



K22 Rapid Inline Derivatization of Primary and Secondary Amine Containing Drugs Using NBD-F and CE-LIF

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After attending this presentation, attendees will be able to understand the mechanism by which drugs containing primary and secondary amine groups are derivatized on — capillary using NBD — F for the purpose of detection using capillary electrophoresis with laser-induced fluorescence.

This presentation will impact the forensic community by explaining how this method allows for the sensitive and rapid detection of drugs in bodily fluids for the purpose trace analysis and general drug screening.

Capillary electrophoresis has become an increasingly common analytical method in forensics due to its flexibility and the wide variety of detection systems which can be used. One particularly useful method of detection in CE is laser-induced fluorescence, LIF. The application of LIF permits highly sensitive detection of compounds using CE in spite of the narrow pathlength inherent in the procedure. However, the number of compounds in which fluorescence occurs naturally are few and in order for most compounds to fluoresce, derivatization is necessary.

There are a variety of fluorescent dyes which can be coupled to primary and secondary amines. Derivatization can take place through reactions with dyes linked to reactive groups such as isothiocyanates, succinamidyl esters, and other amine reactive groups. These derivatizations can be performed before the analyte enters the capillary, while it is on the capillary during the separation or after the separation has been completed, post-capillary. On-capillary derivatization is not used as commonly as pre-capillary or post-capillary derivatization due to difficulties in reproducibility and optimization of the derivative yield. If these issues could be overcome it would greatly increase the throughput of analyses given and permit the use of inline, microfluidic techniques.

A feature common to many drugs of abuse are primary, secondary, or tertiary amine moieties. NBD-F is a non-fluorescent compound which reacts to primary and secondary amines by losing the fluorine attached to the benzene ring and joining to the analyte at the nitrogen which loses a hydrogen atom. The resulting derivative is strongly fluorescent and has an emission wavelength around 530 nm. Given that prior to derivatization NBD-F is not fluorescent, the excess reagent produces minimal interference with the analyte permitting sensitive and specific detection of the drug conjugates. The overall process permits a highly sensitive and rapid screen for drugs in body fluids.

This paper will discuss the development of in-line derivatization techniques for trace detection and screening of phenethylamines and other drugs of abuse.

On-Capillary Derivatization, Phenethylamines, Capillary Electrophoresis