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Purification of antileukemic biopharmaceuticals using carbon xerogels

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L-asparaginase (ASNase) is a vital enzyme successfully used in leukaemia treatment[1] (pharma industries) as well as in food processing (potato, bakery and cereals).[2] However, standard ASNase purification techniques are complex and expensive, increasing their cost and therefore limiting access to lifesaving treatments.[3] To overcome these limitations, new, cheaper, and simpler methods for ASNase purification, yet capable of delivering comparably high purification levels, are needed.[4] This work aims to study a new purification method using carbon xerogels (CXs) as a platform for ASNase purification from cellular extract. Accordingly, the purification capabilities of CXs with different textural properties were initially tested in batch mode. Purification fold (PF) was determined by comparing the specific activity of ASNase present in the cellular extract before and after purification. The effect of total protein in the cellular extract (1.5-20 mg) and average pore size of the CXs (6-30 nm) in the obtained PF was assessed. All results were confirmed by sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE). CXs were noted to preferentially adsorb impurities rather than ASNase, leaving part of the enzyme in the supernatant. Increments in cellular extract total protein concentration hindered the purification process, with the concentration of 1.5 mg/mL being optimal (PF=1.43). In terms of average pore diameter, only the CX with a pore diameter of 9.8 nm was able to purify ASNase with satisfying results, achieving a PF of 2.5. The results of SDS-PAGE confirm that CXs have a higher affinity for the impurities. After the batch purification tests, an adsorption column was packed with the CXs and tested for the purification of ASNase in continuous mode of operation. In the continuous purification process, the packed column with CX was able to continuously purify ASNase from cellular extract for at least 3 hours with a PF equal to 1.5.

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