4: 4: 0.5: 1 (v / v / v / v) for the ethyl acetate and n-butanolic extracts respectively.

#### 2.2.1.2. Antioxidant power screening

The screening of antioxidant power of selective extracts on TLC was carried out according to the work of Takao *et al* [14].

### 2.2.2. Quantitative study

#### 2.2.2.1. Determination of total polyphenols

Folin-Ciocalteu colorimetric method was used for evaluation of the content of total phenolic compounds [15]. The following equation was used:

$$Q = (V \times C \times d) / m \text{ (in } \mu\text{g EAG / g dry matter)}$$

With: V: final extract volume (ml), C: extract concentration ( $\mu\text{g / ml}$ ), d: dilution, m: mass of dry matter (g).

#### 2.2.2.2. Dosage of condensed tannins

Condensed tannin content was determined using the method of Broadhurst and Jones [16] and modified by Heilmer *et al.* [17]. To 50  $\mu\text{l}$  of each previously diluted sample, 3 ml of a 4% vanillin solution in methanol (w / v) and 1.5 ml of concentrated hydrochloric acid are added respectively. The mixture is left to stand for 15 min. The absorbance of mixture is then measured at 500 nm with respect to methanol as a control. Condensed tannin contents are expressed in micrograms of catechin equivalent per milligram of dry matter ( $\mu\text{g ECAT / mg}$ ). They were deduced from a calibration curve performed using a range of catechin concentrations (50 to 600  $\mu\text{g / ml}$ ).

#### 2.2.2.3 Determination of total flavonoids

The method described by Swain *et al.* [18] and taken up by Dif *et al.* [19] was used for quantification of flavonoids. 1.5 ml of distilled water and 0.15 ml of a 5% (w / v) sodium nitrate solution are added to 0.5 ml of each hydromethanolic extract.

The mixture obtained is left to rest for 5 min at room temperature, away from light. To this solution 0.15 ml of a 10% (w / v) aluminium trichloride solution was subsequently added. After resting for 11 min in darkness, 0.5ml of hydroxide sodium (1 M) was added to the reaction mixture. The resulting solution was subjected to vortex. Its optical density was read with UV spectrophotometer at 510 nm. Quantification of flavonoids was obtained from a calibration curve produced with catechin (standard) at different concentrations (0-10-20-30-40-50 mg / l) and prepared under the same operating conditions as those of the samples.

#### 2.2.2.4. Evaluation of antioxidant activity by spectrophotometry

Quantification of antioxidant potential of hydromethanolic extracts was carried out according to Blois' methodology [20]. Vitamin C was used as a reference antioxidant. The percentage reduction of DPPH radical is estimated according to the following formula:

$$PR = [(A_b - A_e) / A_b] \times 100$$

With: PR: Percentage reduction; Ab: Absorbance of negative control; Ae: Absorbance of sample.

## 3. Results and discussion

### 3.1. Phytochemical screening by TLC

Phytochemical screening carried out by means of thin layer chromatography revealed a multitude of phytochemicals (Table I), which were detected by means of specific developers. This is the case of Liebermann-Bürchard's reagent, which was used for detecting sterols and terpenes [21]. The latter reveals sterols in the visible in brown and green; in yellow and yellow-green under UV / 365 nm. While terpenes appear in blue, purple in the visible and red or yellow-orange at 365 nm UV. In addition, certain developers such as Godin's reagent and sulfuric vanillin were used to confirm the presence of these compounds. Indeed, under Godin's reagent action, sterols are identified in blue, purple or brown in the visible; in brown under UV / 365 nm. Terpenes appear in purple with the same developer [12]. Sulfuric vanillin also detects sterols in blue spots, under UV visualization at 365 nm. It also mentions the presence of terpenes in the visible and UV / 365 nm in pink, purple and orange [12]. Orange or orange-yellow spots recorded with Dragendorff's reagent indicate the existence of alkaloids [21]. Then, yellow, green, blue and fluorescent blue molecular fingerprints observed at the visible and at 365 nm with 5% KOH methanolic solution indicate the existence of coumarins. These colorations can intensify or change at 365 nm under action of said developer [22]. Saponins was revealed by means of antimony chloride (SbCl<sub>3</sub>) [11]. The latter, reveals them in the visible in yellow for steroid type saponins and in pink-violet for those of triterpene type. Tannins were detected with the 2% methanolic FeCl<sub>3</sub> solution, in gray or brown to the visible [21]. Finally, flavonoids were identified by means of Neu's reagent, in the visible in yellow and brown; and these colorations can intensify or diversify under UV observation at 365 nm [11]. Moreover, AlCl<sub>3</sub> was used to confirm the existence of certain flavonoids. The latter revealed them in the visible in yellow and under UV at 365 nm, in blue or brown [21] or in yellow-green [11, 23].

