The analysis of drugs and related compounds in biological fluids using LC/MS/MS has become an essential technique in the pharmaceutical industry. This method allows for the quantification of analytes in complex matrices, such as plasma and urine samples. However, matrix effects can significantly affect the performance of the analysis. To mitigate these effects, sample dilution is commonly used, but this can introduce variability and may not always eliminate matrix suppression.

Extractive Electrospray Ionization (EESI) is an indirect electrospray process where analytes are ionized, aerosolized, and transported to the target before ionization. This method is reported to exhibit minimal matrix effects compared to conventional electrospray ionization (ESI). However, EESI requires the use of an auxiliary spray of a pure solvent to generate the ionization source, which can add complexity to the analysis.

The study aims to compare the performance of a thermal gradient focusing (TGF) EESI source configuration with AJS-ES (existing method) in reducing matrix effects. The performance of the TGF-EESI was assessed in the context of a matrix effect study, where a standard was used to prepare samples at different levels in blank plasma, and the analyte response was measured.

### Experimental

#### Sample Preparation

The Agilent LC/MS Toxicology Test Mixture (5160-4471) is a commercial mixture of pharmaceuticals, including benzodiazepines, opiates, and benzodiazepines. This mixture is used to evaluate the performance of various analytical methods. The test mixture is dissolved in methanol, and a reference solution is prepared at 100 ng/µL, 10 fg/µL, and 10 fg/µL for benzodiazepines, opiates, and benzodiazepines, respectively. Dilutions of this solution are prepared for the analysis.

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