DC-SIGN: a target for the design of new antiviral drugs

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VII Jornadas de la Sociedad Española de Química Terapéutica

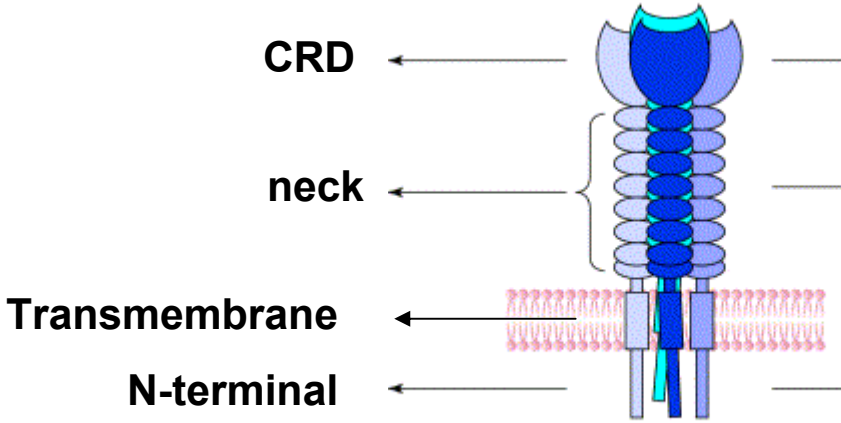
Sitges, 19-21 Octubre 2006

DC-SIGN

- Dendritic cell-specific ICAM-3 grabbing nonintegrin
- Type II transmembrane protein (C-type lectin)
- Discovered in 1992(ref) by B.M. Curtis (Bristol-Myers Squibb)
- Mainly found in immature Dendritic Cells
- Presentation as homo-oligomers

Ref: *Proc. Natl. Acad. Sci. USA*, **1992**, *89*, 8356-8360

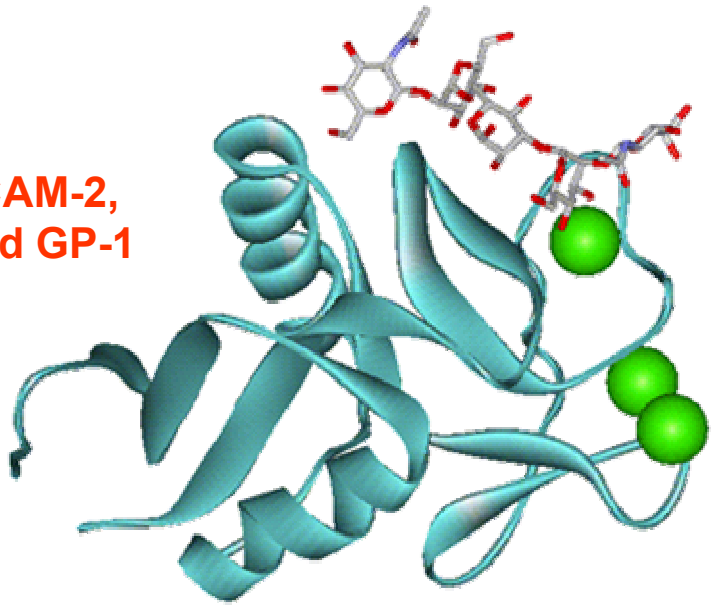
DC-SIGN Domains



Oligomerization

Internalization

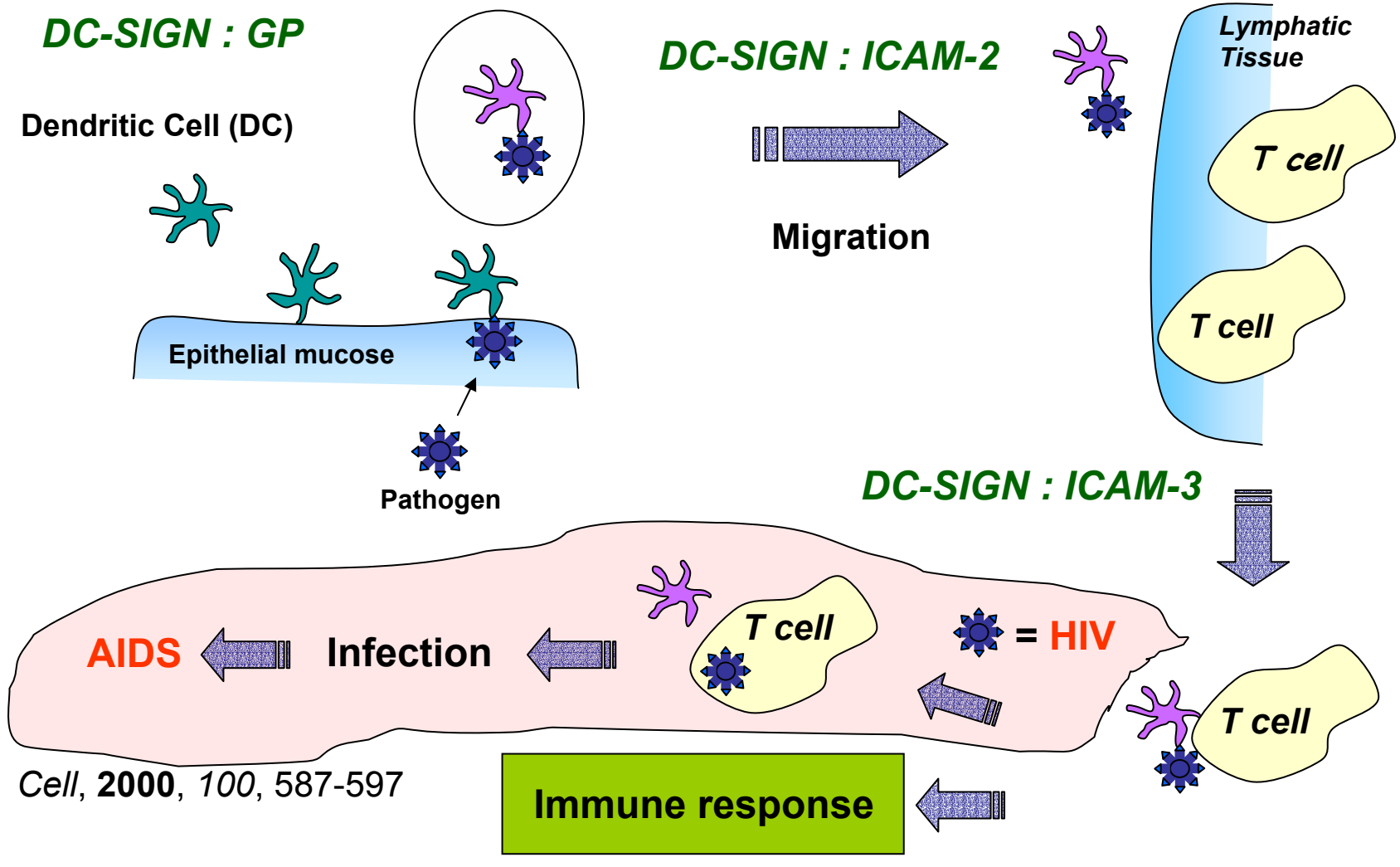
Interaction with ICAM-2, ICAM-3, gp-120 and GP-1



