## Pristine carbon nanotubes for an efficient L-asparaginase immobilization

R.O. Cristóvão,<sup>a</sup> M.R. Almeida,<sup>b</sup> M.A. Barros,<sup>a</sup> J.C.F. Nunes,<sup>a,b</sup> R.A.R. Boaventura,<sup>a</sup> J.M. Loureiro,<sup>a</sup> J.L. Faria,<sup>a</sup> M.C. Neves,<sup>b</sup> M.G. Freire,<sup>b</sup> V.C. Ebinuma-Santos,<sup>c</sup> A.P.M. Tavares<sup>b\*</sup> and C.G. Silva<sup>a</sup>

<sup>a</sup>-Laboratory of Separation and Reaction Engineering - Laboratory of Catalysis and Materials (LSRE-LCM), Department of Chemical Engineering, Faculty of Engineering, University of Porto, Rua do Dr. Roberto Frias, 4200-465, Porto, Portugal.

<sup>b.</sup>CICECO-Aveiro Institute of Materials, Department of Chemistry, University of Aveiro, 3810-193 Aveiro, Portugal.

<sup>c.</sup>Department of Engineering Bioprocess and Biotechnology, School of Pharmaceutical Sciences, UNESP-University Estadual Paulista, Araraquara, Brazil.

The enzyme L-asparaginase (ASNase) presents effective antineoplastic properties for acute lymphoblastic leukemia treatment, besides their potential use in the food sector to decrease acrylamide formation. Considering their applications, the improvement of these enzyme properties by efficient immobilization techniques is in high demand. Carbon nanotubes are promising enzyme immobilization supports since these materials have increased surface area and effective capacity for enzyme loading. Accordingly, in this study, multi-walled carbon nanotubes (MWCNTs) were explored as novel supports for ASNase immobilization by a simple adsorption method. The effect of pH, the contact time of immobilization, and the ASNase to nanoparticles mass ratio were optimized according to the enzyme immobilization yield and relative recovered activity. The enzyme-MWCNTs bioconjugation was confirmed by thermogravimetric analysis (TGA), Fourier transform infrared spectroscopy (FTIR), and transmission electron microscopy (TEM) studies. MWCNTs have a high ASNase loading capacity, with a maximum immobilization yield of 90%. The adsorbed ASNase retains 90% of the initial enzyme activity at the optimized conditions (pH 8, 60 min, and  $1.5 \times 10^{-3}$ g.mL<sup>-1</sup> of ASNase). According to these results, ASNase immobilized onto MWCNTs can find in several areas improved applications, namely biosensors, medicine, and the food industry.

Acknowledgments: This work was financially supported by Base Funding - UIDB/EQU/50020/2020 of the Associate Laboratory LSRE-LCM - funded by national funds through FCT/MCTES (PIDDAC), and POCI-01-0145-FEDER-031268 - funded by FEDER, through COMPETE2020 - Programa Operacional Competitividade e Internacionalização (POCI), and by national funds (OE), through FCT/MCTES. This work was developed within the scope of the project CICECO-Aveiro Institute of Materials, UIDB/50011/2020 & UIDP/50011/2020, financed by national funds through FCT/MCTES

85

## OC42