ANTIOXIDANT ACTIVITY IN SELECTED LOCAL VEGETABLES

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ABSTRACT

A free radical is any atom or molecule that has a single unpaired electron in an outer shell and its damage is closely associated with oxidative damage. Oxidative damage by free radicals in the human body plays an important causative role in disease initiation and progression. Antioxidants provide a measure of protection that slows the process of oxidative damage. In this study, crude petroleum benzene, chloroform, methanol and water extracts of *Allium tuberosum* (garlic chives), *Apium graveolens* (L.) (celery), *Ipomoea batatas* (L.) (sweet potato leaves), *Murraya koenigii* (L.) (curry leaves), *Psophocarpus tetragonolobus* (winged bean), and *Sauropus androgynus* (sweet leaves) were evaluated for antioxidant properties using bioassays namely 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging assay, reducing power assay, metal chelating assay, haemolysate catalytic activity and lipid hydroperoxide assay. In the DPPH assay, petroleum benzene and chloroform extract of *Murraya koenigii* showed good free radical scavenging activities (IC_{50} = 21.5 µg/ml and 22.5 µg/ml respectively) when compared with standard ascorbic acid (IC_{50} = 3.75 µg/ml). IC_{50} is the concentration at which 50% of DPPH radicals are inhibited. In the reducing power assay, chloroform extract of *Murraya koenigii* showed strong reducing powers with absorbance value of 1.833 ± 0.003 when compared with standard butylated hydroxyanisole (BHA) with absorbance value of 2.625 ± 0.004 at 1 mg/ml. In the metal chelating assay, methanol extract of *Murraya koenigii* showed the highest metal chelating capacity of 88.60 ± 0.20% at 1 mg/ml when compared to ethylenediaminetetraacetic acid (EDTA) which was 98.63 ± 0.13%. In the haemolysate catalytic activity, water and chloroform extract of *Psophocarpus tetragonolobus*; methanol and chloroform extract of *Ipomoea batatas*; and chloroform extract of *Murraya koenigii* showed strong H_2O_2 reducing activities of 98.00 ± 0.16%, 92.53 ± 0.99%, 94.03 ± 0.18%, 90.79 ± 0.71% and 87.36 ± 0.77% respectively at 200 µg/ml while the standard absorbance by unblocked catalase is 99.95

(0.949) ± 0.02%. In the lipid hydroperoxide assay, methanol extract of *Ipomoea batatas* showed highest hydroperoxide value of 3.079 nmol whereas methanol extract of *Apium graveolens* showed lowest hydroperoxide value of 0.329 nmol at 25 nmol in comparison with hydroperoxide value of the lipid hydroperoxide standard which was 4.839 nmol obtained at 5 nmol. Thin layer chromatography (TLC) were carried out on four active crude extracts namely crude chloroform and petroleum benzene extract of *Murraya koenigii*, crude water extract of *Psophocarpus tetragonolobus* and *Apium graveolens* to screen secondary compounds present through different type of visualisation. A spot under shortwave UV light and iodine vapour visualisation, orange colour deposition in Dragendorff's reagent test and pink colour spot on TLC plate from sulphuric acid visualisation in all four active crude extracts indicates the presence of organic compounds, unsaturated carbon bonds, alkaloid compounds and oxidisable components, respectively. The presence of these organic compounds, unsaturated carbon bonds, alkaloid compounds and oxidisable component found in the selected vegetables may be linked to the high anti-oxidative activities observed; hence its dietary consumption may be beneficial for general health as well as disease prevention.