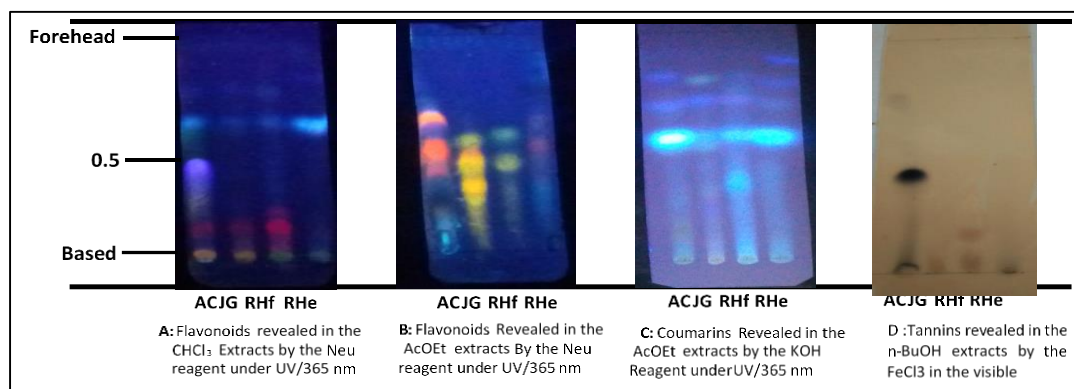
**Table 1:** Secondary metabolites identified in the different samples

