**Conference Abstracts**


Keywords: Ig-binding protein, superantigen, combinatorial crystallization.

Viruses and bacteria have developed strategies for the disruption of immune defences by binding to molecules of the immune system. Staphylococcus aureus protein A (SpA) binds to both the Fc and VH region of antibodies. The crystal structure of the complex between a domain of SpA and a human Fab has enabled a better understanding how this pathogen targets a wide number of human B-cells.

It is a process with similarities to that of T-cell superantigens but with its own peculiarities. This structure and its biological implications will be presented and discussed.

Ig-binding proteins like SpA have been extensively used in immunology, we can extend their use even further to help us set up a combinatorial method for the crystallization of proteins with applications in immunology and structural genomics. The use of combinatorial complex crystallization, stoichiometry variation screening will be presented and their applications discussed.


Keywords: heparin binding protein, chemo attractant, mutagenesis.

Heparin binding protein (HBP), also known as CAP37 or azurocidin, is a multifunctional protein with diverse implications in host defence and inflammation. The antibiotic activity of HBP, directed against Gram-negative bacteria, was one of the first functions of HBP to be elucidated. HBP binds strongly to the lipid A component of endotoxin, and it has been suggested that the bactericidal effect of HBP is due to binding of lipid A. HBP is a chemotactant for monocytes, T-cells and K-cells and induces longevity and differentiation of monocytes towards a macrophage phenotype. In addition, endothelial cells and fibroblasts in monolayers are stimulated by HBP to detachment and aggregation.

HBP is an inactive serine protease homologue, and the inactivity is caused by selective mutations in the active-site triad (Ser for a His at position 41 and Gly for a Ser at position 175) [1]. Despite the inactivity of HBP, the serine protease inhibitor BPTI (bovine pancreatic trypsin inhibitor) is still capable of binding to HBP [2].

Human HBP consists of 225 amino acid residues and is highly glycosylated. The structure of recombinant glycosylated HBP has been determined at 2.3 Å resolution at room temperature [1] and at 1.1 Å resolution at 120 K [3]. The structures confirmed the homology to the serine proteases. However, surface loops and surface charge distribution are significantly different in HBP and in the serine proteases. Based on the structures, a putative lipid A binding site and a putative endothelial cell protein kinase C activation site have been identified.

In order to further elucidate the biological functions of HBP, the structure of native HBP in non-glycosylated form [4] and of two HBP mutants have been determined. The two different mutants ( [R23S,F25E] HBP and [G175Q]HBP) were designed in order to identify amino acid residues of importance for lipid A and BPTI binding, and the structures have been determined to 2.5 and 1.9 Å resolution, respectively. In addition, the structure of porcine HBP has been determined to 2.6 Å resolution, resulting in valuable information on species differences.