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## Investigation of Ethyl Acetate Soluble Proanthocyanidins in the Inflorescence of *Cocos nucifera* L.

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**Abstract**

The immature inflorescence of *Cocos nucifera* L. variety aurantiaca is used in Ayurveda for the treatment of menorrhagia in Sri Lanka. Preliminary phytochemical screening revealed that immature inflorescence predominantly contains proanthocyanidins. The ethyl acetate soluble proanthocyanidin (EASPA) fraction obtained from an acetone/water (7:3) extract of the immature inflorescence was effectively purified and separated from other phenolic compounds by chromatography on sephadex LH-20. EASPA has been fully characterized by chromatographic, spectroscopic and spectrometric methods. Acid catalyzed cleavage followed by TLC indicated that EASPA is composed of (epi)catechin and (epi)afzelechin monomeric units while thiolysis followed by <sup>1</sup>H and <sup>13</sup>C NMR spectroscopy revealed the monomers to be the epi-isomers. <sup>13</sup>C NMR studies of EASPA showed signals characteristic for epicatechin units indicating that EASPA is composed mainly of epicatechin units. Confirming these epicatechin oligomers with degrees of polymerization ranging from 2-5 and mixed oligomers, epicatechin-epiafzelechin dimer and epicatechin-epicatechin-epiafzelechin trimer were observed by ESI-MS.

**Keywords:** *Cocos nucifera* inflorescence, proanthocyanidin, epicatechin, epiafzelechin, thiolysis.

**1. Introduction**

*Cocos nucifera* L., the coconut palm is found widely distributed in the Asian continent and in parts of South America and Africa [1]. It is a member of the monocotyledonous family Arecaceae (Palmae) and is the only species of the genus. Coconut palm is a cultivated crop in Sri Lanka, which is a major producer of coconut in the world.

In Sri Lanka, the immature inflorescence of *Cocos nucifera* L. is used by Ayurvedic and traditional medical practitioners for the treatment of menorrhagia. *Cocos nucifera* L. is classified in to three varieties in Sri Lanka: Typical, Nana and Aurantiaca [2]. Of this, the orange coloured variety aurantiaca, is used for this purpose. Our preliminary phytochemical screening revealed that the inflorescence of *Cocos nucifera* L. contains high level of proanthocyanidins. Presence of proanthocyanidins in husk fibre of coconut [3], green coconut bark [4] and coconut water [5] have also been reported. Proanthocyanidins have recently attracted a considerable amount of attention in the fields of medicine, health and nutrition. They have been reported to exhibit antioxidant [6], anti-inflammatory [6], bacterial anti-adhesion [7], anticancer [8], and cardioprotective [9] activities. There is evidence also to suggest that proanthocyanidins may play a role in the treatment of menorrhagia [10]. Proanthocyanidins are oligomers or polymers made up of flavan-3-ol monomeric units. The most common flavan-3-ol units are (+)-catechin, (-)-epicatechin, (+)-gallocatechin and (-)-epigallocatechin, while (+)-afzelechin and (-)-epiafzelechin have been reported to a lesser extent (Fig. 1) [11]. These monomers may carry acyl/glycosyl substituents linked to the C-3 or the C-5 position. The flavan-3-ol units in proanthocyanidins are mainly linked through C-4 to C-8 or sometimes C-4 to C-6 bonds. Proanthocyanidins that contain only these linkages are named as B-type proanthocyanidins. When additional ether linkages are found (usually between C-2 and C-7), the compounds are named as A-type proanthocyanidins. A large variety of different proanthocyanidins have been reported that differ depending on the monomeric unit, substitution pattern of the monomeric unit and the extent of oligomerization.

In this paper, we report the extraction, purification and characterization of the ethyl acetate soluble proanthocyanidin (EASPA) fraction of the immature *Cocos nucifera* L. inflorescence.