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Proteomic analysis of two *E. coli* samples and sequencing of acetylase mediated from the pMdT1 recombinant plasmid

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Microbial resistance to antibiotics is a worldwide problem in human and veterinary medicine. *Escherichia coli* are commensal microorganism of the gastrointestinal tract of animals and humans. The knowledge about the transfer of genes and specific features of mechanisms such as horizontal gene transfer allows us to understand the acquisition of resistance mechanisms in different organisms. The presence of pMdT1 plasmid containing a gene that encodes a variant of the AAC (6')-lb-cr protein which confers resistance to kanamycin and tobramycin and decreases the susceptibility to ciprofloxacin and norfloxacin has great importance in the study of antibiotic resistance. Two *E. coli* samples were analyzed. Electromax DH10B is a transformation-ready strain and TF-Se20 is a strain that contains the pMdT1 plasmid expressing the acetylase gene. After extraction, fractionation and quantification, proteins were separated by one dimensional and after by two-dimensional electrophoresis and subsequent analysis by matrix-assisted laser desorption/ionization-time of flight/mass spectrometry (MALDI-TOF/MS). To determine the sequence of the protein of interest, liquid chromatography-mass spectrometry was performed (LC-MS). From gels containing TF-Se20 strain it was possible to identify 76 distinct proteins, 71 of which had a known function and from Electromax DH10B strain 72 different proteins were identified of which 71 being associated with a biological process. Proteins identified were related to biological processes such as glycolysis and biosynthesis of proteins. Aminoglycoside N (6')-acetyltransferase type 1 was sequenced. The application of proteomics helped to clarify and to obtain more information about the mechanisms of resistance to determine the total proteome of two samples and analyzing the sequence of acetylase mediated by pMdT1 plasmid.

Biography

Pedro Magalhaes has completed his degree in Genetics and Biotechnology and MSc Biotechnology for Health Sciences, in the University of Trás-os-Montes and Alto Douro. In his Master thesis, he cooperated and worked in Functional Genomics and Proteomics Unit, University of Trás-os-Montes and Alto Douro and Bioscope Group, University of Nova de Lisboa, working on proteomics and mass spectrometry. He has 3 poster presentations and 1 oral communication in national and international conferences.

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