HECUS X-RAY SYSTEMS GMBH GRAZ

Solution Structure of the Protein β2-Glykoprotein I Revealed by Small-Angle X-Ray Scattering (SAXS)

Overview

The low resolution structure of human and bovine β2-Glycoprotein I (β2-GPI) in solution has been restored by smallangle X-ray scattering. β2-Glycoprotein I (molecular mass: 53.5 kDa) is a highly glycosylated phosphat-binding plasma protein, comprised of 4 flexible connected complement control protein domains and a distinct 5th domain. The crystal structure of this protein has been solved by X-ray crystallography (Fig.2, left, PDB-code: 1C1Z) revealing that the protein adopts a J-shaped structure in the protein crystal [1].

Experimental

All small-angle X-ray scattering (SAXS) experiments were carried out with a HECUS SWAX-camera attached to a rotating anode generator (Cu-target, wavelength $\lambda = 0.154$ nm), operating at a power of 2 kW. Scattering curves were monitored in a q-range ($q = 4\pi(\sin\theta)/\lambda$, with 2θ being the scattering angle) from 0.14 to 4.0 nm-1. Protein solutions (60 µl) in a concentration range of 0.5/1/2 and 3%, respectively, were filled in quartz capillaries of 1 mm diameter and measured at 20 °C for typically 1 hour. The scattering of the buffer was subtracted and the resulting scattering curves were normalized to 0.1% and extrapolated to 0% protein concentration which was used for the parameter evaluation, fourier trans-formation and model fitting.

Results

The experimental SAXS curves of β2-Glycoprotein I resulted in a radius of gyration (Rg) of 4.3 nm, and a maximum particle dimension (dmax) of 13.9 nm (Fig.1). They fit poorly the simulated SAXS curves calculated from the crystallographic coordinates (PDB-file 1C1Z) indicating clearly that the protein in solution does not adopt the same Jshaped structure as in the crystal. Low resolution models in real-space that match the experimental SAXS data best were independently constructed by three different ab-initio 3D reconstruction algorithms resulting that an S-shaped molecular structure yielded the best fit (Fig.2, right) [2].

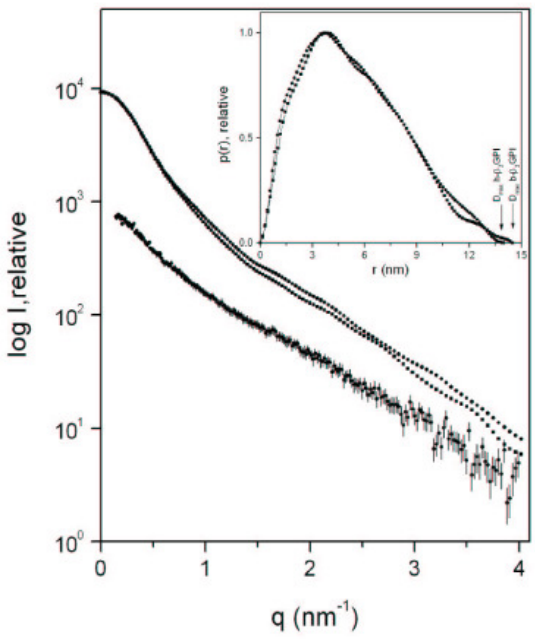

Fig. 1: SAXS-curves (lower curve: smeared rawdata, upper curves: desmeared data) and p(r)-functions (insert) of human and bovine β2-Glycoprotein I.

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Conclusion

The clear evidence that β2-Glycoprotein I can adopt different conformations due to its flexible interlinking domains (Fig.2) – a J-shape, found by X-ray crystallography and an S-shape, found by solution small-angle X-ray scattering – might be an essential feature and biological function for binding antibodies.

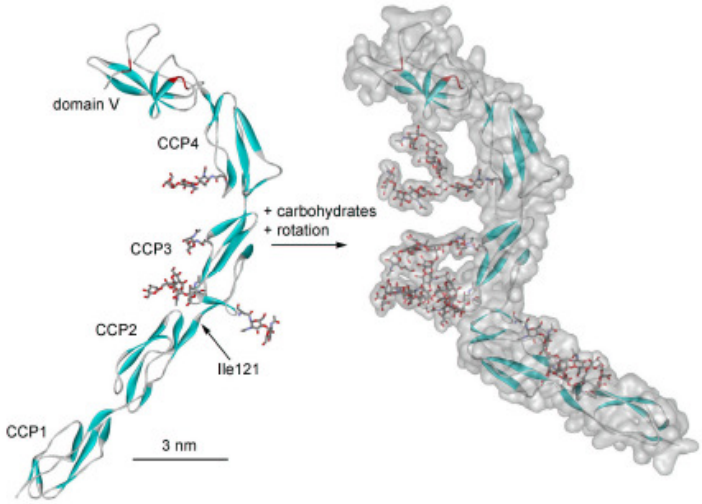

Fig.2: β2-Glycoprotein I: Crystal structure in a protein crystal as obtained by X-ray crystallography (left) and structural model as obtained by SAXS easurements in aqueous solution (right).

References

[1] R.Schwarzenbacher, K.Zeth, K. Diederichs, A.Gries, G.M.Kostner, P. Laggner and R.Prassl, MBO Journal 18/22, 6228-6239 (1999).

[2] M.Hammel, M.Kriechbaum, A.Gries, G.Kostner, P.Laggner and R.Prassl, J.Mol.Biol. 321/1, 85-97 (2002).

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