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Antiplasmodial Effect of Commelina benghalensis/Steganotaenia araliacea Plants Extract on the Human Population in Ngaoundere (Cameroon)

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The study was conducted in the laboratories of the Provincial hospital and the University of Ngaoundere. The study was to assess the frequencies and the parasites densities of *Plasmodium* spp. and the effects of leaves and bark extract of Commelina benghalensis/Steganotaenia araliacea on the malaria parasite. Blood was sampled from patients coming to the hospital for examination. They were submitted for treatment with the plants extract for ten days and with amodiaquine for three days. Sampling on 109 patients revealed that the infestation due to *Plasmodium falciparum* and *Plasmodium malariae* was 73.4%. The results indicated that malaria is endemic in Ngaoundere where the average infestation is 74.3% and where Plasmodium falciparum is the main causative agent. The infestation varied from one quarter to another with elevated values in Haoussa quarter. The parasite density had the same tendency as that of the infestation. Treatment of patients with a mixture of Commelina benghalensis/Steganotaenia araliacea extract revealed a progressive reduction of infestation up to a total dispiriting of the parasites after 10 days. Five days after the beginning of treatment of patients with plants extract, the average parasite density was significantly reduced (p<0.05), whatever were the resident quarter, the sex and age of patient. When patients were treated with amodiaguine, all the parasites were eliminated 3 days after the beginning of treatment.

Key words: Parasite infestation, parasite density, *Steganotaenia araliaceae*, *Commelina benghalensis*

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INTRODUCTION

Protozoa are the causative agents of numerous pathologies in the tropical zone (Eisenhauer et al., 2002). Malaria is one of the pathologies and is responsible for death of people all over the world and in the tropical zones (Adebayo and Krettli, 2011; Eisenhauer et al., 2002; Greenwood et al., 2005; Winter et al., 2006). Malaria affects a large proportion of the world population causing elevated rates of mortality and morbidity (Eisenhauer et al., 2002). The seriousness of the consequences and the epidemicity due to malaria are related to the abundance and the quality of lodging larvae (Bakwo et al., 2010). According to the physiologic status of the patients, malaria manifestations take several aspects (Autino et al., 2012). Patients have developed efficient mechanisms against the parasite invasion which in turn have found means of escaping the humorous host responses (Rang et al., 2002). Plasmodium falciparum is the species responsible for acute malaria and mostly concerned with chemo-resistance (Odhiambo and Odulaja, 2005). The species are frequent to children below 5 years and in pregnant women. The chemo-resistance corresponds to the adaptation of plasmodia to a particular drug like chloroquine for example and can result to an uncontrolled drug cure, an extra auto-medication, or ignorance of medical prescriptions (Randrianarivelojosia et al., 2003). The eradication strategy of malaria defined by who from 1955 to 1969, soon becoming a dogma, was revealed by the mobility means inefficient (Trape and Rogier, 1996). The control of malaria in Africa is prevented by the lack of financial means and mostly the inappropriate hospital structures. efficient medicinal plants allowed the rural population to orient themselves towards traditional treatment (Mesfin et al., 2012; Newman et al., 2003). Hence, many plants extracts are used according to regions to cure different parasitosis (Gathirwa et al., 2008; Gathirwa et al., 2011; Moundipa et al., 2005; Muthaura et al., 2007). In this case, the study was undertaken at the hospital to assess the efficacy of Commelina benghalensis/Steganotaenia araliacea plants extract as anti-malaria on the population of Ngaoundere town.

MATERIALS AND METHODS

Study area: The study was carried out in Ngaoundere environs; both in town and at the University. Ngaoundere is the headquarter of the Adamawa Region and also the Major town in Vina Division. Ngaoundere is a touristic town embroidered with many attraction sites (Lac Tyson, Ranch of Ngaoundaba, Mount Ngaoundere, lamidat of Ngaoundere). It is situated between 7 and 10 degrees Northern latitude and 11 and 15 degrees Eastern

longitude. Ngaoundere is situated at 1200 meters of altitude and represents a transition zone between the forest and the savannah. The climate is the sahelian type which rainfall pattern is monomodal. The rainy season lasts from April to October with annual rainfall of 1500 millimeters.

Subjects: Volunteers' patients with malaria followed up by a Traditional Healer in the city of Ngaoundere were used. These volunteers' patients came from different quarters named: Baladji 1, Centre town, Haoussa, Haut-plateau and Sabongari.

Thick film and Blood smear: Blood was sampled on a finger of each patient after sterilization with 70°C alcohol. A drop of blood was spread on a slide at 10-15 mm diameter for the thick film preparation. On another slide, a drop of blood was placed at 1/3 position for smear blood.

