

ISOLATION OF MORIN FROM THE WOOD OF THAI *ARTOCARPUS HETEROPHYLLUS*

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ABSTRACT

The wood of Artocarpus heterophyllus wildy used in Thailand for medicinal purposes included good health and tonic. One active compound that response for these purposes shall be the antioxidant morin (2',3,4',5,7-pentahydroxy flavone). The method for preparation of morin from the wood of Thai *A. heterophyllus* was developed. Firstly, the wood was macerated in methanol to provided 6.79 % of crude precipitate. Then, the crude precipitate was dissolved in ethyl acetate and the filtrate was extracted with 5% sodium carbonate solution. Finally, the aqueous layer was acidified with 10% hydrochloric acid to precipitate 6.10% of morin. This method used only the chemical being friendly to the environmental and safety for human. It shall be usefull for preparation of morin from *A. heterophyllus* for using in antioxidant therapy.

Keywords: *Artocarpus heterophyllus*, jack-fruit tree, isolation, morin, 2',3,4',5,7-pentahydroxy flavone

INTRODUCTION

The process of lipid peroxidation initiated by free radical, contributes to cell aging and pathological disorders such as atherosclerosis, myocardial infarction, inflammation, and cancer. In order to control lipid peroxidation, a large number of antioxidant have been investigated. Vitamin E, vitamin C and uric acid analogues were synthesized. Also, natural products, some of them belonging to the flavonoid class, have been developed (Repine, 1991; Flora et al, 1991; Elliott and Kandaswami, 1993; Andersson et al, 1996). There were reports that morin (2',3,4',5,7-pentahydroxy flavone)-a number of flavonoids found in plants such as *Chlorophora tinctoria*, *Artocarpus heterophyllus*, *Cudrania javanesis* and *Morus alba*-acts as broad spectrum antioxidant (Prista and Alves, 1958; Spada et al, 1958; Wu et al, 1994; Krol et al, 1994; Wu et al, 1995)and has none toxicity (Cho et al, 2006).

The wood of *A. heterophyllus* (Jack-fruit tree) wildy used in Thailand for many purposes included good health and tonic. One active compound that response for these purposes shall be the antioxidative compound morin (Santiarwon et al, 1998; Saiin, 2006). Structure-activity relationship studies of morin were demonstrated that (i) two hydroxy groups on C-ring and one hydroxyl group at 3-position and (ii) C2-C 3unsaturated bond (C=C) increase antioxidative activity. Moreover, the antioxidative mechanisms of morin were considered. First, morin acted as free radical scavenger. Second, morin could chelate some metal ions such as iron(II) ion in Fenton reaction (Tauber et al, 1984; Pagonis et al, 1986; Hodnick et al, 1986; Decharneux et al, 1992; Mora et al, 1990; Limasset et al, 1993). The recent studies have shown that morin has the important pharmacological activities such as antiangiogenic, *in vivo* anti-inflammatory, antinociceptive activities (Jung et al, 2010) and it has a potential as an anticancer agent (Manna et al, 2007; Yuan et al, 2012; Karimi et al, 2013).

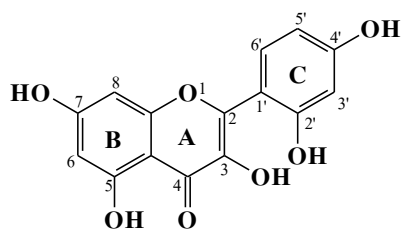


Figure 1: Chemical structure of morin(2',3,4',5,7-pentahydroxy flavone)

Study by using thin layer chromatography indicated that the methanol extract of the wood of Thai *A. heterophyllum* showed the high yield of morin. This result indicated the potential to develop morin from the wood of Thai *A. heterophyllum* for using in antioxidant therapy. Herein, the green method for preparation of morin from the wood of Thai *A. heterophyllum* was carefully reported.

MATERIALS AND METHOD

Plants Material

The wood of *A. heterophyllum* was brought from well known Thai traditional medicine store in Phitsanuloke province, Thailand.

Extraction method

5.2 kg of a pieced wood of *A. heterophyllum* was macerated in 36 L of methanol for one day. Then, it was filtered, concentrated under reduced pressure and let the filtrate precipitate to obtain crude precipitate. The residue plant was extracted again using the same process.

