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NOTE

Free Radical-Scavenging and Antioxidant Activity of Skin of *Dictamnus dasycarpus*

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Various solvent extracts of *Dictamnus dasycarpus* skin, a traditional Chinese medicine material, were screened for antioxidant activities. Three tested systems *in vitro* were employed to investigate the antiradical and antioxidant effect, including DPPH, ABTS radical scavenging assay and reducing power. The results revealed that the acetone extract exhibited outstanding antioxidant activities. The results showed that these extracts, especially acetone extract, could be considered as natural antioxidants.

Key Words: DPPH, ABTS, Reducing power, Skin of *Dictamnus dasycarpus*.

Over-production of reactive oxygen species, the major free radical generated in normal metabolic processes, was induced by exposure to external oxidant substances or a failure in the defense mechanisms[1-3]. Reactive oxygen species will cause a variety of biochemical and physiological lesions and often results in metabolic impairment and cell death[4-6]. Plants, including herbs and spices, have been considered as potential resources for natural antioxidants, because they are rich in components that have antimutagenic, anticarcinogenic, anti-inflammation and antioxidant activities[7-9].

Dictamnus dasycarpus, belonging to the Rutaceae family, is widespread in China[10]. The leaf of *Dictamnus dasycarpus* is one of the most important traditional medicines. It is used as a remedy for a wide range of diseases, for instance, prurigo, icterus, rheumatism, cough and epilepsy. However, there isn't any information on antioxidant activity of *Dictamnus dasycarpus* leaf that has been reported. In the present paper, the antioxidant activities of various solvent extracts from *Phymatopteris hastata* were investigated by employing three tested systems *in vitro*: DPPH, ABTS and reducing power.

Extraction: Fifty grams of air-dried plant materials was immersed in 500 mL of ethanol (95 %), acetone and ethyl acetate, respectively and the filtrate was collected for three times at every 48 h interval. The extract was then concentrated under reduced pressure at 40 ºC using vacuum rotary evaporator. Thus, the acetone extract (AE), ethanol extract (EE) and ethyl acetate extract (EEE) were obtained.

DPPH radical scavenging assay: Extract solution (0.5 mL) in 95 % ethanol was added to 8 mL 0.004 % (w/v) solution of DPPH in 95 % ethanol. The absorbance at 515 nm was measured at 30 min.

ABTS radical scavenging assay: $ABTS^{\bullet+}$ was produced by reacting 7 mM ABTS stock solution with 2.45 mM potassium persulfate and the mixture was left to stand in the dark at room temperature for 12-16 h before use. The $ABTS^{\bullet+}$ solution (stable for 2 days) was diluted with 5 mM phosphate-buffered saline (pH 7.4) to an absorbance of 0.70 ± 0.02 at 730 nm. After addition of 150 µL of sample to 4 mL of diluted $ABTS^{\bullet+}$ solution, an absorbance reading was taken at 0.5 h.

Measurement of reducing power: Fractions solutions (0.5 mL) in 95 % ethanol were mixed with phosphate buffer (2.5 mL, 0.2 M, pH 7.4) and potassium ferricyanide (2.5 mL, 1 %). After the mixture was incubated at 50 ºC for 20 min, 2.5 mL of trichloroacetic acid (10 %, w/v) was added and the mixture was then centrifuged at 3000 rpm for 10 min. 2.5 mL of the upper layer of the above solution was mixed with 2.5 mL of distilled water and 0.5 mL of ferric chloride (0.1 %) and then the absorbance was measured at 700 nm.

DPPH radical scavenging activity: The DPPH radical scavenging activity of the extracts was shown in Fig. 1A. As can be seen, the scavenging effect of the extracts increased with increasing concentration. For ethanol extract, a sharp increase of its DPPH scavenging activity (42.6-75.7 %) was observed, as its concentration ranging from 0.5 to 2.0 mg/mL.

acetone extract, ethanol extract and ethyl acetate extract processed significant scavenging activity on DPPH radical. Hereinto, acetone extract exhibited the lowest highest DPPH scavenging effect, indicating that acetone extract was a prominent scavenger against DPPH radical.

