PURIFICATION OF THE CURCIN (a RNA N-GLYCOSIDASE) FROM JATROPHA CURCAS L. SEEDS PIE

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The use of the Jatropha plant pie has been limited due to the presence of toxic, allergenic and anti-nutritional factors. In part, this toxicity has been attributed to a protein named curcin. The curcin is a RNA N-glicosidase with great biotechnological potential for presenting several biological activities. The objective of this study was to optimize a protocol for the isolation and purification of the curcin from Jatropha seeds pie. Initially, the seeds pie was delipidated with hexane for 12 h and transformed into a fine and homogeneous powder. This powder was submitted to extraction (1: 10 w / v) with 5 mM sodium phosphate buffer (pH 7.0), containing 200 mM NaCl, for 3 hours and centrifuged at 10.000 rpm for 30 min at 10°C. The supernatant was brought to 60% saturation with solid ammonium sulfate for 8 h to 10°C. The protein precipitate was then dissolved in the same extraction buffer, dialyzed and fractionated on a column of Sephadex G-100. Two major protein peaks were obtained, referred to as PI and PII. The fraction corresponding to PII was pooled and analyzed by SDS-PAGE. A single protein band of approximately 28 kDa was observed in fractions of PII, similar to the molecular weight of curcin from seeds. This fraction was also able to depurinate the ribosomal RNA from Pichia pastoris to a concentration of 5 ng, confirming the RNA-N-glycosidase activity of the curcin isolated from seeds pie. As the pure protein yield was 30 μg/g, it can be concluded that the steps optimized at the extraction protocol provided an improvement in the levels of curcin extracted from the total protein content.

Keywords: RIP, Jatropha, curcin

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