B64 Enantiomeric Identification of Pregabalin Via Methylation and Chiral Derivatization, Utilizing Gas Chromatography/Mass Spectrometry (GC/MS)

Mike Hitchcock, MS, U.S. Postal Inspection Service, Forensic Laboratory Services, 22433 Randolph Drive, Dulles, VA 20104-1000; Ioan Marginean, PhD, George Washington University, 2100 Foxhall Road, NW, Somers Hall L14C, Washington, DC 20007; and Peter Nemes, PhD, George Washington University-Chemistry Dept, 725 21st Street, NW, Corcoran Hall, Rm 107, Washington, DC 20052*

After attending the presentation, attendees will understand the need for a viable method to identify pregabalin, or (S)-3-(aminomethyl)-5-methylhexanoic acid, utilizing traditional capillary GC/MS. Attendees will also be exposed to the methylation and chiral derivatization procedure used to prepare pregabalin for enantiomeric identification by GC/MS. Identification of the correct enantiomer of pregabalin by GC/MS will be demonstrated.

The presentation will impact the forensic science community by demonstrating an alternative method of identifying pregabalin through enantiomeric determination utilizing GC/MS, an instrument commonly found in forensic laboratories.

Many available methods for the identification of pregabalin involve Liquid Chromatographic separation with electrospray ionization Mass Spectrometry (LC/MS). LC/MS is not as common in forensic laboratories as the traditional GC/MS. Alternatively, Fourier Transform Infrared (FTIR) spectrophotometry is an effective way to elucidate the structure of pregabalin; however, FTIR cannot distinguish between optical isomers. Therefore, it would be beneficial to provide a method of enantio-selective identification of pregabalin using GC/MS.

Pregabalin, or (S)-3-(aminomethyl)-5-methylhexanoic acid, marketed under the brand name Lyrica®, became a Schedule V controlled substance in the United States in July of 2005 due to its depressant activity. It is used legitimately to treat neuropathy in diabetic patients as well as for fibromyalgia. Pregabalin is an amino acid and an amphoteric compound, which makes it difficult to analyze using traditional GC/MS instrumentation. Problems associated with GC/MS analysis of pregabalin include: (1) ring closure to the corresponding lactam in the injection port or in the transfer line; as well as, (2) difficulty with chiral derivatization due to lack of reactivity of the derivatization agent in the presence of a carboxylic acid moiety. The ring closure and chiral derivatization problems are overcome by methylating (capping) the carboxylic acid portion of the pregabalin molecule and converting it to its corresponding methyl ester.

The simple procedure used to cap the carboxylic acid portion of pregabalin involves small amounts of thionyl chloride and dry methanol using cold temperatures achieved with dry ice/acetone in a typical laboratory fume hood. Once methylation occurs, the carboxylic acid is deactivated and the attachment of the chiral derivatization reagent (S) Trifluoroacetylprolyl chloride, or (S)-TPC, is straightforward. Once the S-TPC is attached, the correct (S) enantiomer of pregabalin can be identified via GC/MS.

Data will be presented showing the corresponding methyl ester of pregabalin as well as the separation of the (S) and (R) enantiomers via GC/MS. This method will allow for the proper identification of pregabalin utilizing GC/MS, the most common instrumentation found in a forensic drug laboratory.

Pregabalin, Chiral Separation, GC/MS