ABTS radical scavenging activity: The ABTS radical scavenging capacities of ethanol extract, BE, PE and BHT were measured and compared in Fig. 1B. The ABTS radical scavenging activities of all extracts increased in a concentration dependent manner, which increased with increasing concentrations. The scavenging capacities decreased as the following order: AE< EE< EEE. In the three extracts, acetone extract exhibited the most effective scavenging ability, while the lowest one was found to be the ethyl acetate extract. The order of scavenging activities on ABTS radical of the three extracts was similar to that on DPPH. The differences of ABTS scavenging activities existed in acetone extract, ethanol extract and ethyl acetate extract indicated that the extracting media significantly influenced the antioxidant abilities of the extracts.

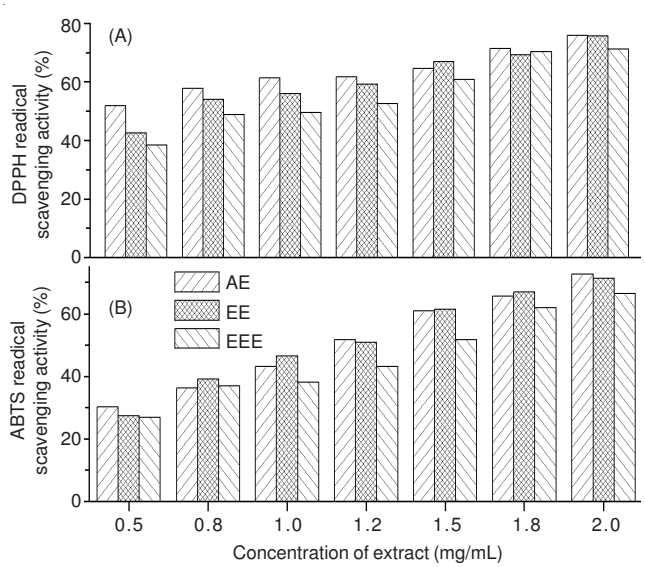

Fig. 1. DPPH radical scavenging activity A) and ABTS radical scavenging activity B) of various solvent extracts from skin of *Dictamnus dasycarpus*

Reducing power: The reducing power of various solvent extracts from skin of *Dictamnus dasycarpus* was shown in Fig. 2. acetone extract was the most outstanding at various concentrations. The reducing power order was: BHT > EE > BE > PE. The tendency for reducing power of various solvent extracts from Phymatopteris hastata was similar to DPPH and ABTS radical scavenging activities, indicating that there is a correspondence in antioxidant activities of various solvent extracts from *Phymatopteris hastata*.

Conclusion

In the present investigation, extracts of skin of *Dictamnus dasycarpus* exhibited outstanding scavenging effect on DPPH and ABTS radical and possessed strong reducing power. Acetone extract was proved to be the most efficient extract. The antioxidant activities in all tested system followed the same order: AE > EE > EEE. Based on the above results, various solvent extracts of *Dictamnus dasycarpus* skin, especially acetone extract extract, were found to be excellent scavengers for free radical and possess remarkable antioxidant ability.

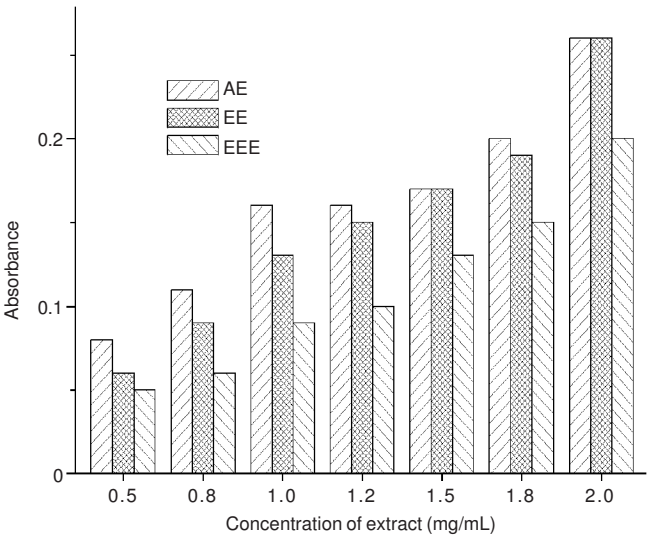

Fig. 2. Reducing power of various solvent extracts from skin of *Dictamnus dasycarpus*

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