ISOLATION AND CHARACTERIZATION OF ANTIBACTERIAL AND ANTIFUNGAL COMPOUNDS IN *TERMINALIA BROWNII*

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DECLARATION

This thesis is my original work and has not been presented for the award of a degree in this or any other university.

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LIST OF ABBREVIATIONS

ATR	Attenuated Total Reflection
BDM	Bark Dichloromethane: Methanol (1:1) Extract
BM	Bark Methanol Extract
br d	Broad Doublet
br s	Broad Singlet
COSY	Correlation Spectroscopy
d	Doublet
dd	Doublet of Doublet
DEPT	Distortionless Enhancement by Polarization Transfer
DMSO	Dimethyl Sulfoxide
FDM	Flower Dichloromethane: Methanol (1:1) Extract
FM	Flower Methanol Extract
FTIR	Fourier Transform Infrared Spectroscopy
HMBC	Heteronuclear Multiple Bond Correlation
HRESIMS	High – Resolution Electrospray Ionization Mass Spectrometry
HSQC	Heteronuclear Single Quantum Coherence Spectroscopy
KEMFRI	*Kenya Marine and Fisheries Research Institute*
m	Multiplet
m-/z	Mass to Charge Ratio
MHz	Megahertz
NMR	Nuclear Magnetic Resonance
s	Singlet
t	Triplet
UV	Ultraviolet Radiation
WHO	World Health Organization

ABSTRACT

In ethnomedicine, *Terminalia brownii* is used to treat and control human illnesses such as yellow fever, rheumatoid arthritis, hepatitis, diabetes, diarrhea, stomach ulcers, and abdominal pain. The study sought to determine the antibacterial activity of crude organic extracts and characterize the components responsible for the action of *T. brownii* from commonly used plant bark and easily regenerated flower material. *T. brownii* plant was randomly identified. The fresh stem bark and flowers of *T. brownii* were collected from Mwingi sub-county, Kitui County in the month of April, 2021. The samples were air dried under shade at room temperature. The dry stem bark and flowers were ground into fine powder using electric grinder, weighed and packed in a clean sample container. The powdered plant materials were sequentially extracted with *n*-hexane, dichloromethane, dichloromethane:methanol (1:1), methanol and hot water (to simulate the conventional preparation process). Phytochemical screening of dichloromethane:methanol (1:1) and methanol crude extracts indicated the presence of alkaloids, flavonoids, tannins, terpenoids, saponins, steroids, phenols and glycosides. Antimicrobial activities of different crude extracts were recorded using agar well diffusion method and all extracts, except those obtained from *n*-hexane, demonstrated variable antibacterial activity against *E. coli, S. aureus*, and *P. aeruginosa* with inhibition zone ranging between 8.0±0.7 mm and 23.6±0.6 mm. Dichloromethane:methanol (1:1) extract (stem bark) exhibited the highest activity against *E. coli* with inhibition zone of 23.6±0.6 mm and 17.3±0.4 mm in flower extract while methanol bark extracts displayed good inhibitory activity against *S. aureus (*21.0±0.5 mm) and *P. aeruginosa (*21.7±0.5 mm) when compared to the other extracts. Only methanol extract exhibited antifungal activity against *C. albicans* with zone of inhibition of 15.0±1.5mm *(stem bark)* and 7.6±0.8mm (*flower).* The activity of the active crude extracts was comparable to commonly used over the counter drugs (Amoxicillin, 24.7±1.4, and fluconazole 19.0±0.3). Purification of the more active dichloromethane: methanol (1:1) and methanol extracts through silica gel column chromatography resulted in isolation of three new compounds: 1,4,7-tri-O-galloyl hept-6-deoxyheptose, 1,2,4-tri-O-galloyl-8,9-dideoxynonose,Rhamnetin-3-*O*-(2,3,6-trigalloyl)-β-D-glucopyranosideand seven known compounds: termiglaucescin, arjunglucoside – I, sericoside, 23-galloyl arjungenin, 28-*O*-β-D-glucopyranosyl-2,3,6-trihydroxy-23-galloylolean-12-dien-28-oate, 3,3',4',5-tetrahydroxy-7-methoxyflavone and 3,3',4',5,7-pentahydroxyflavone. The compound 1,4,7-tri-O-galloyl hept-6-deoxyheptose, was highly active against *E. coli* (16.5±0.7 mm) while termiglaucescin *showed good inhibitory activity against C. albicans (*16±5.7 mm).1,4,7-tri-O-galloyl hept-6-deoxyheptose, 1,2,4-tri-O-galloyl-8,9 dideoxynonose, Rhamnetin-3-*O*-(2,3,6-galloyl)-β-D-glucopyranoside,3,3',4',5-tetrahydroxy-7-methoxyflavone and 3,3',4',5,7-pentahydroxyflavone were isolated and characterized for the first time from dichloromethane:methanol (1:1) extract of *T.brownii* flower. Dichloromethane: methanol (1:1) crude extract of *T. brownii* flowers yielded three new bioactive compounds with remarkable antimicrobial activity against *E. coli, P. aeruginosa, S. aureus* and *C. albicans*. In conclusion this study has shown that flower extracts of *T. brownii*

have antifungal and antibacterial properties associated with stem bark extracts of *T. brownii*. Hence the more sustainable flower extracts are recommended for use in the treatment of bacterial and fungal infections.