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Guest Editorial

UPLC in phytochemical analysis

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Ultra Performance Liquid Chromatography (UPLC), which has been around just over one and a half decade, is an advanced liquid chromatographic technique that offers a significantly short analysis time and small amount of solvent(s) as a mobile phase (Nováková et al., 2006). It also provides much better separation efficiency and resolution of analyte mixtures. This system utilises a special column packed with much smaller particles (typically 1.5-1.7 μm), instead of 3 or 5 μm particles used routinely in column packing for phytochemical analysis. As a result, the run time in an UPLC system can be up to three and nine fold shorter than that of the LC systems using 3 and 5 mm columns, respectively. The column size is usually 50-100 mm. The separation in UPLC is achieved under extremely high pressure (100 MPa or 14.5 K psi). Like any other modern LC systems, UPLC can be hyphenated with various types of detection techniques, UV, PDA or MS. A UPLC system enables the detection of analytes at a very low concentration owing to its improved signal-to-noise ratio, and requires much smaller injection volume without any loss of sensitivity.

Waters was the first manufacturer to introduce commercially this system by the trade name of Acquity in 2004. The driving force behind this introduction of a new LC system was to provide chromatographic run times up to ten times shorter than those of the fastest existing HPLC systems at that time, with up to two times better peak capacity or resolution, and three times better routine sensitivity. These enhanced performance characteristics could be translated into more and higher-quality information per unit of time as well as greater productivity. In the first UPLC system, a particle size of around 1.7 μm was used, which could allow greater speed and peak capacity, but also required a much higher pressure to help move the eluent through the column. The latest UPLC model by Waters, called Acquity

UPLC Plus Series, was unveiled last year to introduce new performance benchmark for chromatographic separations in the analytical laboratories (<https://www.businesswire.com/news/home/20180410005189/en/>).

Because of different obvious advantages over conventional HPLCs or UHPLCs, UPLC has now become a routine technique for chemical, biomedical and pharmaceutical analysis as well as for the analysis of phytochemicals. Since the introduction of UPLC, this technique has been used quite extensively in phytochemical analysis, and to date, several thousand of scientific papers have been published on phytochemical studies using UPLC methods. The application of UPLC in phytochemical analysis is mainly limited to quantification of certain secondary metabolites in plant extracts, fruits and vegetables, as well as plant metabolomics and chemical fingerprinting studies. For example, Cotrut and Bădulescu (2014) carried out the quantification of vitamin C present in several fruits and vegetables. Similarly, 17 commercial teas made from *Chrysanthemum morifolium* and *Coreopsis tinctoria*, have recently been analysed by UPLC coupled with Q-TOF-MS detector (Li et al., 2019a), mainly focusing on their phenolic profiles. Zhou et al. (2019) reported the UPLC-Q-TOF/MS-based untargeted metabolomics coupled with chemometrics (Sarker and Nahar, 2018) approach for Tieguanyin tea with seasonal and year variations.

UPLC is particularly useful in the pharmacokinetic studies of phytochemicals using various animal models. The application of UPLC in pharmacokinetic study of phytochemicals in animal models can be exemplified by the recent report on the determination and pharmacokinetic study of echinatin, a chalcone from *Glycyrrhiza echinata*, in rat plasma (Li et al., 2019b). A classic example of the use of UPLC in fingerprinting

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analysis of phytochemicals is evident in the recent publication by Lu et al. (2019), where they carried out a UPLC fingerprint analysis together with chemometric methods, including similarity analysis and hierarchical clustering analysis (Sarker and Nahar, 2018) to simultaneously identify 29 components including 18 monoterpene glycosides and 11 phenolic compounds in three Chinese Traditional Medicine preparations containing various Paeoniae samples. The effects of nitrogen supply on biosynthesis and accumulation of various flavonols and their glycosides in *Camellia sinensis* have been studied using UPLC-QqQ-MS/MS technique (Dong et al., 2019).

Despite a series of advantages of using UPLC, like more selectivity and sensitivity with high resolution performance and faster resolving power, reduced process cycle time, enhanced end-product quality with reduced cost of operation and decreased run time, decreased consumption of solvent and increased sample throughput, and the convenience of real-time analysis in step with manufacturing processes, UPLC still suffers from some drawbacks. A major disadvantage of UPLC is the higher back pressures compared to conventional HPLC, which decreases the life of the columns, and the particles of less than 2 μm cannot be regenerated and, therefore, have a narrow use.

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