

ABSTRACT

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Title of thesis: Development of HPLC method for evaluation of plant metabolites I

The topic of this thesis is the development and optimization of plant metabolites evaluation method for *Genista tinctoria* L. (Fabaceae) plant (genistein, daidzein, formononetin, biochanin A) using high performance liquid chromatography.

Separation was achieved by ZORBAX ECLIPSE XDV - Phenyl analytical 4,6x150 mm (5 μ) column with the usage of spectrophotometric detection with UV detector at 260 nm.

Mobile phase consisted of two components:

- Mobile phase A: water + phosphoric acid with 99,85 : 0,15 ratio
- Mobile phase B: methanol + phosphoric acid with 99,85 : 0,15 ratio

Chosen elution mode was gradient elution. The flow rate of mobile phase was set at 1,1 ml/min. The column temperature was 25 °C. The volume of injection was 20 μl. Total runtime was 18 minutes.

Principles described in guidelines were used for validation of this method. Validation parameters were selectivity, stability, linearity, accuracy, precision - repeatability. All measured values were within acceptable ranges.

Key words: *Genista tinctoria* L., genistein, daidzein, formononetin, biochanin A, genistin, HPLC