Department of Medicine, Solna

The role of molecular markers in emerging artemether-lumefantrine resistant *Plasmodium falciparum*

AKADEMISK AVHANDLING
som för avläggande av medicine doktorsexamen vid Karolinska Institutet offentligen försvaras i Hillarpsalen, Retzius väg 8, Karolinska Institutet

Fredagen den 18:e januari, 2013, kl 13.00

av

**Maja Malmberg**
MSc, Civillingenjör i bioteknik

**Huvudhandledare:**
Dr José Pedro Gil
Karolinska Institutet
Institutionen för Fysiologi och Farmakologi

**Bihandledare:**
Dr Pedro Eduardo Ferreira
Nagasaki University
Institute of Tropical Medicine

Professor Anders Björkman
Karolinska Institutet
Institutionen för Medicin, Solna

Docent Andreas Mårtensson
Karolinska Institutet
Institutionen för Medicin, Solna

**Fakultetsopponent:**
Professor Harald Noedl
Medical University of Vienna
Institute of Specific Prophylaxis and Tropical Medicine

**Betygsnämnd:**
Professor Elias Arnér
Karolinska Institutet
Institutionen för Medicinsk Biokemi och Biofysik

Professor Dan Andersson
Uppsala Universitet
Institutionen för Medicinsk Biokemi och Mikrobiologi

Dr Kristina Persson
Karolinska Institutet
Institutionen för Mikrobiologi, Tumör- och Cellbiologi

Stockholm 2013
Abstract

Malaria is a devastating disease which kills ~1 million people yearly. The vast majority of lives lost due to malaria are children and pregnant women in sub-Saharan Africa. Although malaria is a treatable disease it continues to be one of the major causes of death, especially in poor settings. Chemotherapy is the key to control the disease, decrease the burden of malaria and save lives. The malaria parasites ability to develop resistance towards antimalarial drugs is therefore a major concern. Artemether-lumefantrine (Coartem®, Novartis) is currently the most used treatment for uncomplicated Plasmodium falciparum malaria. The aim of this thesis was to contribute to the understanding of the role of molecular markers in emerging artemether-lumefantrine resistant P. falciparum.

This thesis is based on artemether-lumefantrine clinical trials designed to evaluate the efficacy and effectiveness of artemether-lumefantrine for treatment of uncomplicated P. falciparum malaria in children in Tanzania. We measured lumefantrine concentrations and investigated their correlation with cure rates and with tolerance/resistance associated markers within the parasite. Our focus was primarily on polymorphisms within P. falciparum multidrug resistance gene 1 (pfmdr1) and P. falciparum chloroquine transporter gene (pfcrt).

One major finding is that lumefantrine blood drug concentrations in combination with pharmacokinetic parameters can be used to assess the relative importance of different single nucleotide polymorphisms for lumefantrine drug susceptibility in vivo. Lumefantrine blood drug concentrations after artemether-lumefantrine treatment were correlated with selection of recurrent infections with specific pfmdr1 N86, 184F and D1246 single nucleotide polymorphisms.

Although artemether-lumefantrine was found to have excellent efficacy and effectiveness according to PCR adjusted cure rates, the number of recurrent infections were high and we observed an up to three week difference in post-treatment prophylactic effect depending on the pfmdr1 polymorphisms among recurrent infections. Since the introduction of artemether-lumefantrine as first line treatment for uncomplicated malaria in Tanzania in 2006, the prevalence of pfmdr1 N86, 184F and D1246 have increased significantly up to 2011.

Overall, the results indicate that pfmdr1 is involved in the mechanism of resistance to lumefantrine. The increased prevalence of parasites carrying the pfmdr1 NFD haplotype could be an early warning of reduced artemether-lumefantrine efficacy.