K15 A Simple Liquid - Liquid Extraction of Carisoprodol and the Metabolite Meprobamate From Suspected Blood and Urine DUI Specimens for GC/MS Analysis

Jamie L. Jouett, BS, Sam Houston State University, 13 FM 1696 East Huntsville, TX 77320*

After attending this presentation, attendees will understand a modified and improved method for analyzing carisoprodol and the metabolite meprobamate in blood and urine specimens. The goal of this presentation is to demonstrate a quick, clean, and effective liquid-liquid extraction method to detect the presence of carisoprodol (Soma®) and the metabolite meprobamate in blood/urine specimens at levels above, below, or at therapeutic concentrations, in turn, providing supportive analytical data for the assessment of suspected DUI cases. Literature data indicates severe driving impairment and intoxication when the combination of the two drugs exceeded 10 mg/L, a level that is still within normal therapeutic range.[1] Ultimately, this extraction method will prove a number of advantages compared to previously reviewed extraction methods.[2]

This presentation will impact the forensic community by demonstrating a clean and effective gas chromatography/mass spectroscopy (GC/MS) based and validated method for detection of carisoprodol (Soma®) and the metabolite meprobamate in suspected driving under the influence (DUI) specimens.

Carisoprodol is a commonly prescribed muscle relaxant that has not been classified as a controlled substance. The Brazoria County Crime Laboratory has observed that in suspected impaired drivers, the frequency of blood/urine specimens testing positive for carisoprodol and the metabolite meprobamate has increased over the past few years. This data reflects the obvious need for a simple, validated extraction method to confirm carisoprodol and the meprobamate in suspected impaired drivers.

To demonstrate a quick, clean, and effective liquid-liquid extraction method to detect the presence of carisoprodol and the metabolite meprobamate in blood/urine specimens at levels above, below, or at therapeutic concentrations, in turn, providing supportive analytical data for the assessment of suspected DUI cases. Literature data indicates severe driving impairment and intoxication when the combination of the two drugs exceeded 10 mg/L, a level that is still within normal therapeutic range.[1]. Ultimately, this extraction method will prove a number of advantages compared to previously reviewed extraction methods.[2]

In this method, samples were prepared by adding buffer, barbital (internal standard) and chloroform to 250 uL of specimen. Barbital is the recommended internal standard due to the fact that it does not co-elute with targeted drugs of interest and is compatible with systems other than GC/MS, such as flame ionization detection (FID). The extraction efficiency and linearity of carisoprodol and meprobamate were analyzed at levels consistent with DUI blood/urine by comparing different buffer systems and adjusting pH levels. Buffer systems and pH adjustments evaluated were 0.1 M acetate buffer pH 4.5 and 0.1 M acetate buffer pH 4.5 saturated with NaCl, 1.0 M acetate buffer pH 4.5, and 0.1 N HCl. A five point calibration curve including 4mg/L, 10mg/L, 30mg/L, 40mg/L, and 60mg/L was utilized to determine linearity. After mixing, the chloroform was pipetted into a clean test tube and evaporated to dryness under nitrogen. The residue was reconstituted with 120uL of ethyl acetate and analyzed using an Agilent Technologies 6890 GC coupled to a 5975 MSD in electron sensitive-selective ion monitoring (EI-SIM) mode for quantitative analysis. GC injection conditions were evaluated under splitless, split, and pulsed-split modes.

The evaluation of this extraction method was based on precision, cleanliness, and chromatographic data. The 0.1 M HCL acidification results in a dirtier extract and tends to build residue in the injector port faster than the acetate buffering systems. Moreover, both 1.0 M acetate buffer and 0.1 M acetate buffer pH 4.5 saturated with NaCl demonstrate a more compacted protein layer between the aqueous and organic layers, resulting in a cleaner extraction. A cleaner extract reduces residue build up and drug decomposition in the GC injector port; thereby, minimizing routine instrument maintenance. However, the 1.0 M acetate buffer pH 4.5 assures the pH stability of blood and urine during extraction and is therefore the preferred buffering system. Chromatographic data were evaluated by comparing split, splitless, and pulsed-split modes. Split and pulsed-split modes offer improved peak symmetry and less column overload. In addition, calibration curves were linear from 4-60 mg/L with R^2 values of 0.995 for carisoprodol and 0.999 for meprobamate. The extraction efficiencies were 48% (barbital), 69% (carisoprodol), and 71% (meprobamate). Thus, 1.0 M acetate buffer pH 4.5 is the optimal buffering system to provide clean extracts with consistent recoveries.

This extraction procedure provides a rapid, clean, and effective method suitable for detecting carisoprodol and meprobamate with the intended purpose of providing analytical data to determine drug concentrations in suspected DUI cases.

References:

1. Journal of Forensic Science 2000; 45(3):619-23
2. Journal of Analytical Toxicology 2006; 30(5):302-5

Carisoprodol, Meprobamate, GC/MS

 ** Presenting Author*