The African Journal of Medicine and Medical Sciences

Volume 14
1985

BLACKWELL SCIENTIFIC PUBLICATIONS
Oxford London Edinburgh Boston Palo Alto Melbourne

Afr. J. Med. med. Sci. (1985) 14, 59–63.

Screening of *Morinda lucida* leaf extract for antimalarial action on *Plasmodium berghei berghei* in mice

J. M. MAKINDE AND P. O. OBIH

Department of Pharmacology and Therapeutics, College of Medicine, University of Ibadan, Ibadan, Nigeria

Summary

The leaf extract of *Morinda lucida* collected in August was administered subcutaneously to albino Swiss mice infected with *P. berghei berghei*. The schizontocidal activity on early infection was assessed by administering chloroquine (standard) distilled water or *Morinda lucida* as single daily dose on day 0–3 to infected mice. On day 4 the degree of parasitaemia and percentage was determined in relation to control. Its schizontocidal activity was also observed on an established infection by administering the drugs 72 h after infecting the mice and the degree of parasitaemia was determined daily. The repository action of pyrimethamine was also compared with *Morinda lucida*.

On the early infection, the chloroquine equivalent of *Morinda lucida* was found to be 1.0 mg/kg. In established infection a daily increase in parasitaemia was observed in control group while the animals that received chloroquine (5 mg/kg) and ⅙ dilution of the stock of *Morinda lucida* extract showed a sharp fall in parasitaemia from the second day of treatment. For the prophylactic test, 1.2 mg/kg of pyrimethamine and ⅙ dilution of stock of extract produced 80.5% and 70% chemosuppression respectively.

It is interesting to note that *Morinda lucida* leaves extract appears to have schizontocidal and repository effects in mice infected with *P. berghei berghei*

Correspondence: J. M. Makinde, Department of Pharmacology and Therapeutics, College of Medicine, University of Ibadan, Ibadan, Nigeria.

Résumé

L'extrait de la feuille de *Morinda lucida* levée en août était administré S.c. chez les souris albinos Swiss infectés de *Plasmodium berghei berghei*. Son activité schizontocidal au début de l'infection était évalué en administrant la chloroquine (étalon) et de l'eau distillé ou *Morinda lucida* à dose unique journellement en jours 0, 1, 2 et 3 chez les souris infectés. Le degré de la parasitaemie était déterminé en jour 4 et le pourcentage en relation au contrôle. Son activité Schizontocidal a été aussi observé sur l'infection fondé en administrant les drogues 72 heures après avoir infecté les souris et le degré de la parasitaemie était determiné journellement. L'action répositoire de la pyrimethamine a été aussi comparé à celle de *Morinda lucida*.

Au début de l'infection l'equivalence de la chloroquine de *Morinda lucida* était 1.0 mg/kg. Quand l'infection était établi un accroissement quotidien en parasitaemie a été observé dans le groupe du contrôle alors que les animaux qui ont reçus la chloroquine (5 mg/kg) et 1/6 dilution due réserve de l'extrait de *Morinda lucida* ont montrés une chute aiguë en parasitaemie à partir de la 2ème jour du traitement. Pour le test prophylactique, 1.2 mg/kg du pyrimethamine et 1/6 dilution du réserve de l'extrait ont produit 80,5% et 70% chemosuppression respectivement.

Il est intéressant à noter que l'extrait de feuilles de *Morinda lucida* paraitre d'avoir les effets schizontocidal et repositoire dans les souris infectés de *Plasmodium berghei berghei*.

Introduction

Malaria is a parasitic disease that has brought misery and death to many millions of people in

tropical and subtropical regions of the world. Despite the huge efforts being made to control malaria it still remains one of the greatest causes of illness and death in the world and particularly in Africa (WHO, 1977).

Most of the standard antimalarial drugs including amodiaquine, chloroquine, proguanil, primaquine and pyrimethamine have been in use for several years. Many authorities in the 1950s considered these drugs so good that there was a general decline in chemotherapy research work and it was expected that malaria would rapidly disappear. According to Howells (1982) this optimism was short lived and during the last 20 years the increasing problem of drug resistance has emphasized the limitations of these drugs and has stimulated a search for new and more effective drugs.

Plants were used in the treatment of malaria long before the synthesis of antimalarial drugs, which was started during the first world war by Germans when normal supplies of quinine and cinchona bark became unavailable. As plants are potential sources of new drugs, and since our forefathers seemed to be able to treat malaria with various herbal decoctions and or plant extracts, it is hoped that more research into medicinal plants would yield some beneficial scientific results.

Morinda lucida is a medium-sized tree which is widespread in tropical Africa and quite abundant in Nigeria, where it has a reputation for antimalarial properties (Dalziel, 1948). The leaves or the stem bark are used in the form of aqueous decoction for drinking. Other alleged properties include antipyretic and some chemotherapeutic actions and warding off evil spirits (Irvine, 1961). This work reports the effect of *Morinda lucida* leaf extract on *Plasmodium berghei berghei* in mice.

