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Two validated methods to measure methadone concentrations in dog plasma and umbilical cord by LC-MS/MS

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Statement of the Problem: Methadone is an opioid μ-receptor agonist commonly used in human and veterinary medicine via systemic or epidural route to achieve intra- and post- operative pain relief. The advantages of the epidural administration of methadone are a lower evidence of side effects, a more profound and prolonged analgesia, and a lower dose required. To date, no information is available about the placental transfer of methadone in dogs and the respective maternal/fetal plasma concentration ratios. The aim of the present study was to develop and validate two methods for the accurate and precise determination of methadone concentrations in bitches plasma and in the umbilical cords of their puppies, after epidural or systemic administration during surgical caesarian section.

Methodology & Theoretical Orientation: Two different techniques were developed for the determination of methadone in the two matrices, using in both methadone-D3 as internal standard. Plasma samples were extracted with acetonitrile and, after centrifugation, the supernatant was evaporated to dryness and reconstituted with mobile phase. Umbilical cords were homogenized, added of formic acid and, after centrifugation, the supernatant underwent a cleanup step on SPE cartridge. The eluted sampled was then evaporated to dryness and reconstituted with mobile phase. All samples were analyzed by liquid chromatography coupled to tandem mass spectrometry.

Findings: The methods have been successfully validated in accordance with current European guidelines, providing satisfying performances over the range 1-250 ng/mL for plasma and 1-250 ng/g for umbilical cord.

Conclusions & Significance: This project raised from the need of determining methadone plasmatic concentration in bitches undergoing surgical caesarian section and in the umbilical cords of their puppies, in order to assess if newborns are less exposed to the drug following epidural administration. The proposed techniques proved to be suitable for the purpose and have been successfully applied to real samples.

Biography

Andrea Barbarossa has his expertise in many aspects of veterinary pharmacology, including antimicrobial resistance, residues and pharmacokinetics/pharmacodynamics studies. In particular, he has been involved in many projects on analgesic and anesthetic drugs in dogs and cats, including ketamine, buprenorphine, medetomidine and methadone. He has years of expertise on analytical chemistry, especially with liquid chromatography-mass spectrometry techniques, which are often the gold standard for this kind of studies.

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