

Bioassay-guided fractionation of  
*Artemisia afra* for *in vitro* antimalarial activity  
against *Plasmodium falciparum*

MERYL A. ABRAHAMS

A project submitted to the Department of Pharmacology,  
University of Cape Town in fulfilment of the requirements  
for the degree

MASTER OF SCIENCE (MEDICINE)

SUPERVISORS

Prof. P.I. Folb

Dr. D.W. Gammon

December 1996

## Abstract

With the increase in recent years in the prevalence of malaria, and in drug resistance of *Plasmodium falciparum*, there has been much interest in natural plant products for new antimalarials with novel modes of action against *Plasmodium*. Artemisinin or Qinghaosu is one such antimalarial isolated from a Chinese herb, *Artemisia annua* (Asteraceae) and it is currently undergoing phase I and II clinical trials. The Southern African species, *Artemisia afra* (African wormwood, wildeals, lengana) is commonly used by local traditional healers for symptoms of malaria, in particular fever. Thus it seemed appropriate to investigate this species for antimalarial activity.

Crude petroleum ether soxhlet extracts of *Artemisia afra* had demonstrated antimalarial activity against *Plasmodium falciparum*, FCR-3, cultured *in vitro*. The IC<sub>50</sub> values ranged from 5-13 µg/ml. The extract from leaves and flowers was then screened against D10 (chloroquine-sensitive) and FAC8 (chloroquine-resistant) *P. falciparum*, *in vitro*, with IC<sub>50</sub> values of 1.03 µg/ml and 1.5 µg/ml respectively. This extract was fractionated by column chromatography using silica gel-60 and the fractions obtained were screened for antimalarial activity. The most active fraction had an IC<sub>50</sub> of 0.5 µg/ml against D10 and FAC8. Using TLC and HPLC-UV analysis with pure artemisinin as a standard, no artemisinin could be detected in this fraction. This result was confirmed by thermospray LC-MS analyses. Purification of this fraction yielded ultimately a single pure compound; a clear colourless oil identified by MS and NMR analyses as hydroxydavanone. The compound was screened against a variety of *P. falciparum* strains with varying degrees of sensitivity and resistance to both chloroquine and mefloquine. Their sensitivity against artemisinin was also established. IC<sub>50</sub> values obtained for the isolated pure compound against *P. falciparum* ranged from 0.87 to 2.54 µg/ml. The IC<sub>50</sub> values obtained for general cytotoxicity of the crude extract

and isolated pure compound against RAT-1 fibroblast cells were  $34.78 \pm 8.23$  and  $6.29 \pm 0.95 \mu\text{g/ml}$  ( $n=4$ ) respectively. Thus the crude extract and isolated pure compound exhibited a greater antimalarial than cytotoxic effect. Hence, there are implications for *A. afra* to be used as a phytomedicine for the treatment of malaria. *In vivo* studies are recommended for hydroxydavanone in order to fully assess its potential for clinical use.