

# Research Journal of **Parasitology**

ISSN 1816-4943



# Antiplasmodial Activity of Ethanolic Root Extract of Telfairia occidentalis

<sup>1</sup>E. Okokon Jude, <sup>2</sup>J. Ekpo Arit and <sup>3</sup>A. Eseyin Olorunfemi <sup>1</sup>Department of Pharmacology and Toxicology, Faculty of Pharmacy, <sup>2</sup>Department of Biochemistry, College of Health Sciences, <sup>3</sup>Department of Pharmaceutical and Medicinal Chemistry, Faculty of Pharmacy, University of Uyo, Uyo, Nigeria

**Abstract:** The *in vivo* antiplasmodial activity of the ethanol root extract of *Telfairia occidentalis* grown particularly for the leaf and seed in Niger Delta region of Nigeria was evaluated in *Plasmodium berghei* infected mice. *Telfairia occidentalis* (200-600 mg kg $^{-1}$  day $^{-1}$ ) exhibited significant (p<0.05) blood schizonticidal activity both in 4 day early infection test and in established infection with a considerable mean survival time comparable to that of the standard drug, chloroquine, 5 mg kg $^{-1}$  day $^{-1}$ . The leaf extract possesses significant (p<0.05) antiplasmodial activity which can be exploited in malarial chemotherapy.

Key words: Antiplasmodial, malaria, Telfairia occidentalis, Plasmodium berghei berghei

# INTRODUCTION

Malaria is a major health problem in Nigeria and other tropical countries where transmission of the diseases is rarely control. It is estimated that half a million cases of malaria is recorded each year (Kwiatkowski, 2005) and an average of 1 million children in Africa die annually from the disease (Jeremiah, 2007). Eighty percent of Nigerians are exposed to malaria with at least 60 million people having repeated attacks in a year (Jeremiah, 2007). Drug resistance, fake drug syndrome and high cost of newer effective drugs have been the major factor affecting the poor populace, thus making their choice of herbal remedies inevitable and economical. The severity of the disease and other factors have made the search for an effective, cheap and readily available antimalarial of plant origin a challenge.

Telfairia occidentalis, Hook, F. (Cucurbitaceae) popularly known as fluted pumpkin is cultivated in the Southern part of Nigeria (Bosa and Mgbeogwu, 1983). Both leaves and seeds of the plant are consumed because of their high content of protein, vitamins and minerals (Johnson and Johnson, 1976). Ethnobotanically, the leaves are useful in the treatment of convulsion, anaemia, atherosclerotic cardiovascular disease, hypertension, malaria and impotence (Iwu, 1983; Sofowora, 1996; Gbile, 1986; Odoemena and Onyenneke, 1998). The leaf extract is useful in the management of cholesterolemia, liver problems and impaired defense immune systems (Eseyin et al., 2005a, b). The hypoglycaemic and antidiabetic activity of the leaf have been reported (Eseyin et al., 2000, 2005a, c; Nwozo et al., 2004). The antioxidant and antimicrobial activity of the leaf have also been reported by Oboh et al. (2006). There is a general belief that the roots are highly poisonous. The root is reported to contain alkaloids, saponins, glycosides and triterpenes (Akubue et al., 1980; Odoemena and Essien, 1995). Antibacterial activity of the root extract against Staphylococcus aureus, Streptococcus pyogenes, Shiegella dysenterae and Kliebsiella pneumoniae has been reported by Odoemena and Essien (1995).

In this study, we evaluate the antiplasmodial activity of the ethanolic root extract of *Telfairia occidentalis* against *Plasmodium berghei* infection in mice.

# MATERIALS AND METHODS

### **Plant Materials**

Fresh roots of *Telfairia occidentalis* were collected in August, 2006 from a farmland in Uruan, Akwa Ibom State, Nigeria. The plant was identified and authenticated by Dr. Margaret Bassey, a taxonomist in the Department of Botany, University of Uyo, Uyo, Nigeria. Hebarium specimen was deposited at Faculty of Pharmacy Hebarium. The fresh roots (2 kg) of the plant were dried on laboratory table for 2 weeks and reduced to powder. The powder 100 g was macerated in 95% ethanol (300 mL) for 72 h. The liquid filtrate obtained was concentrated in vacuo at 40°C. The yield was 0.98% w/w. The extract was stored in a refrigerator at 4°C until used for experiment reported in this study.

