

This work describes a method for the simultaneous determination of primary d- and l-amino acids and secondary amino acids such as d- and l-proline. In order to remove interferences in the simultaneous determination of primary and secondary amines, the primary amines were derivatized with o-phthalaldehyde/N-acetyl-l-cysteine (OPA/NAC) and subsequently with 1-(9-fluorenyl)ethyl chloroformate (FLEC) for secondary amines, in a pre-column separation derivatization technique. These fluorescent diastereomers of the amino acids were obtained within 3 min at room temperature and determined simultaneously by changing wavelengths during analysis in a single eluting run in the high-performance liquid chromatography column. This method, referred to as the "two-step labelling method," is effective for the simultaneous determination of d- and l-amino acids.

