Crystal structure of C1 inhibitor

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C1 inhibitor is a serpin type serine protease inhibitor in blood plasma, showing relatively low sequence homology with serpins of known structure [1,2]. C1 inhibitor is the only inhibitor known that acts on enzymes involved in early steps of the classical and lectin pathways of complement activation. Its physiologically crucial targets also include plasma kallikrein, as well as fXIIa and fXIIa in the intrinsic coagulation. It binds endotoxins from bacteria and E-, P-selectin adhesion proteins on endothelial cells that also contribute to its anti-inflammatory activity. Its deficiency results in hereditary angioedema [3]. In the cases when the risk of inflammation is high, such as organ transplantation, heart attack or sepsis, application of C1 inhibitor in the therapy was found beneficial [4].

Our aim is to understand how heparin [5] and related polyanions increase activity on some targets and not in others. We also explore the structural basis of the effect of specific mutations [6] of this physiologically important protein.

We expressed the serpin domain of human C1 inhibitor in Pichia pastoris and purified for crystallization. Crystals were grown in 1.6M K-Na-phosphate (pH 7.5). Data were collected from the native and Xe derivative crystals at 100 K. Data collections were carried out on the EMBL beamline X11. Space group: P6₃, unit cell dimensions: a,b=98.9 Å, c=94.7 Å resolution: 2.65Å. The Xe derivatisation was not successful. The structure was solved by molecular replacement using prealigned structures of various serpins as search model. The R_work and R_free values of the refined model are 0.174 and 0.218, respectively.

Figure 1: Crystal structure of C1 inhibitor. The sheets of the molecule are colour coded. The RCL region is coloured magenta with the P1 arginine shown in sticks.
The structure shows a proteolytically intact, yet non-inhibitory form of C1 inhibitor (Figure 1). The reactive center loop region (RCL) forms an extra β-strand in the central β-sheet, which is a unique feature of C1 inhibitor structure. Unexpected conformation of the C-terminal tail illustrates how it prevents reactive center loop insertion to trap serpins in metastable state. The pattern of electrostatic potential distribution over the molecular surface suggests that the heparin binding region is located in a different region of the molecule than that of other serpins. As a consequence the mechanism of heparin enhanced inhibitory mechanism could be also different than the mechanism in the thrombin-antithrombin-heparin system [7].

The structure of C1 inhibitor will help understanding structural details of its binding to its targets, and its pathological activity. Based on the structure rational design of inherently enhanced therapeutic C1 inhibitor variants can be carried out.

References