# Animals

Albino swiss mice (21-28 g) of either sex were obtained from the University of Uyo animal house. They were maintained on standard animal pellets and water ad libitum. Permission and approval for animal studies were obtained from the College of Health Sciences Animal Ethics Committee, University of Uyo.

### **Parasite Inoculation**

The chloroquine-sensitive *Plasmodium berghei berghei* was obtained from National Institute of Medical Research, Lagos, Nigeria and maintained in mice. The inoculum consisted of  $5\times10^7$  *P. berghei berghei* parasitized erythrocytes  $mL^{-1}$ . This was prepared by determining both the percentage parasitaemia and the erythrocytes count of the donor mouse and diluting the blood with isotonic saline in proportions indicated by both determinations. Each mouse was inoculated on day 0, intraperitoneally, with 0.2 mL of infected blood containing about  $1\times10^7$  *P. berghei beghei* parasitized red blood cells.

### Determination of LD<sub>50</sub>

The  $LD_{50}$  of the extract was determined using albino mice by intraperitoneal (I.P.) route using the method of Lorke (1983).

# Evaluation of Schizontocidal Activity on Early Infection (4 Day Test)

Schizontocidal activity of the extract was evaluated using the method described by Knight and Peters (1980). Each mouse was inoculated on the first day (day 0), intraperitoneally, with 0.2 mL of infected blood containing about  $1\times10^7 P$ . berghei berghei parasitized erythrocytes. The animals were divided into five groups of five mice each and orally administered, shortly after inoculation with 200, 400 and 600 mg kg<sup>-1</sup> day<sup>-1</sup> doses of the *Telfairia occidentalis* extract, chloroquine 5 mg kg<sup>-1</sup> day<sup>-1</sup> and an equivalent volume of distilled water (negative control) for four consecutive days (day 0 to day 3). On the fifth day (day 4), thin films were made from the tail blood of each mouse and the parasitaemia level was determined by counting the number of parasitised erythrocytes out of 200 erythrocytes in random fields of the microscope. Average percentage chemosuppression was calculated as:

$$100 \left[ \frac{(A-B)}{A} \right]$$

where,

A = Average percentage parasitaemia in the negative control.

Group A and B = Average percentage parasitaemia in the test group.

### Evaluation of Schizontocidal Activity Established Infection (Curative or Rane Test)

Evaluation of curative potential of the extract was done using a method similar to that described by Ryley and Peters (1970). The mice were injected intraperitoneally with standard inoculum of  $1\times10^7$  *P. berghei berghei* infected erythrocytes on the first day (day 0). Seventy-two hours later, the mice were divided into five groups of 5 mice each. The groups were orally administered with *T. occidentalis* extract (200, 400 and 600 mg kg<sup>-1</sup> day<sup>-1</sup>), chloroquine (5 mg kg<sup>-1</sup>) was given to the positive control group and an equal volume of distilled water to the negative control group. The drug/extract was given once daily for 5 days. Thin films stained with Giemsa stain were prepared from tail blood of each mouse daily for 5 days to monitor the parasitaemia level. The mean survival time for each group was determined arithmetically by finding the average survival time (days) of the mice (post inoculation) in each group over a period of 28 days (day 0 to day 27).

### Statistical Analysis

Data obtained from the study were analyzed statistically using Student's test and values of p<0.05 were considered significant.

### RESULTS

### **Acute Toxicity**

The mice were treated intraperitoneally with a single dose of 1-5 g kg<sup>-1</sup> of T. occidentalis root extract after being starved for 24 h. The route was chosen because of its sensitivity and rapid results. T. occidentalis (1-5 g kg<sup>-1</sup>) produced physical signs of toxicity 30 min to 1 h after administration. These signs include writhing, decreased motor activity, gasping, decreased respiratory and body/limb tone and death. The intensities of these effects were proportional to the dose administered. All the animals treated with 3 and 4 g kg<sup>-1</sup> of the extract died and the LD<sub>50</sub> was calculated to be 2.45 g kg<sup>-1</sup>.

