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# Ethnopharmacological study of plants used to treat malaria, in traditional medicine, by Bete Populations of Issia (Côte d'Ivoire)

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**Abstract :**

Malaria is the major tropical pathology in developing countries and man used the medicinal properties of many plants to fight this disease. During an ethnopharmacological survey in 13 villages of Issia, a Department of Côte-d'Ivoire, 37 traditional healers were interviewed. They used 32 plants to develop 32 monospecific receipts to treat malaria. Leaves and stem barks are more requested. The preparation of the medicamentous receipts utilizes mainly decoctions (81.81 %). The majority of the remedies are employed by oral way, particularly out of drink (93.93 %). For each plant, 100 g of powdered material were extracted three times in ethanol. The combined extracts were filtered and concentrated to dryness using a rotary evaporator. The ethanolic extracts were tested against chloroquine-resistant FcB1/ Colombia strain of *Plasmodium falciparum*. Extracts were prepared in DMSO at a concentration of 10 mg/ml and serially diluted with culture medium before to be added to asynchronous parasite cultures (1% antiparasitemia and 1% final hematocrite) in 96-well microplates. The growth inhibition for each extract concentration was determined by comparison of the radioactivity incorpored into the treated culture with that in the control culture maintained on the plate.The results of plants extracts evaluation showed that fourteen plants (43.75 %) had good antiplasmodial activity *in vitro* with $IC_{50}$ values ranging from 2.3 to 14.1 µg/ml. The ethnopharmacological informations and the results *in vitro* of antiplasmodial activities of the selected plants indicate the rightfulness of the traditional use of the studied plants as antipaludics.

**Keywords**: *Issia, Côte-d'Ivoire, Ethnopharmacology, Medicinal plants, Traditional Healers*

## 1. Introduction

Malaria is the major tropical pathology in the tropical world. A dramatic recrudescence of this disease is ongoing due to the increasing resistance of parasites. According to World Heath Organization estimations [1], malaria is a several disease causing 300 to 500 millions of cases and 1.1 million deaths per year in tropical zones worldwide. Despite advances in modern medicine, malaria remains a disease which is difficult to be eradicated and is therefore a major health problem, for one main reason: all anti-malarial drugs are expensive for the populations in endemic countries [2]. In the search of means of fighting, people recognized and used the medicinal properties of many plants to fight this disease. Indeed, indigenous plants play an important role in the treatment of many diseases and 80% of people in Africa and in most of the developing countries are estimated to use herbal remedies [3]. Plants' properties are empirically appreciated despite the lack of data concerning their efficiency and safety. Also, many urban populations turn to treatments using plants. The reason is that traditional medicine is a medicine of proximity, less constraining and non expensive [4, 5]. In the search of fighting means against malaria, ethnopharmacological investigations were conducted in Africa and in most of the developing countries [6, 7, 8, 9, 10]. Bete people in Issia, recognized the medicinal virtues of several species used to treat malaria. During an ethnopharmacological survey in Issia, Central-West of Côte d'Ivoire, 32 plants were selected, according to their commonly use by traditional healers for treatment of malaria [11]. This study aims at presenting the botanical and ethnomedical characteristics of selected medicinal plants and evaluating their *in vitro* antimalarial activities.

## 2. Materials and methods

### 2.1. Study site

Our investigations took place in villages of Issia Department (figure 1). Located at 360 km of Abidjan, Issia is part of the Central-West forest of Côte-d'Ivoire (West Africa), in the guinea field of the mesophilic sector, characterized by dense moist semi-deciduous forest [12].

