

Characterization of nucleotide pool in bacterial cells

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Various bacterial responses to environmental stimuli lead into changes in intracellular concentration of small molecules (nucleotides, nucleosides and their derivatives). Robust, sensitive and simple method for characterization of these changes is therefore necessary. E.g. in the case of stringent response – a potential novel drug target - intracellular levels of alarmone nucleotides guanosine tetraphosphate (ppGpp) and pentaphosphate (pppGpp), need to be determined.

There are two key steps in the process of determining nucleotide levels in bacterial biomass (complex samples with strong matrix effect): 1) extraction method with quantitative and reproducible yield, and 2) good analytical method capable to distinguish and quantify individual nucleotides.

To characterize nucleotide pools in bacteria we have chosen LC-MS in HILIC mode of separation that exhibits many advantages over commonly used ion pair (IP) LC coupled with UV-VIS detector. Moreover, the ballast mass from bacteria are well separated from majority of analytes and do not disturb the analysis.

Biography:

After the period of pesticide analysis in certified laboratories, I switched to medicinal chemistry and continue working with liquid chromatography and mass spectrometry techniques. After several years, I have started PhD studies. Modern analytical chemistry is my hobby and could be often quite challenging.

2003-2008: Faculty of Food and Biochemical Technology, UCT, Prague

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