Materials and methods

Preparation of Morinda lucida *leaf extract*

Fifteen g of fresh *Morinda lucida* leaves, collected in August, were pounded in a mortar with a pestle. The finely crushed leaves were then squeezed to extract the jiuce. (Usually 6 ml was obtained from 15 g). This was regarded as the stock solution and various dilutions were prepared from it as required with distilled water.

Administration of the drug or extract

The drugs or extract were administered to the mice subcutaneously.

Evaluation of blood schizontocidal activity in vivo *using an early infection — 4-day test*

The method was based on that described by Peters (1965) and Porters and Peters (1975). The blood schizontocidal activity of chloroquine and the *Morinda lucida* leaf extract against the drug sensitive *P. berghei berghei* was tested on parasitized albino Swiss mice. The animals were divided into groups of 5 or 10 mice. The day of inoculation was termed D_0. At the same time the animals received once daily treatment of 5 mg/kg chloroquine given in 0.2 ml/20 g mouse, 0.2 ml/20 g mouse of the distilled water (control) and the decoction on days 0–3 (D_0–D_3). Thin blood films were made from the tail on D_4 and used in assessing the activity of the drugs. The average percentage chemosuppression of parasitaemia was calculated using the following formula:

$$\left(\frac{\text{Average \% parasitaemia in controls} - \text{Average \% parasitaemia test}}{\text{Average \% parasitaemia in control}}\right) \times 100$$

Evaluation of the schizontocidal activity in vivo *using an established infection*

The method employed was that described by Ryley and Peters (1970). Details of this method has been described in an early publication (Makinde & Obih, 1984).

Repository activity in vivo

This method is similar to that described by Peter (1965). The full report of this appeared in an earlier publication (Makinde & Obih 1984).

Results

Blood schizontocidal activity of Morinda lucida *leaf juice and chloroquine on an established infection* in vivo

Fig. 1 represents percentage parasitaemia obtained in each group against the days after infection. In the control, the level of parasitaemia increased throughout the period of observation. In animals treated with 5 mg/kg chloroquine subcutaneously, a gradual fall in parasitaemia was observed from the second day of treatment (D_5) and parasites disappeared completely from the blood on D_8. With the subcutaneous administration of ⅙ dilution *Morinda lucida*, and initial rise in parasitaemia was observed on D_5 but this gave way to a sharp fall on D_6 and subsequently, there was a gradual diminution of parasites from the blood until D_8 when they were completely eliminated. Animals that received ⅛ dilution of the original *Morinda lucida* juice also exhibited an initial rise in parasitaemia followed by a sharp fall on D_6. Few parasites were observed on D_8.

Effect of chloroquine and Morinda lucida *on early infection (4-day test)*

In this work, treatment started from the day of infection with *P. berghei berghei* and was continued for 4 days. Figure 2 is the graph of percentage suppression of parasitaemia against log dose of chloroquine subcutaneously. The chloroquine equivalent of *Morinda lucida* was

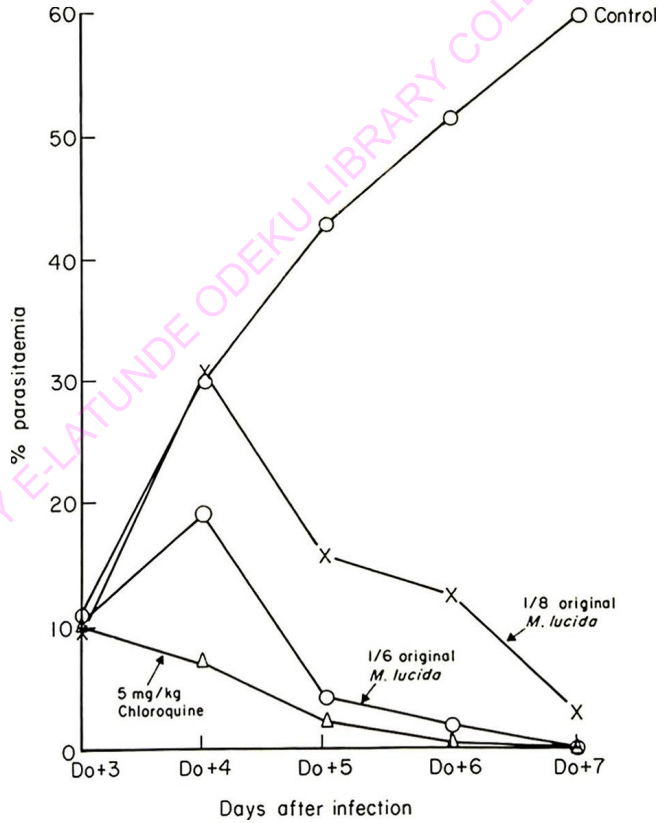

Fig. 1. Percentage parasitaemia in each group *v.* time after infection, Do = day after infection.

