

## HPTLC ANALYSIS OF LIME FLOWERS (TILIAE FLOS)

Natalia Melnyk<sup>1</sup>, Sebastian Granica<sup>2</sup>, Agnieszka Bazylko<sup>2</sup>, Oleh Koshovyi<sup>1</sup><sup>1</sup>National University of Pharmacy, Kharkiv, Ukraine, Department of Pharmacognosy<sup>2</sup>Medical University of Warsaw, Warsaw, Poland, Department of Pharmacognosy and Molecular

Basis of Phytotherapy

melniknataly97@gmail.com

**Introduction.** Lime flowers have a prominent importance in a folk medicine. It is stated to possess expectorant, diuretic, diaphoretic, antispasmodic activities. *Tilia* is used in traditional medicine of many European countries primarily as a non-narcotic sedative for sleep disorders or anxiety. Essential oils, vitamins, mucilage and flavonoids components are known as the active ingredients. The officinal raw material is flowers (inflorescences) with flower bunds (*Tiliae flos*), which are introduced into many pharmacopoeis of the world.

**Aim.** The aim of the study was to investigate the differences in a polyphenolic composition of five chosen species (*T.cordata*, *T.platyphyllos*, *T.tomentosa*, *T.americana*, *T.europaea*) using HPTLC method.

**Materials and methods.** *Tilia flos* was used as a plant material for analysis, collected in Poland and Europe (2015-2017). The analysis was carried out at the Department of Pharmacognosy and Molecular Basis of Phytotherapy of Medical University of Warsaw. For analysis used: CAMAG Linomat 5, CAMAG ADC 2 Automatic Developing Chamber 2, CAMAG TLC plate heater III, CAMAG derivatizer, CAMAG TLC Visualizer 2; HPTLC plates, silica gel 60 F254, Merck. Standard samples of flavonoids which were used for comparison: quercetin derivatives (isoquercetin, rutin, avicularin), keampferol derivatives (keampferol 3-O-glucosid-7-rhamnozide, trans-tilirozyd, astragaline) and acacetin derivatives (linarin). Tetrahydrofuran – dichloromethane – formic acid – acetic acid – water P (9:9:4:2:3) was chosen as mobile phase.

**Results.** The results showed significant differences between HPTLC profiles of analyzed samples. Based on obtained results a key for the phytochemical identification was developed:

- 1a. Dark yellow (c)  $R_f$  0.64 after NA  
 Light green (linarin)  $R_f$  0.35-0.36 after NA  
 Yellow (j)  $R_f$  0.2 after NA  
 Green (p)  $R_f$  0.1 after NA  
 Yellow (q)  $R_f$  0.06 after NA  
 No dark yellow (b)  $R_f$  0.67 after NA  
 No light blue (a)  $R_f$  0.87 after NA  
 No dark yellow (f)  $R_f$  0.39-0.4 after NA  
 No blue (k)  $R_f$  0.19 before NA.....*T.cordata*
- 1b. No dark yellow (c)  $R_f$  0.64 after NA  
 No light green (linarin)  $R_f$  0.35-0.36 after NA  
 No yellow (j)  $R_f$  0.2 after NA  
 No green (p)  $R_f$  0.1 after NA  
 No yellow (q)  $R_f$  0.06 after NA  
 Dark yellow (b)  $R_f$  0.67 after NA  
 Dark yellow (f)  $R_f$  0.39-0.4 after NA...*T.americana*, *T.tomentosa*, *T.platyphyllos*, *T.europaea*
- 2a. Dark blue (d)  $R_f$  0.53 after NA.....*T.americana*
- 2b. No dark blue (d)  $R_f$  0.53 after NA.....*T.tomentosa*, *T.platyphyllos*, *T.europaea*
- 3a. Blue (k)  $R_f$  0.19 before NA  
 Light blue (n)  $R_f$  0.14 after NA  
 Light blue (o)  $R_f$  0.12 after NA.....*T.tomentosa*

- 3b. No blue (k)  $R_f$  0.19 before NA  
 No light blue (n)  $R_f$  0.14 after NA  
 No light blue (o)  $R_f$  0.12 after NA  
 Dark yellow (m)  $R_f$  0.16 after NA.....*T.platyphyllos*, *T. europaea*

