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LC/PDA/ESI-MS/MS polyphenols profiling of the bioactive fractions of *Croton zambesicus* fruits against *Madurella mycetomatis*

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Croton zambesicus Müll. Arg. locally known as Umgelagla belongs to the Euphorbiaceae family. It has a wide application in African traditional medicine. Ethnobotanically, the leaf decoction is used as an antimicrobial to treat various infections and for treating fever associated with malaria [1]. Different types of diterpenes including phorbol esters, clerodane, labdane, kaurane, trachylobane, and pimarane types have been isolated from this genus.

Air dried ground fruits of *C. zambesicus* were extracted using 70% methanol. The concentrated methanolic extract was sequentially fractionated with petroleum ether, chloroform and ethyl acetate. The crude extracts together with respective fractions and its seeds oil were tested against *Madurella mycetomatis* employing a microtitre plate-based antimycetomal assay incorporating resazurin as an indicator of cell growth [2].

The ethyl acetate fraction and seed oil exhibited a significant activity against *M. mycetomatis* with MICs of 39 and 78 µg/mL, respectively.

Reverse phase HPLC-DAD coupled with ESI tandem mass spectrometry employing CID experiments at an alternating mode led to the identification of four methoxylated derivatives of kaempferol and isorhamnetin together with the C-glycosides, vitexin and isoorientin in the ethyl acetate fraction of *C. zambesicus* fruits. We assume that the identified polyphenols, at least to a certain extent, accounts for the activity of *C. zambesicus* against *M. mycetomatis* and this result may validate its traditional use as an antimicrobial.

[1] Neuwinger, H.D.. African traditional medicine. A dictionary of plant use and application. Med. Pharm. Press Stuttgart, 2000 Germany.

[2] Khalid SA. Development of microtiter plate-based method for the determination of the MIC of antimycetomal agents against *Madurella mycetomatis*. II ResNet NPND workshop on natural products against neglected diseases, Nov. 25-28th, 2014, Rio de Janeiro, Brazil.