Excerpts	Rf, Color, Identified compounds
<i>Alchornea cordifolia</i> (AC)	n-C <sub>6</sub> H <sub>14</sub> 0.21 : jv <sup>g</sup> , Sté; 0.44 : jo <sup>a</sup> , jv <sup>g</sup> , vi <sup>i</sup> , Sté <sup>ag</sup> ; 0.52 : j <sup>g</sup> , vi <sup>i</sup> , Sté <sup>g</sup> ; 0.67 : j <sup>f</sup> , vi <sup>i</sup> , Cou <sup>f</sup> , Sté <sup>i</sup> ; 0.73 : jc <sup>a</sup> , jo <sup>h</sup> , jv <sup>f</sup> , Ter <sup>ah</sup> , Cou <sup>f</sup> ; 0.82 : v <sup>a</sup> , jo <sup>h</sup> , j <sup>f</sup> , Ter <sup>ah</sup> , Cou <sup>f</sup> ; 0.9 : jo <sup>a</sup> , Sté
	CHCl <sub>3</sub> 0.04 : jo <sup>l</sup> , Sap Tri; 0.08 : j <sup>l</sup> , Sap Sté; 0.13 : v <sup>g</sup> , or <sup>d</sup> , Sté <sup>g</sup> , Al <sup>d</sup> ; 0.15 : j <sup>l</sup> , Sap Sté; 0.21 : jo <sup>l</sup> , Sap Tri; 0.26 : v <sup>g</sup> , jo <sup>f</sup> , Sté <sup>g</sup> , Cou <sup>f</sup> ; 0.4 : jo <sup>a</sup> , Cou; 0.5 : or <sup>h</sup> , Ter; 0.66 : v <sup>f</sup> , Cou; 0.7 : j <sup>a</sup> , Sap Sté; 0.84 : v <sup>a</sup> , jv <sup>g</sup> , or <sup>d</sup> , blvi <sup>h</sup> , Sté <sup>ag</sup> , Al <sup>d</sup> , Ter <sup>h</sup> ; 0.9 : or <sup>h</sup> , Ter; 0.95 : jv <sup>f</sup> , Cou
	AcOEt 0.14 : jc <sup>b</sup> , Fla; 0.23 : bl <sup>c</sup> , Fla; 0.37 : jo <sup>b</sup> , Fla; 0.39 : gr <sup>ae</sup> , Ta <sup>ae</sup> ; 0.41 : v <sup>a</sup> , jo <sup>bc</sup> , br <sup>e</sup> , Fla <sup>abc</sup> , Ta <sup>e</sup> ; 0.52 : jo <sup>c</sup> , jc <sup>b</sup> , Fla <sup>bc</sup> ; 0.64 : jv <sup>c</sup> , Fla; 0.75 : jv <sup>c</sup> , jo <sup>b</sup> , Fla <sup>bc</sup> ; 0.82 : jo <sup>a</sup> , Fla; 0.91 : br <sup>e</sup> , Ta
	n-BuOH 0.13 : bl <sup>c</sup> , br <sup>e</sup> , Fla <sup>c</sup> , Ta <sup>e</sup> ; 0.18 : jv <sup>c</sup> , blv <sup>b</sup> , br <sup>e</sup> , Fla <sup>bc</sup> , Ta <sup>e</sup> ; 0.24 : jv <sup>c</sup> , Fla; 0.47 : jv <sup>c</sup> , gr <sup>e</sup> , Fla <sup>c</sup> , Ta <sup>e</sup> ; 0.52 : jo <sup>c</sup> , jv <sup>b</sup> , Fla <sup>bc</sup> ; 0.60 : bl <sup>f</sup> , blvi <sup>c</sup> , jv <sup>b</sup> , Cou <sup>f</sup> , Fla <sup>bc</sup> ; 0.65 : jo <sup>c</sup> , Fla; 0.71 : j <sup>f</sup> , jv <sup>c</sup> , jv <sup>b</sup> , Cou <sup>f</sup> , Fla <sup>bc</sup> ; 0.75 : br <sup>e</sup> , Ta
<i>Jatropha gossypifolia</i> (JG)	n-C <sub>6</sub> H <sub>14</sub> 0.34 : j <sup>f</sup> , jv <sup>g</sup> , vi <sup>i</sup> , Cou <sup>f</sup> , Sté <sup>gi</sup> ; 0.43 : j <sup>a</sup> , j <sup>f</sup> , jv <sup>g</sup> , Cou <sup>af</sup> , Sté <sup>g</sup> ; 0.5 : bl <sup>a</sup> , vi <sup>i</sup> , Cou <sup>a</sup> , Sté <sup>i</sup> ; 0.60 : jc <sup>f</sup> , Cou; 0.75 : jo <sup>h</sup> , bl <sup>f</sup> , j <sup>a</sup> , Ter <sup>h</sup> , Cou <sup>af</sup> ; 0.82 : jv <sup>a</sup> , jo <sup>h</sup> , bl <sup>f</sup> , Ter <sup>ah</sup> , Cou <sup>f</sup> ; 0.9 : jo <sup>a</sup> , Ter
	CHCl <sub>3</sub> 0.02 : jo <sup>l</sup> , Sap Tri; 0.06 : j <sup>l</sup> , Sap Sté; 0.13 : jv <sup>f</sup> , v <sup>g</sup> , jo <sup>l</sup> , Cou <sup>f</sup> , Sté <sup>g</sup> , Sap Tri; 0.21 : jo <sup>l</sup> , Sap Tri; 0.26 : v <sup>f</sup> , Cou; 0.37 : jo <sup>a</sup> , v <sup>g</sup> , Ter <sup>a</sup> , Sté <sup>g</sup> ; 0.4 : jo <sup>a</sup> , Ter; 0.67 : jo <sup>h</sup> , or <sup>d</sup> , v <sup>f</sup> , Ter <sup>h</sup> , Al <sup>d</sup> , Cou <sup>f</sup> ; 0.71 : jo <sup>h</sup> , jv <sup>f</sup> , j <sup>a</sup> , Ter <sup>h</sup> , Cou <sup>f</sup> , Sap Sté <sup>a</sup> ; 0.82 : jv <sup>a</sup> , Cou; 0.90 : vic <sup>h</sup> , Ter; 0.95 : blv <sup>f</sup> , Cou
	AcOEt 0.28 : jc <sup>c</sup> , jp <sup>b</sup> , Fla <sup>bc</sup> ; 0.34 : jo <sup>a</sup> , j <sup>bc</sup> , jv <sup>f</sup> , Fla <sup>abc</sup> , Cou <sup>f</sup> ; 0.45 : jv <sup>f</sup> , jo <sup>ac</sup> , j <sup>b</sup> , br <sup>e</sup> , Cou <sup>f</sup> , Fla <sup>abc</sup> , Ta <sup>e</sup> ; 0.51 : jv <sup>c</sup> , Fla; 0.76 : jo <sup>b</sup> , br <sup>e</sup> , Fla <sup>b</sup> , Ta <sup>e</sup> ; 0.9 : jo <sup>a</sup> , Fla
	n-BuOH 0.12 : br <sup>e</sup> , Ta; 0.31 : jc <sup>c</sup> , blv <sup>b</sup> , Fla <sup>bc</sup> ; 0.45 : jc <sup>c</sup> , blv <sup>b</sup> , Fla <sup>bc</sup> ; 0.50 : jc <sup>c</sup> , Fla; 0.56 : jo <sup>c</sup> , j <sup>b</sup> , Fla <sup>bc</sup> ; 0.63 : j <sup>f</sup> , j <sup>b</sup> , Cou <sup>f</sup> , Fla <sup>b</sup> ; 0.74 : j <sup>f</sup> , Cou; 0.80 : jv <sup>b</sup> , Fla
<i>Ricinodendron heudelotii</i> (RHf)	n-C <sub>6</sub> H <sub>14</sub> 0.31 : vi <sup>a</sup> , br <sup>g</sup> , vi <sup>i</sup> , Ter <sup>a</sup> , Sté <sup>gi</sup> ; 0.44 : jo <sup>a</sup> , Cou; 0.53 : vi <sup>a</sup> , jv <sup>g</sup> , vic <sup>i</sup> , Ter <sup>a</sup> , Sté <sup>gi</sup> ; 0.61 : vic <sup>a</sup> , jo <sup>f</sup> , Sté <sup>a</sup> , Cou <sup>f</sup> ; 0.74 : vi <sup>h</sup> , j <sup>f</sup> , Ter <sup>h</sup> , Cou <sup>f</sup> ; 0.85 : jo <sup>a</sup> , vi <sup>h</sup> , Cou <sup>a</sup> , Ter <sup>h</sup> ; 0.9 : vi <sup>a</sup> , jc <sup>f</sup> , Ter <sup>a</sup> , Cou <sup>f</sup>
	CHCl <sub>3</sub> 0.06 : jo <sup>l</sup> , Sap Tri; 0.15 : j <sup>l</sup> , Sap Sté; 0.21 : jo <sup>l</sup> , Sap Tri; 0.27 : v <sup>f</sup> , Cou; 0.43 : jo <sup>a</sup> , v <sup>f</sup> , Sap Tri <sup>a</sup> , Cou <sup>f</sup> ; 0.65 : jo <sup>a</sup> , or <sup>d</sup> , Sap Tri <sup>a</sup> , Al <sup>d</sup> ; 0.70 : j <sup>a</sup> , Sap Sté; 0.81 : or <sup>d</sup> , Al
	AcOEt 0.32 : jo <sup>b</sup> , Fla; 0.43 : jc <sup>f</sup> , jo <sup>a</sup> , jv <sup>c</sup> , j <sup>b</sup> , Cou <sup>f</sup> , Fla <sup>abc</sup> ; 0.54 : jv <sup>c</sup> , gr <sup>e</sup> , Fla <sup>c</sup> , Ta <sup>e</sup> ; 0.75 : jo <sup>b</sup> , Fla
	n-BuOH 0.11 : blv <sup>b</sup> , br <sup>e</sup> , Fla <sup>b</sup> , Ta <sup>e</sup> ; 0.20 : bl <sup>b</sup> , br <sup>e</sup> , Fla <sup>b</sup> , Ta <sup>e</sup> ; 0.35 : jo <sup>c</sup> , Fla; 0.40 : jc <sup>c</sup> , Fla; 0.56 : jc <sup>c</sup> , Fla; 0.62 : j <sup>f</sup> , Cou; 0.68 : jv <sup>b</sup> , Fla; 0.74 : j <sup>f</sup> , Cou
<i>Ricinodendron heudelotii</i> (RHe)	n-C <sub>6</sub> H <sub>14</sub> 0.36 : j <sup>g</sup> , vi <sup>i</sup> , Sté <sup>gi</sup> ; 0.44 : vic <sup>i</sup> , Sté; 0.61 : j <sup>f</sup> , bl <sup>i</sup> , jv <sup>g</sup> , Cou <sup>f</sup> , Sté <sup>gi</sup> ; 0.72 : bl <sup>a</sup> , blv <sup>f</sup> , Cou <sup>af</sup> ; 0.83 : bl <sup>f</sup> , Cou
	CHCl <sub>3</sub> 0.14 : v <sup>f</sup> , Cou; 0.42 : v <sup>f</sup> , Cou; 0.53 : jv <sup>g</sup> , Sté; 0.77 : v <sup>f</sup> , Cou; 0.86 : jo <sup>h</sup> , Ter; 0.95 : v <sup>f</sup> , Cou
	AcOEt 0.27 : bl <sup>f</sup> , Fla; 0.36 : jv <sup>c</sup> , Fla; 0.44 : blv <sup>b</sup> , Fla; 0.50 : jo <sup>c</sup> , Fla; 0.66 : jv <sup>c</sup> , Fla; 0.76 : jo <sup>f</sup> , bl <sup>f</sup> , Cou <sup>f</sup> , Fla <sup>b</sup>
	n-BuOH 0.10 : gr <sup>e</sup> , Ta; 0.32 : jv <sup>c</sup> , Fla; 0.56 : jv <sup>c</sup> , Fla; 0.73 : j <sup>f</sup> , jc <sup>c</sup> , Cou <sup>f</sup> , Fla <sup>c</sup>