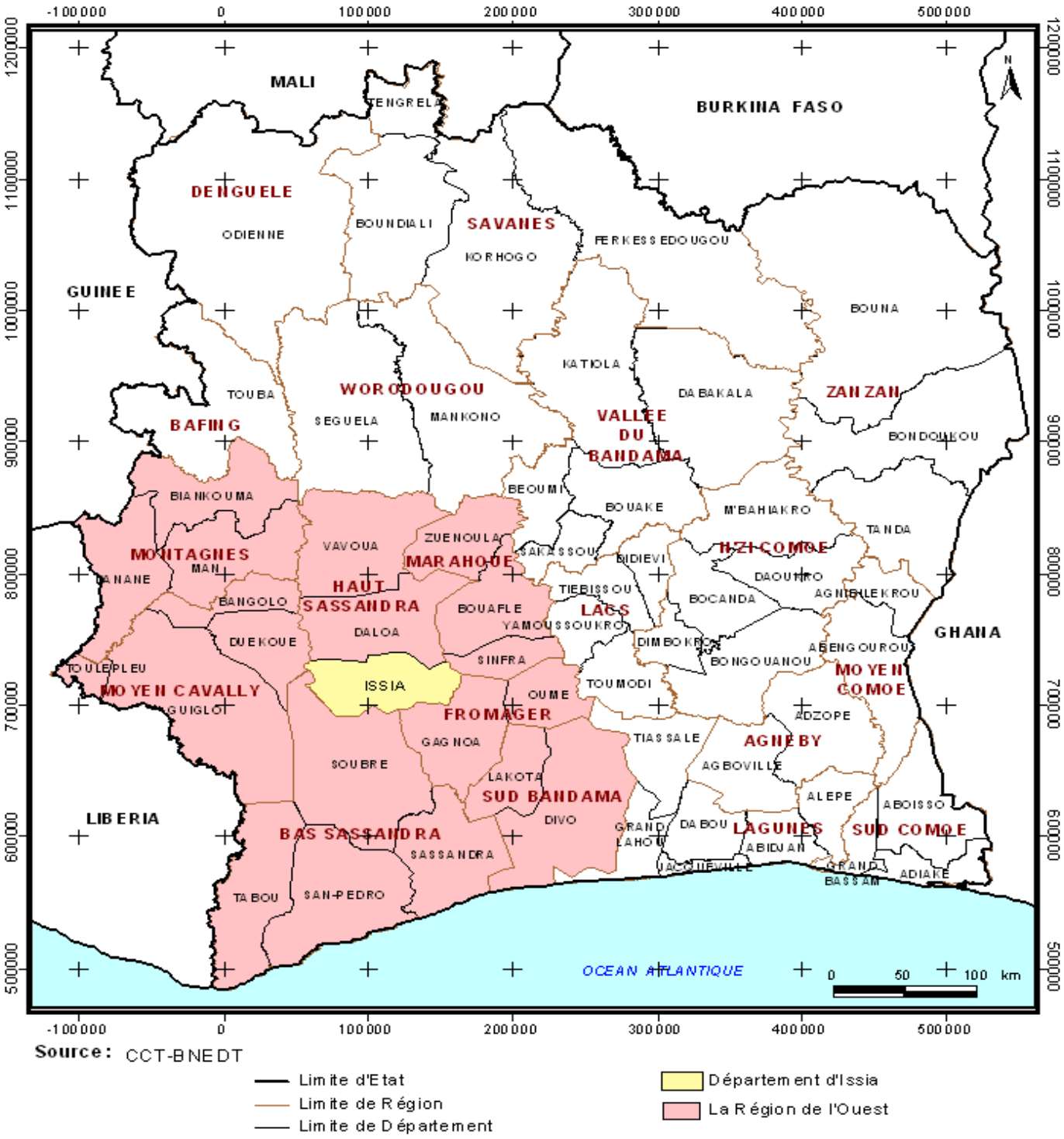


Fig. 1: Geographical situation of the Department of Issia in Côte d'Ivoire.

region is warm and humid and characterized by two seasons: a dry season from December to March and a long rainy season from April to November. The Department of Issia had 105 000 inhabitants. Bete is the native people of Issia; there is a community of non-natives coming from all the other areas of Côte-d'Ivoire and also a community of foreigners for the most part coming from the West African Sub-region and non-

African communities (French, Lebanese and Syrian).

**2.2. Plants and equipment for collection**

The vegetable material is represented by all the plants that are subject of this study. As technical equipment, we used a classic material that allowed us to have access to the plants and take some samples to build up a collection of dried plants.

