

showed uniform green cells in the control HepG2 cells whereas apoptotic cells in the early stage were marked by yellow-green (10 ppm of CME, 5, 10 ppm of MPLCBA-3) and apoptotic cells in the late stage were marked with concentrated and asymmetrically yellow-orange nuclei (20, 40, 80 ppm of CME, 20, 40 ppm of MPLCBA-3, 10 ppm of thymoquinone [control]) under fluorescence microscope. Thus, HepG2 cells after treatment with CME and MPLCBA-3 fractions indicated cell apoptosis. Control had intact nuclei with uniformly dispersed chromatin in HepG2 cell lines. DNA fragmentation is a feature of apoptosis. HepG2 cells treated with CME and MPLCBA-3 fraction of *B. asiatica* and thymoquinone indicated DNA fragmentation, confirmed due to smearing observed in the gel electrophoresis when compared to control. When comparing CME and MPLCBA-3 fraction higher fragmentation was observed with the MPLCBA-3 fraction. The cell membrane damage as indicated by high LDH activity also correlates to the observation made under fluorescence microscopic pictures. When considering fluorescence microscopic pictures stained with Hoechst stain, at higher concentrations of

MPLCBA-3 fraction (20 ppm), condensed nuclei were observed in the HepG2 cells. These correlated with gel picture of DNA fragmentation of MPLCBA-3 fraction in HepG2 cell lines where smearing indicated that DNA were fragmented. The morphological characteristics indicated that these extracts cause apoptosis while biochemical changes linked with apoptosis included leaching of LDH indicating membrane damage and DNA fragmentation. Thus, both crude methanolic extract and MPLCBA-3 fraction have shown high cytotoxic potential due to membrane damage and DNA fragmentation causing apoptosis of HepG2 cells.

#### Keywords

*Barringtonia asiatica*, DNA fragmentation, LDH assay, Fluorescence microscopic analysis

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### Technical Sessions : A - 25

## Durian and rambutan peels as potential sources of antioxidants

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Rambutan (*Nephelium lappaceum* L.) and Durian (*Durian zibethinus* Murr.) are popular seasonal fruits grown in tropical countries, enriched in varieties of phytochemicals.<sup>1,2</sup> However, peels of these fruits cause unpleasant odors and serious environmental problems. As the exploration of bioactive compounds may lead to novel environmentally friendly drug discovery, the main objective of this study was to investigate the use of waste materials, durian and rambutan peels, as potential sources of antioxidants. Therefore, this work focused on determination of antioxidant activities of methanol extract of durian and rambutan peels using 1,1-diphenyl-2-picrylhydrazyl (DPPH) radical scavenging assay and determination of its total polyphenolic contents (TPC) and total flavonoid contents (TFC).

The chemical constituents of durian and rambutan peels were first extracted using methanol. Then the methanol extract was sequentially extracted with hexane, chloroform and methanol. The antioxidant activity of extracts of durian and rambutan peels and its fractions

were investigated using DPPH radical scavenging assay.<sup>3</sup> TPC of methanol extracts of durian and rambutan peels were determined using Folin-Ciocalteu method<sup>4</sup> and TFC of the extracts were analyzed using aluminium chloride method.<sup>4</sup> According to the results, all the extracts and its fractions showed antioxidant activity. The IC<sub>50</sub> values of methanol extract of rambutan peels (7.86±0.22 µg/mL), its hexane (13.49±0.52 µg/mL), chloroform (26.99±0.20 µg/mL) and methanol (30.85±0.97 µg/mL) fractions were lower than that of the control, butylated hydroxytoluene (BHT) (43.70±0.89 µg/mL). IC<sub>50</sub> values of methanol extract of durian peels (100.48±4.16 µg/mL), its hexane (>1000 µg/mL), chloroform (161.99±6.23 µg/mL) and methanol (>1000 µg/mL) fractions were higher than that of the control, butylated hydroxytoluene (BHT) (43.70±0.89 µg/mL). TPC of methanol extracts of durian and rambutan peels were found to be 2.98±0.03 and 14.80±0.21 mg GAE/g dry weight respectively. Higher TFC was observed in methanol extract of durian peels (30.87 mg Catechin /g dry weight) than in methanol

extract of rambutan peels (20.52 mg Catechin /g dry weight). The preliminary results of this study showed that the extracts of durian and rambutan peels are rich in compounds with potential antioxidant activity.

#### Keywords

Antioxidant activity, durian peels, rambutan peels, TPC, TFC

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### Technical Sessions : A - 26

## Complexation between Fe<sup>2+</sup> and 1,10-Phenanthroline-5-amine and the quenching mechanism

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1,10-Phenanthroline and its derivatives are very important chelating bidentate ligands for transition metal ions.<sup>1</sup> From previous studies, it has been investigated that Fe<sup>2+</sup> make stable complexes with 1,10-Phenanthroline and its derivatives.<sup>2</sup>

1,10-Phenanthroline-5-amine, which is a derivative of 1,10-Phenanthroline has an increased fluorescence quantum yield than its parent compound. Hence, to develop a fluorimetric method to analyze Fe<sup>2+</sup>, the behavior of 1,10-Phenanthroline-5-amine in the presence of Fe<sup>2+</sup> was studied. Furthermore, the limit of detection, limit of quantification and the quenching mechanism were found using a calibration plot between Fe<sup>2+</sup> and 1,10-Phenanthroline-5-amine.

Experiments were carried out in 95% acetonitrile solutions (pH 8.21). Excitation and emission wavelengths were at 267 nm and 515 nm, respectively. The fluorescence peak at 515 nm was quenched by Fe<sup>2+</sup>. The linear range was from 225.5 nM to 2850 nM with a detection limit of 7.8 nM at 3.3σ. Calculated limit of quantification was 235.36 nM.

Temperature effect was an evidence for the static quenching of 1,10-Phenanthroline-5-amine by Fe<sup>2+</sup>. Quenching of the probe by Fe<sup>2+</sup> at 278K, 288K, 298K, 303K and 313K were studied.

The study showed that 1,10-Phenanthroline-5-amine

can be used as a sensitive fluorescence sensor to detect Fe<sup>2+</sup> ions in nano molar level. No observable interference is observed upon addition of 225.5 - 2850 nM of Fe<sup>3+</sup> into 8 μM of 1,10-Phenanthroline-5-amine solutions.

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