Staining and observation of smears: Slides were stained with 1/10 diluted Giemsa dye for 15 min and was then rinsed with tape water and let to dry off for 20 min at room temperature. Slides were observed with immersion oil under an ordinary microscope to determine the frequencies and parasite densities. When parasites were detected, they were identified in smear blood (Waldenstrom *et al.*, 2004; Trape, 1985).

Preparation of plants extract: Leaves and bark of *Commelina benghalensis* (30%) and bark of *Steganotaenia araliacea* (70%) were sampled in Ngaoundere. These plant samples were washed with water and dried at room temperature. It was then transformed into powder using a blinder. This powder was passed through a fine pores sieve to obtain a composite sample. The extract was obtained by mixing 200 g of plants powder in 2 L of boiling water for 30 min and then cooled down.

Treatments: One group (Group 1) of patients with malaria was treated with the plants extract of 250 mL twice a day (in the morning and afternoon) for 10 days. A second group of patients (Group 2) was submitted to amodiaquine treatment for 3 days. During these periods, patients were not supposed to take any other treatment. This was to ensure the accuracy of results. The decoction was drunk at 9 am and 3 pm to respect the administration given by WHO.

Statistical analysis: The statistical comparison of parasite density before and after treatment of patients was realized using an analysis of variance. The different prevalence between parasite species and the infestation rate were determined using Fisher exact test two tails. From $p \le 0.05$, the difference was considered significant.

RESULTS

Species of *Plasmodium*: From 109 patients examined, 80 were revealed to be infested with parasites of malaria (*Plasmodium falciparum* and *Plasmodium malariae*). The infestation rate was 73.4%. In addition, *Plasmodium falciparum* was involved in 80% of malaria cases, while *Plasmodium malariae* involved only 20% (Table 1).

Infestation rate in studied areas in Ngaoundere

Infestation rate before treatment: Examination of patients in the Regional hospital of Ngaoundere during survey has shown that the infestation frequency is high and do not significantly varied from one quarter to another. Thus, the lower infestation rate was registered in quarters such as Sabongari (66.67%), Haut-plateau (68.75%), Centre town quarter (70.83%), Baladji 1 (73.08%) and the inhabitants of Haoussa quarter (84.00%) had the highest infestation rate, though the difference was not significant (Table 2). The infestation rate very slightly increased in female (75.86%)

Tablel: Number of parasitized patients at the Regional hospital of Ngaoundere, from May to August 2006

	No. of patients	Percentage of patients
Plasmodium species	with parasites	with parasites
Plasmodium falciparum	64	80
Plasmodium malariae	16	20
Total	80	100

of infestation) when compared to male (70.59% of infestation), Table 3. The frequencies of infestation decrease with the age of patients. Patients of more than 30 years old were slightly less infested (70.0%) than those with ages varying from 0 to 15 years (85.71%) (Table 4).

Infestation rate after treatment: Patients treated with amodiaguine were found without any parasites after 3 days of treatment, their infestation rate was zero. After 10 days of treatment with the plants extract, parasites were eliminated differently according to the quarter, sex and age. In the Haut Plateau quarter, parasites eliminated from all the individuals treated with Commelina benghalensis and Steganotaenia araliacea extract. The infestation rate was also zero. In the four other quarters, the infestation rate after 10 days of treatment was 26.67% for Haoussa, 9.09% for Centre Town, 22.22 for Baladji 1 and 100% for Sabongari Table 2. So, all patients from Sabongari were still parasitized. According to sex there was no significant difference of the infestation rate between female (30.00%) and male (36.84%) after 10 days of treatment with the plants extract (Table 3). Controversially, the extract of Commelina benghalensis and Steganotaenia araliacea treated 100% of patients of less than 15 years old, 55.56% of patients between 15 to 30 years old and 84.00% of patients of more than 30 years old (Table 4).

Table 2: Infestation rate in different quarters and the effect of the plants decoction

				No. of patients with parasites	No. of patients with parasites
		No. of patients		before treatment with the plant	after 10 days of treatment
Quarters	Total no. of patients	with parasites	Percentage of patients with parasites	with parasites after	with the plant
Baladji 1	26	19	73.08	9	2°
Centre town	24	17	70.83	11	1°
Haoussa	25	21	84	15	4°
Haut plateau	16	11	68.75	4	O_c
Sabongari	18	12	66.67	9	9

The number of patients with parasites before treatment with the plant is compared to the number of patients with parasites after 10 days of treatment with the plant, °p<0.001, Fisher exact test two tails

Table 3: Infestation rate according to sex and the effect of the plants decoction

