Oral

GOLD GLYCONANOPARTICLES AS POTENTIAL MICROBICIDES AND AS CONTRAST AGENTS IN MRI

M. Marradi, O. Martinez, P. Di Gianvincenzo, D. Alcántara, J. M. de la Fuente and S. Penadés
Laboratory of Glyconanotechnology, CICbiomaGUNE/CIBER-BBN
Pº de Miramon 182, 20009 Donostia-San Sebastián (Guipuzcoa), Spain
mmarradi@cicbiomagune.es

The role of carbohydrates in living systems has been strongly reconsidered in the last 20 years: once thought to be only energetic sources, nowadays it is getting clearer their implication in cellular recognition processes. [1] Especially, carbohydrate-carbohydrate interactions seem to be responsible of the initial steps in cell adhesion and communication; these transient interactions may facilitate the formation of low-avidity interactions between biological partners. This first contact, in case of matching, can be followed by carbohydrate-protein and/or protein-protein interactions which give rise to a further stabilization and mediate the success of a particular biological process. A characteristic feature of the biological interactions where carbohydrates are involved is their extreme low affinity that has to be compensated by multivalent presentation of the ligands. Gold nanoparticles functionalised with biologically relevant (oligo)saccharides have been designed and prepared in our laboratories in order to mimic the natural presentation of the carbohydrate coating which is present at the cell surface. These glyconanoparticles (GNPs), prepared by an integrated strategy named Glyconanotechnology, [2,3,4] were used to study and intervene in carbohydrate-mediated interactions. The biofunctional GNPs are water soluble, globular shaped and with well defined composition. [4,5] Furthermore, the gold cluster can serve as a platform to insert not only carbohydrates, but also a selected set of ligands (DNA, RNA, peptides, fluorescent probes, etc). The preparation of multifunctional GNPs functionalised, in a control manner, with different ligands allow us to intervene in cell adhesion and recognition processes addressing, by means of basic research, some of the major challenges of current nanomedicine.

This lecture will focus on the presentation of our results and applications of GNPs as potential microbicides and/or vaccines against HIV-1 infection and as contrast agents in MRI for brain tumor targeting.

We have synthesised GNPs in which the gold cluster is functionalised with the mannose structural motives present in the envelope glycoprotein gp120 of the HIV-1. One of the mechanisms of HIV vaginal infection seems to be mediated by the interaction between the high-mannose oligosaccharides which are present in the gp120 and the DC-SIGN receptor of dendritic cells. [6] The inhibition power of our GNPs towards the binding of DC-SIGN to gp120 was tested by Surface Plasmon Resonance (SPR) experiments. First, we tested different mannose neoglycoconjugates and then the corresponding GNPs (Figure 1). All of them showed dose-dependent inhibition. Free oligomannosides need millimolar concentrations to give 100% inhibition, while mannose GNPs require micromolar concentrations. The best inhibitor was a GNP capped with the disaccharide Manα1-2Manα: the disaccharide man1-2man is able to inhibit the binding between DC-SIGN and gp120 at ~500 μM, while the corresponding GNPs inhibit at ~10 nM (Figure 2). This shows the multivalent effect of the GNPs. The evaluation of this inhibition in cell based models and the effect in dissemination of HIV-1 from cells bearing DC-SIGN to T-cell populations will be also illustrated.
The very same GNPs are being used to study their interaction with the monoclonal antibody 2G12, one of the few broadly neutralising antibodies isolated from HIV infected patients. In this way, we are trying to individuate a candidate for the design of a potential vaccine. Preliminary results, based on biosensors assays, will be presented.

On the other hand, the introduction in the gold cluster of additional ligands, other than carbohydrates, has allowed us to address other biomedical challenges. In particular, the functionalisation of GNPs, decorated with simple sugars (like lactose and glucose), with well-known contrast agents in T_1 (such as Gadolinium-complexes [8]) afforded novel paramagnetic nanoparticles which have been used in MRI. Relaxation times T_1 and T_2 were measured with a Minispec at 1.5 T to calculate r_1 and r_2 relaxivity values. Phantoms containing solutions of the compounds were also imaged at 7.0 T. The evaluation of the cytotoxicity of this kind of GNPs will be presented. *In vivo* imaging of intracerebral glioma in mice was run and the images were compared with the ones obtained with commercial Magnevist® (gadolinium DTPA).

References:

[1] R.A. Dwek, Chem. Rev., **96** (1996) 683.
[2] J.M. de la Fuente, et al. Angew. Chem. Int. Ed. Engl., **40** (2001) 2258.
[3] A.G. Barrientos, et al. Chem. Eur. J., **9** (2003) 1909.
[4] J.M. de la Fuente, S. Penadés, Glycoconj. J., **21** (2004) 149.
[5] J.M. de la Fuente, S. Penadés, BBA, **1760** (2006) 636.
[6] H. Feinberg, et al. Science, **294** (2001) 2163.
[7] D.A. Calarese, et al. Science, **300** (2003) 2065.
[8] J.J. Caravan, et al. Chem. Rev., **99** (1999) 2293.