### 4 Day Test

Ethanolic root extract of T. occidentalis produced a dose dependent chemosuppressive effect at various doses employed in this study. The chemosuppression were 69.33, 71.02 and 74.83% for 200, 400 and 600 mg kg<sup>-1</sup> day<sup>-1</sup> doses. The chemosupression produced by the extract were significant (p<0.05) compared to control and uncomparable to that of the standard drug (chloroquine 5 mg kg<sup>-1</sup> day<sup>-1</sup>) with a chemosuppression of 88.48% (Table 1).

### **Curative Test**

On established infection, it was observed that there was a daily increase in parasitaemia of the control group. However, there was a daily reduction in the parasitaemia levels of the extract treated group as well as that of positive control (chloroquine).

On day 7, the average percentage parasitaemia for the groups were 16.0, 14.0, 9.0, 7.0 and 86.0% for 200, 400, 600 mg kg<sup>-1</sup> day<sup>-1</sup> of the extract, chloroquine and control groups respectively (Fig. 1). The mean survival time (m.s.t) of the extract treated groups were significantly (p<0.05) longer than that of control and was comparable to that of the standard drug, chloroquine. The values are given in Table 2.

Table 1: Antiplasmodial activity of T. occidentalis root extract during 4 day test

Drug/Extract	Dose (mg kg <sup>-1</sup> day <sup>-1</sup> )	Average parasitaemia (%)	Average suppression (%)
T. occidentalis extract	200	12.36±1.63*	69.33
	400	11.68±4.49*	71.02
	600	10.14±2.94*	74.83
Chloroquine (standard)	5	4.64±1.72*	88.48
Distilled water (control)	0.2 mL	40.30±2.32	<del>-</del>

Data are expressed as mean±SD for five animals per group. \*: p<0.05 when compared to control

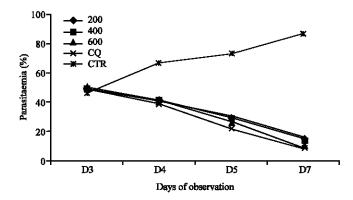


Fig. 1: Effect of T. occidentalis root extract on established infection (curative test)

Table 2: Mean survival time of mice receiving various doses of ethanolic root extract of T. occidentalis

Table 2. Wear survival time of fince receiving various doses of entatione root extract of 1. Occube nices.			
Drug/Extract	Dose (mg $kg^{-1} day^{-1}$ )	Mean survival time (Day)	
T. occidentalis extract	200	20.8±4.42*	
	400	25.3±2.04*	
	600	27.4±1.38*	
Chloroquine (standard)	5	28.0±0.00*	
Distilled water (control)	0.2 mL	9.5±2.04	

Data are expressed as mean±SD for five animals per group. \*: p<0.05 when compared to control

# DISCUSSION

In this study, prelimary phytochemical screening and acute toxicity studies as well as evaluation of antiplasmodial activity of ethanolic root extract of T. occidentalis were carried out. The results show that T. occidentalis root is moderately toxic as shown in its  $LD_{50}$  value of 938.08 kg<sup>-1</sup> (Homburger, 1989). This against the popular belief that it is highly poisonous. The root was found to contain terpenes and flavonoids which have been reported to be responsible for antimalarial activities of plants (Philipson and Wright, 1991; Christenen and Kharazmi, 2001). These compounds could have elicited the observed antiplasmodial activity either singly or in synergy with each other.

The results also indicated that the root extract possess blood schizontocidal activity as evident from the chemosuppression obtained during the 4 day early infection test. The activity of the extract was comparable to that of the standard drug, chloroquine (5 mg kg<sup>-1</sup> day<sup>-1</sup>) at the doses studied. On established infection, the extract exhibited a significant curative activity though not as much as the standard drug, chloroquine. The curative effect of the extract was further demonstrated in the significant mean survival time of the extract -treated groups compared to control.

The chloroquine and extract doses employed in this study could not offer 100% parasitaemia clearance in the infected mice, this is due to the low doses used and the crude nature of the extract. This activity could be improved by further purification of the drug and/or the extract.

# CONCLUSION

The result of this study is encouraging revealing the antiplasmodial potential of the root of *T. occidentalis*. Therefore, it would be interesting if the active principle could be isolated, identified and characterized from this promising medicinal plant.

# ACKNOWLEDGMENT

The authors are grateful to Mr. Nsikan Malachy for his technical assistance.

