

19676 | Carbon xerogels for the purification of anti-leukemic drugs

Marramaque, Teresa P., Faculdade de Engenharia

Barros, Rita A. M., Faculdade de Engenharia

Carabineiro, Sónia A. C., LAQV-REQUIMTE

Freire, Mara G., CICECO

Faria, Joaquim L., Faculdade de Engenharia

Santos-Ebinuma, Valéria C., UNESP

Tavares, Ana P. M., CICECO

Silva, Cláudia G., Faculdade de Engenharia

Cristóvão, Raquel O., Faculdade de Engenharia

Abstract

Nowadays, L-asparaginase (ASNase, EC 3.5.1.1) is an enzyme used for leukemia treatment, in the pharmaceutical industry, and for starchy foods pre-treatment, in the food industry. These applications require an enzyme purification process with a vast number of processing steps, resulting in high operating costs. In addition, many cases of adverse reactions in patients have been reported due to ASNase instability and thermolability.

In order to increase the stability of this enzyme and reduce its purification costs, non-functionalized and functionalized carbon xerogels (CXs) were studied as a purification platform of ASNase from a cell extract of *Bacillus subtilis*. These materials were selected due to its unique properties, such as tunable porosity, high surface area and adsorption capacity.

In this work, different operating conditions were studied during cell extract adsorption onto CXs, such as: cell extract concentration (1-15 mg mL⁻¹), material's type and mass (12, 18 and 24 mg), and net adsorption volume (1.5, 2 and 15 mL tubes). SDS-PAGE analysis was carried out to complement the results.

The results showed that high extract concentrations (7.5, 10, 12.5 and 15 mg mL⁻¹) hamper the separation between ASNase and the remaining proteins. However, adsorption of 3 mg mL⁻¹ of extract onto functionalized CXs seems to be unfavorable, since it decreased the purity of the enzyme in most cases. On the other hand, adsorption of 3 mg mL⁻¹ extract onto non-functionalized CXs in 15 mL tubes allowed a 1.63-fold increase in the ASNase specific activity in the supernatant, when compared to the initial extract. SDS-PAGE analysis confirmed that these conditions seem to be the ideal relationship between cell extract concentration/material's type and mass/net adsorption volume.

In summary, the results obtained in this work revealed that the use of non-functionalized CXs is a promising alternative to traditional ASNase purification processes.