

Analysis of non-covalent complexes – chitinase chito-oligosaccharide interactions – by use of nano-ESI and IR-MALDI mass spectrometry.

A. L. Norberg¹, A. I. Dybvik², V. Schute³, M. Mormann³, J. Soltwisch³, K. Dreisewerd³, S. Berkenkamp⁴, J. Peter-Katalinić³, K. M. Vårum², V. G. H. Eijssink¹, and M. Sørli¹

¹*Department of Chemistry, Biotechnology and Food Science, Norwegian University of Life Sciences, PO 5003, N-1432 Ås, Norway.* ²*Department of Biotechnology, Norwegian University of Science and Technology, Trondheim, Norway.* ³*University of Münster, Institute for Medical Physics and Biophysics, Robert-Koch- Str. 31, D-48149 Münster, Germany.*

⁴*Sequenom GmbH, Mendelssohnstr. 15d, 22761 Hamburg, Germany.*

Mass spectrometry can provide a versatile tool for studying non-covalent complexes in the gas-phase. However, transferring intact ionized non-covalent complexes from the condensed into the gas phase represents a challenging task due to the weak interactions within the complex that have to be maintained during the desorption/ionization process. Typically, electrospray ionization (ESI), is the method of choice to study non-covalent complexes due to its gentle desorption/ionization properties, allowing even very weakly bound complexes to be transferred into the gas phase. Analysis of such complexes using (UV) matrix assisted laser desorption/ionization (MALDI) represents greater challenges due to the acidic and organic nature of most (UV) MALDI matrices and the often rather high internal energy deposited in the analyte upon desorption/ionization. Together, this typically results in dissociation of the non-covalent contacts.

Here, we compare two different mass spectrometric approaches for desorption/ionization, detection and characterization of non-covalent complex formation between the inactive chitinase A mutant, E315Q, from *Serratia marcescens* and chito-oligosaccharides. We find that the use of infrared (IR-) MALDI-o-TOF mass spectrometry employing an Er:YAG laser ($\lambda = 2.94 \mu\text{m}$) and a glycerol matrix represents an alternative method for the analysis of intact non-covalently bound chitinase chito-oligosaccharide complexes. The observed pattern of gaseous ionic species is almost identical to the results obtained by nano-ESI-q-TOF and those

obtained in previous experiments on similar systems [1]. These findings indicate that IR-MALDI-o-TOF-mass spectrometry allows monitoring the liquid phase complex formation between enzyme and ligand.

1. Cederkvist, F., et al., *Identification of a high-affinity-binding oligosaccharide by (+) nanoelectrospray quadrupole time-of-flight tandem mass spectrometry of a noncovalent enzyme-ligand complex*. *Angew. Chem.-Int. Ed.*, 2006. **45**. 2429-2434.