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*8th
meeting on
medicinal
biotechnology*

BOOK OF
ABSTRACTS

MAY 29TH, 2026
SCHOOL OF HEALTH
POLYTECHNIC OF PORTO



BOOK OF ABSTRACTS OF THE 8TH MEETING ON MEDICINAL BIOTECHNOLOGY

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ISBN: 978-989-9226-20-3
DOI: [HTTPS://DOI.ORG/10.26537/ED.P.PORTO.251](https://doi.org/10.26537/ED.P.PORTO.251)

May, 2026



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Optimization of SPE-HPLC-FLD methodologies for the extraction and detection of steroid hormones in plasma

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The quantification of steroid hormones, such as estradiol and ethinylestradiol, in plasma requires sensitive and reliable analytical methodologies¹. This study presents the optimization of a bioanalytical method based on Solid-Phase Extraction (SPE) coupled with High-Performance Liquid Chromatography with Fluorescence Detection (HPLC-FLD). For sample extraction, Strata-X (polymeric reversed-phase) cartridges were used, where critical SPE parameters, including cartridge conditioning, matrix washing, and analyte elution, were evaluated. Chromatographic separation was achieved on a C18 stationary phase using a mobile phase of acetonitrile and 0.1% formic acid in ultrapure water. Both isocratic and gradient elution programs were tested to maximize resolution and peak shape. The method was validated for linearity, recovery, and precision (intra- and inter-day), meeting all established criteria for bioanalytical applications. This combined

optimization resulted in improved selectivity and sensitivity for the determination of estradiol and ethinylestradiol in plasma (Figure 1), demonstrating a robust bioanalytical tool for monitoring these compounds in complex biological matrices.

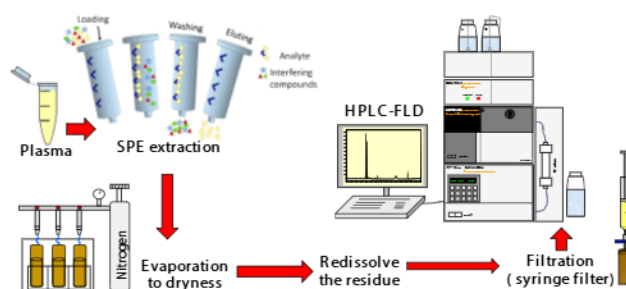


Figure 1. Overview of the SPE-HPLC-FLD methodology for steroid hormones analysis in plasma.

KEYWORDS: high-performance liquid chromatography; steroid hormones, solid-phase extraction

ACKNOWLEDGEMENTS:

This work received financial support from the Northern Regional Operational Program under the HEALTH-UNORTE project (NORTE-01-0145-FEDER-000039) and the FCT Fundação para a Ciência e a Tecnologia and by FCT/MCTES (Foundation for Science and Technology and Ministry of Science, Technology and Higher Education) (2023.03003.BD). Authors also thank the portuguese national funds (FCT/MECI, Fundação para a Ciência e a Tecnologia and Ministério da Educação, Ciência e Inovação) through the project UID/50006/2025 - Laboratório Associado para a Química Verde - Tecnologias e Processos Limpos (DOI: 10.54499/UID/50006/2025).

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