G115 Postmortem Evidence of Methadone Cytotoxicity

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After attending this presentation, attendees will have a greater understanding of the fundamental process underlying death secondary to methadone, and possibly other opiates as well. Knowledge of these changes may be generalizable to other forms of drug-related death as well.

This presentation will impact the forensic science community by alerting them to the possibility that there may be alternate cause of death in cases of apparent overdose.

Background: Methadone concentrations in those dying of methadone toxicity totally overlap in situations where the presence of methadone is only an incidental finding. In practice, causation may be difficult to determine. The identification of an anatomic biomarker for methadone toxicity would be a useful tool. Evidence suggests that the abuse of many different drugs can lead to the occurrence of inappropriate apoptosis. Since the neurons controlling respiration are mainly located in the rostral venteral portion of the medulla oblongata, it was hypothesized that a detailed investigation of this area might disclose an anatomic marker for methadone-related death.

Objective: This study attempted to determine if methadone induced more apoptosis in the brainstems of methadone users than in the brainstems of drug-free individuals also suffering from Sudden Cardiac Death (SCD).

Design, Setting, and Participants: A single cohort of decedents with SCD who had been autopsied at a large university hospital in Palermo, Italy, were studied. In every case, a complete autopsy with toxicology testing was performed. The brains were only examined after two weeks of formalin fixation — this allowed for easier dissection of the brainstem. Multiple blocks of tissue were prepared from the area lying immediately between the inferior and the superior colliculi. These blocks included the area of the Rostral Ventrolateral Medulla (RVLM), known to contain the nucleus solitarius, which is thought to contain most of the brain's respiratory center. There were 11 participants and five age-matched controls. The slides were read separately by two individuals, both trained in neuropathology. No attempt was made at grading the anatomic changes, the only criteria being that the apoptotic process be recognizable. This determination was based entirely on the appearance of neurons in Hematoxylin-Eosin (H&E) stained sections.

Main Outcomes and Measures: The primary outcome measure was detection of the presence or absence of neuronal apoptosis and/or necrosis within the nucleus solitarius.

Results: Cells displaying evidence of both early and advanced apoptosis, consisting primarily of nuclear condensation, nuclear fragmentation, and even nuclear absence were found. These dying neurons were admixed with other neurons displaying the features of classic ischemic necrosis with eosinophilia and nuclear fragmentation. Evidence of classic necrosis was identifiable in most of the controls, though apoptosis was not.

Study Limitations: This study did not have access to the routine immuno-staining procedures used for the identification apoptosis, but it is believed the visual identification was accurate. Since the changes of both ischemic necrosis and apoptosis develop at different rates in different parts of the brain, it is difficult to be sure whether similar changes were or were not present elsewhere.

Conclusions and Relevance: This study shows that neurons, primarily along the tractus solitarius but occasionally in other cell nuclei (even controls), are vulnerable, presumably to direct methadone directly (via apoptosis) and indirectly methadone (via hypoxia). In instances where methadone is present in significant concentrations, but apoptotic lesions are absent, it may be reasonable to assume that methadone was not the cause of respiratory arrest, though this conclusion will require considerably more confirmation.

Methadone, Apoptosis, Biomarker

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