ISOLATION AND PURIFICATION OF LENTIL LECTIN FOR ANALYTICAL APPLICATION IN CARBOHYDRATES RESEARCH

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Carbohydrates are highly diverse and ubiquitous natural molecules, with significant roles in many biological processes. Lectins, as carbohydrate-binding proteins that can specifically recognize and reversibly bind different mono- and oligosaccharides, and glycoconjugates, have proven to be an excellent analytical tool in investigating these glycans structures.
The aim of this study was to introduce a relatively low-cost protocol for the isolation and purification of lectin from lentil seeds, whose agglutinating activity could be used in different glycans studies. To characterize samples after each purification step, the Bradford assay for determination of protein concentration, hemagglutinating activity analysis and PAGE electrophoresis were used. After lentil extraction, lectin crude extract was purified by different purification steps including precipitation with ammonium sulfate and ultrafiltration. The highest results in terms of hemagglutinating activity and protein concentration were determined for fraction isolated after precipitation with ammonium sulfate (saturation 40-80%) and whose molecular weight exceeding 50 kDa. In addition, it was shown that isoelectric precipitation of contaminant proteins after precipitation and ultrafiltration enhanced both the agglutinating activity and the purity of the resulting lectin preparation.