- bl / blue; blf / fluorescent blue; blv / blue-green; blvi / blue-violet; br / brown; brc / light brown; gr / gray; gv / gray-violet; j / yellow; jc / light yellow; jo / yellow-orange; jp / pale yellow; jv / yellow-green; gold / orange; v / green; vf / green-fluorescent; vi / violet; vic / light purple.
- a / without developer; b / AlCl<sub>3</sub>; c / Neu reagent; d / Dragendorff's reagent; e / FeCl<sub>3</sub>; f / KOH; g / Lieberman Burchard's reagent; h / Sulfuric vanillin; i / Godin's reagent; j / SbCl<sub>3</sub>
- Al: Alkaloids; Cou: Coumarins; Fla: Flavonoids; Sté: Sterols; Ta: Tannins; Ter: Terpenes; Sap Sté: steroid-type saponins; Sap Tri: Triterpene type saponins

Thus, the existence of sterols, terpenes and certain coumarins has been observed in hexanic extract. However, a profusion of sterols was noted in *Ricinodendron heudelotii* (RHf) and *Jatropha gossypifolia* (JG) leaf extracts. Chloroformic fraction contains, in addition to the compounds detected in the previous extract, flavonoids; alkaloids whose presence has been remarked in *Alchornea cordifolia* (AC) (Rf = 0.13; 0.84), *Jatropha gossypifolia* (JG) (Rf = 0.67) and *Ricinodendron heudelotii* leaves (RHf) (Rf = 0.65; 0.81); steroid-type saponins, in AC (Rf = 0.08; 0.15), JG (Rf = 0.06)

and RHf (Rf = 0.15); and those of triterpene type, also in AC (Rf = 0.04; 0.21), JG (Rf = 0.02; 0.13; 0.21) and RHf (Rf = 0.06; 0.21). *Ricinodendron heudelotii* (RHe) stem bark is free from saponins and alkaloids. Finally, in ethyl acetate and n-butanol extracts, we identified flavonoids (Figure 1A and B), coumarins (Figure 1C) and tannins (Figure 1D). Overall, a significant presence of flavonoids was reported in n-butanol extract, particularly in the leaves of *Alchornea cordifolia*, *Jatropha gossypifolia* and *Ricinodendron heudelotii*.

