



Diphenhydramine HCL Analyzed with LCMS - AppNote

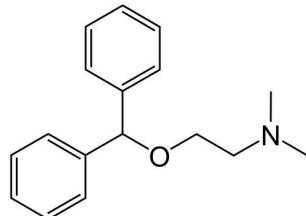
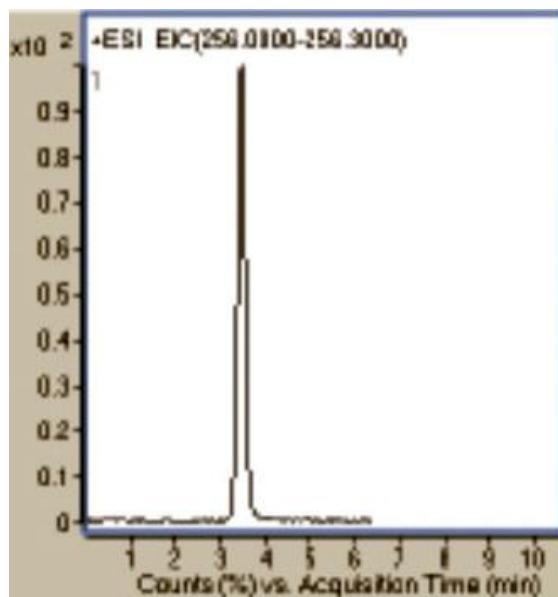
Online SPE with a Simple Mobile Phase Step Gradient

When a Mobile Phase containing 80% Solvent B (with 10mM Pyridine in Solvent A and B) is used Diphenhydramine Hydrochloride (DPH) is fully retained on the Column, meaning it does not elute and continues to be retained until it would overload the Column if injections were continued.

When the Mobile Phase was changed to 25% Solvent B, the same DPH Peak elutes at 3.5 minutes.

This wide range of Retention Time can give analysts many possibilities of developing Methods for Quantitative Analysis of Drug Formulations. With this Method, you can perform On-Column Solid Phase Extraction (*SPE*) Methods for Isolation of DPH from complex matrices. Simply use 80% Solvent B to load the DPH on the Column and 25% Solvent B to elute the compound.

The Method below is very Reproducible and Sensitive with a Lower Limit of Quantitation (LLOQ) of 5ng / mL for DPH, with good Linearity in the range 1-500ng / mL ($r^2 > 0.9990$).



Peak:

Diphenhydramine 256 m/z (M-H)⁻ RT = 3.49 min

Method Conditions

Column: Cogent Diamond Hydride™, 4µm, 100Å

Catalog No.: [70000-15P](#)

Dimensions: 4.6 x 150mm

Solvents:

A: DI Water / 10mM Pyridine

B: Acetonitrile / 10mM Pyridine

Mobile Phase: *Isocratic* 25:75 Solvent B / Solvent A

Flow rate: 0.4mL / minute

Detection: ESI – neg - Agilent 6210 MSD TOF Mass Spectrometer

Sample Preparation: 50ng / mL prepared in 50:50 Solvent A / Solvent B

Notes: Diphenhydramine (DPH) is an antihistaminic drug mainly used as a sedative, hypnotic and antiemetic most known by the trade name, Benadryl®. It is available over-the-counter in many countries and is very common. Generally regarded as a harmless drug, there have been sixty-eight non-fatal and fifty five fatal poisonings with DPH or in combination with other drugs investigated in the past.



Attachment No 79 Diphenhydramine HCL Analyzed with LCMS pdf 0.1 Mb [Download File](#)

Printed from the Chrom Resource Center

Tel: (732) 380-8900

Copyright 2025, All Rights Apply

Fax: (910) 769-9435

MicroSolv Technology Corporation

Email: customers@mtc-usa.com

9158 Industrial Blvd. NE, Leland, NC 28451

Website: www.mtc-usa.com