



UNIVERSITY OF NAIROBI

**PHYTOCHEMISTRY AND BIOACTIVITY INVESTIGATIONS
OF THREE KENYAN *CROTON* SPECIES**

BY

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DECLARATION

This is original work by the author except where reference is made. It has never been submitted anywhere for award of any degree or diploma.

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DEDICATION

I dedicate this work to my children (Evans, Lewis and Michelle Mbithi).

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ABSTRACT

Three Kenyan *Croton* species, *C. megalocarpoides* Friis and Gilbert, *C. alienus* Pax and *C. sylvaticus* Hochst were investigated for their phytochemistry and biological activity relevancies. Anti-microbial activity evaluation was done on aqueous and methanol crude plant extracts to enable selection of most active parts. Documented procedures were used to profile the selected extracts for their phytochemical concentrations followed by fractionation using column chromatography. The phytochemicals obtained were identified using NMR spectroscopic techniques and subjected to various biological activity tests. Forty one compounds (fifteen of them new) were isolated. *C. megalocarpoides* roots produced twenty diterpenoids belonging to, *ent*-clerodane (thirteen, twelve new), abietane (three, one new) and *ent*-trachylobane (four known) series. Two known triterpenoids (lupeol and acetyl aleuritolic acid) and common phytosterols (stigmasterol and sitosterol) were also isolated. Two novel compounds (alienusolin, a 4 α -deoxyphorbol ester and crotonimide C, a glutarimide alkaloid derivative) and nine known compounds (an alkaloid, six methylcyclohexane derivatives of crotepoxide, a triterpenoid and a phytosterol) were isolated from *C. alienus* leaves and roots. From *C. sylvaticus* roots, seven diterpenoids belonging to clerodane (four, one new), halimane (two known) and labdane (one known) series and a phytosterol were isolated.

Anti-microbial activity tests were done using different strains of bacteria and fungi. *Candida albicans* was the most susceptible micro-organism to the crude plant extracts. *C. alienus* and *C. sylvaticus* (root and stem bark aqueous extracts) were active at the lowest concentration tested (25 mg / mL). *C. sylvaticus* stem bark (methanol extract) was the only crude extract that inhibited the growth of a bacteria strain (*Bacillus subtilis*) at a concentration of 10 mg / mL. The compounds that were isolated and assayed from *C. alienus* and *C. megalocarpoides* were inactive to all microorganisms used ($IC_{50} > 20\mu\text{g} / \text{mL}$). *C. alienus* leaves (MeOH: DCM, 1:1 v / v extract) is the only crude extract that showed activity against *Leishmania donovani* ($IC_{50} = 80\mu\text{g} / \text{mL}$). The compounds isolated from it were however inactive against the same, *L. donovani* (IC_{50} and $IC_{90} > 40\mu\text{g} / \text{mL}$). All the crude extracts and compounds isolated and tested from *C. alienus* and *C. megalocarpoides* were inactive against D6 and W2 strains of *Plasmodium falciparum* ($IC_{50} > 4760 \text{ ng} / \text{mL}$); VERO ($IC_{50} > 4760 \text{ ng} / \text{mL}$) and *Aedes aegypti* and *Anopheles gambiae* larvae (LC_{50} and $LC_{95} > 100 \text{ ppm}$). The methanol extract of *C. megalocarpoides* and *C. sylvaticus* stem barks had a low total phenolic content ($1.89 \pm 0.02\%$ - $1.14 \pm 0.01\%$ w / w equivalent of gallic acid) and anti-oxidant activity ($IC_{50} > 1000 \mu\text{g} / \text{mL}$ compared to ascorbic acid, $IC_{50} = 9.51 \pm 0.22 \mu\text{g}/\text{mL}$).

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