Figures:

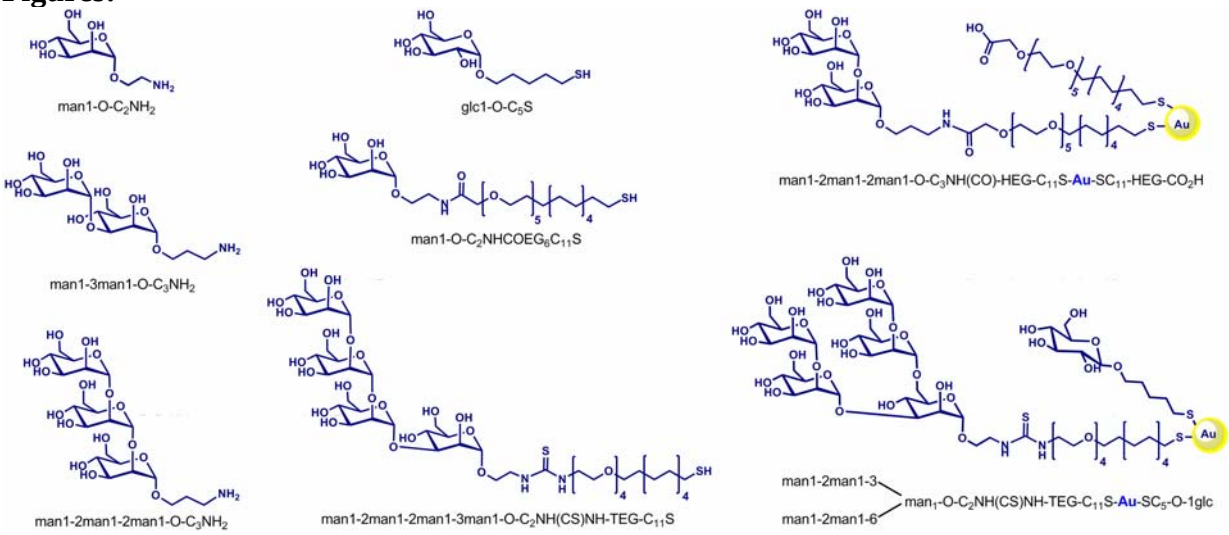

Figure 1. Some examples of neoglycoconjugates and gold GNPs used in this work.

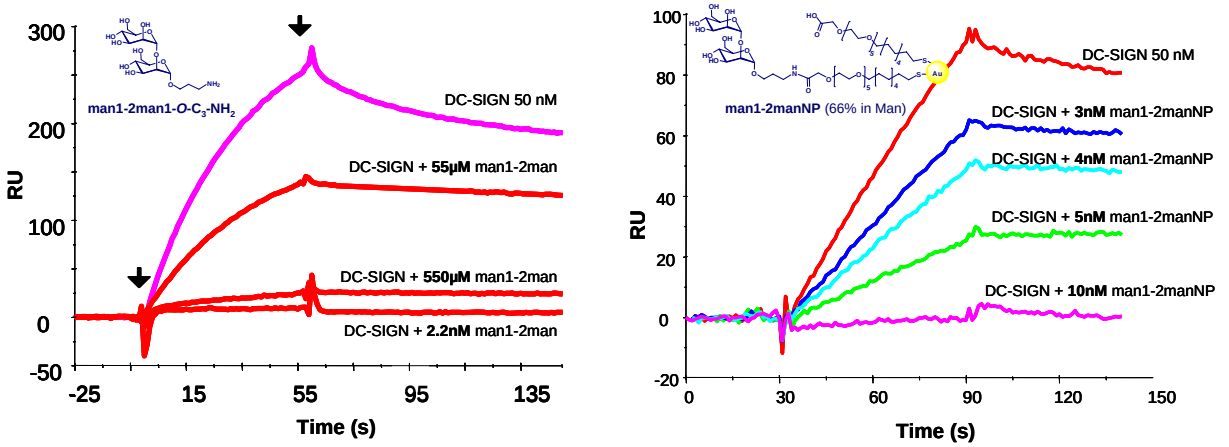

Figure 2. Dose dependent inhibition of a dimannose neoglycoconjugate (left) and the corresponding GNPs (right) on the binding of DC-SIGN to gp120 immobilized on a CM5 chip surface as measured in a BIACORE (DC-SIGN and GNPs are in fluid phase).