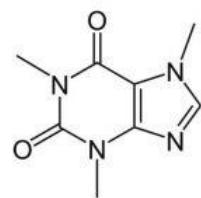
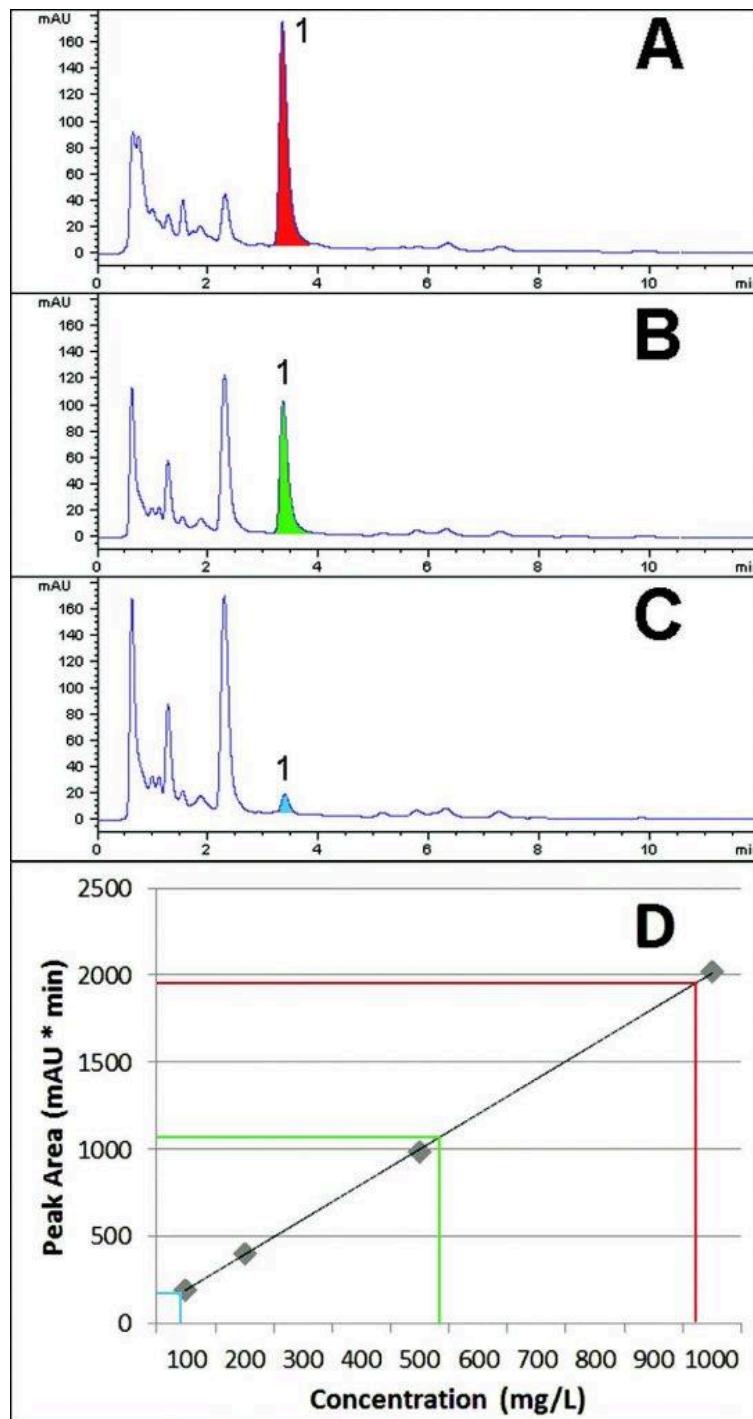


## Caffeine Content in Coffee Analyzed with HPLC - AppNote

### Espresso, Regular, and De-Caffeinated Coffee Caffeine Content

Caffeine is separated from other matrix components in three common Coffee beverages in this AppNote. A shot of Espresso (*Figure A*) had the highest content, followed by regular drip Coffee (*Figure B*) as expected. In De-Caffeinated Coffee, Caffeine is extracted from the coffee beans but even after numerous extractions, Caffeine is not completely removed. This is illustrated by the chromatogram for the De-Caffeinated Coffee sample (*Figure C*), where a Caffeine peak was detected.

A calibration curve was constructed in the range of 100 – 1000mg / L (*Figure D*), which provided estimates of caffeine content in the Beverages: Espresso: 964mg / L, Regular Coffee: 539mg / L, and Decaf Coffee: 85mg / L.



Caffeine

Peak:

Caffeine

## Method Conditions

**Column:** Cogent Bidentate C18 2.0<sup>TM</sup>, 2.2 $\mu$ m, 120 $\text{\AA}$

**Catalog No.:** [40218-05P-2](#)

**Dimensions:** 2.1 x 50mm

**Mobile Phase:** 90:10 A: DI Water with 0.1% Formic Acid (v/v), / B: Acetonitrile with 0.1% Formic Acid (v/v)

**Injection vol.:** 0.5 $\mu$ L

**Flow rate:** 0.3mL / minute

**Detection:** UV @ 254nm

**Sample Preparation:** Espresso, Regular, and Decaf Coffee were purchased from a local coffeehouse. The samples were filtered with 0.45 $\mu$ m Nylon Syringe Filters (MICROSOLV Tech Corp.). A 1000ppm Caffeine reference standard solution was prepared in a diluent of 50:50 Solvent A / Solvent B. Dilutions were made from this Stock Solution to obtain concentrations of 100, 200, and 500ppm.

**t<sub>0</sub>** : 0.8 minutes

---

**Note:** Caffeine is a xanthine alkaloid found in the coffee plant, the tea bush, the kola nut, and other plants. It is the most commonly consumed psychoactive drug in the world.

---



**Attachment No 318 Caffeine Content in Coffee Analyzed with HPL pdf 0.5 Mb** [Download File](#)

Printed from the Chrom Resource Center

Copyright 2025, All Rights Apply

**MicroSolv Technology Corporation**

9158 Industrial Blvd. NE, Leland, NC 28451

Tel: (732) 380-8900

Fax: (910) 769-9435

Email: [customers@mtc-usa.com](mailto:customers@mtc-usa.com)

Website: [www.mtc-usa.com](http://www.mtc-usa.com)