				No. of patients with parasites	No. of patients with parasites
		No. of patients		before treatment with the plant	after 10 days of treatment
Sex	Total no. of patients	with parasites	Percentage of patients with parasites	with parasites after	with the plant
Female	58	44	75.86	30	9°
Male	51	36	70.59	19	7°

The number of patients with parasites before treatment with the plant is compared to the number of patients with parasites after 10 days of treatment with the plant, \$\circ{c}{2} = 0.001\$, Fisher exact test two tails

Table 4: Infestation rate with age and the effect of the plants decoction

				No. of patients with parasites	No. of patients with parasites
		No. of patients		before treatment with the plant	after 10 days of treatment
Age (year)	Total no. of patients	with parasites	Percentage of patients with parasites	with parasites after	with the plant
0-15	14	12	85.71	6	Oc
15-30	55	40	72.72	18	8°
>30	40	28	70.00	25	4^{c}

The number of patients with parasites before treatment with the plant is compared to the number of patients with parasites after 10 days of treatment with the plant, \$\circ{c}{2} < 0.001\$, fisher exact test two tails

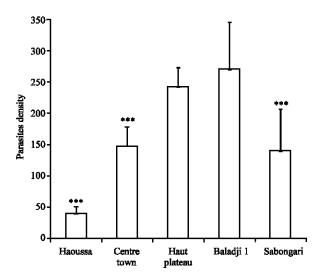


Fig. 1: Variation of parasite density in different quarters in Ngaoundere, The results are expressed as Mean±SEM. ***p<0.001 vs. Baladji 1, ANOVA followed by Dunnett (HSD)

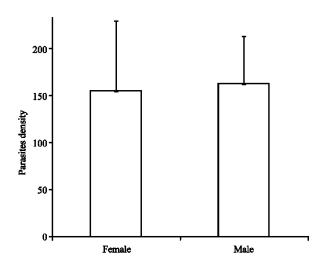


Fig. 2: Variation of parasite density in female and male in Ngaoundere, The results are expressed as Mean±SEM

Parasite density in studied areas in Ngaoundere: The parasite density varied from one quarter to another. Parasite densities (number of parasites per mL of blood) were 270 in baladji 1, 242 in haut plateau, 147 in centre town, 139 in sabongari and 40 in haoussa (Fig. 1). The parasite densities were the highest in patients from baladji 1 (270), haut plateau (242) and the lowest in haoussa (40). There was no difference in parasite densities between female and male (Fig. 2). Controversially, the parasite

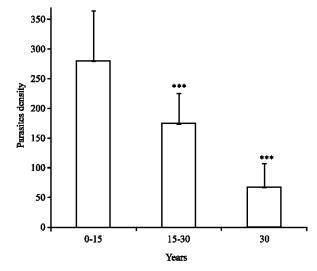


Fig. 3: Variation of parasite density with age in Ngaoundere, The results are expressed as Mean±SEM, ***p<0.001 vs. years below 15, ANOVA followed by Dunnett (HSD)

density decreased with age; 279.7 in patients under 15 years, 173.7 for patients between 15 and 30 years old and 67.1 in patients above 30 years old (Fig. 3).

Parasite densities after treatment: As expected, patients treated with amodiaquine had their blood without any parasites after the three days treatment. The extract of Commelina benghalensis and Steganotaenia araliacea also reduced efficiently the parasites densities of patients. The efficacy of the plants extract was 100% in Haut Plateau after 10 days of treatments. The percentage of reduction of parasites by the plants extract was 98.32% for Haoussa, 99.78% for Centre Town, 99.58% for Baladji 1 and 90.54 for Sabongari after 10 days of treatment (Fig. 4). The reduction of the densities parasites by the plants extract were the same for female (97.7%) and male (98.4%) (Fig. 5). All patients of less than 15 years old were completely treated with the plants extract after 10 days of treatment. In the same direction, the reduction of the densities parasites by the plants extract were the same for patients between 15 and 30 years old (97.60%) and patients above 30 years old (97.12%) after 10 days of treatment (Fig. 6). But after 5 days of treatment, the reduction of the densities parasites by the plants extract were 73.60, 72.60 and 57.50% for patients of less than 15 years, patients between 15 and 30 years old and patients above 30 years old, respectively.

