PEPTIDOMICS ANALYSIS OF THE NITROGEN-FIXING BACTERIUM <i>GLUCONACETOBACTER DIAZOTROPHICUS</i>

Lery, L.M.S.; Oliveira-Carvalho, A.L.; Bisch, P.M.; Zingali, R. Unidade de Espectrometria de Massas e Proteômica – UFRJ Rede Proteômica do Rio de Janeiro

One important application of mass spectrometry (MS) is the direct analysis of complex samples allowing the identification of small proteins and peptides in a single step. Proteomics techniques were used to characterize the major peptides expressed by <i>Gluconacetobacter diazotrophicus</i>. This bacterium, found within plants such as sugarcane and sweet potato, fixes atmospheric nitrogen, produces plant growth-promoting hormones and bacteriocins and solubilizes zinccompounds. In the experimental setup, <i>G. diazotrophicus</i> grown in LGIP medium until exponential phase were lysed with Tris pH 11. Urea 8M. SDS 2% and DTT 200mM. Proteins were precipitated with TCA and the resulting soluble sample was dialysed in water using 1kDa membrane. Also, the culture supernatant filtered in a 0.22µm membrane was ultrafiltrated using 1 and 10kDa cut-off membranes. Both samples were concentrated in Sep-Pak C18 cartridges previously to MS analysis. Samples were analyzed in MALDITOF in the mass range 1.000-10.000 m/z in linear mode. The first peptidomic view of $\langle i \rangle G$. diazotrophicus</i> PAL5 is presented herein, showing 11 major peptides in whole cell lysate of <i>G. diazotrophicus</i> cells and 13 in the culture supernatant. We identified in the MS spectra several peptides with molecular mass < 2.500Da. However, the main peaks of the lysate spectra were 5.030 and 5.162 m/z. We will further sequence these peptides by MALDFTOF/TOF in order to elucidate their role in <i>G. diazotrophicus</i> physiology. Support: FAPERJ, CNPg and FINEP.