Comparative Study of Conventional HPLC, Rapid Resolution LC and Nano-liquid Chromatography Coupled to Electrospray Ionization Time-of- Flight Mass Spectrometry for the Analysis of Olive Oil Polyphenols

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Considering the importance of the phenolic fraction of extra virgin olive oil due to its antioxidant activity, organoleptic properties and healthy effects, a comparative study using different liquid chromatography approaches: conventional HPLC, rapid resolution LC (RRLC) and nanoLC was developed for the analysis of this kind of analytes. Adequate conditions for chromatographic separation, electrospray ionization and mass spectrometry detection were investigated. Firstly, based on the chromatographic conditions of a previous HPLC method (column with 5 µm particle size, 0.5 ml/min, 25ºC), the optimization of a new RRLC method was carried out. Using columns of very small particle diameter (1.8 µm) and higher flows (1.5 ml/min at 30ºC) withstanding high pressures, the analysis time was reduced from 60 minutes to less than 20 minutes, without compromising chromatographic quality, with good resolution and reproducibility. The coupling with a TOF-MS analyzer provided information concerning high mass accuracy and isotopic pattern and allowed the identification of about 45 different phenols. Secondly, by first time the applicability of the nanoLC for the analysis of olive oil polyphenols was studied and compared with the RRLC method. We calculated the most important analytical parameters of both methods (linearity, calibration range, detection limit, repeatability and reproducibility, etc) to establish a comparison. Positive and negative polarities were used in both cases to check the behaviour of the phenols under study in the source, and quantitative analyses were made by using the two methodologies. The new nanoLC method provides comparable analysis time and offers better sensitivity with less consumption of mobile phases and samples; however it presents worse repeatability and it results a bit more difficult to operate.

NanoLC showed the potential to become a very promising alternative, in particular, for studies where the determination of extremely low concentrations of analytes is required (biological samples, for instance).