CRD

Feinberg *et al., Science*, **2001**

DC-SIGN and Dendritic Cells



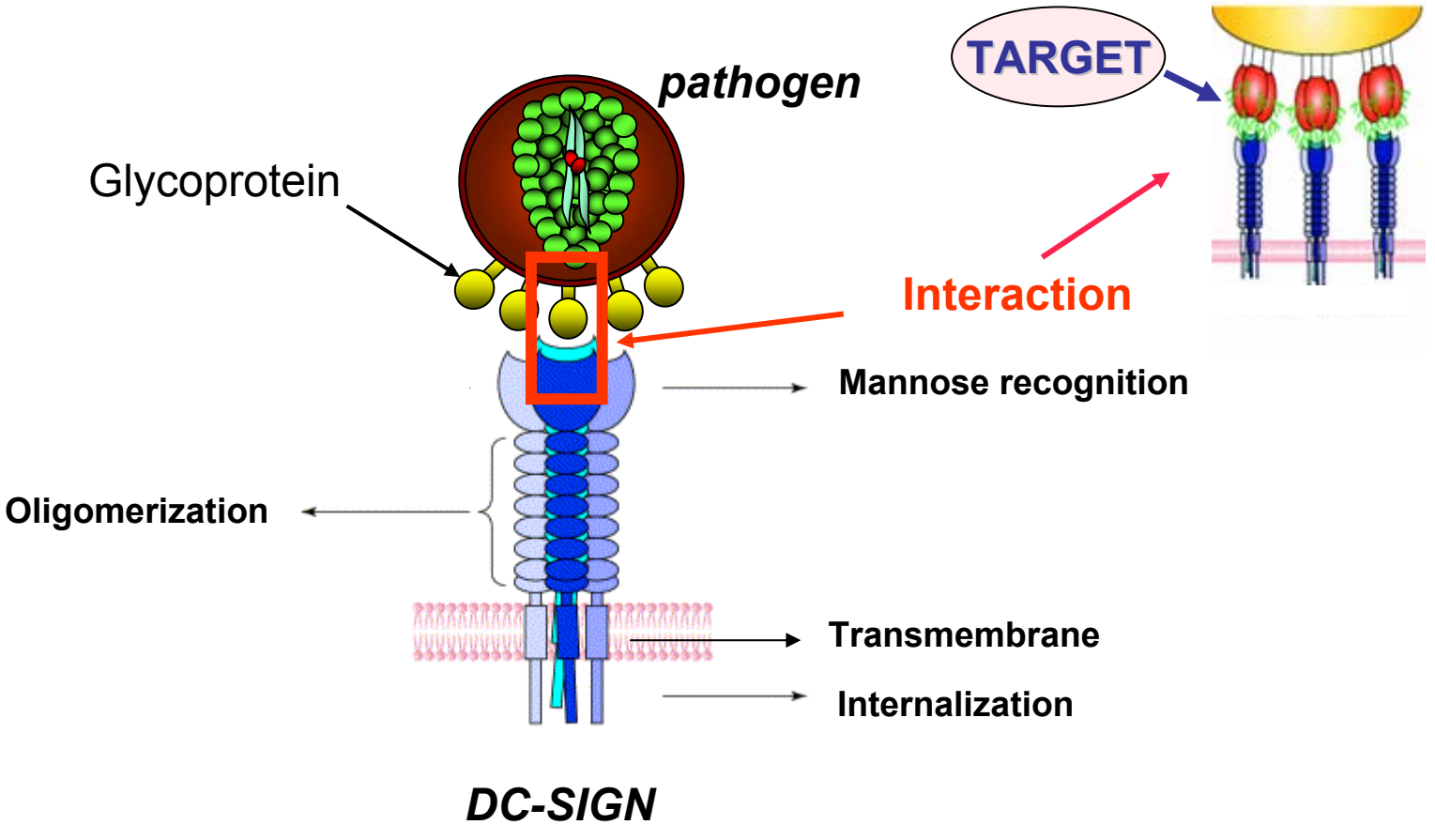
Cell, **2000**, *100*, 587-597

DC-SIGN: Universal Pathogen Receptor

CSIC

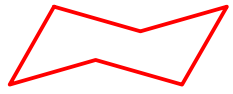
- ***HIV*** (*Cell*, **2000**, *100*, 587-597)
- ***Ebola*** (*J. Virol*., **2002**, 76, 6841-6844)
- ***Leishmania*** (*J. Biol. Chem.*,**2002**, *277*, 36766-36769)
- ***Tuberculosis*** (*J. Exp. Med*., **2003**, *197*, 121-127)
- ***Candida albicans*** (*Eur. J. Virol*., **2003**, *33*, 532-538)
- ***Hepatitis C*** (*J. Biol. Chem*., **2003**, *278*, 20358-20366)
- ***SARS*** (*J. Virol*., **2004**, *78*, 12090-12095)

Target for inhibitors design



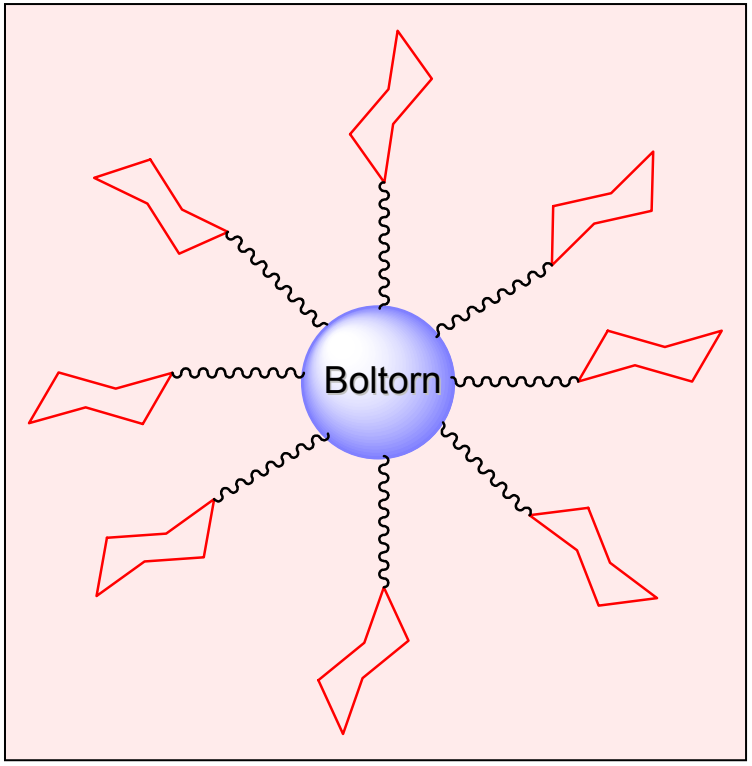
Carbohydrate multivalent system

Carbohydrate: Mannose
o derivatives

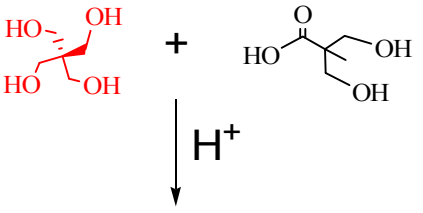


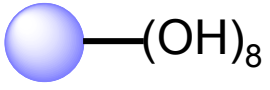
Linker:

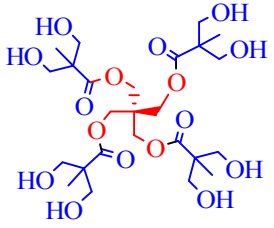
Multivalente
Support: Boltorn



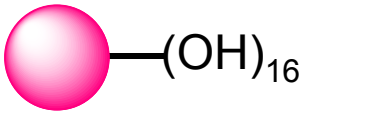
BOLTORN hyperbranched dendritic polymers

