Development of an Analytical Method for Nootropic "Smart" Drugs in Biological Fluids

After attending this presentation, attendees will be aware of the class of nootropic ("smart") drugs, be able to describe the relative merits of different models of analysis, and implement a method to identify, confirm, and quantitate the drugs in biological fluids.

This presentation impacts the forensic science community by describing the development of an analytical method for newly emerging drugs that are subject to misuse and abuse, and can cause mind altering effects. The drugs are not currently regulated and are emerging online and in illicit supply chains.

Smart drugs, also known as nootropics, are an emerging area of growth in recreational drug use with implications for forensic toxicology. The drugs have stimulant properties and are alleged to boost brain function and cognition. The media attention on these drugs has increased within the last few years. The drugs have developed an underground following and are commonly sold online and in illicit supply chains. Most have not been approved or scheduled in the US, and are therefore of concern to regulators such as the Food and Drug Administration (FDA) and Drug Enforcement Administration (DEA). There are ongoing investigations in the applications of smart drugs in the treatment of Alzheimer's disease, Huntington's disease, and attention deficit hyperactivity disorder (ADHD). The stimulant properties of the drugs have led to their use in academic doping and as drugs of abuse. Some drugs are also prohibited by the World Anti-Doping Agency (WADA).

The goal of this project was to develop a single analytical method for screening, confirmation, and quantification of a series of the more widely known smart drugs in blood and urine. Gas chromatography/mass spectrometry (GC/MS) and liquid chromatography/mass spectrometry (LC/MS) were investigated to determine the optimum approach for sensitivity and ability to detect a broad range of compounds, specifically are piracetam, pramiracetam, aniracetam, modafinil, adrafinil, ciproxifan, and noopept.

Analytical methods included the use of GC/MS. Analysis was performed using an Agilent 6890/5973 GC/MS system with an Agilent DB-5MS column (15 m x 0.25 mm; 0.25 µm). Temperature programs starting at 70 °C and injection port temperatures as low as 175 °C were evaluated. The racetam class of drugs which contain a pyrrolidineacetamide structure (e.g. piracetam, pramiracetam, and aniracetam) were thermally unstable and degraded into 3-5 derivative structures. The structures of the degradation products were tentatively identified based on molecular weight, and their mass spectra were added to a database to assist with identification of breakdown artifacts in future toxicological analysis. Adrafinil, a modafinil prodrug, was found to degrade to modafinil during GC/MS analysis by water loss, resulting in an inability to distinguish between the two substances. Even at low injection port temperatures, degradation persisted. Derivatization of the compounds using BSTFA with 1% TMCS to form TMS derivatives as a means of stabilizing the compounds was also investigated, but also failed to produce single stable chromatographable analytes.

As a result of the demonstrated thermal instability of these drugs, LC/MS was investigated. A successful analytical method was developed using an Agilent 1100 series system with an Eclipse Plus C18 column (4.6 mm x100 mm; 3.5 µm), a gradient consisting of 10 mM ammonium acetate buffer (pH 4) and 50:50 acetonitrile:isopropanol (v/v), a flow rate of 0.6 mL/min, and positive (aniracetam, piracetam, pramiracetam, noopept, ciproxifan) and negative (modafinil and adrafinil) ionization. The ions that were monitored are as follows: aniracetam 220.1 m/z; piracetam 143.1 m/z; pramiracetam 270.1 m/z; noopept 319.1 m/z; ciproxifan; 271.1 m/z; modafinil 272.1 m/z; adrafinil 288.2 m/z. All of the peaks were baseline resolved.

Following characterization of the above conditions, extracts of blood and urine samples were prepared using supported liquid extraction procedures and evaluated for recovery cleanliness, reproducibility, limits of detection and quantitation, accuracy, and precision. In conclusion, LC/MS provided a superior means of identification for the target compounds due to the stability of the compounds under these analytical conditions. The methods evaluated were effective for the detection of the targeted compounds in biological fluids, such as whole blood and urine, at toxicologically meaningful concentrations.

Keywords: smart drugs, nootropics, LC/MS