



## Simultaneous Quantification of Protocatechuic Acid, Caffeic Acid, Quercetin and Kaempferol in *Urena lobata* L. by HPLC-VWD

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**SUMMARY.** A simple, rapid and specific HPLC method was developed with variable wavelength UV detection (VWD) for simultaneously quantification of four analytes in *Urena lobata* L., namely protocatechuic acid, caffeic acid, quercetin and kaempferol, with the help of Doehlert matrix and desirability function approach to optimize the extract procedure conditions for sample preparation. The chromatographic separation was performed on a Thermo Synchronis - C<sub>18</sub> (250 × 4.6mm, 5 μm) by gradient elution of methanol and 0.5 % acetic acid at 25°C, 258, 320 and 360 nm of VWD which were switched to determine the corresponding analytes. Protocatechuic acid was determined at 258 nm; caffeic acid was determined at 320nm; kaempferol and quercetin were determined at 360 nm. The method was validated through the following performance criteria: linearity, precision, repeatability, stability, recovery, limit of detection (LOD) and quantification (LOQ). This assay was successfully used for determination of four ingredients in 10 raw herbs collected from different regions in Guangxi province, China. Significant variations were demonstrated in the contents of four ingredients in these samples. This HPLC method provided a feasible and convenient technique for simultaneous determination of four analytes with different maximum UV absorption in one assay by VWD.

**KEY WORDS:** Caffeic acid, HPLC-VWD, Kaempferol, Protocatechuic acid, Quercetin, *Urena lobata* L.

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