UNIVERSITI TEKNOLOGI MARA

THE DOCTORAL RESEARCH ABSTRACTS

Volume: 8, Issue 8 November 2015

EIGHTH ISSUE

INSTITUTE of GRADUATE STUDIES

IGS Biannual Publication

26

Name :
Ummul Hanan Mohamad

Title :
NADH Dehydrogenase as a Molecular Target for Artemisinin Related Antimalarian Drug Screening in a Yeast Model

Supervisor :
Associate Prof. Dr. Mohamad Faiz Foong Abdullah (MS)
Dr. Umi Marshida Abd Hamid (CS)

Artemisinin is currently the only effective drug against malaria. However, artemisinin-resistant *Plasmodium* had begun to emerge in many malaria endemic areas. Discovery of new anti-malarial with artemisinin-like activity had been slow as the molecular target of artemisinin was yet to be established. In addition, studies on the malaria causative agent were also hampered because *Plasmodium* was difficult to culture *in-vitro.* Therefore, this research aims to develop a reliable yeast screening system to help clarify artemisinin mode of action as well as accelerate the discovery of potential anti-malaria with artemisinin-like properties. The NADH dehydrogenase enzyme, which was coded by *NDE1* and *NDI1*, in *Saccharomyces cerevisiae* as thought to be a major molecular target for artemisinin. A yeast system was constructed in which the efflux pump regulator genes, *PDR1* and *PDR3*, and either *NDE1* or *NDI1* were deleted. Absence of the *PDR1* and *PDR3* genes minimized the ability of yeast to remove the test drugs into the extracellular environment, thus the drug effect could be clearly observed. From the study, Δ*pdr1*Δ*pdr3*Δ*nde1* or Δ*pdr1*Δ*pdr3*Δ*ndi1* knock-out tolerated 12 μM artemisinin and 4 μM dihydroartemisinin in contrast to Δpdr1Δpdr3. Hence, Δ*pdr1*Δ*pdr3* and Δ*pdr1*Δ*pdr3*Δ*nde1* were chosen to serve as the screening panels. Several compounds were found to possess artemisinin-like activities. These included black seed oil, black pepper and mangosteen. Since NADH dehydrogenase genes in yeast were homologous to *Plasmodium NDH2* gene, it was assumed that any effect towards the yeast proteins may be reflective of a similar effect towards *Plasmodium* protein. Further validation demonstrated that the cloned *NDE1* gene partially restored the yeast susceptibility to artemisinin derivative, dihydroartemisinin. Real-time PCR revealed that the yeast with cloned *NDE1* expressed NADH dehydrogenase albeit at 32-fold lower than the wild-type. Following that, random mutation to *NDE1* gene showed that most mutation was single nucleotide deletion that altered the protein sequence to produce non-functional (due to stop codons) or missense (due to different amino acid sequence) protein to resist artemisinin derivative.

*** (MS) = Main Supervisor (CS) = Co Supervisor**