Studies of multivalent interactions between DC-SIGN and glycomimetic ligands by NMR and computational techniques

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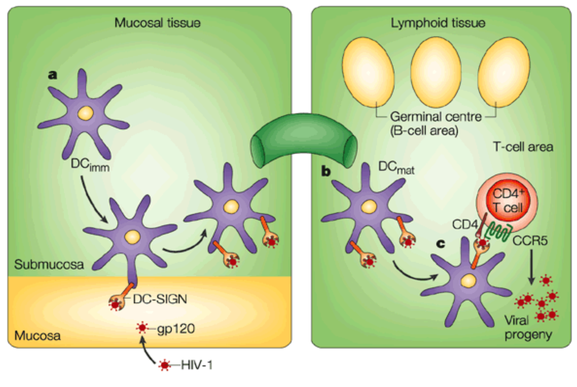
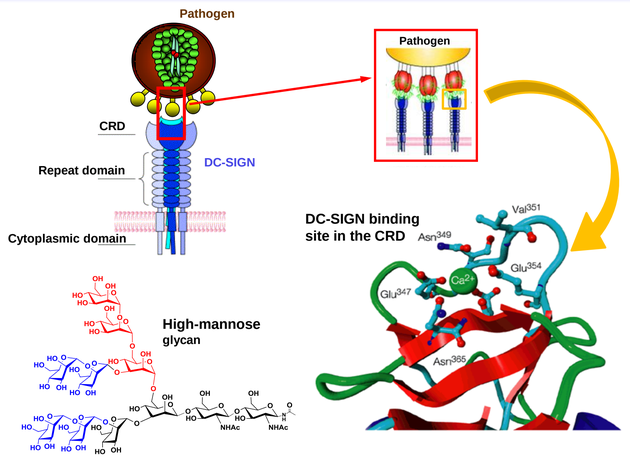
Introduction

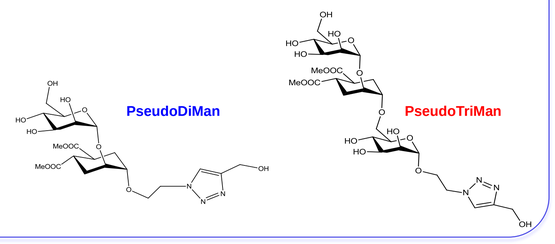


DC-SIGN is a C-type lectin with a Carbohydrate Recognition Domain (CRD) that specifically recognizes highly glycosylated structures of several pathogens such as viruses (HIV, Dengue, Hepatitis C), bacteria, yeasts, and parasites. DC-SIGN plays a key role in the infection processes of some of these pathogens, as they are recognized by interactions of the lectin with carbohydrate structures from their glycoproteins (gp120, GP1, etc.). The natural ligands of DC-SIGN are mannose oligosaccharides or fucose-containing Lewis-type determinants. We are interested in obtaining stable small-molecule glycomimetics of the natural ligands as attractive candidates for drug development.

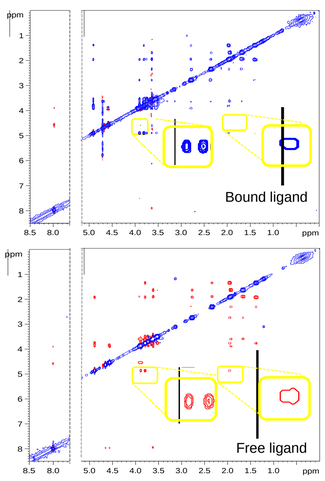


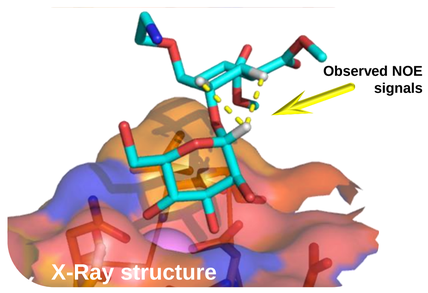
Pseudo-mannosides

Based on the structure of the natural high-mannose $Man_9(GlcN)_2$ two oligomannoside mimics, a pseudo-mannobioside, and a pseudo-mannotrioside, have been designed.
In this work we have studied their binding activities towards DC-SIGN, by NMR spectroscopy and molecular modelling.