**Fig 1:** chromatographic profiles of antimarial plant extracts

Ultimately, phytochemical screening revealed in the different extracts the co-presence of a plethora of compounds,

including sterols, terpenes, alkaloids, flavonoids, saponins, coumarins and tannins. An abundance of flavonoids has been

noticed in the leaves of *Alchornea cordifolia*. For other extracts, a non-negligible presence was noticed. These results are conform to those obtained by other researchers. Indeed, some have reported the existence of flavonoids, tannins, sterols, triterpenes and coumarins in leaves of *A. cordifolia* [24, 25]. Concerning tannins, terpenoids, glycosides and alkaloids, their presence has been reported in leaves of *Ricinodendron heudelotii* by Omolara *et al.* [26]. Finally, some researchers have highlighted alkaloids and flavonoids in *Jatropha gossypifolia* leaves [27, 28]. Thus, we can argue that the antimalarial activity of these plant matrices is conditioned by

their composition in phytochemicals. Some compounds such as alkaloids are known for their antimalarial potential [29-32]. Also, certain classes of flavonoids (artemetine and casticin) and terpenes (triterpenoids and sesquiterpenes) possess antiplasmodic and antimalarial activities [33, 34].

### 3.2. Antioxidant power screening

Anti-radical zones are materialized by yellow spots on a purple background on chromatograms (Figure 2), which are subordinated by the coexistence of phytochemicals endowed with antioxidant potential.

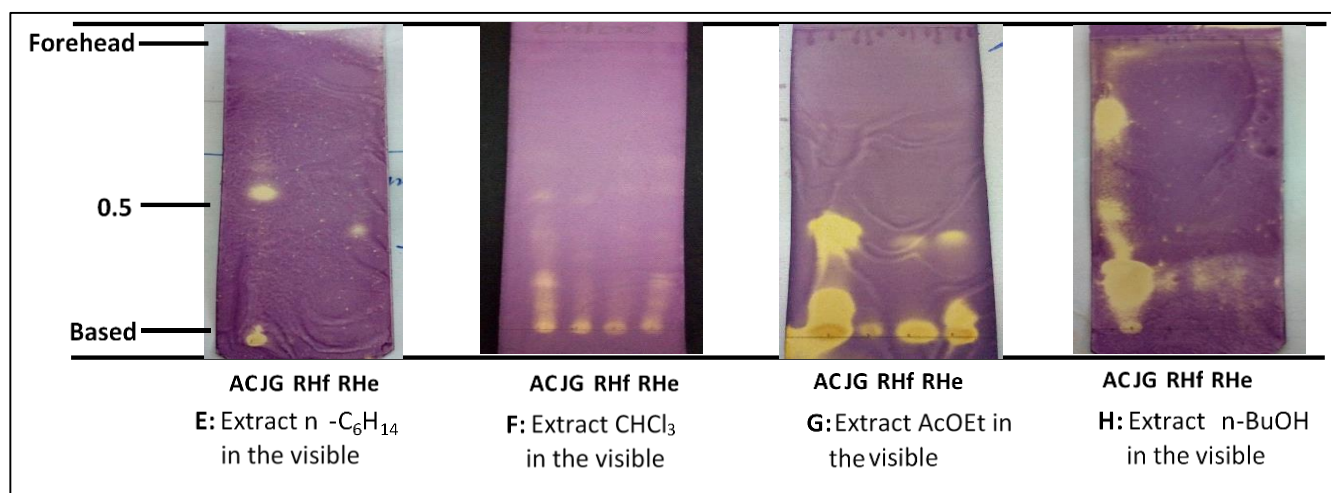


Fig 2: Chromatographic profiles of the antioxidant activity of antimalarial plant extracts

When comparing chromatograms of phytochemical screening (Figure 1 and Table I) and those of antioxidant power (Figure 2), we can deduce that in hexanic extracts, terpenes ( $R_f = 0.01$  and  $0.52$ ) and coumarins ( $R_f = 0.67$ ) are responsible for antioxidant activity observed in AC. Similarly, terpenes ( $R_f = 0.44$ ) are dependent on antioxidant potential in RHe (Figure 2E). For chloroformic extracts (Figure 2F), the phytochemicals scavenging DPPH radical are sterols ( $R_f = 0.13$ ), coumarins ( $R_f = 0.26$  and  $0.50$ ) and flavonoids ( $R_f = 0.40$ ) for AC; then, flavonoids ( $R_f = 0.12$  and  $0.37$ ) and coumarins ( $R_f = 0.40$ ) for JG; flavonoids ( $R_f = 0.16$ ;  $0.43$  and  $0.65$ ) for RHf; at last, flavonoids ( $R_f = 0.14$  and  $0.20$ ;  $0.42$  and  $0.60$ ) and sterols ( $R_f = 0.50$ ) for RHe. On the other hand, the antioxidant potential observed in ethyl acetate extracts (Figure 2G) of antimalarial plants is due to the co-presence of different phytoconstituents. Thus, zones of antiradical activity were visualized in AC (flavonoids:  $R_f = 0.14$ ;  $0.37$  and  $0.64$ ), JG (flavonoids:  $R_f = 0.34$ ), RHf (flavonoids:  $R_f = 0, 32$ ) and RHe (Coumarins:  $R_f = 0.27$  and flavonoids:  $R_f = 0.27, 0.36$  and  $0.66$ ). Finally, for n-butanol extracts (Figure 2H), the molecular fingerprints of flavonoids ( $R_f = 0.40$  and  $0.71$ ) and tannins ( $R_f = 0.13$  and  $0.18$ ) would be responsible for AC's antiradical capacity. This is the case for JG (tannins ( $R_f = 0.12$ ) and flavonoids ( $R_f = 0.31$ )) and RHf (tannins ( $R_f = 0.20$ ) and flavonoids ( $R_f = 0.35$ )). For RHe, flavonoids ( $R_f = 0.32$ ) are responsible. In summary, among the extracts analysed, *Alchornea cordifolia* leaves (control plant) appear to be most active against DPPH radical, followed by *Ricinodendron heudelotii* stem bark. This potential detected in these extracts gives them a host of pharmacological

properties, particularly antimalarial. Indeed, there is a correlation between oxidative stress and malaria. Once infected, we see hypoglycaemia in patients, caused by the parasites' use of blood glucose. This situation increases the population of parasites in the body, leading to elevation of the level of lipoperoxidation, which is a marker of oxidative stress [35, 36]. Thus, antioxidant compounds in the different plants could contribute to prevent oxidative stress, which is a cause of malaria. Among samples studied, we can deduce that *R. heudelotii* stem bark could have the status of an antimalarial plant.