### REFERENCES

- Akubue, P.I., A. Kar and F.N. Nncheita, 1980. Toxicity of extracts of roots and leaves of *Telfairia occidentalis*. Planta Medica, 38: 339-343.
- Bosa, E.O. and C.M. Mgbeogwu, 1983. Fluted pumpkin, *Telfairia occidentalis*, West African vegetable crop. Econ. Bot., 37: 145-149.
- Christenen, S.B. and A. Kharazmi, 2001. Antimalarial Natural Products. Isolation, Characterization and Biological Properties. In: Bioactive Compounds from Natural Sources: Isolation, Characterization and Biological Properties. Tringali, C. (Ed.), Taylor and Francis, London, pp: 379-432.
- Eseyin, O.A., E. Oforah and B.A. Dooka, 2000. Preliminary study of the hypoglycaemic action of the extract of the leaf of *Telfairia occidentalis* in normoglycaemic guinea pigs. Global J. Pure Applied Sci., 6: 639-641.
- Eseyin, O.A., A.C. Igboasoiyi, E. Oforah, P. Ching and B.C. Okoli, 2005a. Effects of leaf extract of *Telfairia occidentalis* on some biochemical parameters in rats. Global J. Pure Applied Sci., 11: 77-79.
- Eseyin, O.A., A.C. Igboasoiyi, E. Oforah, H. Mbagwu, E. Umoh and J.F. Ekpe, 2005b. Studies of the effects of alcohol extract of *Telfairia occidentalis* on alloxan-induced diabetic rats. Global J. Pure Applied Sci., 11: 85-87.
- Eseyin, O.A., A.C. Igboasoiyi, E. Oforah, N. Nkop and A. Agboke, 2005c. Hypoglycaemic activity of *Telfairia occidentalis* in rats. J. Pharm. Bioresour., 2: 36-42.
- Gbile, Z.O., 1986. Ethnobotany, Taxonomy and Conservation of Medicinal Plants. In: State of Medicinal Plants Research in Nigeria. Sofowora, A. (Ed.), Proceedings of Conference, University of Ile-Ife, Nigeria, pp. 19.
- Homburger, F., 1989. *In vivo* Testing in the Study of Toxicity and Safety Evaluation. In: A Guide to General Toxicology. Marquis, J.K. (Ed.), 2nd Edn., Karger, New York.
- Iwu, M.M., 1983. Report of a sponsored project by the Institute of African Studies. University of Nigeria, Nsukka.
- Johnson, E.J. and T.J. Johnson, 1996. Economic plants in a rural Nigerian market. Econo. Bot., 30: 375-381.
- Knight, D.J. and W. Peters, 1980. The antimalarial action of N-benzyloxy dihydrotriazines. The action of cycolguanil (BRL50216) against rodent malaria and studies on its mode of action. Ann. Trop. Med. Parasitol., 74: 393-404.
- Lorke, D., 1983. A new approach to practical acute toxicity test. Arch. Toxicol., 54: 275-286.
- Nwozo, S.O., O.A. Adaramoye and E.O. Ajaiyeoba, 2004. Antidiabetic and hypolipidaemic studies of *Telfairia occidentalis* on alloxan induced diabetic rabbits. Nig. J. Nat. Prod. Med., 8: 45-47.
- Oboh, G., E.E. Nwanna and C.A. Elusiyan, 2006. Antioxidant and antimicrobial properties of *Telfairia occidentalis* (fluted pumpkin) leaf extract. J. Pharmacol. Toxicol., 1: 167-175.
- Odoemena, C.S. and J.P. Essien, 1995. Antibacterial activity of the root extract of *Telfairia occidentalis* (fluted pumpkin). West Afr. J. Biol. Applied Chem., 40: 29-32.
- Odoemena, C.S. and E.C. Onyeneke, 1998. Lipids of fluted pumpkin (*Telfairia occidentalis*) seeds. 1st African Conference on Biochemistry of Lipids, pp. 147-151.
- Philipson, J.D. and C.W. Wright, 1991. Antiprotozoal compounds from plants sources. Planta Medica, 57: 553-559.
- Ryley, J.F. and W. Peters, 1970. The antimalarial activity of some quinone esters. Ann. Trop. Med. Parasitol., 84: 209-222.
- Sofowora, A., 1996. Medicinal Plant and Traditional Medicine in Africa. 2nd Edn., Spectrum Books, Ibadan, Nigeria, pp. 112.