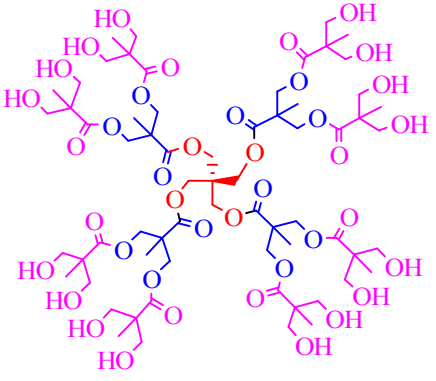




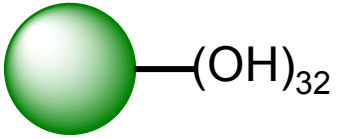


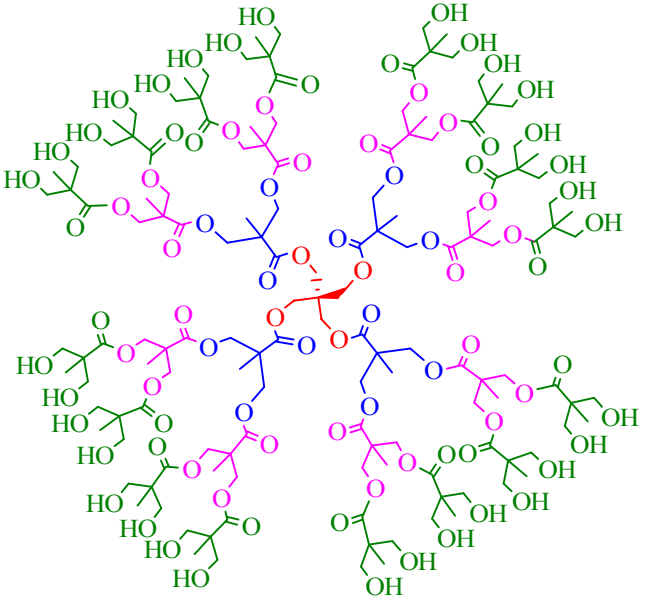
BH10
1st Generation
8 OH groups





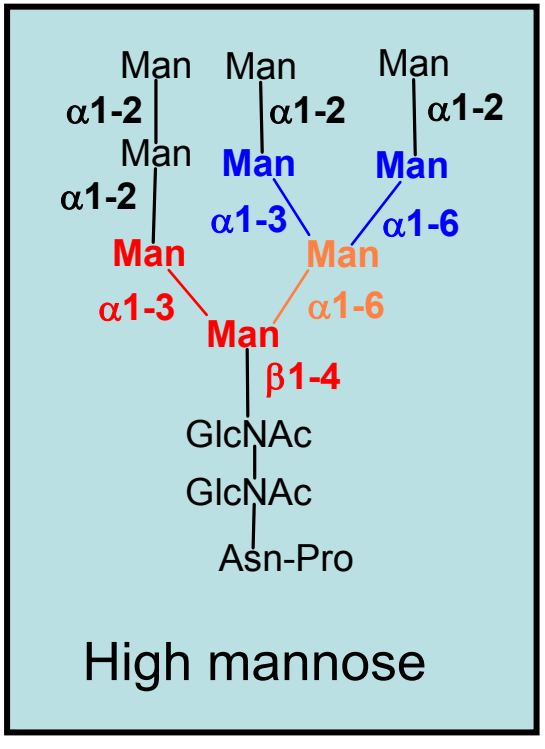
BH20
2nd Generation
16 OH groups



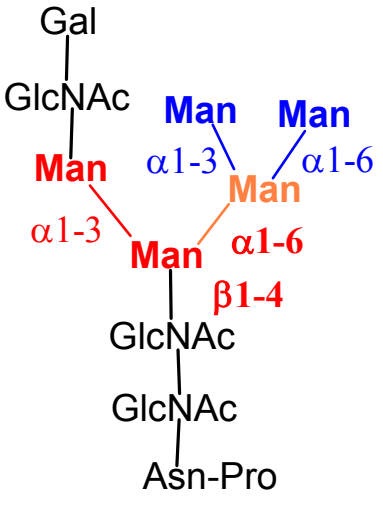


BH30
3rd Generation
32 OH groups

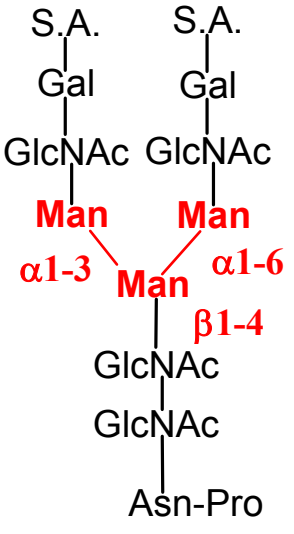
Pathogen Glycoproteins



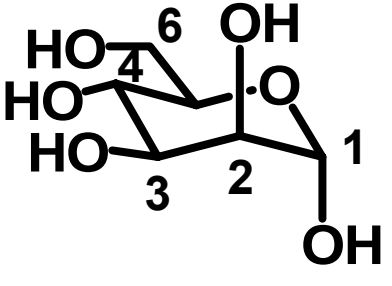
High mannose



Hybrid

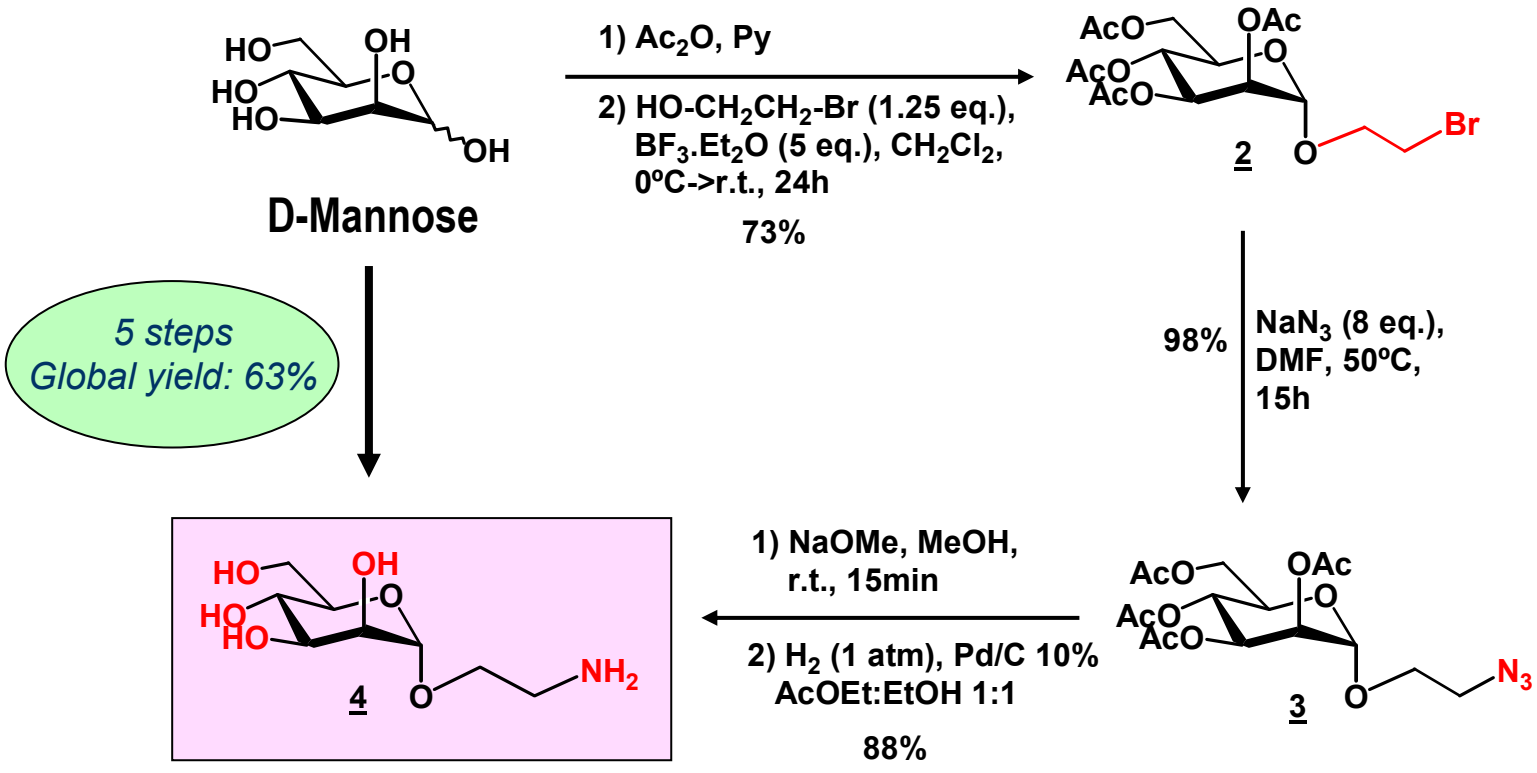


Biantenary

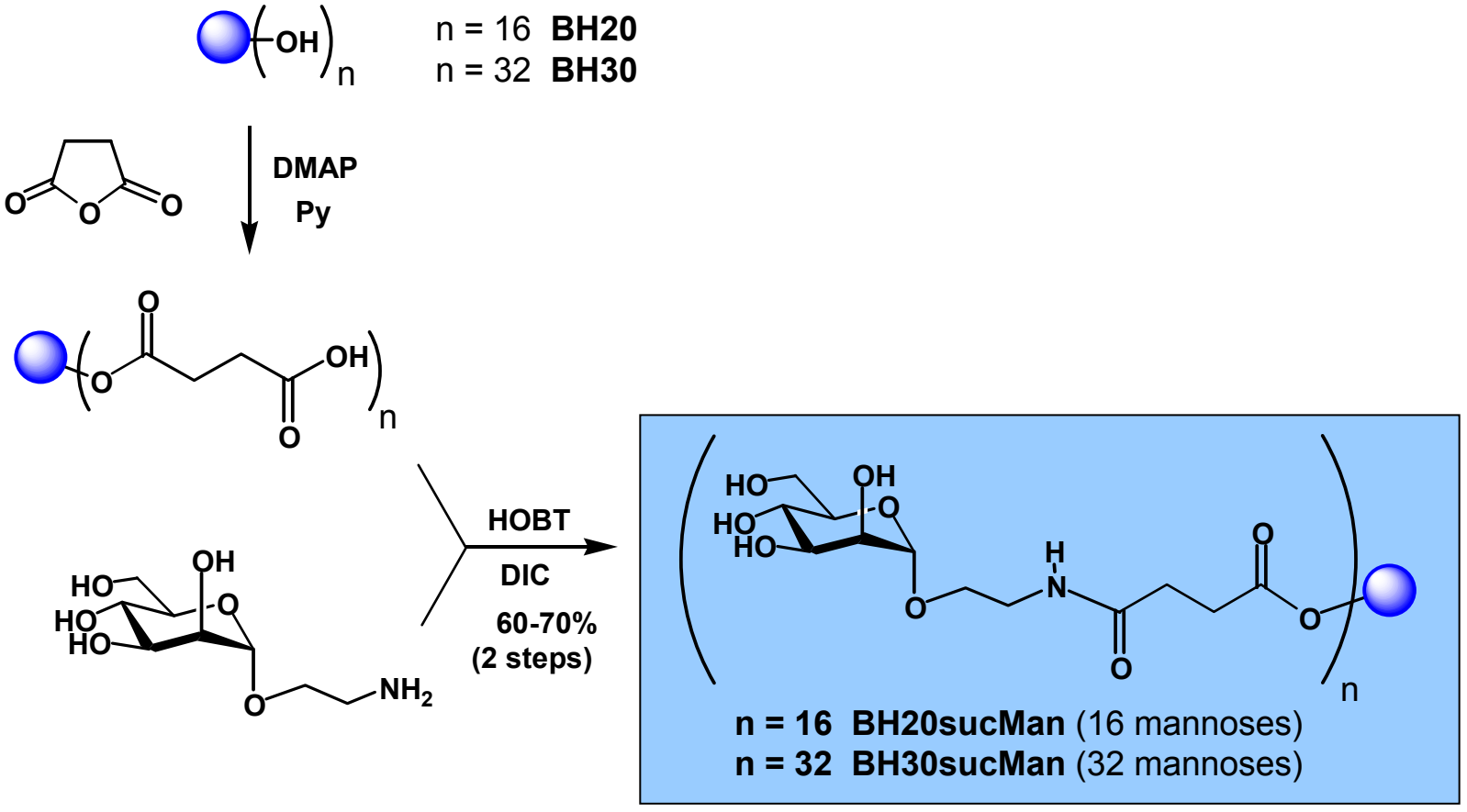


= Man

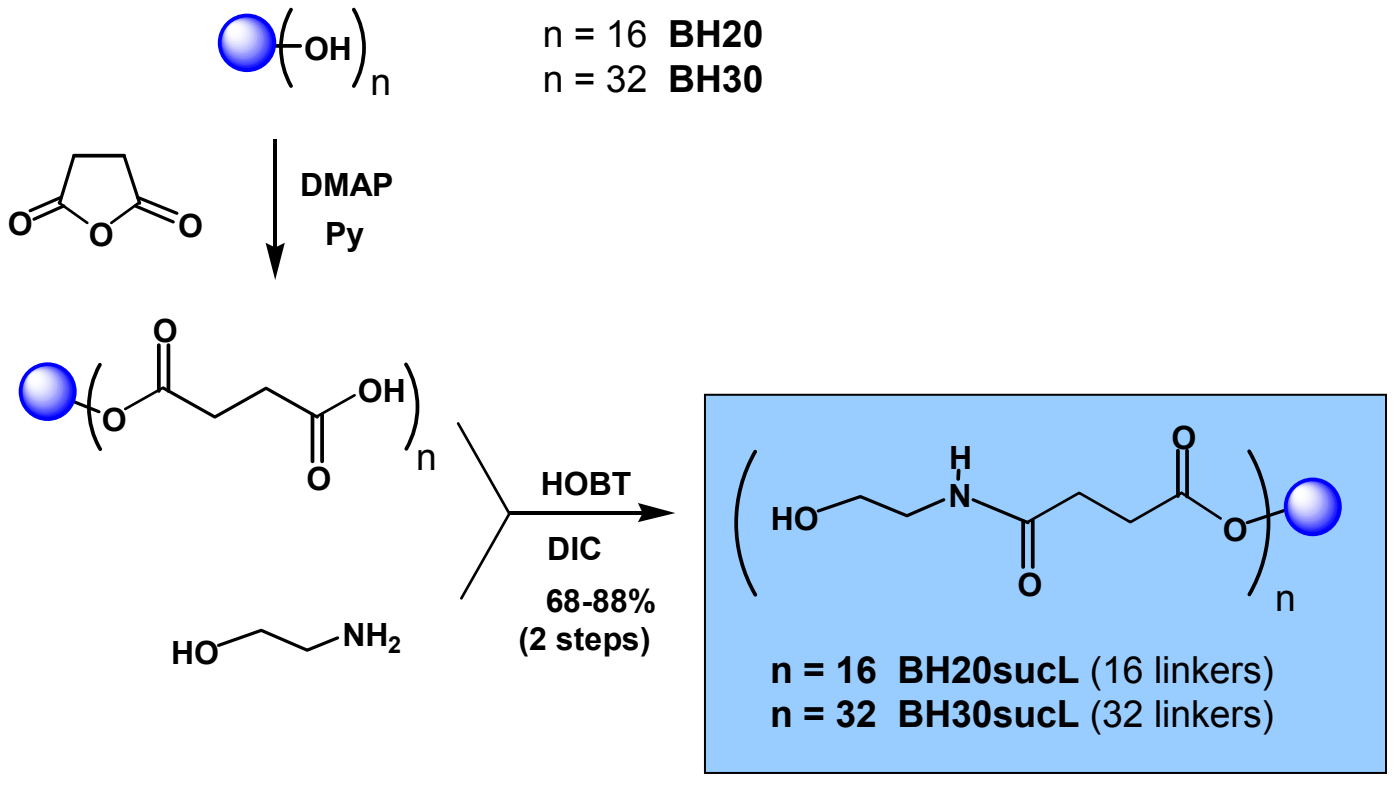
Mannose derivative synthesis



Synthesis of glycodendritic polymers



Synthesis of dendritic controls



Dendritic structures

- **High Solubility in physiological conditions**