**2.3. Ethnomedicinal survey**

The investigation on the traditional use of plants having antimalarial effect was conducted among 13 native villages in the Department of Issia (Côte-d'Ivoire). As approach, we met the healers and organized semi-structured interviews. Each of them was met twice, at different moments, to answer the same questions. During this ethnomedicinal investigation, we collected informations relating to the plants used to treat malaria: different parts used as drugs, methods of collection and modes of preparation and administration of the medicamentous receipts. From the collected samples and specimens of the herbarium of the National Floristic Center (C.N.F.), we identified the plants, by their scientific name and we determined their botanical characteristics.

**2.4. Preparation of plants extracts**

Part of collected plants were air-adried during 7 or 10 days in the shade at room temperature and powdered. For each plant, 100g of material were extracted three times in ethanol at room tempature. The combined extracts were filtered and concentrated to dryness using a rotary evaporator. Dry extracts were stored at 4° C until analysis.

**2.5. In vitro antiplasmodial activity**

Extracts were tested against chloroquine-resistant FcB1/ Colombia strain of *Plasmodium falciparum* ($IC_{50}$ value for chloroquine of 0.1Um [13]. The antiplasmodial activity was determined according to Desjardins *et al*. (1979). Extracts were prepared in DMSO at a concentration of 10 mg/ml and serially diluted with culture medium before to be added to asynchronous parasite cultures (1% antiparasitemia and 1% final hematocrite) in 96-well microplates. Plates were maintained for 24h at 37°C. 0.5 uCi of $^{3}H$ hypoxanthine was then added to each well and parasites were maintened for further 24 h. The growth inhibition for each extract concentration was determined by comparison of the radioactivity incorpored into the treated culture with that in the control culture maintained on the plate. The concentration causing 50% inhibition of parasite growth ($IC_{50}$) were calculated from the drug concentration-response curves.

**3. Results and discussion**

**3.1. Botanical characteristics of the studied plants**

The ethnopharmacological investigations that we conducted in Issia (Côte-d'Ivoire), made it possible to identify 32 species of plants used in traditional medicine to treat malaria. These species of plants (table 1) belong to 19 families, 13 orders, 05 sub-classes, 1 class (Dicotyledon). The families of Apocynaceae and Euphorbiaceae, with respectivly 05 and 04 individuals, are the best represented. From the viewpoint of Morphological Types, taxons can be divided into 4 groups (table 2): trees, shrubs, lianas and herbs. The trees, 13 of them, representing 40.62%, are mainly used. Concerning the Biological Types, the Phanerophytes are in the majority (92.80%). Among Phanerophytes, the Microphanerophytes are the best represented. Thirty-one species (96.87 %) of our listed plants are wild taxons; only one of them is introduced. The intensive wild plants sampling cause regression of spontaneous wild species.

**3.2. Ethnopharmacological survey and characteristics of the studied plants**

During this ethnopharmacological study conducted in the Department of Issia (Côte d'Ivoire), we met 37 traditional healers, native of 13 villages who accepted our collaboration and gave informations on anti-malarial plants (table 3). The ethnopharmacological characteristics of the plants, the different parts used as

drugs, the methods of preparation and administration of medicamentous receipts, are consigned in table 4. We note that 32 medicamentous receipts are developed to treat malaria by Bete people in Issia. All the receipts are monospecific. These results are similar to that of Ouattara [14] who indicated that all the medicinal formulas are monospecific. The following organs are used as drugs: leaf, stem bark, root bark and whole plant. There are different methods of sampling these organs: for roots and whole plants, the sampling is made with a hoe; leaves are usually picked by hand; concerning the stem and roots bark, the sampling is done with machetes. Easily accessible specimens (leaves) are usually picked by hand. Methods of sampling are very important; they must prevent plant species extinction [15].

There are different modes of preparation: decoction, infusion, chewing, kneading and roasting. In this study decoction (100%) is the most widespread method of preparation (table 4). The drink (100%) is the most used method of administration. This result is similar to [14] who showed that the decoction and the drink are applied in 100 % of cases in the Department of Divo. Of the 32 receipts, 12 (37.5 %) are made with leaves, 10 (31.25%) made with stem barks, 7 (21.87%) made with whole plants and 3 (9.375%) with root barks. This result is in the line with OUATTARA's study [14]; the author showed that the leaves are mostly used in 37.50 % of the cases. We noticed that 5 receipts are made with plants belonging to Apocynaceae Family, 4 made with Euphorbiaceae species and 3 receipts are prepared with Rubiaceae species. These Families contain alkaloids with antimalarial properties [16]. According to some studies, we notice interesting similarities.

