# Phytochemical Profiling, Cytotoxicity Assessment and Molecular Mechanism of Action of South African Traditional “uMhlabelo” Methanolic Extracts

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South African traditional medicine, “uMhlabelo”, is widely used within the South African population to aid in the healing of wounds; however, there are no studies that elucidate on the chemical profiling and mechanism of action of these extracts. In this study we investigated two “uMhlabelo” herbal medicines to identify and characterize the potential compounds that ellicit proangiogenic functions leading to wound healing. The toxicity profiles of the extracts were assessed in liver cells and angiogenic profiles were assessed on HUVEC cells. HPLC-HRMS operating in $MS^E$ mode was used to identify, and compare the chemical compositions of methanol extracts Predictive pharmacokinetic and toxicological profiles of the compounds were assessed using the SwissADME and Protox II webservers, whilst the potential biological targets of the compounds were identified using the SwissTargetPredict online tool. UCSF Chimera was then used to predict the binding affinities of the bioactive compounds to their respective biological targets. C3A liver cells and HUVEC cells were cultured and treated with 11 extract doses (0.1 to 1000ug/ml) for 24 hours. Cell viability was then assessed via ATP assay. HPLC-HRMS analysis revealed six and five flavonoids in two of the methanolic extracts.No composition similarity was noted between the two extracts. Computational analysis revealed all compounds to be non-hepatotoxic. Swiss ADME prediction revealed that all compounds could be drug leads. Compounds (1, 11, 7, and 8) were identified as the most biologically relevant compounds (biological targets with P>0.50). Docking scores showed increased affinity to VEGF R2 and Cox-2 enzyme. ATP results showed extracts to be non-toxic at doses below 200ug/ml. uMhlabelo extracts were predicted and validated to be non-toxic in liver cells. The identified phytochemicals demonstrated a high binding affinity to proangiogenic initiator, VEGF-R2, and anti-inflammatory marker, Cox-2 indicating a potential mechanism of action of the extracts.