



Quantification of Caffeine in different Coffee samples

Abstract

HPTLC (High Performance Thin Layer Chromatography) is a very useful analytical technique with very low sample preparation. Shown in the example: the quantification of caffeine in different coffee samples. The boiled coffee samples were extracted and filtered with a 0.45 μm syringe filter. The plate was pre-conditioned with the mobile phase. After chromatographic separation the plate was scanned with a TLC-Scanner and subsequently quantified.

Results

Caffeine was detected under UV light (254 nm, $hR_f = 56$, Fig. 1), (quantification information Fig. 2). A triple determination of each sample is possible in parallel.

Chromatographic data

Track	Compound	hR_f	Conc. [mg/mL]	Application volume [μL]	Amount of Caffeine in samples
1, 2, 3, 4, 9, 10, 11, 12 17, 18, 19, 20	Caffeine standard	56	1.0	0.1, 0.5, 1.0, 2.0	-
5, 13, 21	Filter Coffee 1	56	50	1	27.54 mg/g
6, 14, 22	Filter Coffee 2	56	50	1	15.25 mg/g
7, 15, 23	Pad Coffee	56	50	1	15.52 mg/g
8, 16, 24	Chai Latte	56	100	1	4.94 mg/g

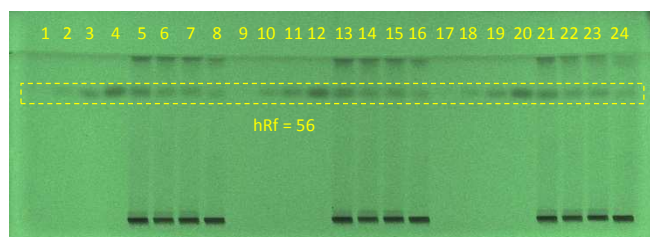


Fig. 1: Developed plate at 254 nm. Chromatographic data shown on next page.

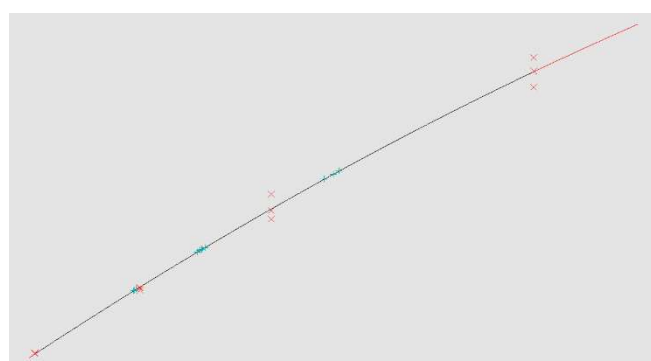


Fig. 2: graph from the polynomial regression mode ($r = 0.9970$).

Conclusion

These results demonstrate that caffeine can be easily quantitated by TLC with a three-fold determination and low sample preparation.

Application data

Chromatography	Plate	HPTLC Silica gel 60 F ₂₅₄ , 20x10cm
	Sample preparation	Caffeine standard diluted in Methanol; Coffee boiled with water and filtered with a 0.45 µm syringe filter.
	Sample application	ATS4 (Camag)
	Application volume	0.1 to 2.0 µL
	Pre-conditioning and mobile phase	Acetonitrile / Methanol / n-Heptane 6/4/1
	Pre-conditioning distance migration distance	7 cm 5 cm
	Migration time	9 min
Detection	Wavelength	UV light at 254 nm
	Staining	None

Ordering Information

HPTLC Si60 F ₂₅₄ , 20x10 cm	1.005642
Acetonitrile gradient grade for liquid chromatography LiChrosolv®	1.00030
n-Heptane for liquid chromatography LiChrosolv®	1.04390
Methanol gradient grade for liquid chromatography LiChrosolv®	1.06018
Millex®-FH filter, 0.45 µm hydrophobic PTFE, 25 mm, non-sterile	SLFH025NS