**Table 1: Systematic groups of studied plants**

| Plants species used | Family | Order | S/C | Classes |
|---|---|---|---|---|
| *Albizia ferruginea* | Fabaceae | Fabales | Rosidae | Dicotyledons |
| *Alchornea cordifolia* | Euphorbiaceae | Euphorbiales | Rosidae | Dicotyledons |
| *Alstonia boonei* | Apocynaceae | Gentinales | Asteridae | Dicotyledons |
| *Alternanthera pungens* | Amaranthaceae | Caryophyllales | Caryophyllidae | Dicotyledons |
| *Anthocleista djalonensis* | Loganiacee | Gentinales | Asteridae | Dicotyledons |
| *Anthonotha macrophylla* | Caesalpiniaceae | Fabales | Rosidae | Dicotyledons |
| *Bersama abyssinica* | Melianthaceae | Sapindales | Rosidae | Dicotyledons |
| *Cassia alata* | Caesalpiniaceae | Fabales | Rosidae | Dicotyledons |
| *Chrysophyllum perpulchrum* | Sapotaceae | Ebenales | Dilleniidae | Dicotyledons |
| *Clerodendrum splendens* | Verbenaceae | Lamiales | Asteridae | Dicotyledons |

| | | | | |
|---|---|---|---|---|
| *Cyathula prostrate* | Acanthaceae | Scrophulariales | Asteridae | Dicotyledons |
| *Elytraria marginata* | Acanthaceae | Scrophulariales | Asteridae | Dicotyledons |
| *Erythrococca anomala* | Euphorbiaceae | Euphorbiales | Rosidae | Dicotyledons |
| *Funtumia elastica* | Apocynaceae | Gentianales | Asteridae | Dicotyledons |
| *Hunteria eburnean* | Apocynaceae | Gentianales | Asteridae | Dicotyledons |
| *Hura crepitans* | Euphorbiaceae | Euphorbiales | Rosidae | Dicotyledons |
| *Irvingia gabonensis* | Irvingiaceae | Sapindales | Rosidae | Dicotyledons |
| *Mareya micrantha* | Euphorbiaceae | Euphorbiales | Rosidae | Dicotyledons |
| *Melanthera scandens* | Asteraceae | Asterales | Asteridae | Dicotyledons |
| *Microglossa pyrifolia* | Asteraceae | Asterales | Asteridae | Dicotyledons |
| *Millettia zechiana* | Fabaceae | Fabales | Rosidae | Dicotyledons |
| *Morinda morindoides* | Rubiaceae | Rubiales | Asteridae | Dicotyledons |
| *Motandra guineensis* | Apocynaceae | Gentianales | Asteridae | Dicotyledons |
| *Nauclea latifolia* | Rubiaceae | Rubiales | Asteridae | Dicotyledons |

**Table 1 (End): Systematic groups of studied plants**

| | | | | |
|---|---|---|---|---|
| *Olax gambecola* | Olacaceae | Santalales | Rosidae | Dicotyledons |
| *Parquetina nigrescens* | Perplocaceae | gentianales | Asteridae | Dicotyledons |
| *Phyllanthus muellerianus* | Euphorbiaceae | Euphorbiales | Rosidae | Dicotyledons |
| *Physalis angulata* | Solanaceae | Solanales | Asteridae | Dicotyledons |
| *Rauvolfia vomitoria* | Apocynaceae | Gentianales | Asteridae | Dicotyledons |
| *Rhigiocarya racemifera* | Menispermaceae | Ranunculales | Magnoliidae | Dicotyledons |
| *Scherbournia bignoniiflora* | Rubiaceae | rubiales | Rosidae | Dicotyledons |

| | | | | |
|---|---|---|---|---|
| *Zanthoxylum gilletii* | Rutaceae | Sapindales | Rosidae | Dicotyledons |
| **Total: 32 Species** | **19 Families** | **13 Orders** | **05 S/Classes** | **01 Class** |

