

Application Data Sheet

No.46

LC-MS

Liquid Chromatograph Mass Spectrometer

Analysis of Drugs in Putrefied Human Pleural Fluid using Triple Quadrupole LC/MS/MS

This application illustrates a drug screening method in putrefied human pleural fluid using the Shimadzu UFMS triple quadrupole mass spectrometer, LCMS-8050.

In forensic and toxicology fields, it is important to develop a highly sensitive and exhaustive methodology for screening and identifying drug substances. A wide range of these compounds in various biological matrices, such as whole blood, urine and tissue, need to undergo a simple and uniform sample pretreatment protocol prior to Liquid Chromatography Triple Quadrupole Mass Spectrometry (LC-MS/MS) analysis. This data sheet presents a drug screening method using the LCMS-8050 with newly developed sample preparation protocol in putrefied human pleural fluid.

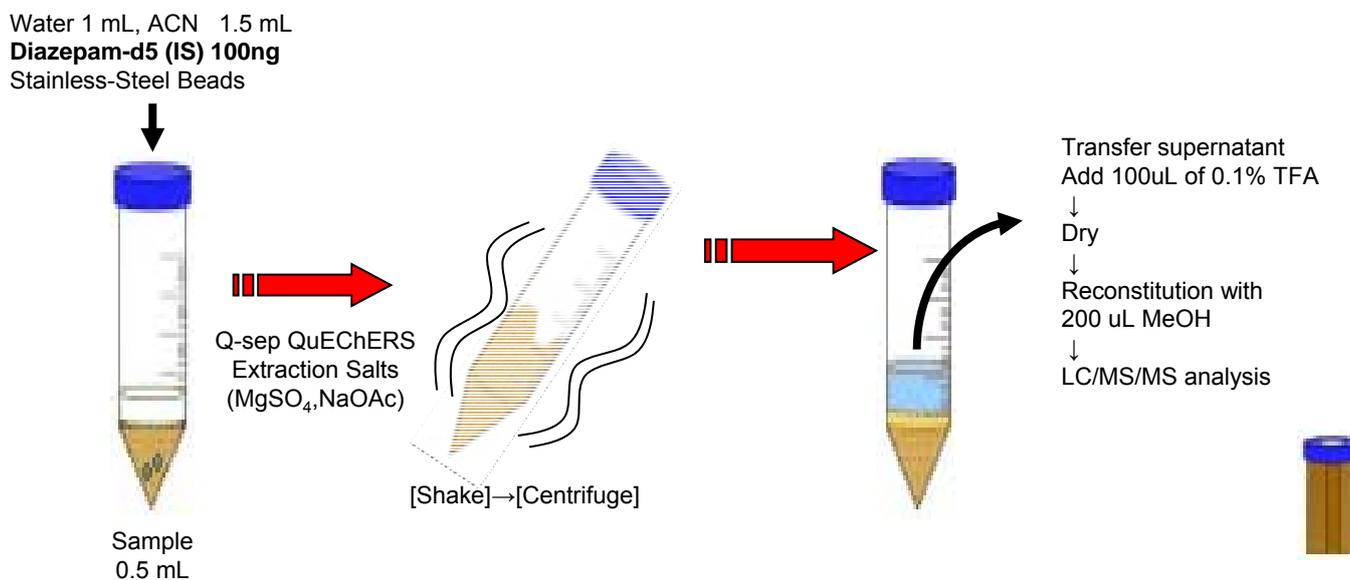


Figure 1: Scheme of the modified QuEChERS procedure

Table 1: Analytical Conditions

Liquid Chromatography

- Column : Shim-Pack FC ODS (150x2mm, 3µm)
- Temperature : 40 °C
- Injected volume : 5 µL
- Mobile phases : A: Water + 10 mM Ammonium Formate
B: Methanol
- Flow rate : 0.3 mL/min
- Gradient : 5%B (0 min) – 95%B (15 min – 20min) – 5%B (20.1 min – 30 min)

Mass Spectrometry

- Configuration : LCMS-8050
- Ionization mode : Heated ESI positive and negative
- Nebulizing gas flow : 2 L/min
- Drying gas flow : 10 L/min
- Heating gas flow : 10 L/min
- DL temperature : 250 °C
- HB temperature : 400 °C
- Analysis mode : MRM

Traditional sample preparation strategies for biological fluids, such as protein precipitation and solid phase extraction, require multiple time-consuming steps. In addition, commercially available sample preparation techniques lack the ability to extract all compounds of interest. A modified QuEChERS protocol for drug screening in biological fluids described here illustrates stable recoveries for drug substances regardless of sample or chemical properties.

