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Thermal and chemical denaturation of *Colocasia esculenta* tuber agglutinin from $\alpha 2\beta 2$ to unfolded state, its 1.74 Å crystal structure and a complex crystal structure at 1.85 Å with mannose

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The major tuber storage protein of *Colocasia esculenta* is a monocot mannose-binding, widely used dietary lectin. This tuber agglutinin contains two polypeptides of 12.0 and 12.4 kDa by MALDI-TOF analysis. By both gel filtration and dynamic light scattering at pH 7.2 show the lectin has a $\alpha 2\beta 2$ form; however, at pH 3, it converts to $\alpha\beta$ form. Our circular dichroism spectroscopy studies show that the lectin retains approximately 100% of its secondary structure between pH 2-8. The fluorescence emission maxima of 346 to 350 nm for pH 4 to 10 show that the tryptophan residues are relatively exposed. The unfolding is a simple two-state process, $N \leftrightarrow U$, as seen in our denaturation scan profiles, when monitored by fluorescence, far-UV CD and near-UV CD are completely super imposable. Analyses of these profiles provide an estimate of several thermodynamic parameters at each guanidinium chloride concentration, including the melting temperature T_g , which is 346.9 K in 0 M, but lowers to 321.8 K in 3.6 M. Mannose-free mannose-bound lectin crystals were obtained by hanging-drop, vapor-diffusion method at room temperature and high-resolution X-ray diffraction data were collected using a home X-ray source. The mannose-free structure (5D5G) and mannose-bound structure (5D9Z) are both available in the PDB, along with the X-ray data. Some highlights of both structures will be presented. Such high resolution structures obtained from a home X-ray source is rare among this class of lectins and it has not been possible to crystallize simple mannose with such lectins before.

Biography

Rajagopal Chattopadhyaya (b.1957) completed his PhD (UCLA,1987) under Richard Dickerson, who as a postdoc solved the 2Å structure of myoglobin under John Kendrew. Rajagopal did his postdoctoral work at UC Berkeley and Baylor College of Medicine. He has been a faculty member at Bose Institute since 1993, professor since 2006. His work at Bose Institute is mentioned in the Encycl. Britannica. Book of the Year, 1996 and LexA model in Burton E. Tropp's Molecular Biology : Genes to Proteins, 3rd edn, a continuation of David Freifelder's classic undergraduate textbook. Structural Biologist, biochemist, religious historian & author (as a hobby).

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