

The Isolation of Hydroxychavicol from *P. betel* L. and its Effect on DNA Fragmentation

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Introduction: *Piper betel* is a vine widely grown in South East Asia. It was traditionally used as an ingredient for betel chewing as well as traditional herbal medicine for dermal irritation and fungal infection. Previous studies revealed that hydroxychavicol, a major phenolic compound from *P. betel* leaf, possessed antioxidative, antimutagenic and anticarcinogenic activities. Therefore, this study aimed to isolate this compound and to evaluate hydroxychavicol's effect on PUC19 plasmid DNA fragmentation. **Materials and Method:** Dried *Piper betel* L. leaves were macerated with 96% ethanol (EtOH). The EtOH extract was subsequently partitioned with *n*-hexane, dichloromethane (CH₂Cl₂) and ethyl acetate (EtOAc) respectively. The CH₂Cl₂ extract was further isolated on silica-columns. Fractions were collected and pooled according to TLC pattern. The fraction containing major compound was further purified on a Sephadex LH20 column. Structure elucidation of the compound was done by means of NMR and MS. Various concentrations of this isolated compound were subsequently tested for its effects on PUC19 plasmid DNA breaks. **Results:** By means of chromatographic techniques, a pure compound, hydroxychavicol (yield 0.008% dw), was isolated from *P. betel* leaf extract. Structural identification was accomplished by ¹H-, ¹³C-NMR and MS. Hydroxychavicol by itself at concentrations ranging 5, 8, 16 and 32 μM did not induce PUC19 DNA breaks but hydroxychavicol at concentrations of 0.5, 1, 2, and 5 mM respectively led to DNA damage. Interestingly, H₂O₂-induced DNA fragmentation was suppressed by hydroxychavicol at low concentration but DNA damage was markedly enhanced by hydroxychavicol at high concentration (>1mM). **Conclusion:** This study shows that hydroxychavicol is isolated from *P. betel* and its effect on DNA fragmentation depends on the exposed concentrations. Therefore, further study should be conducted to investigate its underlying mechanisms.

Keywords: hydroxychavicol, *Piper betel* L., DNA fragmentation

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Determination of Biomarker in Cassava leaves and the Minimization of Cyanogenic Glycosides

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Introduction: Cassava is one of the major crops of Thailand cultivated mainly for food and tapioca flour. Cassava root is the main part-used of cassava whereas the leaves are by-products used in animal feed as a source of protein. The current study aims to promote the use of cassava leaves focusing on the biomarker compounds of 3 selected cassava cultivars (Rayong 9, Huay Bong 60 and Mun Ha natee) and the minimization of cyanogenic glycosides (linamarin) in cassava leaves (Huay Bong 60). **Materials and Method:** All 3 cultivars of cassava leaves were collected from KhonKaen field crops research center. Fresh leaves of three cassava cultivars (Rayong 9, Huay Bong 60 and Mun Ha natee) were extracted with 80 % ethanol. The extracts were used to study TLC chromatogram and to identify the biomarker compound of each cultivar. Fresh leaves of Huay Bong 60 were fermented with various traditional recipes using sugar and salt at various ratio. The extracts were analyzed for the linamarin and biomarker compound using the validated TLC densitometer. **Results:** Rutin was found as a biomarker in leaves of three cassava cultivars at R_f 0.40 ± 0.02. The solvent system was ethyl acetate: formic acid: glacial acetic acid: water with the ratio 100:10:10:12, respectively. The optimal ferment recipe for the leaves of Huay Bong 60 was fresh leaves with sugar and salt in the ratio of 4:1:1 by weight, respectively. Linamarin was not detected in both fresh leaves and fermented leaves. Whereas the content of rutin in fresh leaves and fermented leaves were 3.17 ± 0.07 % and 0.87 ± 0.04% fresh weight, respectively. **Conclusion:** TLC densitometry is a rapid and convenient technique to determine and compare rutin content in various cultivars of cassava leaves. The existence of rutin, an antioxidant compound with the capillary-protective action, as a biomarker in cassava leaves is particularly encouraging. Cassava leaves can be used as new source of rutin or develop them to the health food supplement in the future.

Keywords: Cassava leaves, Biomarker, Cyanides

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