



## Evaluation of 13 selected medicinal plants from Burkina Faso for their antiplasmodial properties

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### ABSTRACT

**Aim of the study:** The aim of this study was to evaluate the antiplasmodial properties of 13 plants used against malaria in traditional medicine in Burkina Faso.

**Materials and methods:** *In vitro* antiplasmodial activity of dichloromethane, methanol and aqueous crude extracts obtained from vegetal samples collected in Burkina Faso was first evaluated on the *Plasmodium falciparum* 3D7 chloroquine-sensitive strain using a colorimetric method.

**Results:** Thirteen extracts obtained from 8 different species were found to exhibit antiplasmodial activity ( $IC_{50} < 50 \mu\text{g/ml}$ ). Five species demonstrated a moderate activity ( $15 \mu\text{g/ml} < IC_{50} < 50 \mu\text{g/ml}$ ): *Boswellia dalzielii* (leaves), *Waltheria indica* (roots and aerial parts), *Bergia suffruticosa* (whole plant), *Vitellaria paradoxa* (bark) and *Jatropha gossypifolia* (leaves). The best results were obtained with extracts from the *Dicoma tomentosa* whole plant, from *Psorospermum senegalense* leaves and from *Gardenia sokotensis* leaves. These extracts found to display promising antiplasmodial activity, with  $IC_{50}$  values ranging from 7.0 to 14.0  $\mu\text{g/ml}$ .

The most active plant extracts were then tested for *in vitro* activity on the *Plasmodium falciparum* W2 chloroquine-resistant strain and also for *in vitro* cytotoxicity on normal human fibroblasts (WI-38) in order to determine the selectivity index.

**Conclusions:** *Dicoma tomentosa* (Asteraceae) and *Psorospermum senegalense* (Clusiaceae) appeared to be the best candidates for further investigation of their antiplasmodial properties, reported for the first time by this study.

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### 1. Introduction

Malaria, a parasitic disease caused by *Plasmodium* sp. and transmitted by *Anopheles mosquitoes*, currently ranks highly among the most important infectious diseases around the world. The disease mainly affects Southern countries where malaria is a major public health problem.

According to the last World Malaria Report (WHO, 2009), there are almost 250 million malaria cases and about one million people dying of malaria each year. The majority of deaths from malaria occur in the sub-Saharan African region (89%), particularly in children under 5 years of age (88%). The World Health Organization (WHO) has estimated that one child dies of malaria every 40 s.

The situation is all the more critical because a growing problem of parasite resistance towards available drugs, particularly

chloroquine, has occurred in recent years. The WHO is now promoting ACT (artemisinin-based combination therapy) as the reference drugs for health care management of uncomplicated falciparum malaria in order to reduce the resistance risk (WHO, 2006). However, resistance to artemisinin has been recently described in Asia (Burki, 2009; Egan, 2009). It is thus now more necessary than ever to continue the search for new antimalarial drugs.

The vegetal kingdom remains a good source of pharmacologically active compounds and especially antiplasmodial agents, as reviewed by Bero et al. (2009) and Kaur et al. (2009).

In this context, the pharmacological and phytochemical study of plants from traditional pharmacopoeias could lead not only to the discovery of new antimalarial "lead compounds", but also to the valorization of local vegetal species whose efficacy and safety would have been demonstrated in laboratory investigations. Indeed, people from developing countries often do not have access to modern therapeutics such as ACT to treat malaria because of financial, geographical and/or cultural obstacles. The WHO estimates that up to

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80% of the world's population relies on traditional medicinal products for some aspects of primary health care. Better knowledge of plants from traditional pharmacopoeias and local valorization of validated traditional remedies in ITM (Improved Traditional Medicine) could lead to access to effective, standardized, available and affordable therapeutics for management of malaria by local populations.

Moreover, the pharmacological value detected for tropical species represents a huge argument for the conservation of biodiversity through the protection of species of medicinal interest. It is therefore important to report scientific evidence of the usefulness of plants in order to prevent the extinction of valuable species.

We decided to focus our study on the flora of Burkina Faso, to search for plants which could be active against *Plasmodium falciparum*.