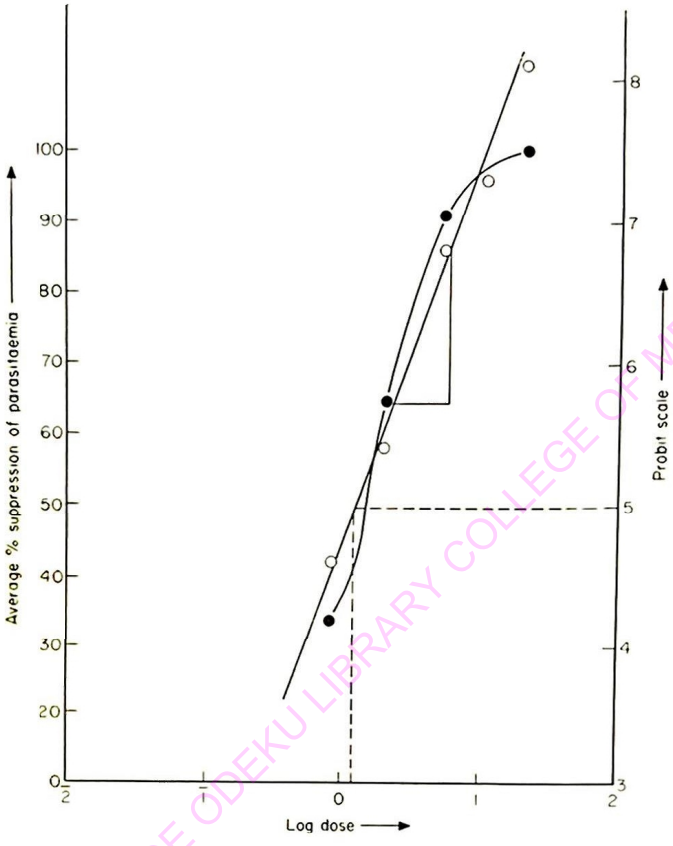

Fig. 2. Percentage suppression of parasitaemia *v.* log dose of chloroquine.

extrapolated from this graph. ED_{50} of chloroquine subcutaneously was found to be 1.3 mg/kg. The ⅙ dilution of *Morinda lucida* juice used had a chloroquine equivalent of 1.0 mg/kg (chemosuppression 44.4%). Higher concentrations of *Morinda lucida* such as the stock, ½ or ¼ dilutions were found toxic to the animals when administered subcutaneously.

Repository action of Morinda lucida *juice and pyrimethamine on* P. berghei berghei

A sixth dilution of *Morinda lucida* gave a chemosuppression of 70% while that of pyrimethamine 1.2 mg/kg gave 80.5%.

Discussion

Morinda lucida leaf extract has been shown to possess schizontocidal activity on an established infection when administered subcutaneously. It was observed that there was a delay in the onset of schizontocidal action of both ⅙ and ⅛ dilution, given subcutaneously, the effect did not commence until the third day of treatment. This delay in onset of effect observed to *Morinda lucida* leaf extract might probably be attributed to inadequate levels of the active principle in the blood stream within the first 2 days of treament.

The schizontocidal effect of the ⅙ dilution was observed to be more than that of ⅛

dilution. Thus the *Morinda lucida* leaf juice probably exhibited a dose-dependent schizontocidal effect. When compared to the effect of a standard antimalarial agent, chloroquine, 5 mg/kg completely cleared the blood of the malaria parasites by D_4 while the ⅛ dilution of *Morinda lucida* leaf extract did not produce complete eradication of the parasites from the blood until after the last dose.

In this work *Morinda lucida* leaf juice has also been shown to possess schizontocidal activity in an early infection.

In the repository test, *Morinda lucida* was observed to show great therapeutic potentials, producing 70% chemosuppression. When this was compared to pyrimethamine which produced 80.5% chemosuppression, the results observed with *Morinda lucida* is encouraging.

From the results obtained in our work, it is interesting to note that *Morinda lucida* leaf extract collected in August was observed to have some schizontocidal and repository actions in mice. This calls for further investigations.

References

Dalziel, J.M. (1948) *The Useful Plants of West Tropical Africa*. 2nd edn. pp. 403–404, London: Crown Agents for the colonies.

Howells, R.E. (1982) Advances in chemotherapy. *Br. Med. Bull.* **38** (2), 193–199.

Irvine, F.R. (1961) *Woody Plants of Ghana* Oxford University Press, Oxford.

Makinde, J.M. & Obih, P.O. (1984) Lack of schizontocidal activity of three Herbal decoctions on *Plasmodium berghei berghei* in mice. *In Press.*

Peters, W. (1965) Drug resistance in *Plasmodium berghei*, Vincka and Lips, 1948: I chloroquine resistance. *Exp. Parasitol.* **17**, 80–89.

Porters J.A. and Peters, W. (1975) Chemotherapy of rodent malaria. Antimalarial ativity of WR 122, 453 (a-phenantrene methanol) *in vivo* and *in vitro*. *Ann. Trop. Med. Parasit.* **70**, W0 3P 259–270.

Ryley, J.F. & Peters, W. (1970) The antimalarial activity of some quioline esters. *Am. J. Trop. Med. Parasit.* **84**(2), 209.

World Health Organisation (1977) Information on the world malaria situation Geneva (reprinted from the Weekly epidemological record).

(*Received* 30 *April* 1984; *accepted* 31 *May* 1984)