

DEVELOPMENT OF A SIMPLE MULTI-RESIDUE DETERMINATION METHOD OF 80 VETERINARY DRUGS IN *OPLEGNATHUS PUNCTATUS* BY LIQUID CHROMATOGRAPHY COUPLED TO QUADRUPOLE-ORBITRAP MASS SPECTROMETRY

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The residues of veterinary drug in fish tissues are harmful to the public health. High resolution mass spectrometry (HRMS) is a very promising tool in the area of multi-residue analytical methods and has gained wider recognition. In this article, a simple, rapid and sensitive multi-residue analytical method was developed and validated for 80 veterinary drugs in *Oplegnathus punctatus* using ultrahigh performance liquid chromatography-Orbitrap high resolution mass spectrometry (LC-HRMS). The analytes belong to 12 different families include benzimidazoles, β -lactams, lincosamides, macrolides, nitromidazoles, quinolones, sulfonamides and trimethoprim, tetracyclines, triphenylmethane dyes, amphenicols, nonsteroidal estrogens and steroid hormones. Particular attention was devoted to the optimization of sample extraction and UHPLC-HRMS conditions. A very simple and sufficient preparation procedure without salting-out and complex cleanup process was studied. Finally, a mixture of acetonitrile, methanol and water (3:1:1, v/v/v) which includes 1% acetic acid and 10 mM ethylenediaminetetraacetic acid disodium salt 2-hydrate was selected as the extraction solvent,

and the cleanup step consisted of a low temperature procedure and two times of high-speed centrifugation to deproteinize and remove lipids. The detection and quantification of all compounds were performed by ultrahigh performance liquid chromatography coupled with electrospray ionization quadrupole-Orbitrap high resolution mass spectrometry in positive and negative ion mode. The developed method was validated with selectivity, linearity, recovery, precision, sensitivity, matrix effect and tested the CC α and CC β for each target analyte according to Commission Decision 2002/657/EC and SANTE/11945/2015. The satisfactory performances achieved, mainly depended on the high efficiency of extraction solution and the excellent selectivity and sensitivity of Orbitrap. This validated method has been successfully applied on the determination of veterinary drugs in real commercial *Oplegnathus punctatus* samples. The results indicated that the proposed method is a very simple, rapid and efficient multi-residue and multi-class analytical method for the simultaneous determination of 80 veterinary drugs in fish tissue.

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