

High-Performance Liquid Chromatographic Method for Separation and Quantitative Analysis of Arbutin in Plant Tissue Cultures

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ABSTRACT

*A simple and sensitive isocratic high-performance liquid chromatographic (HPLC) method for the quantitation of arbutin, a tyrosinase-enzyme inhibitor, commonly used as a whitening agent in cosmetic products, is reported. A system which consisted of a reverse-phase Apollo C-18 column (4.6x150 mm), a methanol-water (10:90) mobile phase, a flow rate of 0.9 mL/min and a UV detection at 280 nm provided a highly-sensitive and reproducible assay. Simultaneous separation and determination of arbutin from its precursor, hydroquinone, in plant cell suspension culture samples at amounts as low as 0.16 µg was achieved within 10 minutes. Sharp symmetrical peaks were obtained and a linear relationship between the amount of arbutin injected and the area under the peak was achieved over the range of 0.016 to 1.024 mg/mL with the relative standard deviation of less than 0.4%. Using this method of analysis, it was demonstrated that cell suspension cultures of *Capsicum annuum* L., *Solanum aculeatissimum* Jacq., and *Datura fastuosa* L. (Solanaceae) were able to biotransform exogenous hydroquinone into arbutin, while those of *Ocimum basilicum* L. (Lamiaceae) and *Allamanda cathartica* L. (Apocynaceae) did not show such ability.*

Key words: High-performance liquid chromatography (HPLC), arbutin, hydroquinone, plant cell suspension culture

INTRODUCTION

Arbutin [1], a phenolic glucoside found in many plants such as bearberry (*Arctostaphylos uva-ursi*), cranberry (*Vaccinium macrocarpon*) and blueberry (*Vaccinium corymbosum*) (Ericaceae), as well as bergenia (*Bergenia crassifolia*) (Saxifragaceae) and pears (*Pyrus communis*) (Rosaceae), is a known inhibitor of tyrosinase enzyme and has been employed as a whitening agent in many cosmetic preparations. It is structurally related to hydroquinone (Figure 1[2]), a compound which also possesses a tyrosinase-inhibitory effect but the legal use as an ingredient in topical cosmetic products has been prohibited in Thailand due to its toxicity and side effects. The advantages of arbutin over hydroquinone are that it is more stable,