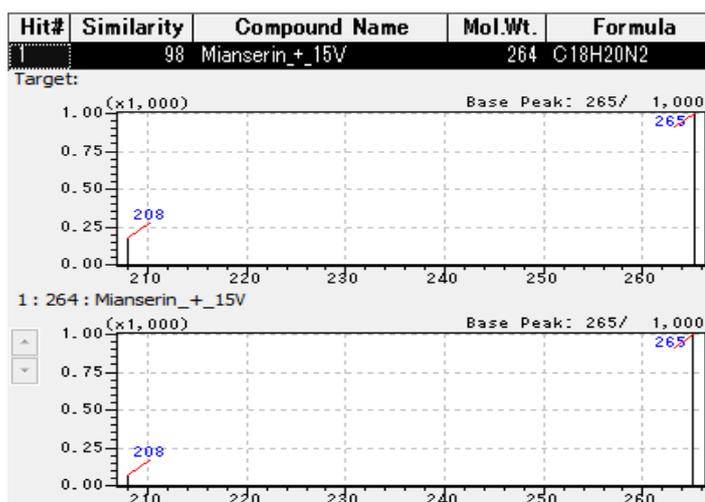
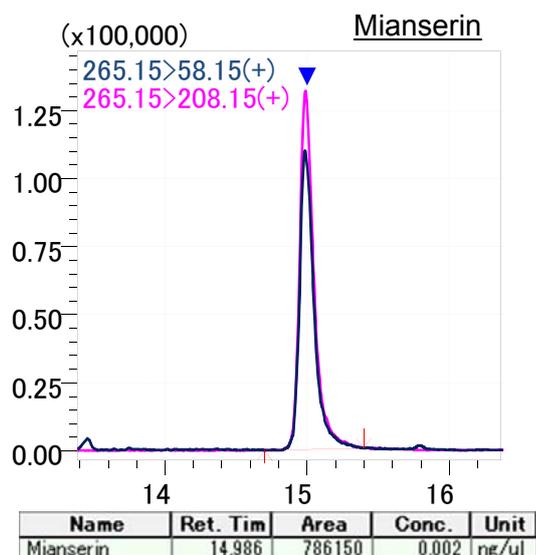
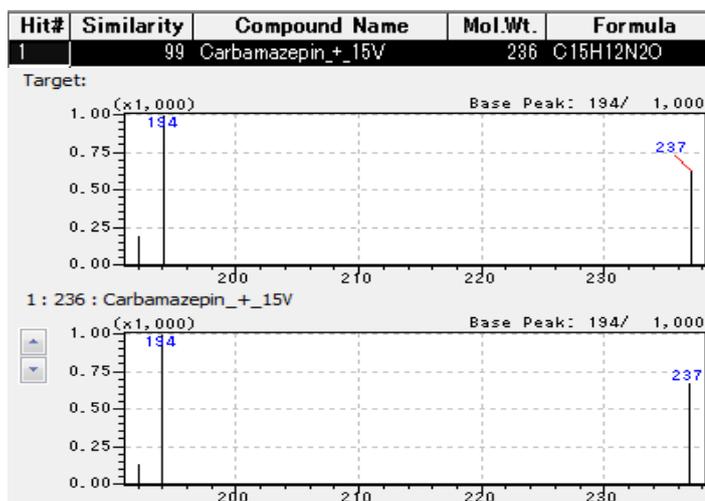
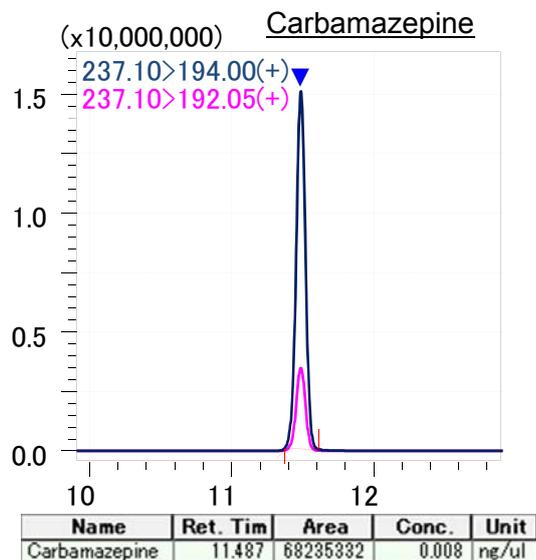


Figure 3: MRM Chromatograms, semi-quantitative value and Library Search results of Carbamazepine and Mianserin in putrefied human pleural fluid

Shimadzu's LCMS-8050 UFMS can perform Synchronized Survey Scan® (SSS), which automatically conducts a product ion scan triggered by preset MRM intensity thresholds. SSS provides both quantitative (MRM chromatograms) and qualitative data (Product ion spectrum) in a single run.

Furthermore, utilizing diazepam-d5 as an internal standard, semi-quantitative results can be determined using the method's built-in calibration curves (slope and intersection).

This software functionality is a very effective way for one to understand the quantitative values obtained during a simultaneous analysis.