ABSTRACT

Aims: In our search for new antiplasmodial agents, *in vitro* antiplasmodial activities of the crude extracts and isolated pure compounds were determined. In addition to the *in vitro* assays, *in vivo* acute toxicity of the crude extracts was investigated to assess the safety of the plants. Furthermore, structure elucidation of the pure compounds was also carried out to determine the identity of the isolated compounds.

Study Design: Extraction of the root crude extracts of *Euclea latideus* was done using four solvents: hexane, dichloromethane, ethyl acetate and methanol. Isolation and purification were carried out on only the dichloromethane and ethyl acetate crude extracts.

Methodology: Four solvent; hexane, dichloromethane, ethyl acetate and methanol were used to carry out the extraction process of the crude samples. Isolation and purification of crude extracts were achieved using chromatographic techniques which included column and thin layer chromatography (TLC). The characterization of the isolated compounds was determined using NMR spectroscopic techniques.

In vitro antiplasmodial activity was performed on two strains of *Plasmodium falciparum* (chloroquine [CQ]-sensitive 3D7 and CQ-resistant Dd2 strains) using a non-radioactive fluorescence-based SYBR Green 1 assay technique. Lorke's method of acute toxicity was used to determine the *in vivo* acute toxicity of the crude extracts in mice.

Results: Results of acute toxicity studies showed that all crude extracts of *E. latideus* had LD_{50} > 5000 mg/kg and therefore regarded as a non-toxic plant. The four crude extracts of *E. latideus* had good activity

With range of (IC_{50}) 3D7: (9.75-38.21) µg/mL and Dd2: (2.78-38.93) µg/mL. The resistance indices for *E. latideus* crude extracts ranged between 0.10- 1.43, suggesting that some of the extracts had equal promise against the CQ resistant strain of *P. falciparum*. Isolation resulted in the identification of three known compounds which include; three triterpenoids Lupeol (EL1), betulin (EL2), 3β-(5-hydroxyferuloyl)lup-20(30)-ene (EL3). Among the pure compounds EL2 had the highest activity against on both strains (IC_{50}) 3D7: 1.64 ± 0.02 µg/mL and Dd2: 7.69 ± 1.21 µg/mL while Lupeol (EL1) displayed moderate activity with (IC_{50}) 3D7: 23.91 ± 0.05

µg/mL, Dd2: 25.14 ± 0.01 µg/mL. The antiplasmodial activity of the crude extracts and pure compounds were significantly different ($P < 0.05$) from that of the reference standards (chloroquine diphosphate and mefloquine hydrochloride). Both the crude extracts IC_{50} (2.78-38.93) µg/mL and pure compounds IC_{50} (1.64-25.14) µg/mL showed a significant decrease in activity compared to the reference standards (0.0056-0.0440) µg/mL. Significant difference ($P < 0.05$) also existed between the antiplasmodial activities of the crude extracts, which showed the same trend with that of the pure compounds.

Conclusion: The results show that the root crude extracts and pure compounds of the plant have good antiplasmodial activity and low toxicity which can be exploited for malaria therapy. Therefore, this justifies their ethnomedicinal use of the plant by the local communities of Butebo Sub-County, in Pallisa District in Eastern Uganda in the treatment of malaria.

Keywords: Euclea latideus, antiplasmodial, acute toxicity, betulin, in vitro, Butebo