

## K39 Use of Serotonin Metabolites in Postmortem Alcohol Determinations

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After attending this presentation, attendees will more fully understand postmortem ethanol formation and ways to identify it.

Specimens from aviation accident victims are submitted to the

FAA's Civil Aerospace Medical Institute (CAMI) for toxicological analysis. During toxicological evaluations, ethanol analysis is performed on each such case. Care must be taken when interpreting a positive ethanol result due to the potential for postmortem ethanol formation. Historically, ethanol distribution in various tissues and fluids from the same case and/or the presence of other volatile organic compounds at abnormal concentrations in these fluids and tissues has been employed as an indicator of postmortem microbial ethanol formation. However, these methods are not always reliable. The consumption of ethanol has been shown to alter the concentration of two major serotonin metabolites, 5-hydroxytryptophol (5-HTOL) and 5-hydroxytindole-3-acetic acid (5-HIAA). While the 5-HTOL/5-HIAA ratio is normally low, previous studies have demonstrated that the urinary 5-HTOL/5-HIAA ratio is significantly elevated following ethanol ingestion. The 5-HTOL/5-HIAA ratio is not affected by the microbial formation of ethanol, by consumption of serotonin-rich foods or by the use of SSRI's.

A single analytical approach has been developed to determine concentrations of both 5-HTOL and 5-HIAA that has provided a convenient, rapid and reliable solution to this problem. This novel methodology eliminates the need for two separate and unrelated analytical techniques, GC/MS and LC/EC, for the determination of these metabolites. The simultaneous determination of 5-HTOL and 5-HIAA in forensic urine specimens was achieved using a liquid/liquid extraction technique in conjunction with LC/MS. The ion trap MS used allowed us to perform MS/MS/MS on both 5-HTOL and 5-HIAA, and afforded limits of quantitation below 1 ng/mL for each compound. After development of this method, the previously established, antemortem, 15 pmol/nmol 5-HTOL/5-HIAA ratio cutoff was investigated and subsequently validated for use with forensic specimens.

The FAA laboratory utilizes this method to examine all postmortem ethanol-positive urines, where the source of ethanol is unclear. This presentation will discuss the difficulties in determining the source of ethanol in postmortem cases, markers of ethanol ingestion, and the application of this novel methodology in elucidating ethanol origin. Multiple case studies that involved postmortem alcohol formation will be presented.

## Postmortem Ethanol, LC/MS, Serotonin Metabolites