Title: Improved Specificity for Targeted LC-MS/MS Measurements of 2,3-dinor-11β-Prostaglandin F2α in Urine using FAIMS Technology

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Abstract:

Background:

Mast cells are crucial for immune responses, wound healing, and allergic reactions. Mast cell activation disorders occur due to excessive activation, releasing inflammatory mediators and leading to conditions like mast cell activation syndrome. Urine testing of mediator metabolites, such as prostaglandin metabolite 2,3-dinor-11β prostaglandin F2α (BPG), by liquid chromatography-tandem mass spectrometry (LC-MS/MS) offers a noninvasive alternative for the detection of mast cell activation. However, chromatographic interferences can severely impair detection accuracy. Field asymmetric ion mobility spectrometry (FAIMS) technology, a type of differential mobility spectrometry (DMS), enhances method selectivity and robustness. In this work, we demonstrate the improved signal-to-noise ratio for BPG quantification from patient urine samples using the Thermo Scientific™ FAIMS Pro Duo interface on a Thermo Scientific™ TSQ Altis™ Plus MS. The enhanced BPG specificity suggests FAIMS selectivity could reduce interferences without compromising analytical or clinical performance.

Methods:

After extraction on an SPE plate, BPG was quantified on a Thermo Scientific™ Vanquish Horizon UHPLC system coupled to a TSQ Altis Plus MS in the selected ion monitoring (SRM) mode, with the FAIMS Pro Duo interface. Intra-assay and inter-assay precision were tested using 20 replicate measurements of four urine pools. Accuracy was confirmed by comparing results with a reference LC-MS/MS method currently performed clinically without an ion mobility device. Results were compared using Passing-Bablok regression analysis.

Results:

Quantifying BPG in urine samples is analytically challenging due to co-eluting background interferences, even after extensive sample preparation. Incorporating FAIMS selectivity markedly reduced matrix interferences in chromatograms of 114 patient samples. Furthermore, FAIMS improved signal-to-noise ratios for low calibration samples, achieving strong linearity ($R^2 > 0.99$). Without FAIMS, 46 of 114 patient urine samples exhibited BPG quantifier/qualifier ion pair differences exceeding 20%, whereas FAIMS selectivity ensured measurements within 20% for all samples, streamlining reporting and reducing turnaround time. Precision and accuracy assessments demonstrate inter- and intraday imprecision below 6.5% and good agreement with a reference method.

Conclusion:

FAIMS improved BPG quantification in urine samples via LC-MS/MS by enhancing peak shape and reducing background, improving specificity, and demonstrating excellent performance in validation studies.