**Table 1**  
Selected plants and collected samples.

Species	Family	Plant part	Place of collection	Voucher number	Traditional use (associated with malaria and related symptoms)	Previous report of antiplasmodial activity
<i>Bauhinia rufescens</i> Lam.	Fabaceae	Le.	Koudougou	BR0000005012077	Malaria (leafy stem decoction)	No report for <i>Bauhinia rufescens</i> but reported for other <i>Bauhinia</i> sp. Muñoz et al. (2000) and Kittakoop et al. (2000)
<i>Bergia suffruticosa</i> Fenzl.	Elatinaceae	Tw. Wh.Pl.	Koudougou Koudougou	BR0000005012077 07-BFOJ-06	Childhood malaria – tonic (whole plant decoction)	No report
<i>Boswellia dalzielii</i> Hutch.	Burseraceae	Le.	Kassou	BR0000005011971	Malaria – fever (leaf and bark decoction)	No report
<i>Crossopteryx febrifuga</i> Benth.	Rubiaceae	Le.	Banfora	07-BFOJ-10	Malaria – fever – icterus (leafy stem decoction – sometimes in combination)	<i>In vitro</i> activity of leaf alkaloid extract ( <i>Plasmodium falciparum</i> ) Sanon et al. (2003) and <i>in vivo</i> activity of stem bark EtOH extract ( <i>Plasmodium berghei</i> ) Elufioye and Agbedahunsi (2004)
<i>Dicoma tomentosa</i> Cass.	Asteraceae	Wh. Pl.	Banfora	07-BFOJ-02	Childhood malaria with spleen inflammation (whole plant decoction); also for adults	No report
<i>Dyschoriste perrottetii</i> O. Kuntze	Acanthaceae	Ae.Pa.	Koudougou	07-BFOJ-01	Childhood malaria (aerial part decoction)	No report
<i>Ficus thonningii</i> Blume	Moraceae	Le.	Godin	07-BFOJ-09	Malaria (pernicious form) – fever (leaf decoction)	No report for <i>Ficus thonningii</i> but reported for other <i>Ficus</i> sp. Sanon et al. (2003) and Muregi et al. (2007)
<i>Gardenia sokotensis</i> Hutch.	Rubiaceae	Le.	Godin	BR0000005012299	Fever – malaria with gastrointestinal disorder (leaf decoction – often in combination)	<i>In vivo</i> activity of DCM:EtOH (7:3) extract ( <i>Plasmodium berghei</i> ) Traoré et al. (2006)
<i>Jatropha gossypifolia</i> L.	Euphorbiaceae	Le.	Bonyolo	BR0000005144952	Malaria – fever – anaemia (leaf decoction)	<i>In vitro</i> activity of hot water extract ( <i>Plasmodium falciparum</i> ) Gbeassor et al. (1989)
<i>Loeseneriella africana</i> (Willd.) N. Hallé Syn: <i>Hippocratea africana</i> Loes ex Engl.	Celastraceae	Le.	Banfora	BR0000005012190	Fever – childhood malaria with spleen inflammation (leaf decoction)	<i>In vivo</i> activity of EtOH roots extract ( <i>Plasmodium berghei</i> ) Okokon et al. (2006)
<i>Psorospermum senegalense</i> Spach.	Clusiaceae	Le.	Banfora	BR0000005012398	Malaria (leaf decoction – sometimes in combination)	No report
<i>Vitellaria paradoxa</i> Gaertn. syn. <i>Butyrospermum paradoxum</i> Hepper	Sapotaceae	Ba. (1)	Godin	07-BFOJ-08	Malaria – fever – icterus (leaf decoction or infusion)	No report
<i>Waltheria indica</i> L.	Sterculiaceae	Ba. (2)	Réo	BR0000005012176	Malaria (aerial part decoction)	<i>In vitro</i> inactivity of water and DCM:MeOH (1:1) extracts Clarkson et al. (2004)
		Le.	Réo	BR0000005012176		
		Ae. Pa.	Réo	07-BFOJ-13		
		Ro.	Réo	07-BFOJ-13		

Le. = leaves; Tw. = twigs; Ba. = bark; Ro. = roots; Ae.Pa. = aerial parts; Wh.Pl. = whole plant.

*parum*. The plants were selected using an ethnopharmacological approach: we first identified and listed the plants used by local people for the traditional healing of malaria. In this West African country, the WHO estimated up to 3.5 million cases of malaria in 2008 – for a population of 15 million inhabitants – with 50% of cases being observed in children under 5 years age (WHO, 2009). Malaria thus has a major socio-economic impact and is also associated with mortality (about 8000 deaths in 2008) and a high incidence of serious complications, mainly in children under 5 years of age. The use of traditional medicine and plants is still widespread in the country for malaria health care management, often in association with modern treatment, especially chloroquine and paracetamol (Beiersmann et al., 2007).

Ethnopharmacological investigations conducted in Burkina Faso allowed us to identify 72 vegetal species used in traditional medicine to treat malaria. From these 72 species, we selected 13 plants with the intention of investigating their antiplasmodial properties. Selection was mainly based on our ethnopharmacological data but also on bibliographic data collected for the listed species. Chemotaxonomic links with species from the same genus or family already known to exhibit antiplasmodial properties and the possibility of local valorization of active and non-toxic plants in “Improved Traditional Medicine” (ITM) was also considered (Jansen et al., 2008). Finally, 17 samples obtained from the 13 selected species were collected in different areas of Burkina Faso (Table 1).