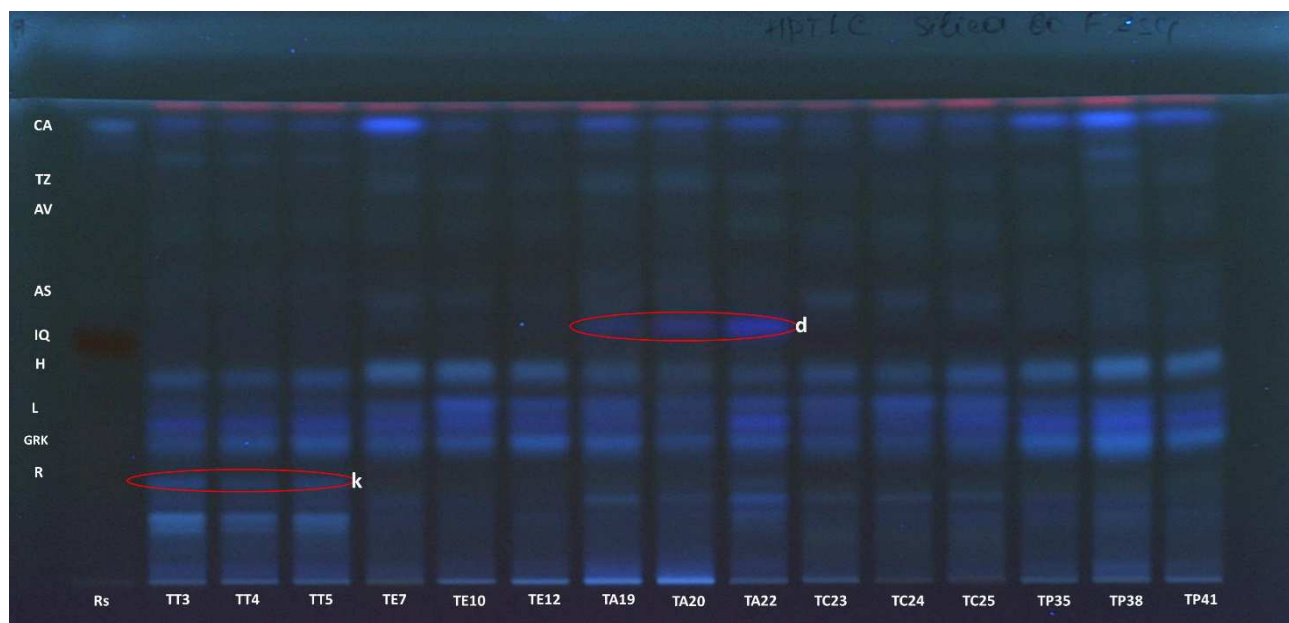


Fig. 1. HPTLC chromatogram at 366 nm of five *Tilia* species before derivatization with 1% NA

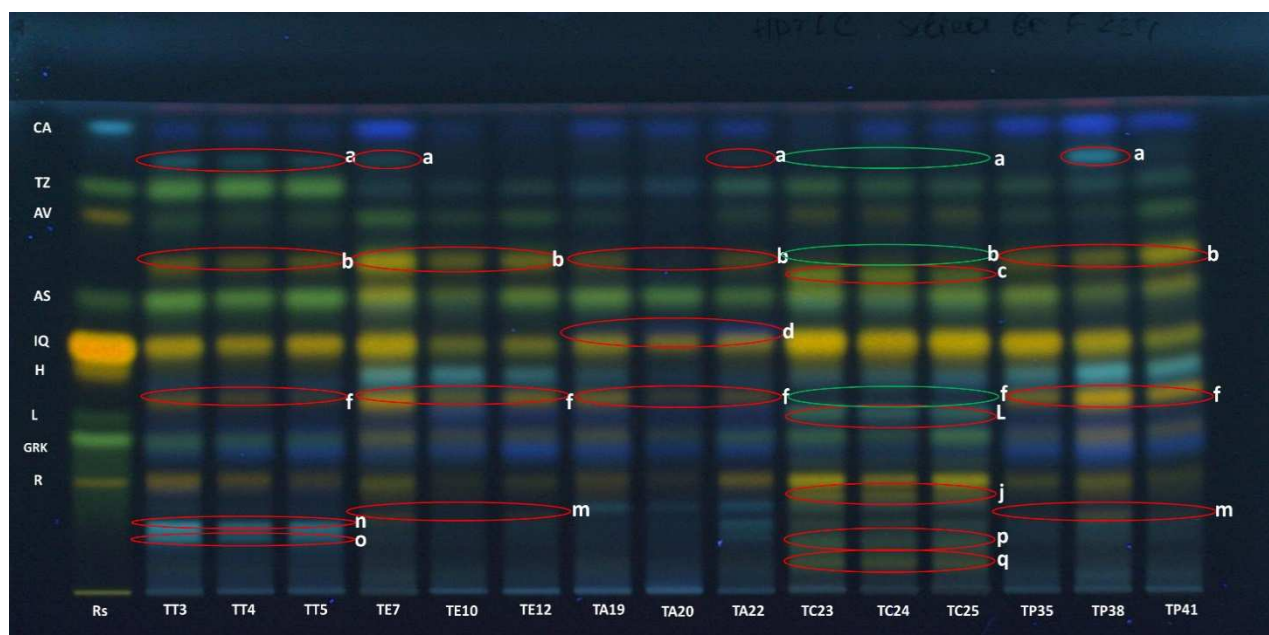


Fig. 2. HPTLC chromatogram at 366 nm of five *Tilia* species after derivatization with 1% NA

For the first time, the study was carried out to optimize the solvent system of TLC analysis of Lime flowers, which would allow the separation of camferol, quercetin and acacetin derivatives, while some of them have similar  $R_f$  values.

For the first time, a comparative analysis of different types of *Tilia* was conducted with the identification of similarities and differences in their chemical composition, on the basis of which the key for identification of *T. tomentosa*, *T. europaea*, *T. americana*, *T. cordata*, *T. rlatyphyllos* was developed.