### 3.3. Determination of total polyphenols

*A. cordifolia* leaves had the highest content ( $14865 \pm 624.49$   $\mu\text{g EAG} / \text{g dry matter}$ ), followed by *R. heudelotii* stem bark ( $6615 \pm 835.16$   $\mu\text{g EAG} / \text{g dry matter}$ ). On the other hand, the lowest levels were recorded at the level of the leaves of *J. gossypifolia* ( $2715 \pm 600$   $\mu\text{g EAG} / \text{g dry matter}$ ) and those of *R. heudelotii* ( $915 \pm 519.61$   $\mu\text{g EAG} / \text{g dry matter}$ ). Overall, we note that the control plant (*A. cordifolia*) contains more phenolic compounds than other plant matrices. However, *R. heudelotii* stem bark stood out among the rest. In addition, the results obtained for *A. cordifolia* leaves are much better than those recorded by other researchers. In fact, the latter obtained an estimated content of  $7011.57$   $\mu\text{g EAG} / \text{g dry matter}$  [37]. This notable difference could be explained by several factors, including those called biogenetic and environmental [38]. Thus, the high content observed could be justified by the coexistence of several phytochemicals (flavonoids, coumarins, and so on.) mentioned above in phytochemical screening.

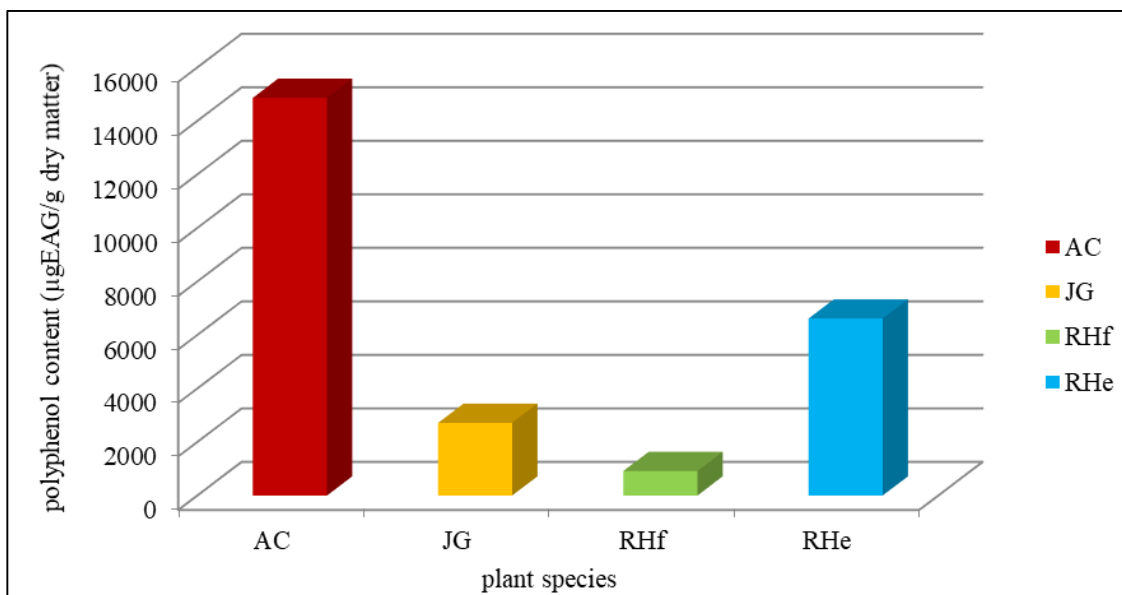


Fig 3: shows total polyphenol contents of different antimalarial plants extracts.

### 3.4. Determination of total condensed tannins

The total condensed tannin contents recorded vary from one plant species to another (Figure 4).

*A. cordifolia* leaves contain more condensed tannins ( $1183.75 \pm 10.89 \mu\text{g ECT} / \text{mg}$ ) than other plant species (*J. gossypifolia* ( $733.43 \pm 12.5 \mu\text{g ECT} / \text{mg}$ ) and *R. heudelotii* (leaves:  $533.61 \pm 8.66 \mu\text{g ECT} / \text{mg}$ ; stem bark:  $183.77 \pm 10.0$

$\mu\text{g ECT} / \text{mg}$ ). These results had already been predicted by phytochemical screening. Additionally, we found that leaves are rich in condensed tannins compared to stem bark. This observation would be due to unequal distribution of secondary metabolites in different plant organs, depending on the species, tissues and physiological stages [39].