- **Toxicity (MTT assays)**
- **Multivalent binding properties (*Lens Culinaris* model)**

Bioconjugate Chem., **2003**, *14*, 817-823.
J. Antimicrob. Chemother., **2004**, *54*, 579-581

EBOLA Virus

- Most lethal pathogen (60-80%)
- Biosafety level 4 containment
- Cytotoxicity?
- Animal reservoir?
- Receptor?
- Absence of effective drugs

Studies of Antiviral Activity

Dr. R. Delgado,
Hospital 12 de Octubre, Madrid

CSIC

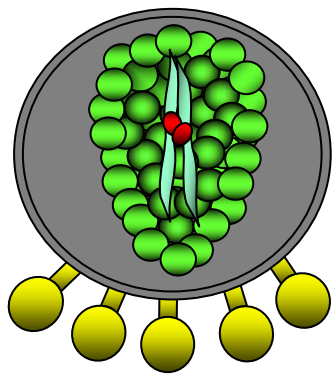
1. Molecular Level : *Inhibition of Interaction DC-SIGN and GP-1*

2. Functional Level : *Inhibition of infection*

Pseudotype viral particles (Artificial Virus)

Modified HIV gene machinery :

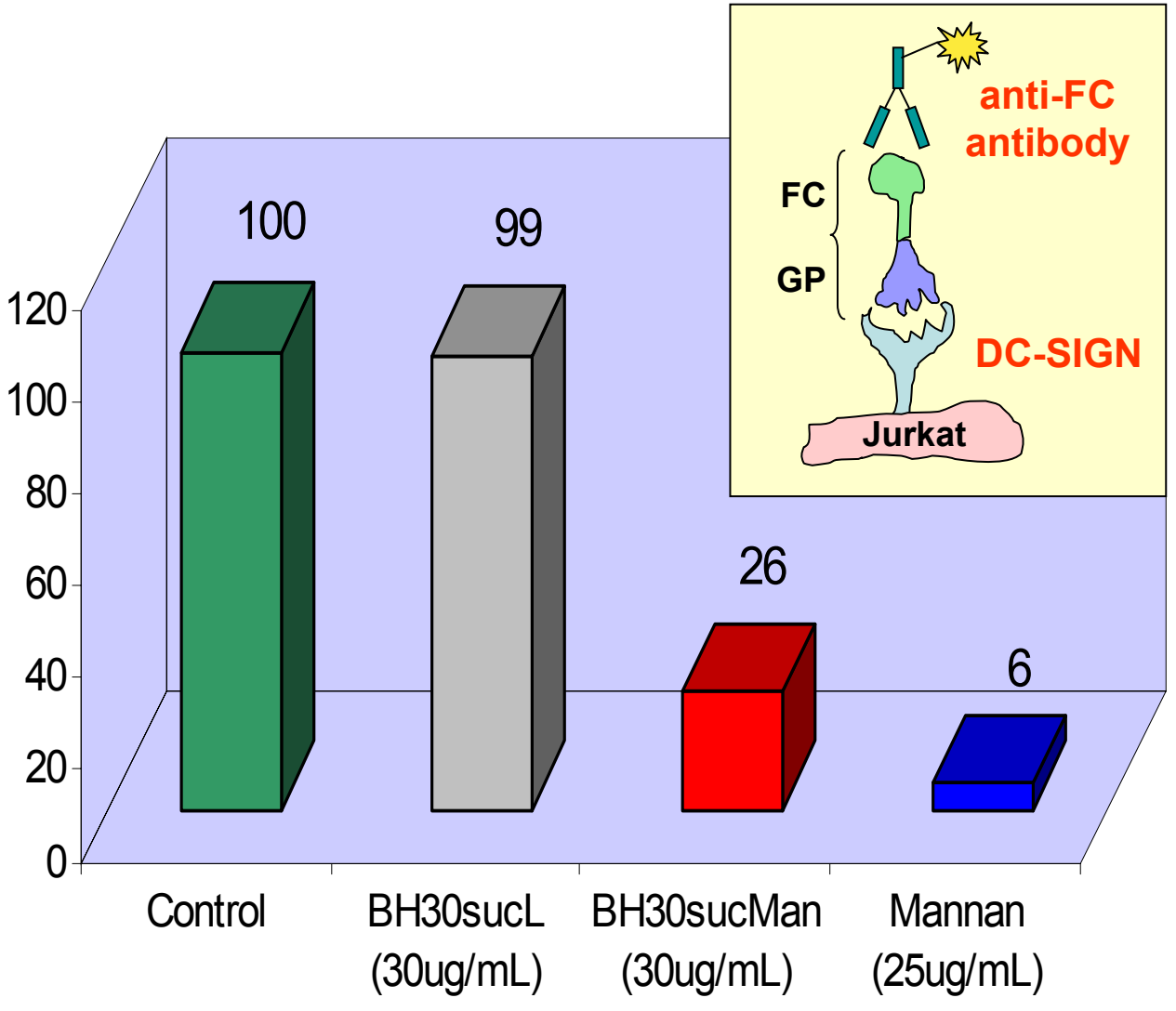
- **Protein GP-1 of Ebola**
- **Luciferase**
- **Transcription**
- **Non replicant**



