

Komunikasi Pendek/Short Communications

In Vitro Screening of *Aralidium pinnatifidum* Extract
for Anti-Malarial Activity

MOHD KAMEL ABD. GHANI, SITI NAJILA MOHD JANIIB, NOOR RAIN
ABDULLAH, SYED ZAHIR SYED IDID, KHOZIRAH SHAARI, LOKMAN
HAKIM SULAIMAN, ZAKIAH ISMAIL & AZIZOL ABD. KADIR

ABSTRAK

Aralidium pinnatifidum disaring untuk aktiviti anti-malaria ke atas *Plasmodium falciparum* secara *in vitro* dengan menggunakan asai laktat dehidrogenase. Ekstrak mentah metanol *Aralidium pinnatifidum* menunjukkan aktiviti skizontisid berpotensi lemah. Ia lebih efektif ke atas *P. falciparum* terasing rintang klorokuina, Gombak A (IC_{50} 173.0 µg/ml), dari strain *P. falciparum* rentan klorokuina, D10 (IC_{50} 417.0 µg/ml).

ABSTRACT

Aralidium pinnatifidum was screened for anti-malarial activity against *Plasmodium falciparum* *in vitro* using the lactate dehydrogenase (LDH) assay. The crude methanol extract of *Aralidium pinnatifidum* exhibited some schizonticidal activity of weak potency. It was more effective against a chloroquine resistant *P. falciparum* isolate, Gombak A (IC_{50} of 173.01 µg/ml), than a chloroquine sensitive *P. falciparum* strain, D10 (IC_{50} of 417.04 µg/ml).

In an effort to overcome the growing problem of chloroquine resistant *Plasmodium falciparum* strain, attention has been focused on medicinal plants as sources of new and novel anti-malarial agents. Several natural products isolated from plants used in traditional medicine showed potent anti-plasmodial activity in preliminary studies (Benoit et al. 1998; Kuria et al. 2001). Recently we reported the anti-malarial activity of the extracts of two *Goniothalamus* species viz. *G. macrophyllus* and *G. scortechinii*. The latter exhibited a stronger activity with IC_{50} value of 9 µg/ml for the chloroquine resistant strain and 40 µg/ml for the chloroquine sensitive strain (Siti Najila et al. 2000; Mohamed Kamel et al. 2001).

Aralidium pinnatifidum Miq. (Family Cornaceae), locally known as hempedu buaya, has been selected for this study on the basis of its

ethnomedicinal use to treat fever (Burkill 1966). This study was carried out to screen the extract of *A. pinnatifidum* for anti-malarial activity against *Plasmodium falciparum* *in vitro* using the lactate dehydrogenase (LDH) assay.

The methanol extract of *Aralidium pinnatifidum* showed some activities of weak potency towards the target strains as shown in Table 1. Its inhibition of the resistant strain, Gombak A (IC_{50} 173.0 $\mu\text{g/ml}$) was better compared to its inhibition of the sensitive strain, D10 (417.0 $\mu\text{g/ml}$). All the tested extracts showed a concentration dependent growth inhibition of the malaria parasite. Chloroquine and artemisinin functioned well as positive controls. Though *A. pinnatifidum* has shown some anti-malarial activity, its activity is considered of weak potency. Its inhibition of the resistant strain, Gombak A (IC_{50} 173.01 $\mu\text{g/ml}$) was better compared to its inhibition of the sensitive strain, D10 (417.04 $\mu\text{g/ml}$). However, these values were much higher compared to the inhibition by other medicinal plants previously studied by us (Siti Najila et al. 2000; Mohamed Kamel et al. 2001).

TABLE 1. IC_{50} values of each extracts obtained with both the sensitive (D10) and resistant (Gombak A) strains of *Plasmodium falciparum*

Sample	IC_{50} ($\mu\text{g/ml}$)	
	D10	Gombak A
1. <i>A. pinnatifidum</i>	417.0	173.0
2. Chloroquine	0.017 (\pm 0.0009)	1.78 (\pm 0.116)
3. Artemisinin	0.0009	0.009

MATERIALS AND METHODS

Aralidium pinnatifidum was collected from Taman Rimba Templer, Selangor, Malaysia. A voucher specimen (FRI45577) was deposited at the Herbarium of the Forest Research Institute of Malaysia (FRIM). The stems were dried and ground by a Wiley mill to pass through a 40-60 mesh screen. The sample was soaked in methanol and the resulting extract was concentrated and dried. The crude methanol extract was then assayed for anti-malarial activity by the lactate dehydrogenase (LDH) method as described by Makler et al. (1993). Two strains of *Plasmodium falciparum* were used in this study. Gombak A was a local isolate and was known to be resistant to chloroquine (Slamet et al. 1991). It was originally isolated from an Orang Asli patient who was admitted to the Gombak Hospital, Selangor in 1982. The chloroquine sensitive strain,

D10, was obtained from the Institute of Medical Microbiology, University of Milan, through the courtesy of Prof. Donatella Taramelli.

The lactate dehydrogenase method was performed using 96-well microtitre plate (flat bottom). The stock solution of the extract was prepared at 1000 µg/ml while the positive controls, chloroquine (Sigma Chemicals, USA) and artemisinin (Sigma Chemicals, USA) were both prepared at 1µg/ml. For each test, a blood suspension of 1% parasitaemia and 2% haematocrit was prepared. Control readings of parasitised red blood cells devoid of plant extracts/ controls and non-parasitised red blood cells were done simultaneously. After the plates have been prepared, they were placed in a candle jar and incubated for 48 hrs at 37°C. Absorbance was read at 630nm using an ELISA plate reader (MRX Microplate Reader, Dynex Technologies, USA). Inhibition percentages of parasite viability were determined and the mean of at least three IC₅₀ values was calculated using the curve fitting analysis (Grafit v.4.09, Erithacus Software Limited).

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Mohamed Kamel Abd. Ghani
Siti Najila Mohd Janib
Syed Zahir Syed Iddid
Department of Biomedical Science
Faculty of Allied Health Sciences
Universiti Kebangsaan Malaysia
50300 Jalan Raja Muda Abdul Aziz
Kuala Lumpur, Malaysia

Noor Rain Abdullah
Lokman Hakim Sulaiman
Zakiah Ismail
Herbal Medicine Research Centre
Infectious Disease Research Centre
Institute for Medical Research
Jalan Pahang
50588 Kuala Lumpur
Malaysia

Khozirah Shaari
Azizol Abd.Kadir
Forest Research Institute of Malaysia
Kepong
52109 Kuala Lumpur
Malaysia