FERM domain associates with the plasmamembrane and directly binds the juxtamembrane region of the cytoplasmic tail of CD44, whereas precise details concerning with the FERM-CD44 interaction remain unclear. Recently, we have successfully crystallized the radixin FERM domain bound to the CD44 cytoplasmic tail peptide (Mori *et al*.,2007). The structure revealed the CD44 binding to the groove between helix α1C and strand β5C of FERM subdomain C. The CD44 peptide forms a short β strand that associates with strand β5C by anti-parallel β-β interactions. In addition, the C-terminal tail of the peptide binds to the hydrophobic groove between helix α1C and strand β5C. The binding mode found in the present structure is compared with those of the previously determined structures of the FERM domain bound to ICAM-2 (Hamada *et al*.,2003) and NEP (Terawaki *et al*.,2007).

Hamada *et al*., ***EMBO J.*** **22**, 502-514 (2003).
Terawaki *et al*., ***JBC*** **282**, 19854-19862 (2007).
Mori *et al*., ***Acta Crystallogr. F*** **63**, 844-847 (2007).

Keywords: CD44, ERM proteins, FERM domain

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Expression, purification and crystallization of Aurora kinase C

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The Aurora family of serine/threonine kinases plays key roles in cell division. Three Aurora kinases (Aurora A, Aurora B and Aurora C) are expressed in mammals and have highly conserved catalytic domains. Despite these similarities, they differ in function, subcellular localization, and timing of activity during cell division. Aurora A and Aurora B are present in mitosis. In contrast, the expression of Aurora C is restricted to meiosis. Although Aurora C is indicated to work as a key regulator in meiosis, its function is poorly understood. We attempted to determine the structure of Aurora C in order to obtain the insight into its function. Active-Aurora C as a C-terminal His6-tagged fusion protein was overexpressed in *E. coli* and purified by Ni-NTA column chromatography. The purity was confirmed by SDS-PAGE. Initial crystallization trials using 1-2 mg/ml of purified protein gave small crystals. Optimization of crystallization conditions for X-ray crystallography is currently in progress.

Keywords: aurora, kinase, crystallization

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Structural insights into the SM protein-syntaxin interactions

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The Sec1/Munc18 (SM) proteins and the soluble N-ethymaleimide-sensitive factor attachment protein receptors (SNAREs) play an essential role in vesicle docking and fusion. The SNARE complex is composed of the target membrane SNAREs (t-SNAREs Sx and SNAP) and the vesicle membrane SNARE (v-SNARE VAMP). The formation of the SNARE complex leads to membrane fusion. Three mammalian SM proteins (Munc18a, Munc18b and Munc18c) are suggested to regulate vesicle transport to the plasma membrane. Munc18a is a predominantly neuronal protein, which binds to Sx1-3. Munc18b interacts with the same Sxs as Munc18a but is expressed mainly in epithelial cells. Munc18c binds to Sx2 and Sx4, involved in insulin-regulated trafficking of glucose transporter (GLUT4). Previous structural studies showed that neuronal Munc18a binds to cognate Sx1a in a closed conformation that is incompatible with SNARE complex formation (1). We recently showed that Munc18c binds to SNARE complex as well as Sx4 alone (2). Furthermore, we found that Munc18c required the N-terminal 29 residues (N-peptide) of Sx4 for its interaction and determined the crystal structure of Munc18c bound to Sx4 N-peptide (3). Taken together, our findings suggest that the N-terminal binding mode is a conserved molecular mechanism that mediates membrane fusion.

References
1. Misura K, Scheller R, Weis W. (2000) Nature 404, 355-62.
2. Latham, C.F., Lopez, J.A., Hu., S.-H., Gee, C.L., Westbury, E., Duncan Blair, D., Armishaw, C., Alewood, P.F., Bryant, N.J., James, D.E. and Martin, J.L. (2006) Traffic 7, 1408-1419.
3. Hu, S.-H., Latham, C.F., Gee, C.L., James, D.E., and Martin, J.L. (2007) PNAS 104, 8773-8778.

Keywords: protein crystallography, vesicle trafficking, protein-protein interactions

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Nucleant-mediated protein crystallization with microporous zeolite showing heteroepitaxial growth

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Protein crystallization is a major bottleneck in X-ray crystallography to determine the three-dimensional structure which is indispensable for understanding the chemical mechanism of protein function. Because the current methodology of protein crystallization is a kind of screening, medically or biologically important targets such as human membrane proteins are usually difficult to be crystallized due to their poor crystallizability. The hetero-epitaxial growth from the surface of mineral crystal as a nucleant had been thought to be effective to enhance the chance of protein crystallization. However, generally applicable hetero-epitaxial nucleants for protein crystallization have never been found. Here we introduce the first candidate of universal hetero-epitaxial nucleant, microporous zeolite, which is a synthetic aluminosilicate crystalline polymer with regular micropores and generally promotes a form-selective crystal nucleation of proteins as a crystallization catalyst. The most successful zeolite nucleant was Molecular Sieves 5A with pore size of 5 Å and with bound calcium ion, suggesting that the versatility of the microporous zeolite is likely to be derived from a general regularity in proteins such as secondary structures, or the high frequency of calcium ion recognition on the protein surface.

Keywords: X-ray crystallography, protein crystallization, epitaxial growth