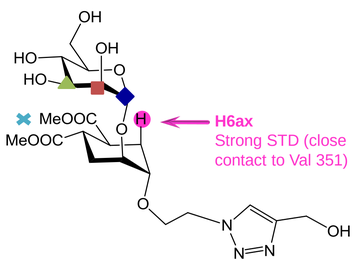


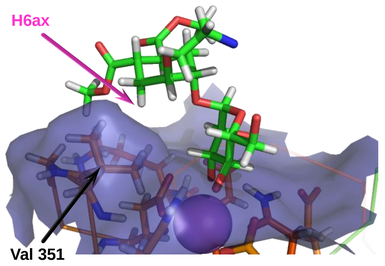
PseudoDiMan



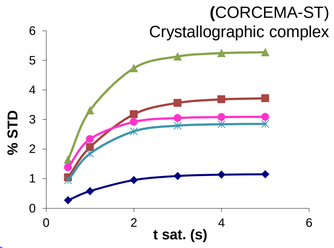


Good agreement with NMR data

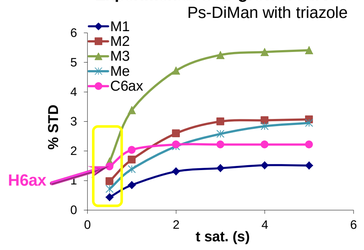




Theoretical STD Intensities (CORCEMA-ST)
Crystallographic complex

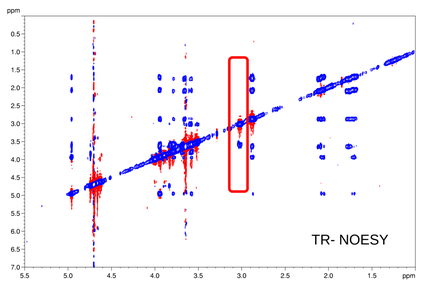


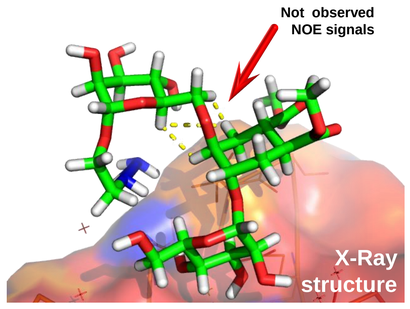
Experimental STD growth curves
Ps-DiMan with triazole



STD-NMR and CORCEMA – ST studies [1, 2]

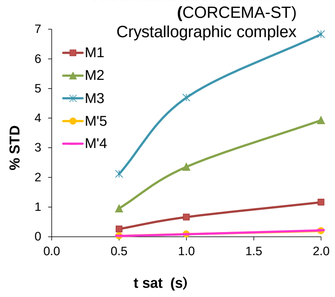
PseudoTriMan



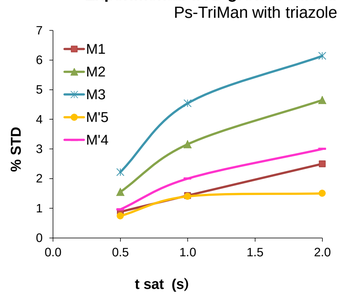


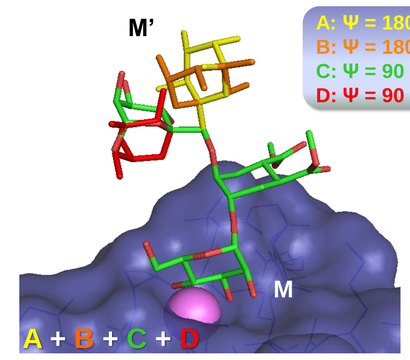
- The bound conformation determined by Transfer NOESY is comparable to that of the free state.
- The absence of some key NOE cross peaks cannot be justified by the conformation around 1-6 linkage shown in the X-ray structure.
- Interpretation of STD NMR intensities with CORCEMA-ST reveals two modes of ligand interaction in solution

Theoretical STD Intensities (CORCEMA-ST)
Crystallographic complex



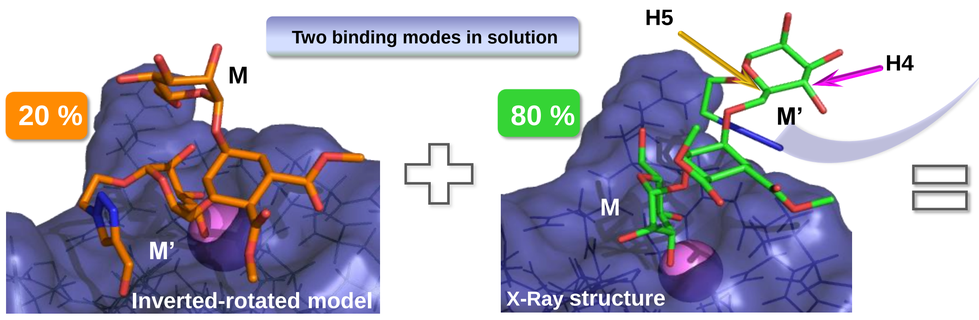
Experimental STD growth curves
Ps-TriMan with triazole