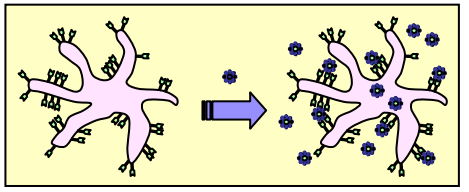
Artificial Virus

J. Virol., **2002**, *76*, 6841-6844.

Inhibition of Interaction GP/DC-SIGN



Inhibition of infection in cis



Infectious agent:

Pseudotyped particles with Ebola Zaire GP-1(EBO-Z)

infection (DC-SIGN dependent) of Jurkat cells

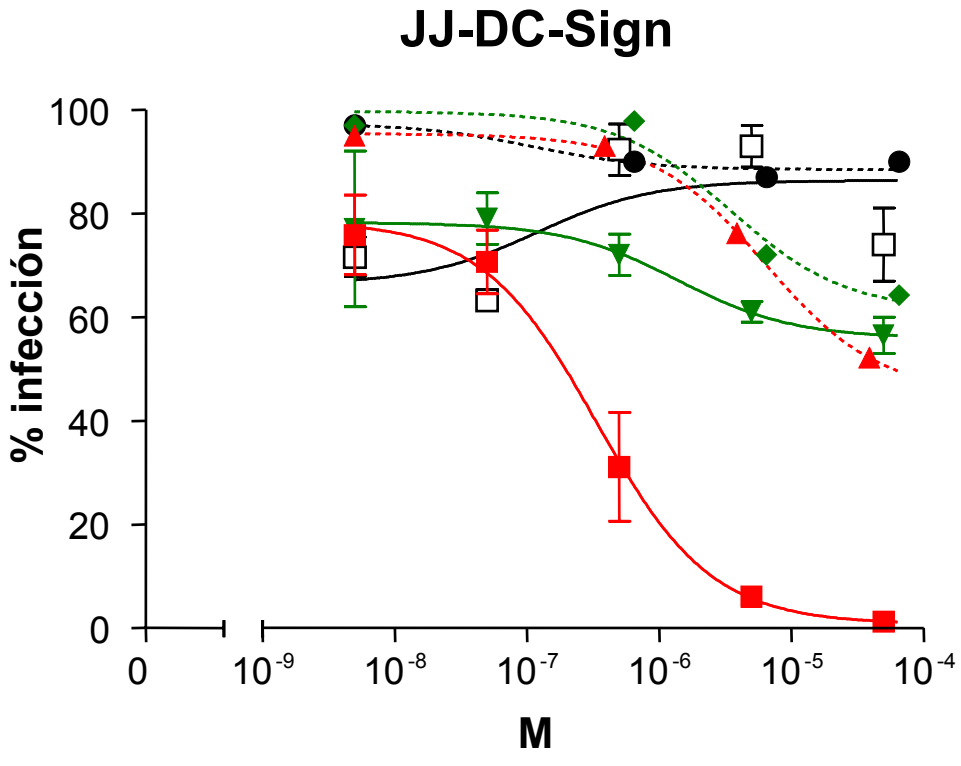
Target Cells:

Jurkat cells expressing DC-SIGN

Control: Pseudotyped particles with vesicular estomatitis virus protein (VSV)

Jurkat cell infection DC-SIGN non dependent

Inhibition of infection: results



Viral Particle + Inhibitor

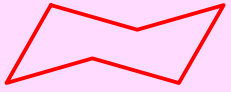
- **VSV + BH20sucMan**
- **VSV + BH30sucMan**
- **EBO-Z + BH20sucL**
- **EBO-Z + BH30sucL**
- **EBO-Z + BH20sucMan**
- **EBO-Z + BH30sucMan**

Antimicrob. Agents Chemother., **2003**, *47*, 3970-3972.

Multivalent effect

ManOMe

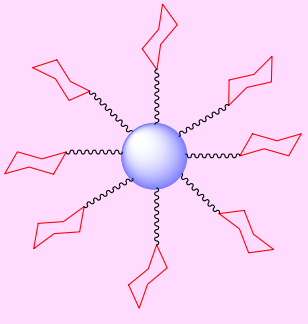
(1 mannose)



EC_{50} = 1mM

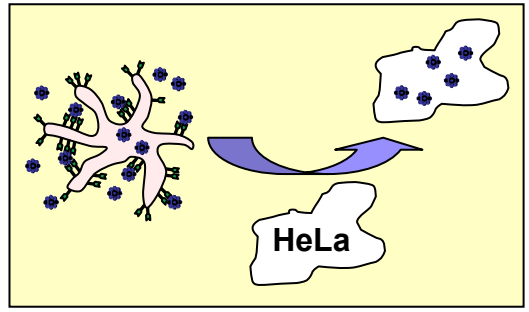
BH30sucMan

(32 mannoses)



EC_{50} = 0.3 μM

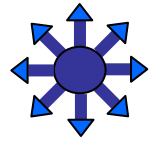
Inhibition of infection in trans



CSIC

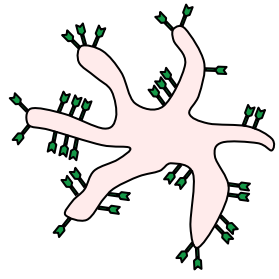
Infectious agent:

Pseudotyped particles with Ebola Zaire GP-1(EBO-Z)



Intermediate cells:

K562 cells expressing DC-SIGN



Target cells:

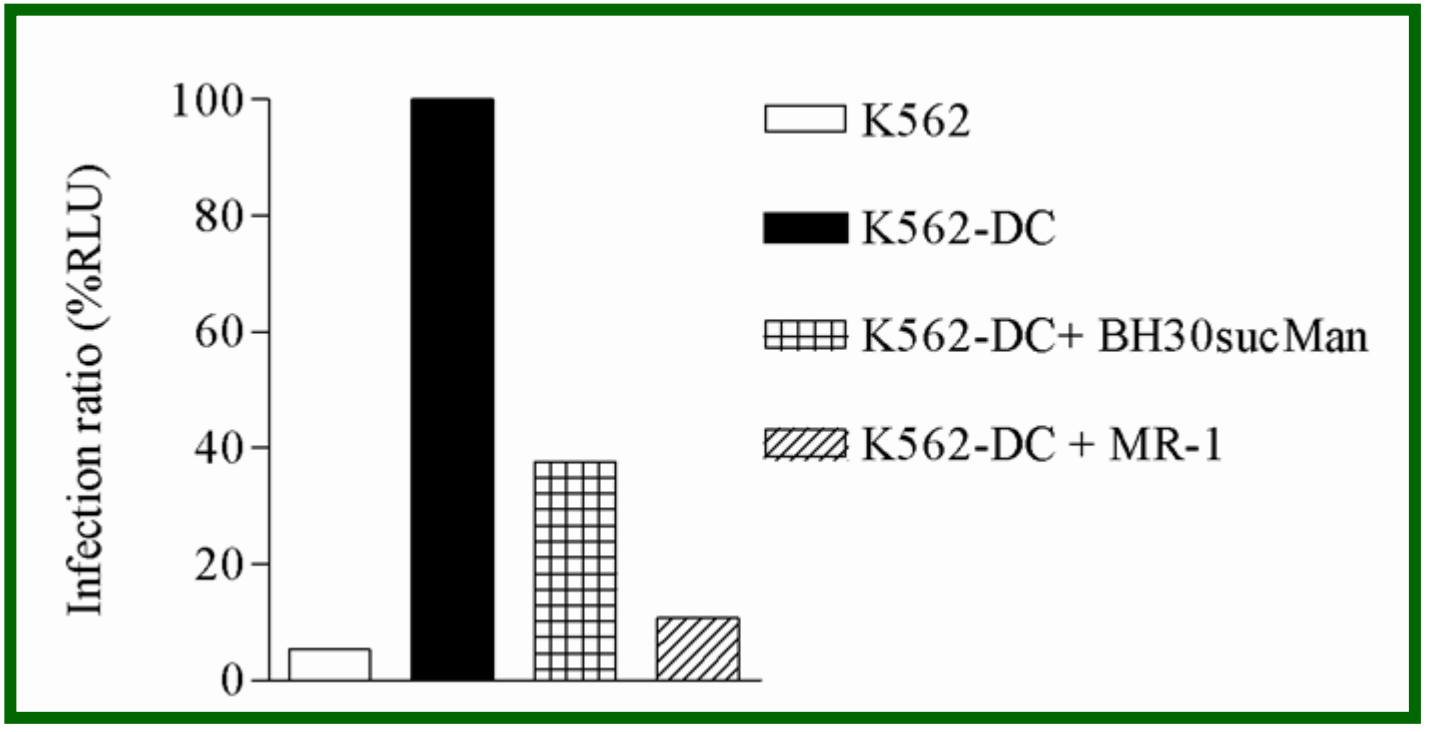
HeLa cells

Controls:

