ACTIVITY-GUIDED PARTIAL PURIFICATION OF FREE RADICAL SCAVENGING COMPONENTS OF HYGROCYBE CONICA

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ABSTRACT

The aim of this study is to partially purify Hygrocybe conica and evaluate the free radical scavenging activities in fractions and sub-fractions with usage of silica gel column chromatography (CC), thin layer chromatography (TLC) and high performance liquid chromatography (HPLC). Evaluations of free radical activities were accomplished using various antioxidant activity assays (DPPH, ABTS and FRAP) and total phenolic content test. Free radical scavengers were extracted using water extraction and purified through liquid-liquid partitioning with organic solvents of hexane, chloroform, n-butanol, and formic acid. Crude extract that showed highest TPC value (6.00 µg GAE/ml of extract), with second highest DPPH radicals scavenging ability (31.43%) was selected to proceed with CC. In increasing gradient, CC was begun with hexane, ethyl acetate, and end with methanol. Among all three sub-fractions, SF-II (23.45%) and SF-III (23.22%) were identified by DPPH assay as the highest, and SF-III indicated highest TPC value which is 3.60 µg GAE/ml of extract. HPLC screening indicated that among all sub fractions, Compound 2 (SF-III, RT 3.059) was a match to catechin (RT 3.097), and quantified amount was 107.99µg/ml. All unknown A (RT 3.040), B (RT 12.082), C (RT 12.092) and D (RT 12.099) are suggested to proceed with LC-MS for further studies. As a conclusion, H. conica in general is not a high source of antioxidant.