FRACTIONATION OF METHANOL EXTRACT FROM THE LEAVES OF *ENTADA SPIRALIS* RIDL. (SINTOK) USING VACUUM LIQUID CHROMATOGRAPHY AND ITS PRELIMINARY PHYTOCHEMICAL INVESTIGATION OF AN ACTIVE COMPONENTS

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ABSTRACT

FRACTIONATION OF METHANOL EXTRACT FROM THE LEAVES OF *ENTADA SPIRALIS* RIDL. (SINTOK) USING VACUUM LIQUID CHROMATOGRAPHY AND ITS PRELIMINARY PHYTOCHEMICAL INVESTIGATION OF AN ACTIVE COMPONENTS

This study was performed to investigate the preliminary phytochemicals and in-vitro antimicrobial properties against *Staphylococcus aureus* ATCC 33591 and *Streptococcus epidermis* ATCC 12228 from the leaves extract of *Entada spiralis* Ridl which can cause skin diseases to human. The fine-powdered leaf samples of *E. spiralis* were soaked with methanol. The soaking solution was evaporated using rotary evaporator at 75°C for 90 (rpm) to get the crude extract. Next the crude was undergoing fractionation process using two different solvents of dichloromethane (DCM) and methanol (MeOH) as the binary system. The fraction system used was fraction F1 (9:1); (DCM:MeOH), (fraction F2 (5:5)); (DCM:MeOH), and fraction F3 (3:7)); (DCM:MeOH). After fractionated, the solution was evaporated once again to get the crude extract based on the fractionated polarities. The crude extract was investigated with antimicrobial assay. The sample containing 400mg/mL of concentration from each three fraction system was pipetted onto a sterile paper disc on a Mueller-Hinton agar plate that had been spread with both *S. aureus* and *S. epidermis*. For the next method, the crude extract was undergoing TLC profiling to determine the compound present inside the leaves. The developing solvent for TLC profiling consisted of chloroform ($CHCl_3$) and methanol (MeOH) solvent with different ratios. The best developing solvent for fraction F1 and F3 was 9:1; ($CHCl_3$: MeOH) and 7:3 for fraction F2. And the last method, the crude extract for the three fraction systems was identified its functional groups using FTIR. As for the result, fraction F2 had the most active compound for both phytochemical investigation and antimicrobial properties. In TLC profiling, fraction F2 had all the compounds responsible for antimicrobial properties which were the antioxidant, terpenoid and phenolic compounds. For fraction F1 and F3, both fractions indicated the absence of phenolic compound. gave a negative result towards $FeCl_3$ only for fraction F3. While for the antimicrobial assay, fraction F2 had the biggest inhibition zone for both *S. aureus* and *S. epidermis* and proved to be the most susceptible compound towards both of the bacteria. As for the other two fractions, both had weak susceptibility towards *S. aureus* and *S. epidermis* due to small inhibition zone. Based on the result for both TLC profiling and antimicrobial assays, it was proved that fraction F2 was the most active fraction from the leaves extract of *E. spiralis* towards phytochemical screening and also antimicrobial properties.