ABSTRAK

Radikal bebas adalah mana-mana atom atau molekul yang mempunyai satu elektron tunggal yang tidak berpasangan di petala luar dan kerosakan yang berkait rapat dengan kerosakan oksidatif. Kerosakan oksidatif oleh radikal bebas dalam tubuh manusia adalah penyebab yang memainkan peranan yang penting dalam permulaan dan perkembangan penyakit. Antioksidan menyediakan langkah perlindungan yang melambatkan proses kerosakan oksidatif. Dalam kajian ini, ekstrak mentah petroleum benzena, kloroform, metanol dan air dari *Allium tuberosum* (daun kucai bawang putih), *Apium graveolens* (L.) (saderi), *Ipomoea batatas* (L.) (daun ubi keledek), *Murraya koenigii* (L.) (daun kari), *Psophocarpus tetragonolobus* (kacang botol), dan *Sauropus androgynus* (daun manis) telah dinilai bagi antioksida dengan menggunakan bioassei iaitu asai penyahbebas radikal 1,1-difenil-2-picrylhydrazyl (DPPH), asai kuasa penurun, asai pengkelat logam, aktiviti katalitik haemolysate dan asai lipid hidroperoksida. Dalam asai DPPH, ekstrak petroleum benzena dan kloroform dari *Murraya koenigii* menunjukkan aktiviti penyahbebas radikal bebas yang baik (IC_{50} = 21.5 μg/ml dan 22.5 μg/ml masing-masing) berbanding dengan asid askorbik standard (IC_{50} = 3.75 μg/ml). IC_{50} ialah kepekatan di mana 50% daripada radikal DPPH dihalang. Dalam asai kuasa penurun, ekstrak kloroform dari *Murraya koenigii* menunjukkan penurunan kuasa yang kuat dengan nilai keserapan 1.833 ± 0.003 berbanding dengan hydroxyanisole butylated standard (BHA) dengan nilai keserapan 2.625 ± 0.004 pada 1 mg/ml. Dalam asai pengkelat logam, ekstrak metanol dari *Murraya koenigii* menunjukkan kapasiti pengkelat logam tertinggi iaitu 88.60 ± 0.20% pada 1 mg/ml berbanding asid atelindiamintetrasetik (EDTA) yang merupakan 98.63 ± 0.13%. Dalam aktiviti haemolysate katalitik, ekstrak air dan klorofom dari *Psophocarpus tetragonolobus*; ekstrak metanol dan kloroform dari *Ipomoea batatas*; dan ekstrak kloroform dari *Murraya koenigii* menunjukkan aktiviti penurunan H_2O_2 yang kuat dari 98.00 ± 0.16%,

92.53 ± 0.99%, 94.03 ± 0.18%, 90.79 ± 0.71% dan 87.36 ± 0.77% masing-masing pada 200 μg/ml manakala keserapan standard oleh katalase yang tidak disekat adalah 99.95 (0.949) ± 0.02%. Dalam asai hidroperoksida lipid, ekstrak metanol dari *Ipomoea batatas* menunjukkan nilai hidroperoksida yang tertinggi iaitu 3.079 nmol manakala ekstrak metanol dari *Apium graveolens* menunjukkan nilai hidroperoksida yang terendah iaitu 0.329 nmol pada 25 nmol berbanding dengan nilai hidroperoksida dari standard lipid hidroperoksida adalah 4.839 nmol diperolehi pada 5 nmol. Kromatografi lapisan nipis (TLC) telah dijalankan ke atas empat ekstrak mentah yang aktif iaitu ekstrak mentah kloroform dan petroleum benzena dari *Murraya koenigii*, ekstrak mentah air dari *Psophocarpus tetragonolobus* dan *Apium graveolens* untuk menyaring sebatian sekunder yang hadir melalui visualisasi yang berlainan jenis. Satu tompok di bawah cahaya UV gelombang pendek dan visualisasi wap iodin, pemendapan warna oren dalam ujian reagen Dragendorff, dan tompok berwarna merah jambu pada plat TLC dari visualisasi asid sulfurik dalam kesemua empat ekstrak mentah aktif menunjukkan kehadiran sebatian organik, ikatan karbon tak tepu, sebatian alkaloid dan komponen oxidisable masing-masing. Kehadiran sebatian organik, ikatan karbon tak tepu, sebatian alkaloid dan komponen oxidisable yang ditemui dalam sayur-sayuran yang terpilih boleh dikaitkan dengan aktiviti anti-oksidatif yang tinggi dapat diperhatikan; maka penggunaan diet ini mungkin bermanfaat untuk kesihatan umum serta pencegahan penyakit.

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Thank you very much.

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NOMENCLATURES

Abbreviations	Description
Abs	Absorbance
BHA	Butylated hydroxyanisole
DPPH	1,1-disphenyl-2-picrylhydrazy)
EDTA	Ethlene diamine tetra acidic acid
et al.	et alia (and other)
e.g.	exempli gratia (example)
$FeCI_2$	Ferrous chloride
$FeCI_3$	Ferric chloride
H_2O_2	Hydrogen peroxide
IC_{50}	Inhibition of concentration
i.e.	id est (it/this is)
MeOH	Methanol
$NaHPO_4$	Sodium phosphate monobasic
Na_2HPO_4	Sodium Phophate dibasic heptahydrate
NaOH	Sodium hydroxide
OH^-	Hydroxyl radical
α	Alpha
RNS	Reactive nitrogen species
ROS	Reactive oxygen species
rpm	revolution per minute
TCA	Tricholoracetic acid