Abstract

Malaria is one of the world's most prominent and life-threatening parasitic diseases which spreads through bites of infected female *Anopheles* mosquitoes. Moreover, due to the increase of resistance to the current antimalarial drug as well as the high mutation rate of this parasite, there is a tendency and preference of herbal plants usage as treatment medicine which open further possible alternatives for more efficacious antimalarial drugs. One of Indonesia's native medicinal plants *Gynura divaricata* is known to contain the active compound β-sitosterol which had been demonstrated to work as an antimalarial agent against *P. falciparum*. Moreover, previously conducted *in-silico* study by molecular docking also reported a strong interaction between PfCDPK1 protein with β-sitosterol in *Gynura divaricata* showing a correlation which require further evidence through *in-vitro* study validating β-sitosterol is responsible for exerting antimalarial properties. This study aims to do isolation of β-sitosterol from the herbal plant *Gynura divaricata* and *in-vitro* study to evaluate β-sitosterol's antimalarial property against *P. falciparum* 3D7 strain and its mechanism of action. This research encompasses the extraction of *Gynura divaricata* leaves and isolation of β -sitosterol. As well as *in vitro* investigations of its antimalarial activities which include testing the extract against cultured *P. falciparum* 3D7 strain to determine the 50% inhibitory concentration (IC50) and further *in-vitro* testing by reactive oxygen species oxidative assay (TBARS) and DNA fragmentation assay using *P. falciparum* culture were performed to characterize and understand the mechanism of β-sitosterol's antimalarial action. The isolation from *Gynura divaricata* crude extract resulted in 31 mg of concentrated β-sitosterol powder. Further potency determination of the compound resulted in β-sitosterol IC50 of 6.734 µg/mL and the standard treatment artemisinin IC50 of 3.357 µg/mL. Determination of oxidative damage activity using TBARS assay showed that concentrated β-sitosterol IC50 resulted in the highest ability in inducing oxidative damage after 24 hour incubation. However, further investigation of β-sitosterol's ability in inducing parasite death through apoptosis could not be determined based on the DNA fragmentation assay result which showed faint smear of DNA fragments. Although it has been proven that β-sitosterol is able to induce oxidative damage, further optimization of DNA isolation and fragmentation methods should be done to investigate whether the oxidative damage resulted in apoptosis

Keywords: *Malaria, Plasmodium falciparum, Gynura divaricata,* β*-sitosterol, antimalarial agent.*