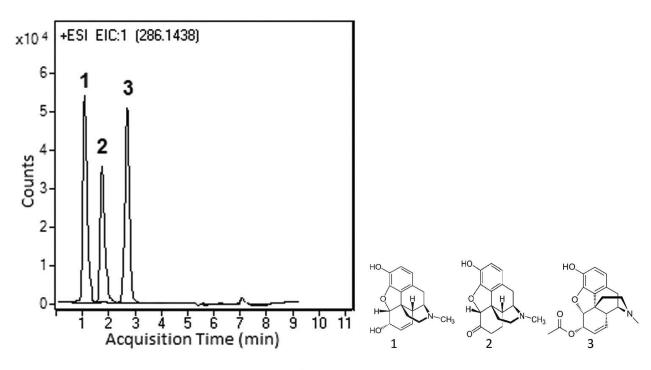


Morphine, Hydromorphone and 6-MAM in Plasma Analyzed with LCMS - AppNote

Morphine, Hydromorphone and 6-Monoacetylmorphine (6-MAM) in Plasma

This AppNote presents a Method that could be used for a routine analysis of Plasma Samples or (whole blood samples after changing the extraction procedure) for the presence of Morphine, Hydromorphone (isobaric compounds) or 6-MAM (an indicator of Heroin use).



Peaks:

- 1. Morphine 286.1438 m/z [M+H]+
- 2. Hydromorphone 286.1438 m/z [M+H]+
- 3. 6-Monoacetylmorphine (6-MAM) 328.1543 m/z [M+H]+

Method Conditions

Column: Cogent Bidentate C18 2.0™, 2.2µm, 120Å

Catalog No.: <u>40218-05P-2</u> **Dimensions**: 2.1 x 50mm

Mobile Phase:

A: DI Water / 0.1% Formic Acid (v/v)

B: 50:50 Acetonitrile / Methanol / 0.1% Formic Acid (v/v)

Gradient:

Time (minutes)	%B
0	5
4	50
5	90

6 90 7 5

Post Time: 3 minutes **Injection vol.**: 1µL

Flow rate: 0.4mL / minute

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

Sample Preparation: To a spiked Plasma Sample (1mL), 1mL of an Ammonia solution (0.1% v/v) was added and vortex-mixed for 30 seconds to alkalize the Plasma. Then, the alkalized Plasma Sample was extracted with two 4mL Ethyl Acetate aliquots by vortex for 5 minutes, and centrifuged at 4000 rpm for 8 minutes at room temperature. The supernatant was separated and evaporated to dryness under a gentle stream of Nitrogen. The residue was reconstituted with 200µL Mobile Phase, and a 1µL aliquot of the reconstituted solution was injected into the LC– ESI-MS for analysis.

Note: Morphine is used as an analgesic drug in the treatment of the severe pain. Hydromorphone, a semi-synthetic derivative of Morphine is used in cases of Morphine-resistant Cancer-related pain and 6-Monoacetylmorphine (6-MAM) is a specific indicator for Heroin use. All three drugs remain the selected substances in monitoring of drug abuse and addiction. Heroin use remains a serious public health issue which accounts for a substantial proportion of the health costs. Analysis of Plasma Samples for the presence of the three substances is essential in assessing short or long term exposure to the drugs.



Attachment

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