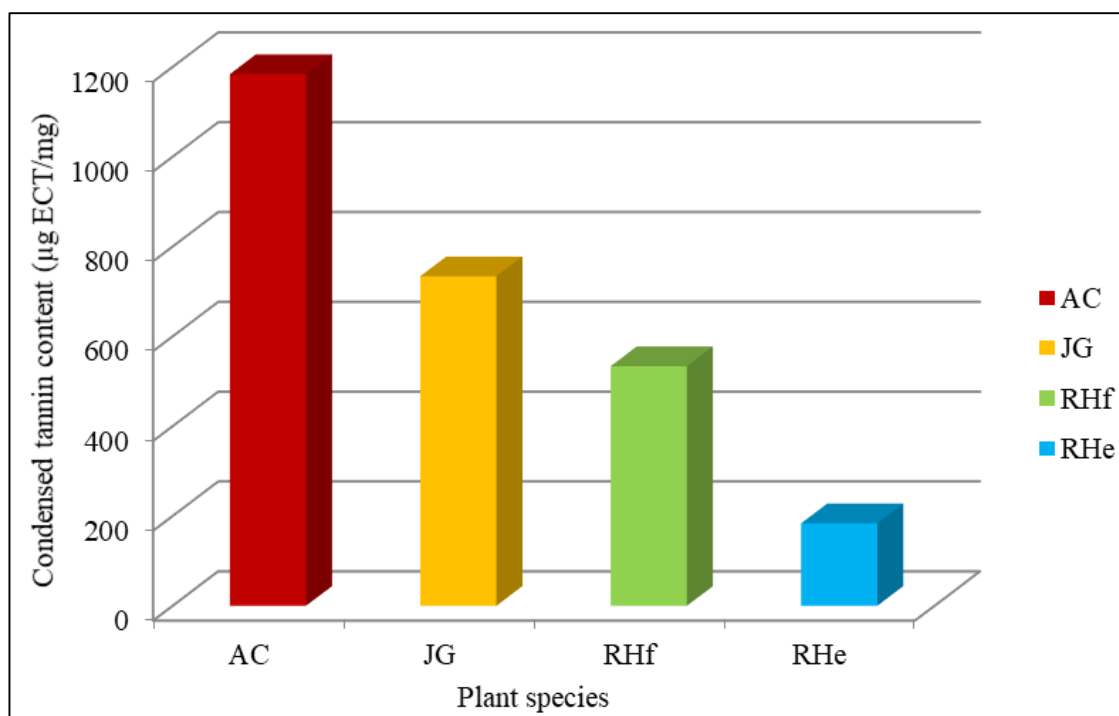


Fig 4: Condensed tannin content ( $\mu\text{g ECT} / \text{mg}$ )

### 3.5. Determination of total flavonoids

*A. cordifolia* leaves ( $82.77 \pm 0.57 \text{ mg/g}$  of dry matter) and *R. heudelotii* stem bark ( $72.28 \pm 0.69\% \text{ mg/g}$  of dry matter) contain more flavonoids than other plant matrices (Figure 5). The low levels were recorded in leaves of *J. gossypifolia* ( $18.78 \pm 0.57\% \text{ mg/g}$  of dry matter) and those of *R. heudelotii*

( $24.56 \pm 0.99\%$ ). These results are in accordance with those obtained during the phytochemical screening. We note that the flavonoid content of *R. heudelotii* stem bark is close to that of the control plant. Thus, it would be advisable to use these in the management of certain pathologies, in particular malaria.

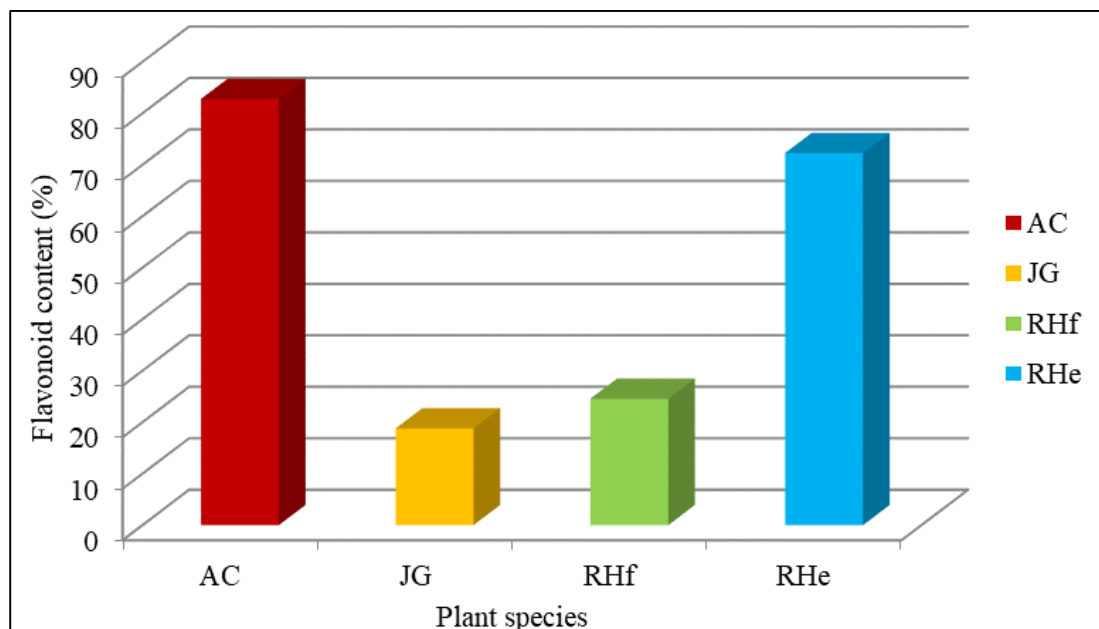


Fig 5: Total flavonoid content

### 3.6. Evaluation of antioxidant activity by spectrophotometry

The estimation of antioxidant potential of crude hydromethanolic extracts vis-à-vis DPPH radical was carried out by spectrophotometry, with vitamin C as a reference antioxidant (Figure 6).