K562 cells without DC-SIGN

MR-1 anti-DC-SIGN antibody

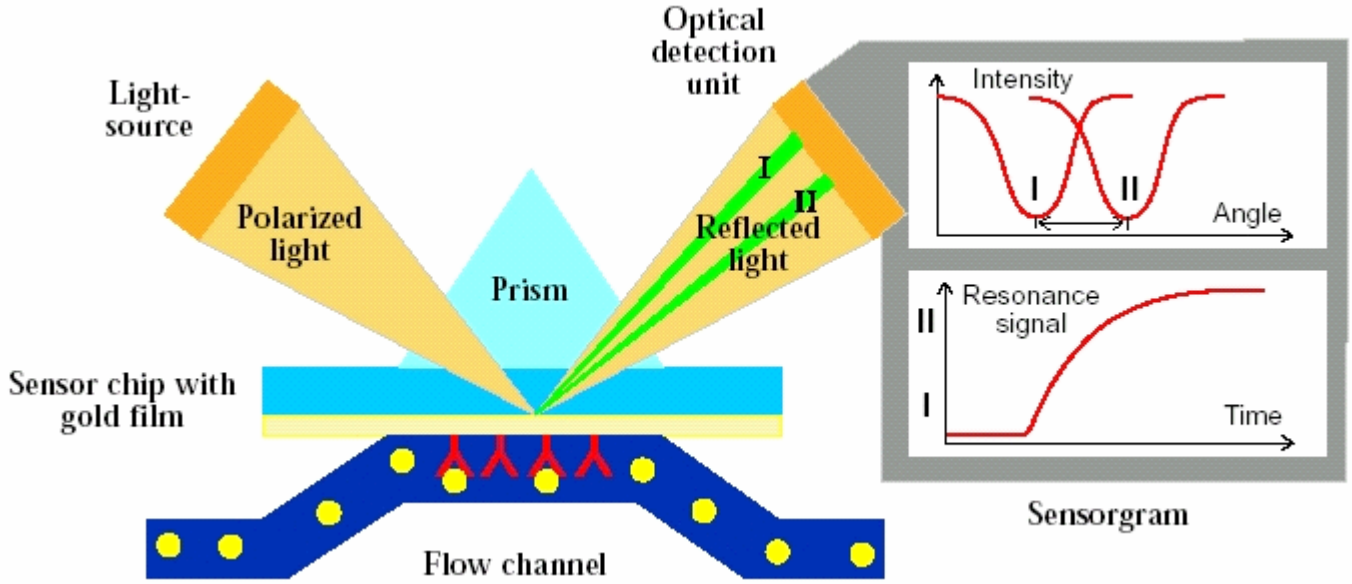
Inhibition of infection in trans

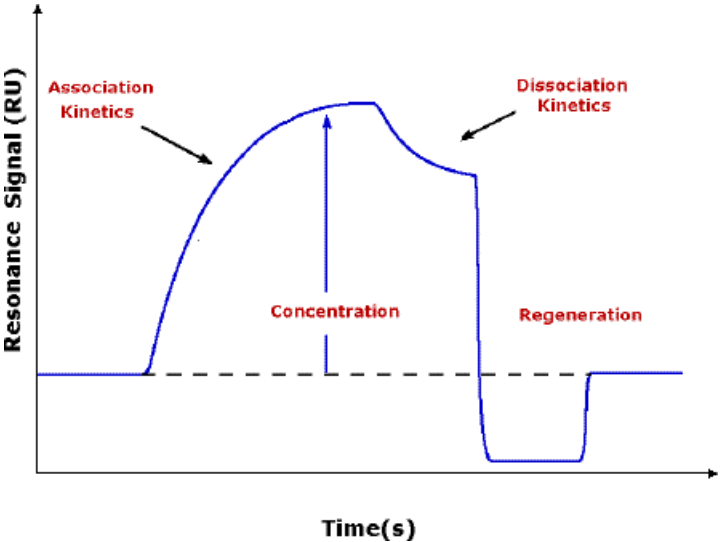


Infection on HeLa Cells

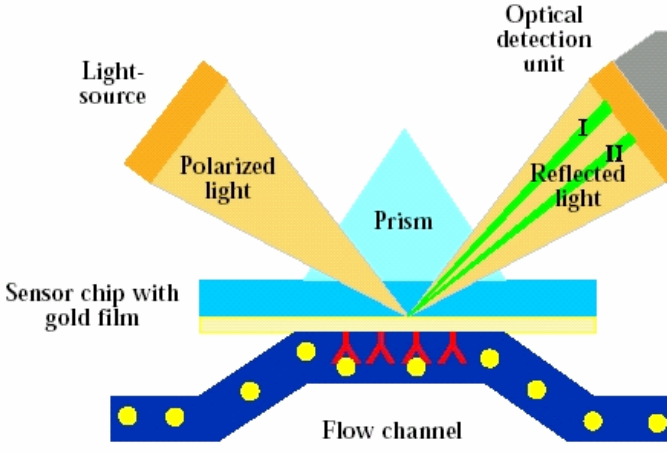
Biosensors (SPR)

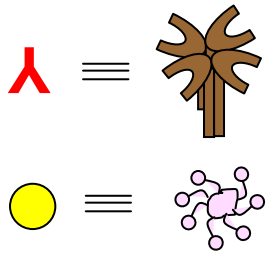
Dr. F. Fieschi, IBS, Grenoble



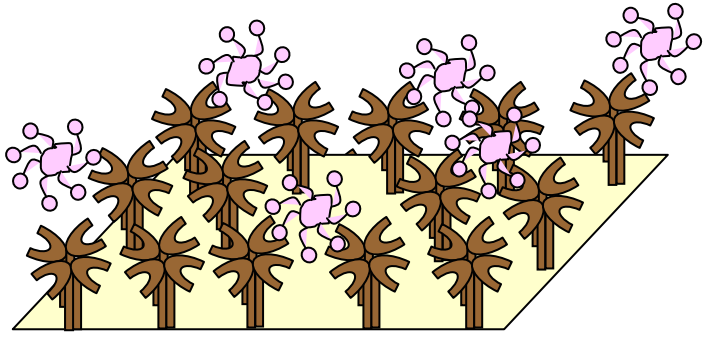


ECD functionalized surface





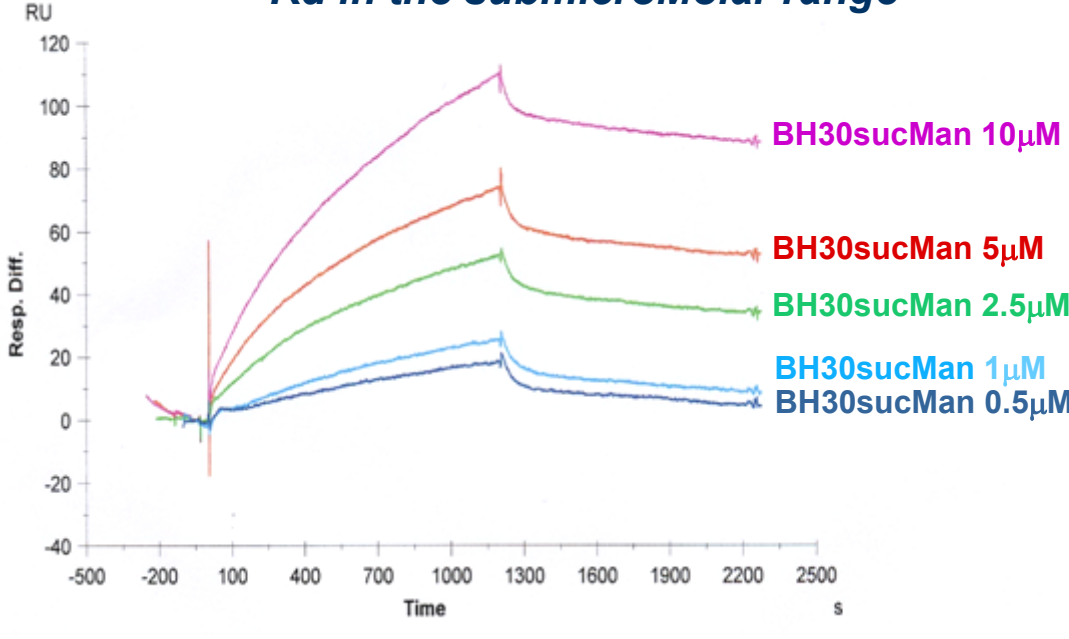
(BH30sucMan)



