Simultaneous Detection of Protocatechuic Acid, Chlorogenic Acid and Caffeic Acid in Honey by HPLC with Ultraviolet and Electrochemical Detectors

Zhongyin Zhang¹, Wanqing Zhang¹, Jianqiu Tu¹, Yin Li¹, Guangri Xu¹, and Yuping Zhang^{1*}

Henan Institute of Science and Technology, Xinxiang, 453003, China

*E-mail: zhangwqzzu@163.com, franklin525@126.com

doi: 10.20964/2018.07.44

Received: 13 January 2018 / Accepted: 1 May 2018 / Published: 5 June 2018

A rapid and sensitive method for the detection of protocatechuic acid (PA), chlorogenic acid (CLA) and caffeic acid (CAA) has been developed by high-performance liquid chromatography (HPLC) with an ultraviolet detector (UV) and an electrochemical detector (ECD). HPLC with UV detection was performed on a C₁₈ column (250×4.6 mm, 5 μm) with methanol (pH 4.0) - acetic acid (v/v, 85/15) as the mobile phase at a flow rate of 1.0 mL/min. ECD detection was performed on a three-electrode system, with a glassy carbon electrode, a platinum foil and a saturated calomel electrode (SCE) as the working electrode, the auxiliary electrode and the reference electrode, respectively. Compared with individual HPLC-UV and ECD methods, the presented HPLC (UV-ECD) technique had the advantage of enhanced capture of electroactive PA, CLA and CAA and achieved obvious detection sensitivity. Under the optimized conditions, the linear concentration ranges were from 0.0005 to 6 mg L⁻¹ for PA and CAA, and from 0.0015 to 18 mg L⁻¹ for CLA, with limits of detection of 10 ng L⁻¹ for PA, 30 ng L⁻¹ for CLA and 13 ng L⁻¹ for CAA (S/N = 3), which were clearly lower than those obtained with the HPLC-UV method. The approach was also successfully applied to detection in honey samples with satisfactory recoveries.

Keywords: HPLC-UV method; HPLC-ECD method; HPLC (UV-ECD) technology; protocatechuic acid; chlorogenic acid; caffeic acid

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