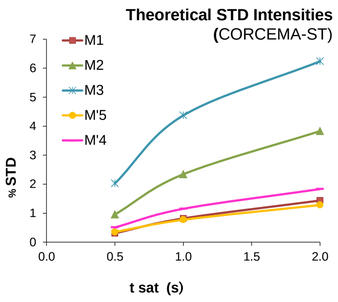


1→6 linkage (M') is undefined in the major conformation

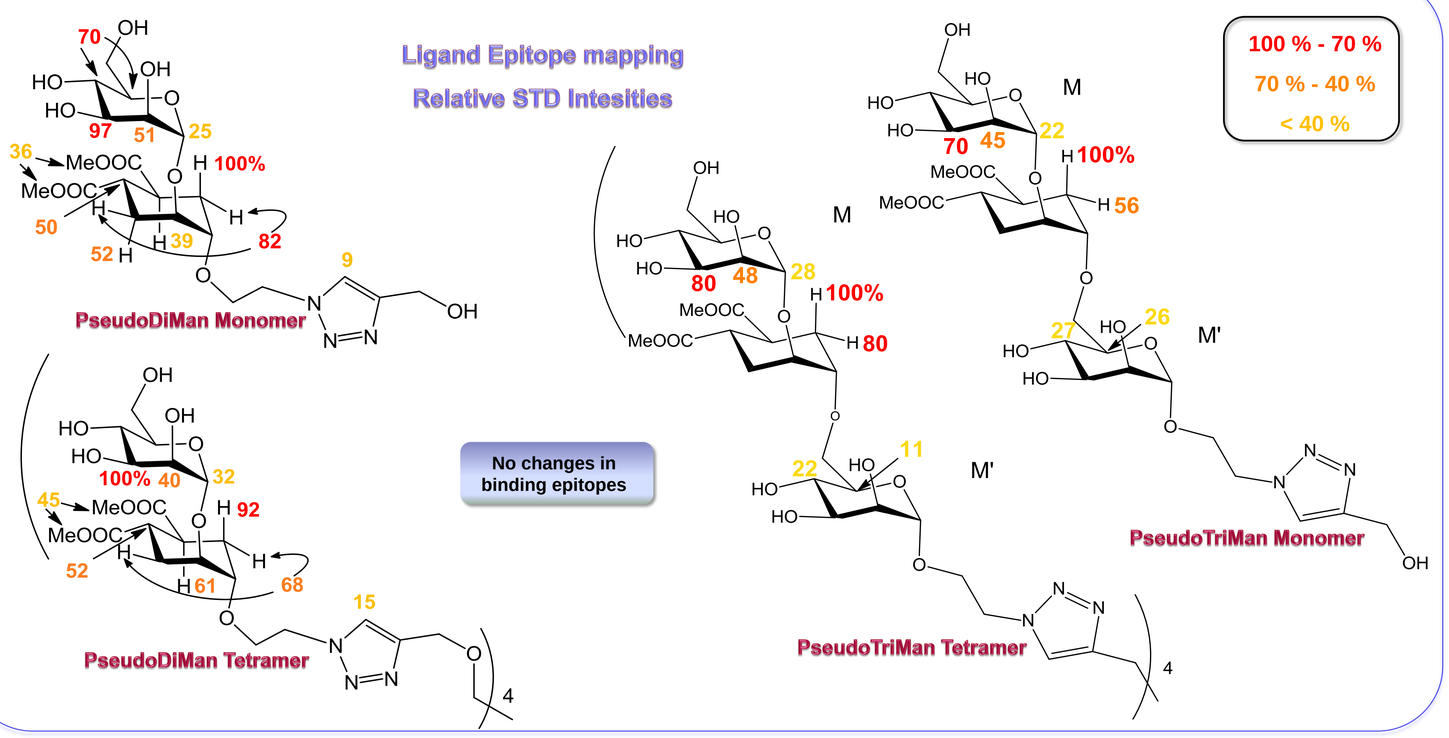
Two binding modes in solution



Theoretical STD Intensities (CORCEMA-ST)



From Mono- to Multivalent Systems



Conclusions

- NOESY experiments show that there are no changes in ligand conformation upon binding. The bound conformation in solution is in accordance with the X-ray result.
- Theoretical STD intensities confirm the agreement between the crystallographic structure of the complex and the NMR data.
- Despite DC-SIGN is known to recognize some carbohydrate ligands in a multimodal fashion, for pseudoDiMan the results clearly explain a single binding mode.
- In contrast, pseudoTriMan is mainly bound as in the X-ray structure, but with a minor contribution of a different orientation (inverted-rotated) in which the other mannose (M') is interacting with calcium ion.
- From monovalent to multivalent systems we do not see changes in the binding modes. Apparently, this is true for both pseudoDiMan and pseudoTriMan cases.
- The triazole moiety does not affect binding to DC-SIGN , both, in terms of affinity and of protein-ligand contacts.

References

[1] Meyer B. and Peters T., *Angew. Chem. Int. Ed. Engl.*, **2003**, 42, 864-890.
[2] Jayalakshmi V. and Krishna N. R., *J. Magn. Reson.*, **2004**, 168, 36-45.
[3] Feinberg H., Castelli R., Drickamer K., Seeberger P.H., Weis W. I., *J. Biol. Chem.*, **2007**, 282, 4202-4209.

Acknowledgments

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