ECD High density

FEBS Lett., ***2006***, *580*, *2402*

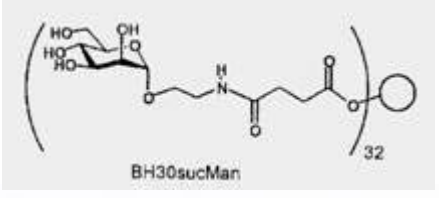
Kd in the submicroMolar range

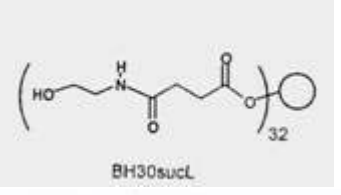


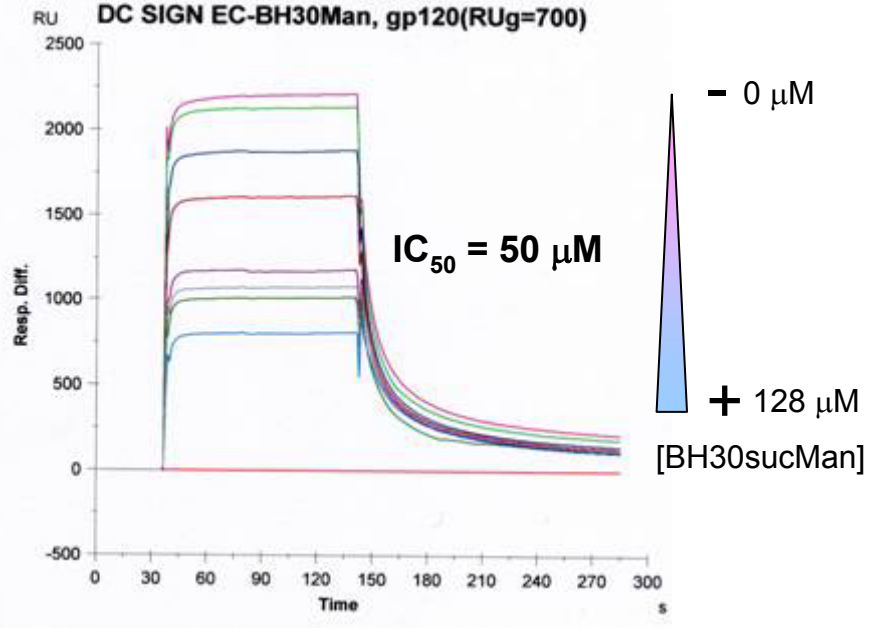
Inhibition of gp-120 binding

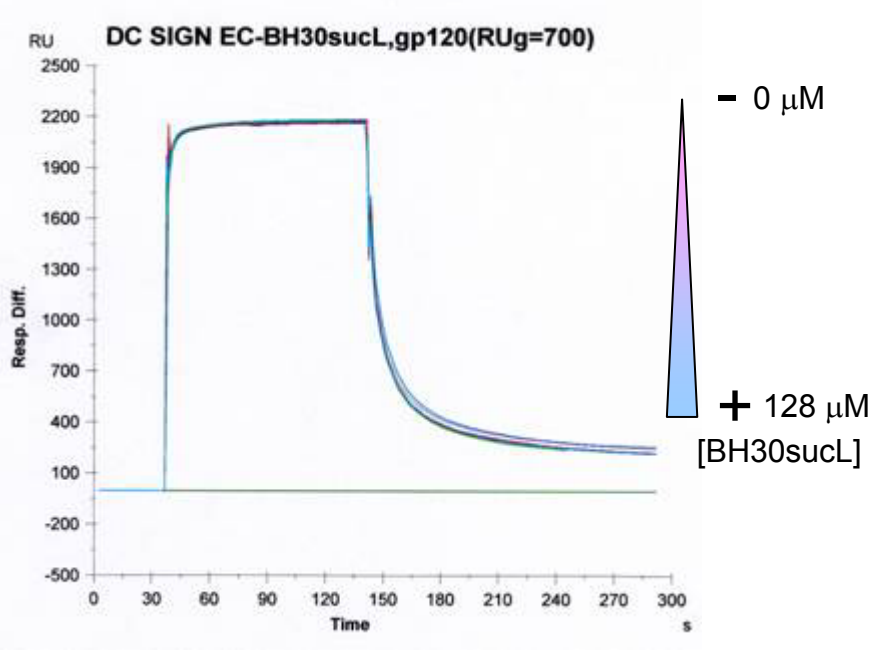
HIV gp120 is covalently linked on biacore surface (CM4)

DC-SIGN is injected alone or with increasing concentration of dendritic compounds