The aim of this study was firstly, to evaluate the antiplasmodial activity of extracts obtained from the 13 selected plants in order to identify active species to be investigated in further studies and secondly, to (un)confirm traditional use of these medicinal plants against malaria.

Dichloromethane, methanol and aqueous crude extracts were prepared for each plant sample and their *in vitro* antiplasmodial activity was evaluated on the *Plasmodium falciparum* 3D7 chloroquine-sensitive strain using a colorimetric method (Kenmogne et al., 2006). The most active plant extracts were then tested for *in vitro* activity on the *Plasmodium falciparum* W2 chloroquine-resistant strain and their selectivity index was also evaluated using the WST-1 assay on WI-38 normal human foetal lung fibroblasts.

## 2. Materials and methods

### 2.1. Plant material

For the 13 selected plant species, a total of 17 vegetal samples (leaves, twigs, bark, aerial parts, roots and whole plants, depending on the species) were collected in different areas of Burkina Faso (Centre-West region and Cascades region), between June 2007 and October 2007. The samples were authenticated by the Botany Department Staff of the “UFR/SVT, Université de Ouagadougou” in Burkina Faso and confirmed by the National Botanic Garden of Belgium (Meise). Voucher specimens were deposited at the National Botanic Garden of Belgium at Meise and at the Herbarium of the Laboratory of Pharmacognosy at the University of Liège (Belgium). Details are shown in Table 1 for each sample. The samples (mainly leaves and aerial parts) were washed, dried in a ventilated room and then powdered.

### 2.2. Crude extracts

Dichloromethane and methanol crude extracts were obtained by macerating 5 g of dried plant powder (leaves, twigs, barks, aerial parts, etc.) three times with 50 ml of solvent, under shaking for 30 min. The preparation was filtered and evaporated under reduced pressure.

For the aqueous crude extracts, we proceeded by decoction of 5 g of dried plant powder in 150 ml distilled water for 1 h in order to try to approximate the traditional preparation method. The preparation was filtered and freeze dried.

Three extracts were finally obtained for each sample with a total of 51 extracts.

### 2.3. Antiplasmodial assays

#### 2.3.1. Culture

Continuous cultures of the *Plasmodium falciparum*, chloroquine-sensitive (3D7) and chloroquine-resistant (W2) strains were maintained following the method of Trager and Jensen (1976), as described by Frédérick et al. (2002). Both strains were obtained from Prof. Grellier (“Museum National d’Histoire Naturelle” in Paris, France).

#### 2.3.2. Assays

Each extract was first dissolved in DMSO (Sigma) to a concentration of 10 mg/ml. *Plasmodium falciparum* culture was placed in contact with a set of 8 twofold dilutions of each extract in medium (final concentrations ranging from 0.8 to 100 µg/ml and final DMSO concentration ≤ 1%) on 2 columns of a 96-well microplate for 48 h. Parasite growth was estimated by colorimetric revelation (630 nm) based on the measurement of the plasmodial lactate dehydrogenase activity according to the method of Makler et al. (1993) and as described previously by Kenmogne et al. (2006). Artemisinin (Sigma–Aldrich) and chloroquine (Sigma–Aldrich) were used as standards, and infected and uninfected erythrocytes were added as positive and negative controls, respectively.

IC<sub>50</sub> values, indicating the concentration of the drug needed to obtain 50% inhibition of parasite growth, were calculated by linear regression from a set of eight concentrations tested for each extract. Each extract was tested in triplicate ( $n = 3$ ).

### 2.4. Cytotoxicity assays

#### 2.4.1. Culture

WI-38 normal human foetal lung fibroblasts were maintained in continuous culture in DMEM medium (Bio Whittaker) in a humid atmosphere at 37 °C and 5.5% CO<sub>2</sub>. Each medium was supplemented with 10% heat-inactivated foetal bovine serum (Bio Whittaker), 1% L-glutamine (200 mM) (Bio Whittaker) and antibiotics: penicillin (100 UI/ml) – streptomycin (100 µg/ml) (Pen-strep<sup>®</sup>, Bio Whittaker).

#### 2.4.2. Assays

Each extract was first dissolved in DMSO (Sigma) to a concentration of 10 mg/ml. For the assays, 96-well tissue culture microplates (Micro Test-96<sup>®</sup>, Falcon, Becton-Dickinson) were seeded with 200 µl medium containing 8000 cells in suspension.

After 24 h incubation, cells were treated with 6 dilutions with final concentrations of crude extracts in culture medium of 1, 5, 10, 25, 50 and 100 µg/ml and a final DMSO concentration ≤ 1%.

After 48 h incubation, cell viability was determined by adding WST-1 (Roche Biomolecular) tetrazolium salt as a cytotoxicity indicator and by reading absorbance at 450 nm with a scanning multiwell spectrophotometer after about a 1-h wait. Tetrazolium salts are cleaved to formazan dye by cellular enzymes. The absorbance directly correlates to the viable cell number. Each condition was reproduced in triplicate and each set of tests was performed twice. Camptothecin (Sigma–Aldrich) was used as a positive cytotoxic control. IC<sub>50</sub> values, indicating the concentration of the drug needed to obtain 50% inhibition of cell growth, were calculated by linear regression from a set of six concentrations tested for each extract.

