

ABSTRACT (EN)

This thesis deals with the determination of nucleotides in bacterial cells. Nucleotides play crucial role in most of the metabolic pathway. Determining their concentrations could give us important clues about the influence of internal and external conditions on the bacterial metabolism.

Previously published papers dealing with the analysis of nucleotides and other intracellular metabolites can be divided into two groups according to the analytical approach: a) metabolomic approach and b) targeted approach dealing with narrow group of target analytes. In the case a) most authors use the state-of-the-art LC-MS/MS technique, whereas in the case b) robust UV detection coupled mainly to IP-LC is widely used. The aim of this study was to combine both approaches to obtain a method for routine analysis that would take advantages of mass detection, such as sensitivity and selectivity, while avoiding the need of demanding optimization of MS/MS transitions and expert service. The main purpose of the newly developed HILIC-MS method is its universal applicability in most biological and biochemical laboratories.