Isolation and Purification

1.0 g of crude precipitate was dissolved in 10 ml of ethyl acetate. The filtrate was collected and evaporated on water bath to 7 ml. Then, the filtrate was extracted with 5 ml of 5% sodium carbonate solution. The aqueous layer was acidified with 10% hydrochloric acid to precipitate. The precipitate was washed with cool water and dry in oven to obtain powder of morin.

Structure elucidation

The chemical structure of pure compound was elucidated from $^1\text{H-NMR}$ spectrum. This spectrum was performed with a 400 MHz $^1\text{H-NMR}$ spectrometer Avance Bucker, Germany. The solvent for this spectrum was deuterated methanol using tetramethylsilane as the internal reference standard. The chemical shifts were reported in the ppm scale.

RESULTS AND DISCUSSIONS

Extraction, isolation and purification

The method for preparation of antioxidant agent morin from the wood of Thai *A. heterophyllum* was developed. The extraction of the wood provided 351.9 g (6.79%) of crude precipitate (Figure 2). Further isolation and purification of 1.0 g of crude precipitate provided 61 mg (6.10%) of pure morin (Figure 3).

Structure elucidation of pured morin

The structure of morin was determined using $^1\text{H-NMR}$ technique. The $^1\text{H-NMR}$ spectrum corresponded to the chemical structure of standard morin (Sigma-Aldrich); $^1\text{H-NMR}$ 400

MHz, CD₃OD) δ 6.17 (1H, t, J=1.90, H-6 or H-8Hz), 6.34 (1H, t, J=1.80, H-6 or H-8Hz), 6.39 (1H, d, J=1.85, H-3Hz), 6.43 (1H, dd, J=8.58, 1.85Hz, H-5,7), 6.47 (1H, d, J=8.54Hz, H-6)

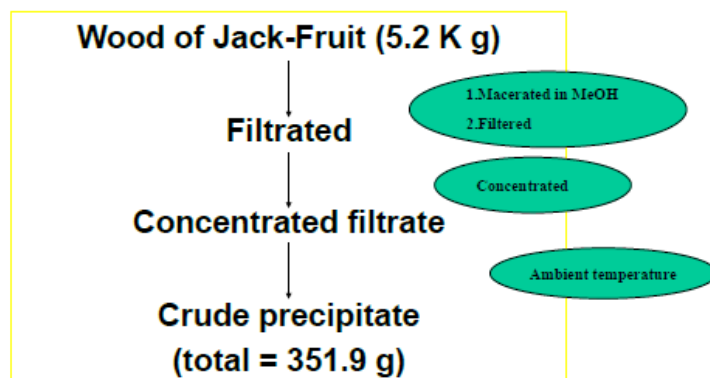


Figure 2. Extraction of morin from the wood of *A. heterophyllus*

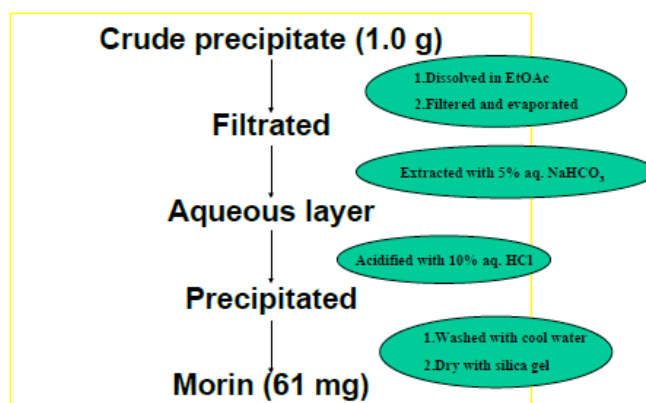


Figure 3. Isolation and purification of morin from the crude precipitate

CONCLUSION

The method for preparation of antioxidant agent morin from the wood of *A. heterophyllus* was developed. This method used only the chemical being friendly to the environmental and safety for human. It shall be useful for preparation of morin from the wood of *A. heterophyllus* for using in antioxidant therapy.

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