and purified. Tm values of each mutant were determined by circular dichroism (CD) spectra. The change of Tm value (ΔTm) from native BstHU was +3.1°C(T13A), +1.7°C(T33L), -2.0°C(E34D), and -4.0°C(K38N) respectively. Each mutant was crystallized for X-ray structure analysis. The crystallization conditions of each mutant were 40% MPD, 80mM phosphate(pH 8.0), and protein concentration was 25mg/ml. E34D mutant was crystallized as above condition plus 6% dioxane. X-ray experiments were done for T13A mutant, and this crystal diffracts at least 2.5Å. Cell parameters are a=65.5 b=37.3 c=65.5 and ß=1 14.5 (that is isomorphous to native *B.st* HU crystal), and overall merging-R was 3.8%. Structure determination was done by a molecular replacement. For model structure, we used the coordinates of native *B.st* HU. Refinement of this mutant structure was done with X-plor and current R-factor is 20%. We discuss the difference of thermal stability between native and mutant based on the structure.

Immune System

MS04.15.01 STRUCTURAL BIOLOGY OF IgSF CELL ADHESION MOLECULES. E. Yvonne Jones. Laboratory of Molecular Biophysics, The Rex Richards Building, South Parks Rd., Oxford, OX1 3QU, UK.

Structural studies on cell adhesion molecules, primarily those belonging to the immunoglobulin superfamily (IgSF), will be described. Direct intercellular and cell-matrix interactions are fundamental to the development and maintenance of multicellular organisms. These interactions are mediated by the extracellular regions of specific cell surface receptors, termed cell adhesion molecules. Sequence analysis indicates that the extracellular regions of many of these molecules contain immunoglobulin-like folds, leading to their classification as members of the IgSF. IgSF molecules variously mediate specific adhesion through a broad range interaction modes, homotypic with the same or closely related IgSF molecules, heterotypic with other types of adhesion molecules or even with specific carbohydrates. We have determined the structures of functional portions of several IgSF adhesion molecules by x-ray crystallography. These include representative examples of molecules which function by widely differing adhesion modes; IgSF/IgSF, IgSF/integrin and IgSF/sialylated glycan. The structural adaptations of the basic Ig-like motif which confer the different binding specificities will be assessed.

Jones E.Y., Davis S.J., Williams A.F., Harlos K. and Stuart D.I. (1992) Crystal structure at 2.8 Å resolution of a soluble form of the cell adhesion molecule CD2. *Nature* **360** 232-239

Bodian D.L., Jones E.Y., Harlos K., Stuart D.I. and Davis S.J. (1994) Crystal structure of the extracellular region of the human cell adhesion molecule CD2 at 2.5 Å resolution. *Structure* **2** 755-766

Jones E.Y., Harlos K., Bottomley M.J., Robinson R.C., Driscoll P.C., Edwards R.M., Clements J.M., Dudgeon T.J. and Stuart D.I. (1995) Crystal structure of an integrin-binding fragment of Vascular cell adhesion molecule 1 (VCAM-1) at 1.8 Å resolution.*Nature* **373** 539-544

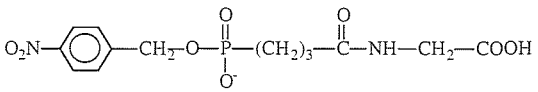
Vinson M., Van der Merwe P.A., Kelm S., May A., Jones E.Y. and Crocker P.R. (1996) Identification of a putative sialic acid binding site within the amino-terminal immunoglobulin-like domain of sialoadhesin: analysis by site-directed mutagenesis. *J. Biol. Chem.* (In the press)

MS04.15.02 THE CRYSTAL STRUCTURE OF ICAM-2, ITS COMPARISON TO OTHER INTEGRIN-BINDING PROTEINS. Wang, J-H.#, Casasnovas, J.*, Springer, T.*, Liu, J.H.#, Harrison, S.C.$. *Center for Blood Research, Harvard Medical School, Boston, MA. 02115; #Dept. of Mol. and Cell. Biol., Harvard University, Cambridge, MA. 02138; $Dept. of Mol. and Cell. Biol. and HHMI, Harvard University, Cambridge, MA. 02138, USA

A crystal structure of the extracellular fragment of ICAM-2 has been determined to 2.2Å by the MIR method. ICAM-2 is an adhesion molecule, constitutively expressed on endothelial cell surface; it binds to integrin LFA-1 on leukocyte surface. It may be important for leukocyte trafficking in uninflamed tissues. The structure consists of two tandomly arranged Ig domains;the compact, N-terminal integrin-binding domain (Di) and the relatively larger C_2-type Ig domain (D_2). Both D_1 and D_2 are structurally homologous to D_1-D_2 of VCAM-1, with two distictions. The integrin-binding site on ICAM-2 is not so prominent a protrusion as in VCAM-1. Rather, significant bending between D_1 and D_2 ensures the exposure of the binding region. There are 5 "extra" residues at the N-terminus, flexibly extended from the tip of D_1. This extension may explain why ICAM-2 does not serve as a rhinovirus receptor despite the expected close structural similarity between ICAM-1 and ICAM-2.

MS04.15.03 STRUCTURAL STUDIES OF CATALYTIC ANTIBODIES WITH ESTERASE ACTIVITY. M. Knossow[1], J.-B. Charbonnier[1], B. Gigant[1], B. Golinelli[1], D. Tawfik[2], Z. Eshhar[2] and B.S. Green[3] [1]Laboratoire de Biologie Structurale, UMR 9920 C.N.R.S., 91198 Gif-sur-Yvette, France; [2]Department of Chemical Immunology, Weizman Institute, Rehovot, Israël; [3]School of Pharmacy, Hebrew University of Jerusalem, Jerusalem, Israël.

The X-ray structures of the complexes of three esterase-like catalytic antibodies with a transition state analog of the reaction they catalyze are reported. These antibodies were identified by screening for catalytic activity the total immune repertoire elicited in response to the phosphonate transition state analog (TSA) hapten **1**. The high resolution (1.9 Å and 2.2 Å) structures suggest that rate acceleration is accounted for by oxyanion stabilization.



1

Most of the abzymes with esterase activities known to date have been raised against phosphonate haptens similar to **1**. Comparison of the newly determined structures to those of other abzymes with similar activities shows that the p-nitrophenol part of the hapten is buried in a hydrophobic pocket that is conserved in these antibodies and that the orientation of the hapten with respect to the antibody is conserved as well. The oxyanion hole motifs fund in Fabs with esterase-like activity whose structures have been determined are varied: a lysine (1), two peptide NHs (2) and an (Asn, Tyr) combination in the three antibodies we have studied. Participation to the oxyanion hole of a tyrosine residue is likely to be related to the well known prevalence of tyrosine residues in antibody combining sites; it is unprecedented in esterases.

(1)W.G. Zhou et al., *Science* **265**, 1049-1064 (1994)
(2)J.-B. Charbonnier et al., *P. N. A. S. USA* **92**, 11721-11725 (1995)