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OP 28 - Antibacterial Screening of Chromatographic Fractions from Dichloromethane Crude Extract of Leaves of *Croton tiglium* (Japala)

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Background: Plant based medicines have over the years contributed to treat diseases and illnesses. Infectious diseases have become a leading cause for premature deaths worldwide. Evidence based validation of medicinal plants is the need of the day for developing better and safe antimicrobial agents. Chromatography is a significant analytical technique that plays an important role in natural product chemistry and it helps finding new therapeutic agents. *Croton tiglium* is a plant belongs to the family Euphorbiaceae that has a long role as traditional medicine.

Objectives: The current study was focused on the fractionation of crude ethyl acetate extract of *C. tiglium*, step by step visual demonstration of fractions and screening of antibacterial activity of those fractions.

Methodology: Crude extract was fractionated using a silica gel column chromatography eluted with hexane and ethyl acetate in stepwise gradients with increasing polarity. Antibacterial activity was evaluated by resazurin-based broth microdilution method against three bacterial strains; *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa*.

Results and conclusions: According to the results obtained, six fractions exhibited significant antibacterial activity against *S. aureus* and *P. aeruginosa* with minimum inhibitory concentration (MIC) in the range of 62.5- 125 μ g/mL. Interestingly, four fractions showed significant activity against *S. aureus* while five fractions showed significant activity against *P. aeruginosa*. The high-performance liquid chromatography (HPLC) and nuclear magnetic resonance (NMR) data indicated the presence of low-polar metabolites in the extract; Thus, further studies are needed to isolate, purify and elucidate the structures of these metabolites.

Keywords: Chromatography, Croton tiglium, resazurin, Staphylococcus aureus