

**ACTIVITY-GUIDED PARTIAL PURIFICATION  
OF FREE RADICAL SCAVENGING AND  
PHENOLIC COMPOUNDS FROM *Pleurotus* sp.**

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## ABSTRACT

Growing interest of scientific community on naturally derived products has fostered research on inhabitant of class Basidiomycetes. Recent studies reveals that *Pleurotus* sp., commonly known as 'oyster mushroom' possesses various pharmacological properties such as antioxidant, antimicrobial, antiviral, prebiotic, antihypertensive, antitumour, immunomodulatory, anti-inflammatory, and antihyperglycemic activities. This study designed at partial purification of *Pleurotus* sp. into its sub-fractions and investigation of their antioxidant activities. Dried *Pleurotus* sp. extracted with water, centrifuged and concentrated to obtain crude water extract, which then subsequently partitioned with solvents (hexane, chloroform, n-butanol and aqueous formic acid) in increasing polarity. Antioxidant activities were determined using 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2,2'-azinobis (3-ethyl-benzothiazoline-6-sulphonic acid) (ABTS), ferric reducing antioxidant power (FRAP) assays, and total phenolic content (TPC) were also evaluated. Antioxidant activity increased parallel with the elevation of concentration of the extract. A linear relationship existed between the DPPH with TPC ( $R^2=0.822$ ) and ABTS<sup>++</sup> with TPC ( $R^2=0.859$ ). Aqueous formic acid fraction exhibited significant high antioxidant activity with TPC of  $47.09 \pm 1.32$  mg of GAE/g of extract, FRAP of  $105.0 \pm 2.80$  mg of TEAC/g of extract, DPPH<sup>\*</sup> scavenging activities of  $64.29 \pm 0.54$  % with EC<sub>50</sub> of 219.69 µg/ml, and ABTS<sup>++</sup> scavenging activities of  $58.23 \pm 0.39$  % with IC<sub>50</sub> of 277.13 µg/ml at concentration of 300 µg/ml. The aqueous formic acid fraction was partially purified using Amberlite XAD-4, Amberlite XAD-7, and Amberlite XAD-16. Amberlite XAD-16 SF II exhibited significant high TPC ( $21.53 \pm 1.18$  mg of GAE/g of extract) and DPPH of  $27.60 \pm 1.20$  mg of TEAC/g of extract with EC<sub>50</sub> of 584.13 µg/ml. Amberlite XAD-16 SF I exhibited highest FRAP ( $22.60 \pm 1.30$  mg of TEAC/g of extract) while Amberlite XAD-7 SF I showed highest ABTS<sup>++</sup> scavenging activities of  $34.51 \pm 0.44$  % with IC<sub>50</sub> of 550.17 µg/ml. Fast Blue B reagent (FBB) and DPPH method revealed the presence of phenolic and free radical scavenging compounds respectively in the sub-fractions. Phytochemical screening performed on crude water extract revealed presence of flavonoids, terpenoids, saponins and tannins. Similar screening for the aqueous formic acid fraction revealed presence of all test compounds except tannins. All Amberlite XAD sub-fractions tested positive for saponins and flavonoids and negative for terpenoids and tannins, except for XAD-4 sub-fractions tested negative for flavonoids. FTIR analysis revealed presence of hydroxyl group, carbon double bond, C-H and C-O groups. Based on these results, *Pleurotus* sp. extract seems to possess antioxidant effect exerted by the phytochemical compounds and functional groups present.