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Title: A VERSATILE TARGETED METABOLIC METHOD FOR THE RAPID QUANTIFICATION OF MULTIPLE CLASSES OF PHENOLICS AND THEIR METABOLITES: ITS APPLICATION IN FOOD METABOLOME AND NUTRITIONAL STUDIES

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Abstract: Compelling evidence of the health benefits of phenolic compounds and their impact on food quality have stimulated the development of analytical methods for the identification and quantification of these compounds in different matrices in recent years. The complexity and remarkable diversity of phenolics has challenged the analytical performances of separation and detection methods in terms of resolving power, selectivity and sensitivity for the identification and quantification of these compounds in different matrices. Targeted metabolomics is a strategy based on the use of predefined metabolite-specific signals, such as MRM transitions, that can be used to accurately determine the concentrations of a wide range of known metabolites. A targeted metabolomics method has been developed for the quantification of >150 phenolics, such as benzoates, phenylpropanoids, coumarins, stilbenes, dihydrochalcones, and flavonoids, and their metabolites/catabolites using UPLC/MS/MS system. Reversed-phase chromatography was optimised to achieve separation of the compounds over 15 min, reducing possible ion suppression effects and resolving many isomeric compounds and MRM transitions were selected for accurate quantification. The validated method was found to be particularly flexible, and was successively applied to the analysis of fruits and wine as well as in nutritional studies providing a valuable tool for food quality evaluation and nutritional relevant bioactive compounds profiling. The short duration of the analysis and the straightforward sample preparation make the methodology suitable for high-throughput screening studies, providing access to a large amount of quantitative compositional data within a single run and at an affordable cost.

The method was shown to be a valuable tool in the breeding studies of apples with the aim to identify uncommon chemotypes with higher concentration of bioactive compounds in the edible parts. It has also proved to be useful in screening fruits of different varieties and species (such as grapes, raspberries, apples) for characterizing the interesting subjects from the physiological or biosynthetic point of view. In nutritional studies the method was applied for an accurate quantification of some polyphenols catabolites associated with the consumption of fruits in animal organs and biofluids. Due to the high sensitivity of the method it was possible to find evidence and/or to provide physiologically relevant interaction with the mammalian metabolism and consumption of fruits.