Molecular Biology and Information - Biopolymer Structure -

Visitors

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Scope of Research

Our research aims to elucidate structure-function relationships of biological macromolecules, mainly proteins, by using X-ray diffraction method and other physicochemical methods. The following attempts have been mainly made in our laboratory for that purpose. (1) X-ray diffraction studies on protein structures in crystal and in solution are carried out by crystallographic and/or small-angle X-ray scattering techniques to elucidate structure-function relationships of proteins. (2) Molecular mechanism for myosin assembly is studied by proteolytic method, electron microscopy, and computer analysis of the amino acid sequence.

Research Activities (Year 2001)

Presentations


Grants


Fujii T, Elucidation of reaction mechanism of high-active-thermostable aspartase by crystallographic analyses of complexes, Grant-in-Aid for Encouragement of Young Scientists, 1 April 2000 - 31 March 2003.

Hata Y, X-Ray crystal structure analyses of functional proteins, Grant of Rice Genome Project PR-2202, MAFF, JAPAN, 1 April 2000 - 31 March 2002.
Structure-based analysis of functional sites of thermostable aspartase

Aspartase catalyzes the reversible conversion of L-aspartate to fumarate and ammonium ion, and plays an important role in the bacterial nitrogen metabolism. The structure of the thermostable enzyme from Bacillus sp. YM55-1 was determined by X-ray diffraction method and compared with those of E. coli aspartase and fumarase C, which belong to the same family. The results revealed four conformationally different regions which were located around putative functional sites, suggesting the involvement of these regions into functions characteristic of the individual enzymes. Moreover, the increase in the number of intersubunit hydrogen-bonds and salt-bridges explains the thermostability of the present aspartase.

Analyses of enzymes involved in biogenesis of iron-sulfur cluster

E. coli CsdB is a dimeric PLP-dependent NifS-homologue and catalyzes the decomposition of L-selenocysteine into selenium and L-alanine with specificity higher than that for cysteine. The structure of the enzyme has been determined by X-ray crystallographic method. The subunit of CsdB comprises a large domain, a small domain, and an N-terminal segment. A remarkable structural feature of CsdB is that an α-helix in the lobe extending from the small domain to the large domain in one subunit of the dimer interacts with a β-hairpin loop protruding from the large domain of the other subunit. Cys364, which is essential for the activity toward cysteine but not toward selenocysteine, is clearly seen on the loop of the extended lobe (Thr362-Arg375).

Direct detection of protein quaternary structure and denatured entity by SAXS

A change in the quaternary structure of the oligomeric protein is directly detectable by a small-angle scattering method. Denaturation process of the chaperonin protein GroEL showed that the disappearance of the quaternary structure can be monitored by the Kratky plot of the scattered intensities, demonstrating the advantage of the SAXS method over other indirect methods. The quaternary structure collapsed at a GdnHCl concentration of 0.8 M. The pair wise plots of the change in J(0)/C and the Rg,z enabled us to estimate the stability and nature of the denatured protein. The GroEL tetradecamer was dissociated directly to unfolded coils. The denatured ensemble is some mixture of entangled aggregates and monomeric coil molecules.