**Table 2**  
Antiplasmodial activity (IC<sub>50</sub>) of the 51 extracts tested against the *Plasmodium falciparum* 3D7 strain.

Species (family)	Samples	CH <sub>2</sub> Cl <sub>2</sub> extracts IC <sub>50</sub> 3D7 ± S.D. (μg/ml)	MeOH extracts IC <sub>50</sub> 3D7 ± S.D. (μg/ml)	H <sub>2</sub> O extracts IC <sub>50</sub> 3D7 ± S.D. (μg/ml)	Yield CH <sub>2</sub> Cl <sub>2</sub> extracts	Yield MeOH extracts	Yield H <sub>2</sub> O extracts
<i>Bauhinia rufescens</i> Lam. (Fabaceae)	Leaves	52.96 ± 7.43	>100	>100	2.2%	24.2%	27.1%
	Twigs	>100	>100	>100	0.7%	7.1%	16.7%
<i>Bergia suffruticosa</i> Fenzl. (Elatinaceae)	Whole plant	<b>19.53 ± 2.50</b>	>100	>100	1.2%	12.7%	15.8%
<i>Boswellia dalzielii</i> Hutch. (Burseraceae)	Leaves	<b>41.01 ± 11.30</b>	<b>18.85 ± 1.93</b>	>100	4.7%	24.7%	33.4%
<i>Crossopteryx febrifuga</i> Benth. (Rubiaceae)	Leaves	56.85 ± 5.15	>100	>100	4.0%	30.0%	31.9%
<i>Dicoma tomentosa</i> Cass. (Asteraceae)	Whole plant	<b>7.90 ± 0.26</b>	<b>7.04 ± 1.15</b>	<b>21.87 ± 6.33</b>	3.9%	4.1%	8.3%
<i>Dyschoriste perrottetii</i> O. Kuntze (Acanthaceae)	Aerial parts	>100	>100	>100	2.6%	7.3%	4.8%
<i>Ficus thonningii</i> Blume (Moraceae)	Leaves	58.74 ± 9.65	>100	>100	1.8%	10.6%	25.6%
<i>Gardenia sokotensis</i> Hutch. (Rubiaceae)	Leaves	<b>14.01 ± 3.60</b>	<b>27.62 ± 7.83</b>	>100	9.1%	17.9%	23.3%
<i>Jatropha gossypifolia</i> L. (Euphorbiaceae)	Leaves	<b>35.66 ± 2.86</b>	87.65 ± 7.84	>100	2.8%	8.8%	27.5%
<i>Loeseneriella africana</i> N. Hallé (Celastraceae)	Leaves	>100	>100	>100	2.9%	13.2%	30.0%
<i>Psorospermum senegalense</i> Spach. (Clusiaceae)	Leaves	<b>10.03 ± 2.00</b>	80.46 ± 13.61	>100	2.8%	22.3%	24.3%
<i>Vitellaria paradoxa</i> Gaertn. (Sapotaceae)	Bark (Godin)	<b>43.94 ± 13.44</b>	78.11 ± 12.25	>100	3.7%	17.1%	16.2%
	Bark (Reo)	81.24 ± 10.19	>100	>100	4.1%	19.1%	16.8%
	Leaves (Reo)	55.45 ± 12.60	>100	>100	5.4%	22.6%	25.2%
<i>Waltheria indica</i> L. (Sterculiaceae)	Aerial parts	<b>33.73 ± 13.44</b>	57.21 ± 11.64	>100	1.3%	10.5%	23.8%
	Roots	<b>29.71 ± 6.23</b>	62.09 ± 14.80	>100	0.6%	15.3%	16.2%

Artemisinin: IC<sub>50</sub> = 0.0067 ± 0.0018 μg/ml; chloroquine: IC<sub>50</sub> = 0.016 ± 0.004 μg/ml. Values in bold show a significant level of activity.

#### 2.4.3. Selectivity index (SI)

The SI value allows the evaluation of the toxicity impact of the extracts against normal human cells compared to the toxicity against the parasite and allows the assessment of the selectivity of extracts for the parasite. The SI is calculated as the ratio between cytotoxic IC<sub>50</sub> values and 3D7 or W2 parasitic IC<sub>50</sub> values.

### 3. Results

The 17 vegetal samples obtained from 13 selected plants traditionally used against malaria were extracted by dichloromethane, methanol and water to give 51 extracts. These extracts were first tested for activity against the *Plasmodium falciparum* 3D7 strain. Antiplasmodial results are presented in Table 2.