**Table 2: Morpho-Biological Types and Chorological Affinities of listed plants**

| **Plants species used** | **Morphological Types** | **Biological Types** | **Chorological affiniti**es | **Status** |
|---|---|---|---|---|
| *Albizia ferruginea* | Tree | Microphanerophyte | GC | Wild |
| *Alchornea cordifolia* | Shrub | Microphanerophyte | GC | Wild |
| *Alstonia boonei* | Tree | Megaphanesophyte | GC-SZ | Wild |
| *Alternanthera pungens* | Herb | Chamephyte | GC-SZ | Wild |
| *Anthocleista djalonensis* | Tree | Microphanerophyte | GC | Wild |
| *Anthonotha macrophylla* | Tree | Microphanerophyte | GC | Wild |
| *Bersama abyssinica* | Tree | Microphanerophyte | GC | Wild |
| *Cassia alata* | Shrub | Nanophnerophyte | GC | Wild |
| *Chrysophyllum perpulchrum* | Tree | Mesophanerophyte | GC | Wild |
| *Clerodendrum splendens* | Liana | Microphanerophyte | GC | Wild |
| *Cyathula prostrate* | Herb | Nanophanerophyte | GC-SZ | Wild |
| *Elytraria marginata* | Herb | Chamephyte | GC | Wild |
| *Erythrococca anomala* | Shrub | Nanophnerophyte | GC | Wild |
| *Funtumia elastica* | Tree | Mesophanerophyte | GC | Wild |
| *Hunteria eburnean* | Tree | Microphanerophyte | GC | Wild |
| *Hura crepitans* | Tree | Mesophanerophyte | I | Cultivated |

| | | | | |
|---|---|---|---|---|
| *Irvingia gabonensis* | Tree | Megaphanerophyte | GC | Wild |
| *Mareya micrantha* | Tree | Nanophnerophyte | GC | Wild |
| *Melanthera scandens* | Liana | Nanophnerophyte | GC | Wild |
| *Microglossa pyrifolia* | Shrub | Microphanerophyte | GC | Wild |
| *Millettia zechiana* | Tree | Microphanerophyte | GC | Wild |

**Signification of Symbols: I: Introduced; GC: Guineo-Congolais; SZ: Soudano-Zambesienne**

**Table 2 (End): Morpho-Biological Types and Chorological Affinities of listed plants**

| | | | | |
|---|---|---|---|---|
| *Morinda morindoides* | Liana | Microphanerophyte | GC | Wild |
| *Motandra guineensis* | Liana | Microphanerophyte | GC-SZ | Wild |
| *Nauclea latifolia* | Liana | Microphanerophyte | GC | Wild |
| *Olax gambecola* | Herb | Nanophnerophyte | GC | Wild |
| *Parquetina nigrescens* | Liana | Microphanerophyte | GC | Wild |
| *Phyllanthus muellerianus* | Shrub | Nanophanerophyte | GC | Wild |
| *Physalis angulata* | Herb | Therophyte | GC-SZ | Wild |
| *Rauvolfia vomitoria* | Tree | Microphanerophyte | GC-SZ | Wild |
| *Rhigiocarya racemifera* | Liana | Microphanerophyte | GC | Wild |
| *Scherbournia bignoniiflora* | Liana | Microphanerophyte | GC | Wild |
| *Zanthoxylum gilletii* | Tree | Mesophanerophyte | GC | Wild |

**Table 3: Overview of interviewed traditional healers per visited village**

| Villages | Traditional healers (Number) | Sex | | Age (years) |
|---|---|---|---|---|
| | | Male | Female | |
| Takouahio | 03 | 02 | 01 | 45-56 |
| Bobam | 03 | 01 | 02 | 56-63 |
| Broma | 02 | 00 | 02 | 56-66 |

| | | | | |
|---|---|---|---|---|
| Dahira | 03 | 01 | 02 | 34-46 |
| Digbam | 04 | 03 | 01 | 55-60 |
| Godoua | 03 | 02 | 01 | 30- 55 |
| Louoboua | 03 | 03 | 00 | 38-40 |
| Madia | 02 | 00 | 02 | 43-48 |
| Nahio | 03 | 02 | 01 | 53-58 |
| Sereguhé | 02 | 01 | 01 | 43-56 |
| Tapéguia | 04 | 02 | 02 | 18-60 |
| Tézié | 03 | 01 | 02 | 27-30 |
| Zadia | 02 | 02 | 00 | 50-60 |
| **Total: 13 villages** | **37** | **20** | **17** | **30-66** |

