INDUCEMENT OF *Plasmodium berghei* ANKA STRAIN RESISTANCE TO LUMEFANTRINE, PIPERAQUINE AND AMODIAQUINE IN A MOUSE MODEL.

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A thesis submitted in partial fulfillment for the Degree of Master of Science in Biochemistry in the Jomo Kenyatta University of Agriculture and Technology.

2008

ABSTRACT

The evolution of drug resistance in *Plasmodium falciparum* the species that causes the most virulent form of malaria in humans is a major public health problem. In response to increasing antimalarials resistance, old drug such as amodiaquine (AQ) have been resurrected while new once such as lumefantrine (LM) and Piperaquine (PQ) are being introduced. LM, PQ and AQ are potent partner drugs in artemether-lumefantrine (ATM/LM), piperaquine-dihydroartemisinin (PQ/DHA) and amodiaquine-artesunate (AQ/ASN) ACTs respectively. ATM, DHA and ASN are short acting drugs with a half-life of less than 2 hours. LM, PQ and AQ active derivative (N-desethylamodiaquine (DEAQ) are long acting drugs with a half-life 6, 19 and 14 days respectively. During elimination period the unprotected LM, PQ and DEAQ remain alone especially at sub-therapeutic concentrations. At this point, selection of re-infecting resistant parasites may occur rapidly especially in African regions with high malaria transmission. Markers coding for LM, PQ and AQ resistance are not clearly understood due to lack of well characterized resistant lines. The objective of the study was to induce and select *Plasmodium berghei* ANKA strains resistant to LM, PQ and AQ *in vivo. P. berghei* ANKA strain in mouse model was exposed to selection pressure from AQ, LM and PQ. Two methods were adopted: first the use of a single dose at every passage and second, the stepwise increase of drug pressure dose. Once every 7-10 days, parasitized erythrocytes were passed to the next group of naive mice. The level of resistance was assessed in the 4-DT at different intervals every 4^{th} or 8^{th} passage, the increase in ED_{50}, ED_{90} and ED_{99} was estimated graphically using version 5.5 of Statistica 2000 and indices of resistance, I_{50}, I_{90} and I_{99} calculated. Resistance was classified into four categories I_{99} =1.0, sensitive, I_{99} =1.01-10.0, slight resistance, I_{99} =10.01-100.0, moderate resistance, and I_{99} = >100.0, high resistance. The ED_{50}, ED_{90} and ED_{99} of AQ against parent *P.*

berghei ANKA strain was 0.95, 4.29 and 5.05 mg/kg.day respectively. Within thirty six passages the ED_{50}, ED_{90} and ED_{99} were 12.01, 19.13 and 20.73 mg/kg.day respectively. Slight AQ resistant line was selected with I_{50}, I_{90} and I_{99} of 12.64, 4.46 and 4.10 respectively. The ED_{50}, ED_{90} and ED_{99} of LM against parent *P. berghei* ANKA strain was 1.67, 3.93 and 4.48 mg/kg.day respectively. Within twenty eight passages, the ED_{50}, ED_{90} and ED_{99} were 9.76, 25.48 and 29.02 respectively. Slight LM resistant line was selected with I_{50}, I_{90} and I_{99} of 5.84, 6.48 and 6.48 respectively. The ED_{50}, ED_{90} and ED_{99} of PQ against parent *P. berghei* ANKA GFP was 1.30, 3.52 and 8.10 mg/kg.day respectively. Within twenty eight passages of selection pressure the ED_{50}, ED_{90} and ED_{99} was 122.00, 193.30 and 210.00 mg/kg.day respectively. Moderate PQ resistant line was selected with I_{50}, I_{90} and I_{99} of 93.85, 54.91 and 25.93 respectively, the line exhibited relative stability with I_{50}, I_{90} and I_{99} of 36.23, 42.76 and 20.85 after drug free five passages. Stable PQ resistant line was selected which could be used to study its mechanism of resistance. Further work is warrant for LM and AQ.