In line with WHO guidelines and with previous results from our team (Jonville et al., 2008; Philippe et al., 2005; Pink et al., 2005), antiplasmodial activity was classified as follows:

IC<sub>50</sub> ≤ 15 μg/ml: promising activity; IC<sub>50</sub> = 15–50 μg/ml: moderate activity; IC<sub>50</sub> > 50 μg/ml: weak activity, but at a level that could not explain the existence of antiplasmodial activity in the plant; IC<sub>50</sub> > 100 μg/ml: inactivity.

**Table 3**  
Antiplasmodial activity against *Plasmodium falciparum* 3D7 and W2 strains and cytotoxicity on WI-38 normal human fibroblasts, for the most active species.

Species	Extracts	IC <sub>50</sub> WI-38 ± S.D. (μg/ml)	IC <sub>50</sub> 3D7 ± S.D. (μg/ml)	SI <sub>3D7</sub> <sup>a</sup>	IC <sub>50</sub> W2 ± S.D. (μg/ml)	SI <sub>W2</sub> <sup>a</sup>
<i>Dicoma tomentosa</i> Cass. (Asteraceae)	CH <sub>2</sub> Cl <sub>2</sub>	17.03 ± 3.01	7.90 ± 0.26	2.2	6.34 ± 1.49	2.7
	MeOH	15.96 ± 3.53	7.04 ± 1.15	2.3	7.25 ± 1.68	2.2
<i>Gardenia sokotensis</i> Hutch. (Rubiaceae)	CH <sub>2</sub> Cl <sub>2</sub>	12.67 ± 1.13	14.01 ± 3.60	0.9	6.75 ± 1.29	1.9
<i>Psorospermum senegalense</i> Spach. (Clusiaceae)	CH <sub>2</sub> Cl <sub>2</sub>	36.39 ± 6.86	10.03 ± 2.00	3.6	3.63 ± 0.49	10.0

Standard drugs: artemisinin (3D7): IC<sub>50</sub> = 0.0067 ± 0.0018 μg/ml; artemisinin (W2): IC<sub>50</sub> = 0.0034 ± 0.0009 μg/ml; chloroquine (3D7): IC<sub>50</sub> = 0.016 ± 0.004 μg/ml; chloroquine (W2): IC<sub>50</sub> = 0.35 ± 0.08 μg/ml; camptothecin (WI-38): IC<sub>50</sub> = 0.029 ± 0.011 μg/ml.

<sup>a</sup> Selectivity index is calculated by SI = IC<sub>50</sub>(WI-38)/IC<sub>50</sub>(3D7 or W2, respectively).

Thirteen extracts obtained from 8 different plants displayed antiplasmodial activity (IC<sub>50</sub> < 50 μg/ml). The dichloromethane extracts were generally more active than the methanol and water extracts. Five species showed moderate activity (15 μg/ml < IC<sub>50</sub> < 50 μg/ml): *Boswellia dalzielii* (leaves), *Waltheria indica* (roots and aerial parts), *Bergia suffruticosa* (whole plant), *Vitellaria paradoxa* (bark) and *Jatropha gossypifolia* (leaves). The best results were obtained with extracts from the *Dicoma tomentosa* whole plant, from *Psorospermum senegalense* leaves and from *Gardenia sokotensis* leaves. These extracts displayed promising antiplasmodial activity, with IC<sub>50</sub> values ranging from 7.0 to 14.0 μg/ml. Among active plants, the *in vitro* antiplasmodial activity of *Jatropha gossypifolia* was previously reported by Gbeassor et al. (1989) and *Gardenia sokotensis* was also reported to exhibit *in vivo* antiplasmodial activity by Traoré et al. (2006).

However, the present study constitutes the first report of antiplasmodial activity against the *Plasmodium falciparum* of *Dicoma tomentosa*, *Psorospermum senegalense* and also of *Boswellia dalzielii*, *Bergia suffruticosa*, *Waltheria indica* and *Vitellaria paradoxa*. *Dicoma tomentosa* was the most active species on the 3D7 strain and the only one for which the aqueous extract showed significant activity against *Plasmodium falciparum*.

The four most active extracts ( $IC_{50} < 15 \mu\text{g/ml}$ ), obtained from three different plant species, were tested for *in vitro* activity against the *Plasmodium falciparum* W2 chloroquine-resistant strain, in order to confirm their activity. All four extracts were at least as active against this second strain.

The selectivity of those extracts was then evaluated using the WST-1 cytotoxicity assay on normal human foetal lung fibroblasts in order to check that their toxicity was specific to the parasite. The impact of the toxicity was established by analysing the selectivity index (SI) values. Only *Psorospermum senegalense* showed some selectivity ( $SI > 3$ ), while *Gardenia sokotensis* dichloromethane extract showed an  $SI_{(WI-38/3D7)} < 1$ .

Additional results obtained with the most active extracts are summarized in Table 3.