**Table 4: Indications on the methods of preparation and administration of medicines**

| Plants species used | Part used | Mode of Preparation | Medicamentous Form | Mode of Administration |
|---|---|---|---|---|
| *Albizia ferruginea* | Leaf | Decoction | Decocte | Drink |
| *Alchornea cordifolia* | Leaf | Decoction | Decocte | Drink |
| *Alstonia boonei* | Stem bark | Decoction | Decocte | Drink |
| *Alternanthera pungens* | Whole plant | Decoction | Decocte | Drink |
| *Anthocleista djalonensis* | Stem bark | Decoction | Decocte | Drink |
| *Anthonotha macrophylla* | Stem bark | Decoction | Decocte | Drink |
| *Bersama abyssinica* | Leaf | Decoction | Decocte | Drink |
| *Cassia alata* | Leaf | Decoction | Decocte | Drink |
| *Chrysophyllum perpulchrum* | Stem bark | Decoction | Decocte | Drink |
| *Clerodendrum splendens* | Whole plant | Decoction | Decocte | Drink |
| *Cyathula prostrate* | Whole plant | Decoction | Decocte | Drink |

| | | | | |
|---|---|---|---|---|
| *Elytraria marginata* | Whole plant | Decoction | Decocte | Drink |
| *Erythrococca anomala* | Leaf | Decoction | Decocte | Drink |
| *Funtumia elastica* | Stem bark | Decoction | Decocte | Drink |
| *Hunteria eburnean* | Stem bark | Decoction | Decocte | Drink |
| *Hura crepitans* | Stem bark | Decoction | Decocte | Drink |
| *Irvingia gabonensis* | Stem bark | Decoction | Decocte | Drink |
| *Mareya micrantha* | Leaf | Decoction | Decocte | Drink |
| *Melanthera scandens* | Whole plant | Decoction | Decocte | Drink |
| *Microglossa pyrifolia* | Root bark | Decoction | Decocte | Drink |
| *Millettia zechiana* | Stem bark | Decoction | Decocte | Drink |

**Table 4 (End): Indications on the methods of preparation and administration of medicines**

| | | | | |
|---|---|---|---|---|
| *Morinda morindoides* | Leaf | Decoction | Decocte | Drink |
| *Motandra guineensis* | Leaf | Decoction | Decocte | Drink |
| *Nauclea latifolia* | Root bark | Decoction | Decocte | Drink |
| *Olax gambecola* | Whole plant | Decoction | Decocte | Drink |
| *Parquetina nigrescens* | Leaf | Decoction | Decocte | Drink |
| *Phyllanthus muellerianus* | Leaf | Decoction | Decocte | Drink |
| *Physalis angulata* | Whole plant | Decoction | Decocte | Drink |
| *Rauvolfia vomitoria* | Root bark | Decoction | Decocte | Drink |
| *Rhigiocarya racemifera* | Leaf | Decoction | Decocte | Drink |
| *Scherbournia bignoniiflora* | Leaf | Decoction | Decocte | Drink |
| *Zanthoxylum gilletii* | Stem bark | Decoction | Decocte | Drink |

