K9 Interpretation of Fentanyl in Postmortem Cases

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After attending this presentation, attendees will have an increased knowledge of the application of High Performance Liquid Chromatogra- phy/Tandem Mass Spectrometry (LC/MS/MS) to the analysis of Fentanyl, in postmortem specimens and of the concentrations of fentanyl encountered during analysis of these specimens.

Fentanyl is a synthetic opiate analgesic. It is frequently administered as a transdermal patch and is a substance of abuse. This presentation will impact the forensic community by increasing awareness of the very high potency of fentanyl and the frequency of its use both alone and in combination with other intoxicants. The presentation will also demonstrate the need for very sensitive analytical methodologies for its detection and quantitation.

Methods: Twenty-three medical examiner cases containing fentanyl were identified by a positive ELISA, by a gas chromatographic/mass spectrometric (GC/MS) screen, or by the presence of a transdermal patch on the body. Fentanyl was quantified in blood by LC/MS/MS using multiple reaction monitoring techniques and fentanyl-D5 as internal standard. Molecular ions (m/z 337.3 and 342.3) were refragmented to yield ions of masses 188, 132 and 105 (Fentanyl) and 188, 137 and 105 (Fentanyl-D5). Specimens, standards and controls (0.5 mL) were basified by addition of 0.05 mL concentrated ammonium hydroxide solution and were extracted with 1.25 mL hexane:ethanol (95:5). The organic solvent was decanted, forced through a 0.2 micron acrodisc syringe filter and evaporated. The residues were reconstituted into 0.25 mL mobile phase and twenty microliters (20µL) were injected. Chromatography was performed on a Varian Pursuit column (C-18, 3 micron, id = 2 mm, l = 50 mm) and an aqueous formic acid : acetonitrile gradient solvent system. The calibration range of the assay was 0.5 to 20 µg/L. Positive controls were run at 2.5 and 15 µg/L. The limit of quantitation (generally 0.5 µg/L) was defined as the lowest standard or control that assayed within twenty percent of target, with acceptable ion qualifier ratios. Samples containing greater than 20 µg/L of fentanyl were diluted prior to analysis.

Results: Fentanyl was the major intoxicant in 13 cases and was present in a further 10 cases in combination with at least one other significant intoxicant, such as methadone, an opiate, cocaine or ethanol. The fentanyl content of the 13 fentanyl-"primary" cases ranged between 3 and 49 µg/L (14 + 12 µg/L). In the 10 cases that contained other significant intoxicants, fentanyl concentrations were between 0.5 and 18 µg/L (5 + 5 µg/L) and seven of these cases contained less than 5 µg/L (0.5 to 4.6 µg/L) of fentanyl.

Conclusion: These data indicate that fentanyl is frequently encountered in combinations with other drug substances and that, in these combinations, even very small amounts of fentanyl may contribute to lethality. Sensitive methods of analysis, such as LC/MS/MS are required for quantitation of fentanyl in these cases. The data also indicate that interpretation of fentanyl levels in postmortem cases must be done on a case-by-case basis and must consider fully the combined effects of all intoxicants present.

Fentanyl, Postmortem, LC/MS/MS

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