#### 4. Discussion

Eight of the 13 selected plants used for traditional malaria healing in Burkina Faso exhibited promising to moderate antiplasmodial activity against *Plasmodium falciparum*, the causative agent of the disease. These findings provide some evidence underlying the traditional use of these plants as antimalarials. However, it is not so easy to draw conclusions from *in vitro* results in order to (un)confirm the use of plants in traditional medicine (Gertsch, 2009), particularly in the field of malaria, a disease which involves many different symptoms and which has a complex physiopathology. Indeed, several plants that are frequently reported to be used as antimalarials in different countries do not necessarily show high activity in *in vitro* screening.

For the 8 active plants in this study, the detected antiplasmodial activity allows the validation, at least partially, of the traditional use of these plants for malaria symptom management. For the inactive species, we cannot conclude categorically to their uselessness in traditional malaria healing.

In fact, several elements might explain the absence of activity for some of the evaluated extracts. Firstly, the *in vitro* test model used in this study reproduces the erythrocytic development stage of the parasite but some plants could also be active against the liver phase of *Plasmodium* development, as shown by Carraz et al. (2006) and documented in a review by Bourdy et al. (2008). This activity against hepatic schizonts could not be detected with our *in vitro* model but such plants could however act as prophylactics as well as treatment of the dormant hypnozoites which can cause relapses sometimes several months after the first infection (*Plasmodium ovale*, *Plasmodium vivax*).

Moreover, traditional remedies often consist of a combination of several different plants and some plants used in the treatment could have therapeutic effects other than an antiparasitic effect, such as antipyretic, analgesic, hepatoprotective, anti-inflammatory or immunomodulatory effects. The different constituents of the mixture can exert a synergistic action against several associated symptoms of malaria, and this synergy has an impact on the patient's recovery. Finally, plants could be effectively more active against *Plasmodium falciparum* in man than *in vitro*, as is the case for plants containing prodrugs. Prodrugs are non-active by themselves and need a metabolization step in order to become active drugs; they cannot therefore be detected in an *in vitro* test model. This could also explain why dichloromethane extracts are generally more active *in vitro* than aqueous extracts; these extracts are even closer to the remedies produced by the traditional preparation method, but contain polar compounds such as heterosides. Like prodrugs, it is possible that these heterosides are inactive *in vitro* but active *in vivo* after metabolization, and that dichloromethane extract contains the corresponding aglycone, which may be capable of being active *in vitro* without the need of a metabolization step.

##### 4.1. *Bauhinia rufescens* Lam.

A decoction of *Bauhinia rufescens* leafy stems is used in traditional medicine in Burkina Faso to treat malaria and fevers. In the present study, no significant activity was found in any extracts obtained from either sample collected for this species. There is no previous report of the antiplasmodial properties of this plant, although other *Bauhinia* species (*Bauhinia guianensis*, *Bauhinia malabrica*) have been found to be active in different studies (Kittakoop et al., 2000; Muñoz et al., 2000).

##### 4.2. *Bergia suffruticosa* Fenzl.

The whole plant is traditionally used in Burkina Faso to treat childhood malaria, and it is also used as a tonic. Our results showed that only dichloromethane extract was moderately active against *Plasmodium falciparum*. The antiplasmodial properties of this plant have been highlighted for the first time in this study. The Elatiaceae family is not very widely studied in the literature. However, *Bergia suffruticosa* is known to exhibit free-radical scavenging activity (Anandjiwala et al., 2007).

##### 4.3. *Boswellia dalzielii* Hutch.

Decoctions of the leaves and bark of this plant are used for the traditional treatment of malaria, fevers and inflammation. We found methanol leaf extract to be more active against *Plasmodium falciparum* than dichloromethane extract. This study is the first report of the antiplasmodial properties of *Boswellia dalzielii*. The resin of this tree has been shown to exhibit anti-inflammatory activity (Duwiejuwa et al., 1993), while stem bark extracts have been shown to possess antioxidant and antibacterial properties (Alemika et al., 2006).

##### 4.4. *Crossopteryx febrifuga* Benth.

The leaves of this plant are used (often in combination with other plants) in Burkina Faso and West Africa to treat malaria and related symptoms such as fevers and icterus but also to treat many different disorders. No significant *in vitro* antiplasmodial activity was detected in our tests for this species. Antiplasmodial properties of this plant have previously been described *in vitro* for alkaloid leaf extract ( $IC_{50} < 10 \mu\text{g/ml}$ ) by Sanon et al. (2003) and also *in vivo* by Elufioye and Agbedahunsi (2004) for ethanol stem bark extract. However, to our knowledge, no active compound has been isolated to date. The lack of activity observed in our study could be explained by the fact that we tested crude extracts and not alkaloid extracts and/or that we tested only leaf extracts (and not stem bark) against an *in vitro* model (and not *in vivo*).

