Biophysical and preliminary crystallographic studies of Lectin from Entada rheedii seeds

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A novel lectin was isolated, purified and characterized from seeds of *Entada rheedii* using ammonium sulphate precipitation followed by lactose affinity chromatography. On SDS-PAGE, the purified Entadin lectin appeared as a single band (monomer in nature) with a molecular mass of approx. 20 kDa both in reducing as well as in nonreducing conditions. Mass spectroscopic analysis confirms the molecular weight of Entadin lectin as 19333 Da.

Entadin lectin showed highest titer value in agglutination against human blood group-B RBC and its Hemagglutination activity was inhibited by lactose, cellobiose, and galactose only. Periodic Acid Schiff's (PAS) stain confirmed the glycoprotein nature of Entadin lectin with an approx. 5 % of carbohydrate content. The lectin is highly stable even after incubation at a wide range of temperatures (30 to 60 °C) and pH (6 to 10). Antiproliferative effect of Entadin lectin against lung cancer cells A549 and cervical cancer cells HeLa showed IC₅₀ value of 38 μ g/mL and 34 μ g/mL and no anti-proliferative activity against normal cells. Cell morphological studies revealed that Entadin lectin induced apoptosis both in A549 and HeLa cancer cells which was confirmed by (AO/EB) and Hoechst (33258) nuclear counter staining. Further, Lectin was crystallized using the hangingdrop vapour-diffusion method with 30% PEG 8000 as precipitating agent, 0.2 M ammonium sulphate and 0.1 M sodium cacodylate pH 6.5.

Keywords: Lectin, Lactose affinity chromatography, Glycoprotein, Antiproliferative, crystallization