

## Detection and quantification of polyphénols in apples tree leaves : UPLC-DAD utilisation

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### Introduction

The objective of this work consisted in developing an analytical method to determine the content of polyphenols of apple tree leaves. The results obtained during these analysis must be correlated with the total polyphenol content given by the Folin-Ciocalteux method on the same samples and be correlated with the potential of resistance of the variety of apple tree to scab.

### Material and methods

A phenolic extract was obtained from apple tree leaves coming from old varieties. The analysis were initially carried out in UPLC-DAD, before developing a UPLC-MSMS method.

### Results

Initially, the method was optimized by modifying the potential mobile phases: the acetonitrile was preferred to the methanol in the gradient of separation. For the six analyzed compounds, a difference in selectivity of 32.91 could be calculated for the two mobile phases (acetonitrile and methanol).

The impact of the temperature of the column was evaluated, in order to reduce the time of analysis of six polyphenols potentially present in the extracts (rutin, catechin, quercetin, quercetin glycoside, phlorizin and phloretin) to 1.1 minute.

The diode array detector was used to confirm the identity of the phlorizine and the glycosylated quercetin present in the extracts.

### Conclusions

A UPLC-DAD method was developed in order to give a quantitative determination of six polyphenols in apple tree leaves. The method suggested is based on a gradient "Acetonitrile - Water", the column being thermostated at 35°C. The time of analysis is 6 minutes, the capacity of PEAK under these conditions being of 57 for the "Acetonitrile" phase against 48 for the "Methanol" phase.

The diode array detector was used to confirm the identity of two compounds present in the extracts.

The later developments relate to the characterization in mass spectrometry of six polyphenols as well as the identification of other molecules, whose presence was revealed by the DAD.