##### 4.5. *Dicoma tomentosa* Cass.

Dichloromethane and methanol *Dicoma tomentosa* whole plant extracts were found to be the most active against the *Plasmodium falciparum* 3D7 strain. This was the only species for which the aqueous extract (the nearest to the method of preparation of traditional remedies) also showed moderate antiplasmodial activity. Activity of *Dicoma tomentosa* was confirmed on the W2 chloroquine-resistant strain. This study is the first report on the antiplasmodial properties of this plant. Named "gômtingda" in the local language (Mooré), this Asteraceae is mainly used for the traditional treatment of malaria in adults and children in Burkina Faso. The plant is known to contain flavonoids (Aqil et al., 1998, 2001; Aqil and Khan, 1999a,b) and sesquiterpen-lactones (Bohlmann et al., 1982; Krishna et al., 2003; Zdero and Bohlmann, 1990). There

are no data concerning antiplasmodial or any other pharmacological properties of this plant or isolated compounds, but another species, *Dicoma anomala*, has already been reported to possess antiplasmodial properties (Matsabisa et al., 2006).

#### 4.6. *Dyschoriste perottetii* O. Kuntze

A decoction of the aerial parts of this plant is used in traditional medicine in Burkina Faso for the treatment of childhood malaria. All extracts were found to be inactive against *Plasmodium falciparum* in our tests. Few data are available regarding this plant and there is no previous report of its antiplasmodial properties. The plant's antioxidant properties were identified by Sawadogo et al. (2006), who also reported its use in fever, malaria and diarrhoea and its rich content of phenolic compounds and flavonoids.

#### 4.7. *Ficus thonningii* Blume

The leaves of this tree are traditionally used in Burkina Faso to treat malaria and related symptoms such as fever, icterus and anaemia but also pernicious malaria. No significant antiplasmodial activity against *Plasmodium falciparum* was detected for this plant in our study. Few data are available regarding this plant and there is no previous report of antiplasmodial properties. However, other *Ficus* sp. have been found to be active as shown *in vivo* by Muregi et al. (2007) for *Ficus sur* (leaves and stem barks) and also *in vitro* by Sanon et al. (2003) for leaf extract of *Ficus sycomorus*.

#### 4.8. *Gardenia sokotensis* Hutch.

Dichloromethane and methanol extracts of *Gardenia sokotensis* leaves showed, respectively, promising and moderate antiplasmodial activity but dichloromethane extract also exhibited a high cytotoxic activity ( $SI_{(WI-38/3D7)} = 0.9$ ). The leaves of this bush tree are used in traditional remedies (often in combination with other plants) in Burkina Faso to treat malaria and some related symptoms such as fevers, weakness, icterus and gastrointestinal disorders.

Few bibliographic data are available regarding this plant but its antiplasmodial properties have already been described by Traoré et al. (2006) with an *in vivo* model of *Plasmodium berghei*. The same study showed the presence of flavonoids, triterpens and carotenoids in the plant, but the active compounds were not identified. Another *Gardenia* species (*Gardenia saxatilis*) has been shown to contain antiplasmodial triterpens (Suksamrarn et al., 2003).

#### 4.9. *Jatropha gossypifolia* L.

This plant is used in traditional medicine to treat malaria and fevers but also various other disorders. Only the dichloromethane extract of our leaf sample was found to be moderately active against *Plasmodium falciparum*. Hot water extract of *Jatropha gossypifolia* leaves has already been shown to exhibit *in vitro* activity against *Plasmodium falciparum* (Gbeassor et al., 1989) for a specimen collected in Togo. The fact that no activity was detected in our aqueous extract could be explained by a different place of collection and/or a different experimental methodology (e.g. proceeding by working on patients' blood and counting parasites).

Many different compounds have been isolated from this plant (mainly terpenoids, lignans, alkaloids, flavonoids, and amino and fatty acids), as reviewed by Kakade et al. (2008). However, to our knowledge, there has been no study on the antiplasmodial properties of isolated compounds from *Jatropha gossypifolia*.

Other pharmacological properties such as antimicrobial, anticoagulant and anticancer activity have been detected for some crude extracts and isolated compounds (Kakade et al., 2008).

#### 4.10. *Loeseneriella africana* (Willd.) N. Hallé (syn. *Hippocratea africana* Loes ex Engl.)

The leaves and roots of this plant are used in traditional medicine by the local population in Burkina Faso to treat malaria and fevers. All extracts (leaves) were found to be inactive against *Plasmodium falciparum* in our tests. However, *in vivo* antiplasmodial properties against *Plasmodium berghei* were previously shown by Okokon et al. (2006) for ethanol roots extract obtained from *Hippocratea africana*, a botanical synonym for *Loeseneriella africana*, but no active compounds were isolated.