**Table 5: *In vitro* antiplasmodial activity of ethanol extracts of plants on *Plasmodium falciparum***

| Plants species | *Plasmodium falciparum*, $IC_{50}$ (µg/ml) |
|---|---|
| *Albizia ferruginea* | >50 |
| *Alchornea cordifolia* | 22.3 ± 3.3 |
| *Alstonia boonei* | 12.3 ± 0.2 |
| *Alternanthera pungens* | >50 |
| *Anthocleista djalonensis* | >50 |
| *Anthonotha macrophylla* | 43.2 ± 0.3 |
| *Bersama abyssinica* | 20.7 ± 3.3 |
| *Cassia alata* | >50 |
| *Chrysophyllum perpulchrum* | 12.8 ± 4.2 |
| *Clerodendrum splendens* | >50 |
| *Cyathula prostrate* | 12.4 ± 3.8 |
| *Elytraria marginata* | >50 |
| *Eryhrococca anomala* | 13.1 ± 3.0 |
| *Funtumia elastica* | 3.6 ± 0.9 |
| *Hunteria eburnean* | 2.2 ± 0.9 |
| *Hura crepitans* | >50 |
| *Irvingia gabonensis* | 22.1 ± 0.8 |
| *Mareya micrantha* | >50 |
| *Melanthera scandens* | 20.7 ± 0.4 |
| *Microglossa pyrifolia* | 31.1 ± 3.0 |
| *Millettia zechiana* | 14.1 ± 3.1 |
| *Morinda morindoides* | 9.8 ± 0.3 |

$IC_{50}$ values are the mean ± the standard deviations from three independent experiments

**Table 5 (End): *In vitro* antiplasmodial activity of ethanolic extracts of plants on *Plasmodium falciparum.***

| | |
|---|---|
| *Motandra guineensis* | 16.3 ± 1.8 |
| *Nauclea latifolia* | 7.3 ± 1.3 |
| *Olax gambecola* | 5.2 ± 0.4 |
| *Parquetina nigrescens* | 20.3 ± 0.9 |
| *Phyllanthus muellerianus* | 10.3 ± 0.7 |
| *Physalis angulata* | 7.9 ± 0.3 |
| *Rauvolfia vomitoria* | 2.5 ± 0.8 |
| *Rhigiocarya racemifera* | >50 |
| *Scherbournia bignoniiflora* | 24.7 ± 0.8 |
| *Zanthoxylum gilletii* | 2.8 ± 1.0 |

Some plants in this study are used by Bulu people from Cameroun in the treatment of malaria: this is the case of *Alstonia boonei* (Apocynaceae), *Morinda morindoides* (Rubiaceae) and *Rauvolfia vomitoria* (Euphorbiaceae) [17]. In Democratic Republic of Congo, people use the decoction of *Alchornea cordifolia* (Euphorbiaceae), *Morinda morindoides* (Rubiaceae) and *Rauvolfia vomitoria* (Apocynaceae) leaves in the treatment of malaria [18] ; in the Mont Koupé region in Cameroun, one uses stem barks decoction of *Alstonia boonei* (Apocynaceae) and *Zanthoxylum gilletii* (Rutaceae) for their anti-malarial effects [19].

**3.3. In vitro antiplasmodial activity of the studied plants**

Of the 32 plants extracts tested, 9 (28.12%) were considered inactive against *Plasmodium falciparum* in culture with $IC_{50}$ values > 50 µg/ml , nine (28.12%) showed a weak antiplasmodial activity with $IC_{50}$ values between 15 and 50 µg/ml and fourteen ( 43.75%) had good in vitro antiplasmodial activity with $IC_{50}$ values ranging from 2.3 to 14.1 µg/ml (tale 5). Many plants included in this study have been described as having antiplasmodial activity; this was the case of *Funtumia elastica*, *Zanthoxyllum gilletii* [20], *Morinda morindoides* [21], *Pycnanthus angolensis* [22] and *Physalis angulata* [23].

**4. Conclusion**

The ethnomedicinal investigations conducted in the Department of Issia (Côte-d'Ivoire) show that 32 species of plants are used by Bete people for the treatment of malaria. The drugs (stem barks, root barks and leaves) are used to develop many medicinal preparations by decotion wich is the often used methods. The drinking is the most widespread mode of administration. According to some studies, we found similarities with many plants with antimalarial effects. The results of plants extracts evaluation showed that fourteen plants (43.75 %) had good in vitro antiplasmodial activity with $IC_{50}$ values ranging from 2.3 to 14.1 µg/ml. The ethnopharmacological informations and the results of *in vitro* antiplasmodial activities of the selected plants indicate the rightfulness of the traditional use of the studied plants as antipaludics.

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