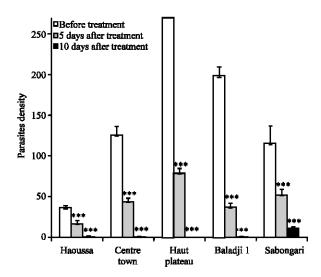


Fig. 4: Effect of the extract on the parasite density in different quarters in Ngaoundere. The results are expressed as Mean±SEM. In each quarter, parasite density before treatment is compare to parasite density after treatment. ***p<0.00 vs. before treatment, ANOVA followed by Dunnett (HSD)

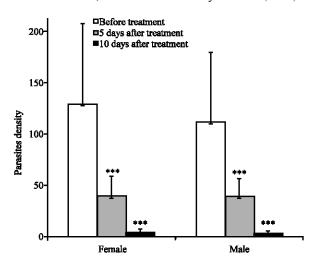


Fig. 5: Effect of the extract on the parasite density in female and male in Ngaoundere. The results are expressed as Mean±SEM. In each sex, parasite density before treatment is compare to parasite density after treatment. ***p<0.00 vs. before treatment, ANOVA followed by Dunnett (HSD)

DISCUSSION

The fact that more than 73% of patients were infested with *Plasmodium* revealed malaria is endemic in the region, since an endemic region is characterized by an

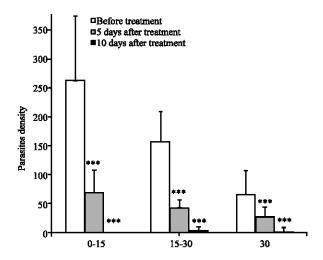


Fig. 6: Effect of the extract on the parasite density with age in Ngaoundere. The results are expressed as Mean±SEM. In each age subdivision, parasite density before treatment is compare to parasite density after treatment. ***p<0.00 vs. before treatment, ANOVA followed by Dunnett (HSD)

infestation rate higher than 50% (Felger et al., 1994; Killen et al., 2006; Nzevimana et al., 2002). This was found even in the hospital area where the infestation evaluation of children in Djamena (Chad) showed an infestation rate higher than 70% (Moore et al., 2012) or in towns like Brazzaville in Congo Ebogo and Yaounde in Cameroon (Fondjo et al., 1992; Peterson et al., 2009; Robert et al., 2003). The Involvement of Plasmodium falciparum in 80% of malaria cases, suggests that Plasmodium falciparum is significantly the main species responsible for malaria in the studied zone. Similar results were obtained by other authors (Eboumbou Moukoko et al., 2009; Nzeyimana et al., 2002)

The slight high infestation rate in Haoussa quarter could be related to the presence of more areas with permanent resting place of larvae and more insalubrities (Keiser et al., 2005; Njan Nloga and Messi, 2005). The infestation rate increased very slightly in female, probably because more women stay the whole day at home (not going outside neither for shopping nor for working) while more men go to their office where there is less mosquitoes. The frequencies of infestation decrease with the age of patients. Though mosquitoes prefer adults than children (Wanji et al., 2003; Le Hesran, 2000), children are more exposed to parasite than adults, since they can go away from their parents, walking around the house, at the same time; adults may have acquired partial

immunity (Nzeyimana et al., 2002; Richard et al., 1988). The reduction of the infestation rate in the patients who are more than 30 years old is similar with the results from others hospitals seen in Ebolowa Regional Hospital in Cameroon (Cot et al., 2003) and could be explained by the fact that people in endemic area of malaria progressively acquired immunity in *Plasmodium*.

The results show the efficiency of both amodiaquine (three days treatment) and the extract of Commelina benghalensis and Steganotaenia araliacea (ten days treatment) in destroying Plasmodium falciparum from patients' blood. Controversially, after 10 days of treatment with the plants extract, all patients from Sabongari were still parasitized. This result can be explained by the fact that these patients are among the less educated in the Region of Ngaoundere. They could not understand well the necessity of following very well the prescriptions of the extract administration. The 100% efficiency of the plant extract in treating patients of less than 15 years old, could be explained since children of this age are very well followed for their treatments by their parents, while that is not the case for young patients between 15 to 30 years.

Re-infestation and longer infestation duration could explain the high parasites density in patients from baladji 1 and haut plateau. The high rate infestation correlating with the low parasitemia in haoussa explained very well the fact that more the population is infected, less their parasitemia is (Nzeyimana et al., 2002; Richard et al., 1988). The decrease with age of the parasite density could be understood since people above 30 years old know and recognize the malaria symptoms. So they would go to the hospital at the beginning of the manifestation of malaria. And also, people from 30 years old possess at least partial immunization (Nzeyimana et al., 2002). The efficiency of the plants extract in the treatment of patients of less than 15 years could be explained since children of this age have not yet developed any resistance to plasmodium parasites and are in addition very well followed for their treatments by their parents.

CONCLUSION

bark The extract αf leaves and of Commelina benghalensis/Steganotaenia araliacea like amodiaquine possess antiplasmodial effect in people infested with Plasmodium falciparum and Plasmodium malariae. The antiplasmodial effect of the extract of Commelina benghalensis/Steganotaenia araliacea explained its use in traditional medicine in Ngaoundere Region in the treatment of malaria.

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