Overall, vitamin C exhibited very strong anti-free radical activity at all concentrations. *A. cordifolia* showed slightly lower percentages of reduction (PR) than vitamin C at certain concentrations (2 mg / ml; 0.5 mg / ml and 0.0625 mg / ml). As a result, *A. cordifolia* is the most active plant, followed by *R. heudelotii* bark stem. The leaves of *J. gossypifolia* and *R. heudelotii* exhibited reduction potentials of less than 50% at all concentrations, except at 0.0625 mg / ml where these obtained respectively  $61.6199 \pm 0.6739\%$  and  $52.7725 \pm 0.3070\%$  as the percentage reduction of DPPH radical. In

addition, *J. gossypifolia* presented at 0.5 mg / ml a PR =  $57.757 \pm 0.571\%$ . In short, we note a similarity in the results of screening and the evaluation of antioxidant power. Thus, the antioxidant power detected in the various plant matrices would justify their use in traditional medicine against many pathologies, in particular malaria. Indeed, several studies have indicated the involvement of antioxidant compounds in preventing malaria. This is the case with vitamin E and trolox, whose use provided partial protection against cerebral malaria [40]. Also, the use of radical scavengers such as catalase or SOD enzymes in the mouse model could prevent cerebral malaria [41]. In addition, butylated hydroxyanisole, an antioxidant, is thought to have anti-malarial properties. Its use would reduce or even annihilate the symptoms of cerebral malaria [41].

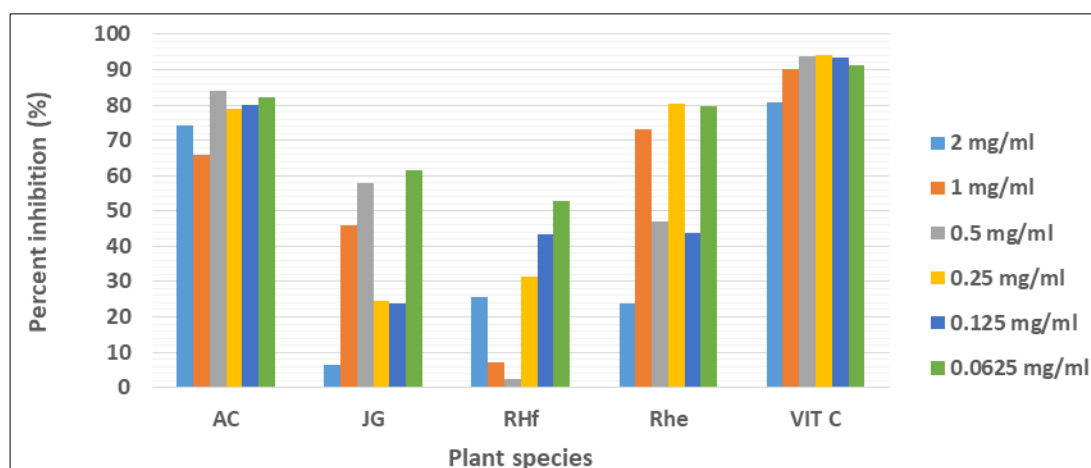


Fig 6: Antioxidant profiles of hydromethanolic extracts and vitamin C

### 4. Conclusion

Qualitative study of three plants traditionally used in the treatment of malaria revealed the presence of several secondary metabolites, including flavonoids, coumarins, sterols, terpenes, tannins, saponins and alkaloids. For quantitative study, the contents obtained ranged from  $14865 \pm 624.49$  to  $915 \pm 519.61$   $\mu\text{g EAG} / \text{g dry matter}$  for total polyphenols;  $82.77 \pm 0.57$  and  $18.78 \pm 0.57$  mg/g of dry

matter for total flavonoids and  $1183.75 \pm 10.89$  and  $183.77 \pm 10.00$   $\mu\text{g ECT} / \text{mg}$  for total condensed tannins. *Alchornea cordifolia* leaves are the richest in phytochemicals, followed by *Ricinodendron heudelotii* stem bark. These results are in agreement with those of phytochemical screening. Also, we note a notable antioxidant potential of *A. cordifolia* leaves and *R. heudelotii* stem bark, which potential is subordinated by the richness in polyphenols, particularly in flavonoids. In

contrast, the leaves of *Jatropha gossypifolia* and *R. heudelotii* showed relatively low antioxidant potentials. Thus, this study provides a rational justification for the use of these plants in treatment of malaria in traditional medicine. In addition, *R. heudelotii* stem bark would be the most appropriate for the treatment of malaria, considering the relative similarity of its antioxidant character to that of *A. cordifolia*, a plant matrix recognized as an antimalarial. Moreover, work on toxicity and evaluation of antimalarial potential of these plants is being carried out with a view to validating the various hypotheses mentioned.

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