#### 4.11. *Psorospermum senegalense* Spach.

Our results showed that dichloromethane extract of *Psorospermum senegalense* leaves demonstrated promising activity against both strains. This extract was found to be the most active on the W2 chloroquine-resistant strain and showed a good  $SI_{(WI-38/W2)}$  equal to 10.

This plant is traditionally used to treat malaria and also different disorders (particularly skin disorders but also epilepsy) in some West African countries. There is no previous report of the antiplasmodial properties of this plant, which, to date, has not been much studied. Recently, anticonvulsive properties were found in ethanol extract from the roots of this plant (Pedersen et al., 2009). We found no phytochemical studies in the literature for this species. However, there is a great deal of ambiguity in the literature regarding the botanical name of this plant. Indeed, *Psorospermum guineense* Hochr. and *Vismia guineensis* (L.) Choisy have sometimes been reported as botanical synonyms for *Psorospermum senegalense* Spach. Nevertheless, according to some specialists in botany (National Botanic Garden of Belgium, Meise) consulted in our study, *Vismia guineensis* cannot be considered as a synonym. Several (bi)anthrones, anthraquinones, flavonoids, benzophenones, vismiones, xanthonones and derivatives have been previously identified from *Vismia guineensis* (as well as other *Vismia* and/or *Psorospermum* species). Some of these compounds exhibit cytotoxic activity (Botta et al., 1986; Bilia et al., 2000; Poli et al., 2004). Vismione H, isolated from *Vismia guineensis* has been found to show promising *in vitro* activity on *Plasmodium falciparum* (François et al., 1999).

#### 4.12. *Vitellaria paradoxa* Gaertn. (syn. *Butyrospermum paradoxum* Hepper)

The present study is the first report of antiplasmodial properties for this tree, which is widespread in the area. Decoctions of leaves and barks are traditionally used in Burkina Faso to treat malaria, fever and many other disorders such as pain, inflammation, and gastrointestinal disorders. The antibacterial and antifungal properties of *Vitellaria paradoxa* have been demonstrated by Ogunwande et al. (2001) for different parts of this plant. In our study, the best results were obtained with dichloromethane extracts. Only the barks collected in Godin showed antiplasmodial properties while those collected in Reo were almost inactive. Leaves obtained from this last specimen showed weak activity against *Plasmodium falciparum*. The difference between the antiplasmodial results obtained for extracts prepared in the same way from two different plant parts of the same specimen and/or for the same plant part but collected on two geographically different specimens reflects the differences between the different samples in the phytochemical content of bioactive compounds.

#### 4.13. *Waltheria indica* L.

Aerial parts and roots of this plant are used in traditional medicine in Burkina Faso to treat malaria, fevers, icterus and

various disorders (particularly gastrointestinal and respiratory disorders). In the present study, both tested plant parts showed comparable antiplasmodial activities: both dichloromethane extracts were moderately active, while both methanol extracts were weakly active against *Plasmodium falciparum*. This study constitutes the first report of antiplasmodial properties in this plant.

However, some extracts obtained with *Waltheria indica* were found to be inactive by Clarkson et al. (2004) with an  $IC_{50} > 100 \mu\text{g/ml}$  for a specimen collected in South Africa. The discordance between results obtained with different samples from the same plant species could be explained by various parameters such as localization, period of collection and method of extract preparation, which could modify the phytochemical content and therefore the pharmacological response of a particular species. Previous phytochemical studies have revealed the presence of various flavonoids and polyphenolic compounds in the plant (Petrus, 1990; Rao et al., 2005; Maheswara et al., 2006). The plant and some isolated compounds have been described as exhibiting anti-inflammatory and antibacterial activity (Rao et al., 2005; Maheswara et al., 2006).

## 5. Conclusion and perspectives

The present study allowed us to identify the antiplasmodial activity of some extracts obtained from 8 plants out of the 13 tested and constitutes the first report of antiplasmodial activity against *Plasmodium falciparum* for crude extracts obtained from *Dicoma tomentosa*, *Psorospermum senegalense*, *Boswellia dalzielii*, *Bergia suffruticosa*, *Waltheria indica* and *Vitellaria paradoxa*.

*Dicoma tomentosa* (Asteraceae) and *Psorospermum senegalense* (Clusiaceae) combine the best antiplasmodial activity against two *Plasmodium falciparum* strains and an acceptable cytotoxicity against normal human cells. As such, these two plants could be selected as the best candidates for further investigations in the field of new antimalarial drug discovery as well as valorization of the use of these plants in traditional medicine.

Further pharmacological and phytochemical studies regarding these active plants are needed. In our own future studies, we plan to focus on the evaluation of the *in vivo* activity of these plants against a mouse model of *Plasmodium berghei*, on the isolation and identification of active compounds through a bioguided fractionation and on the study of the activity and toxicity of these active isolated compounds. From these additional results, we will also consider the possibilities of local valorization of active and non-toxic plants in standardized Improved Traditional Medicine.

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