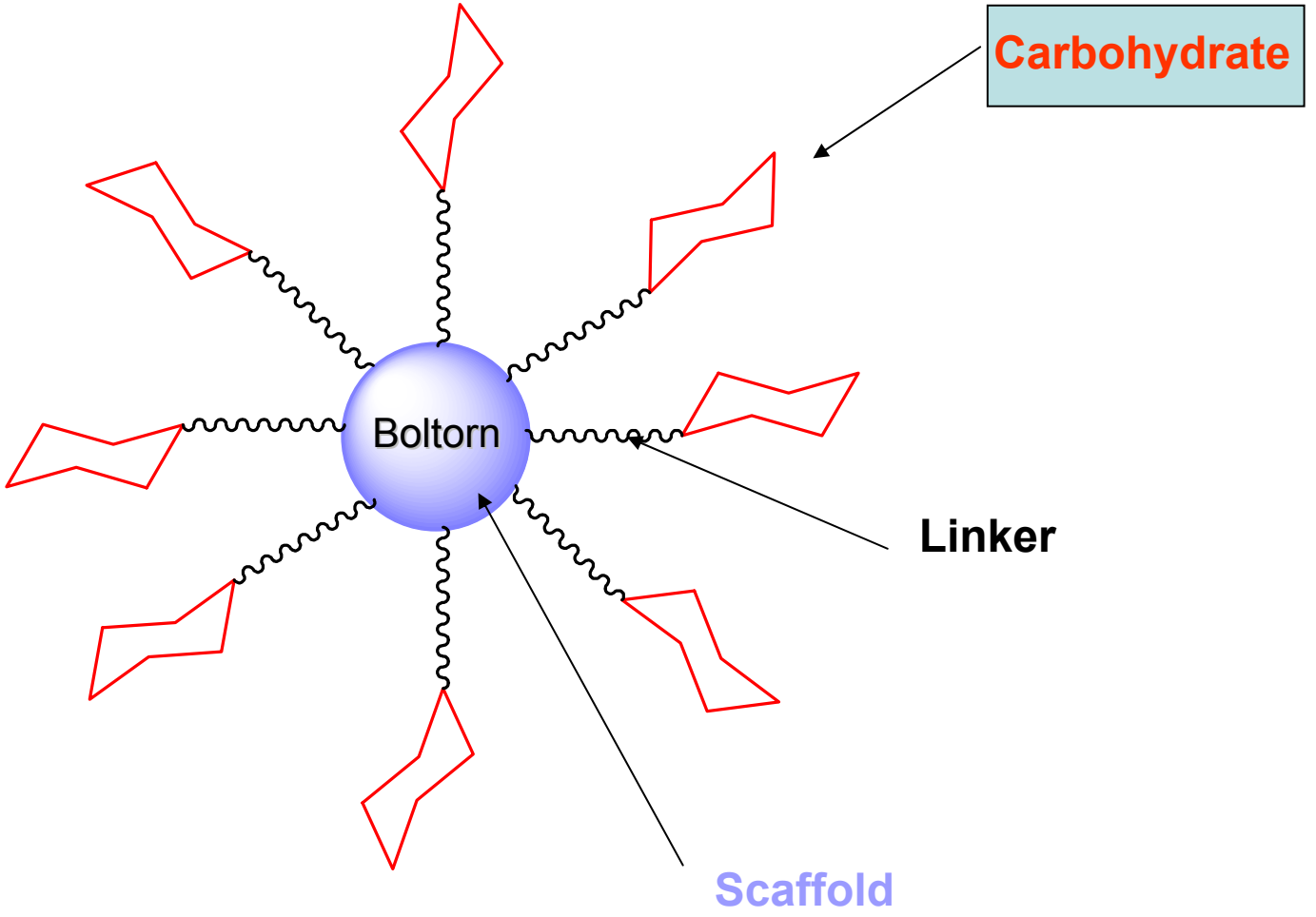




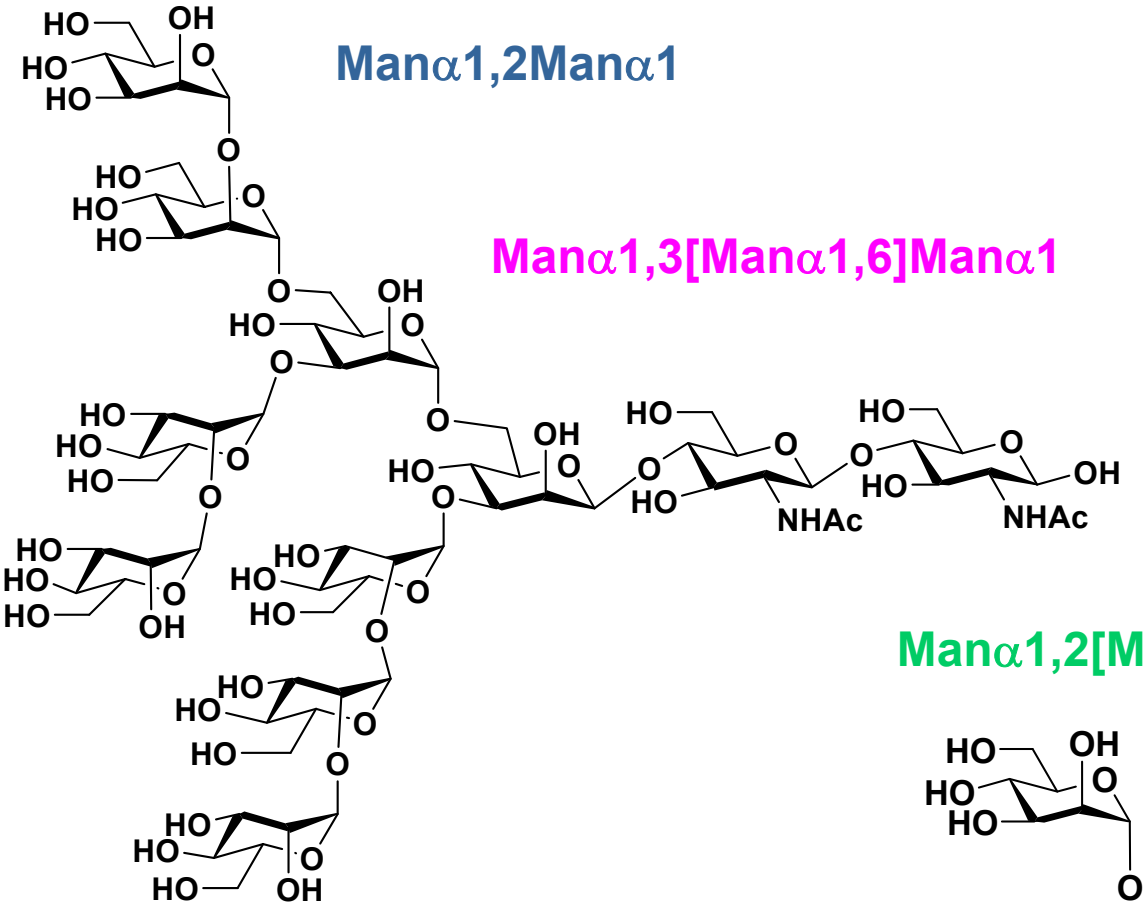




Structural modifications

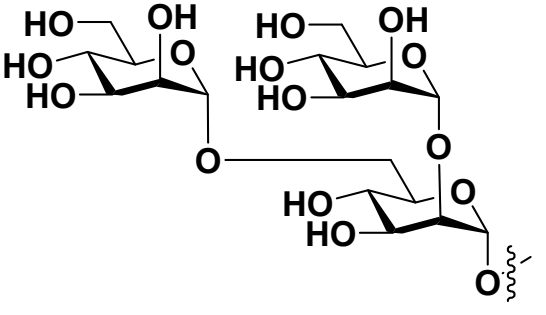


Mannose di- and trisaccharides



Manα1,2Manα1

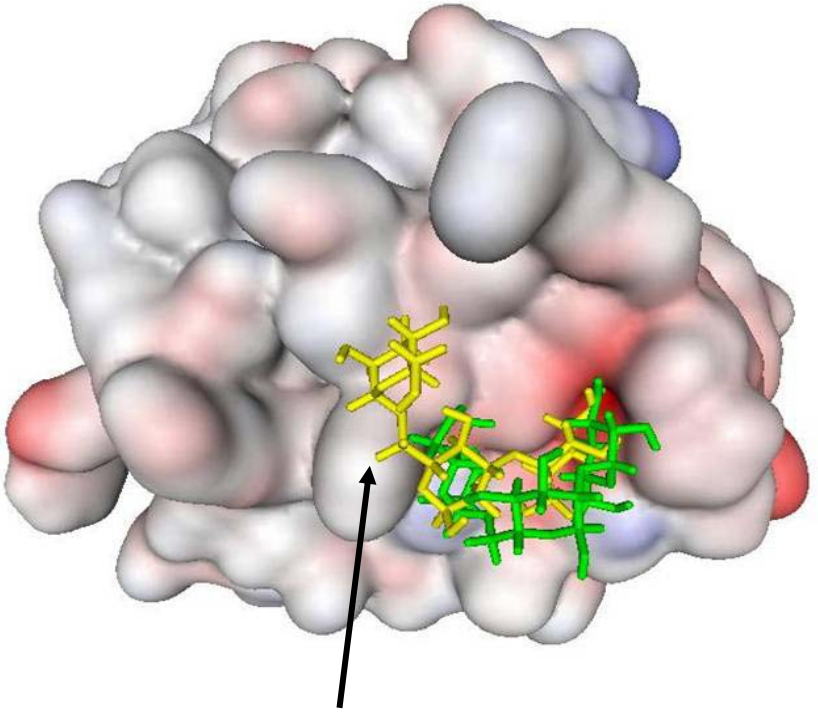
Manα1,3[Manα1,6]Manα1



Manα1,2[Manα1,6]Manα1

Docking Studies

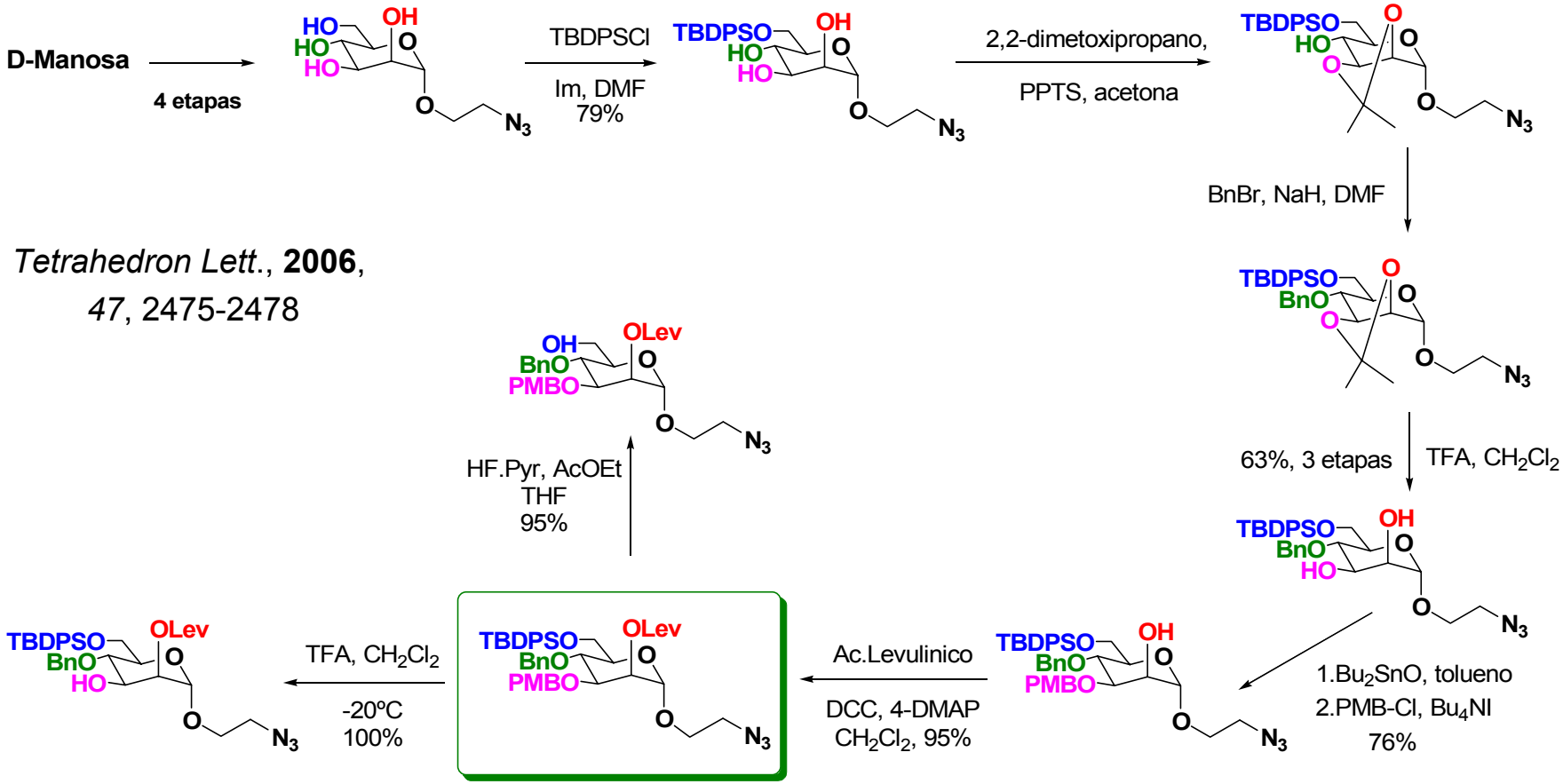
Dra. N. Campillo, IQM, Madrid



Manα1,2[Manα1,6]Man (green)

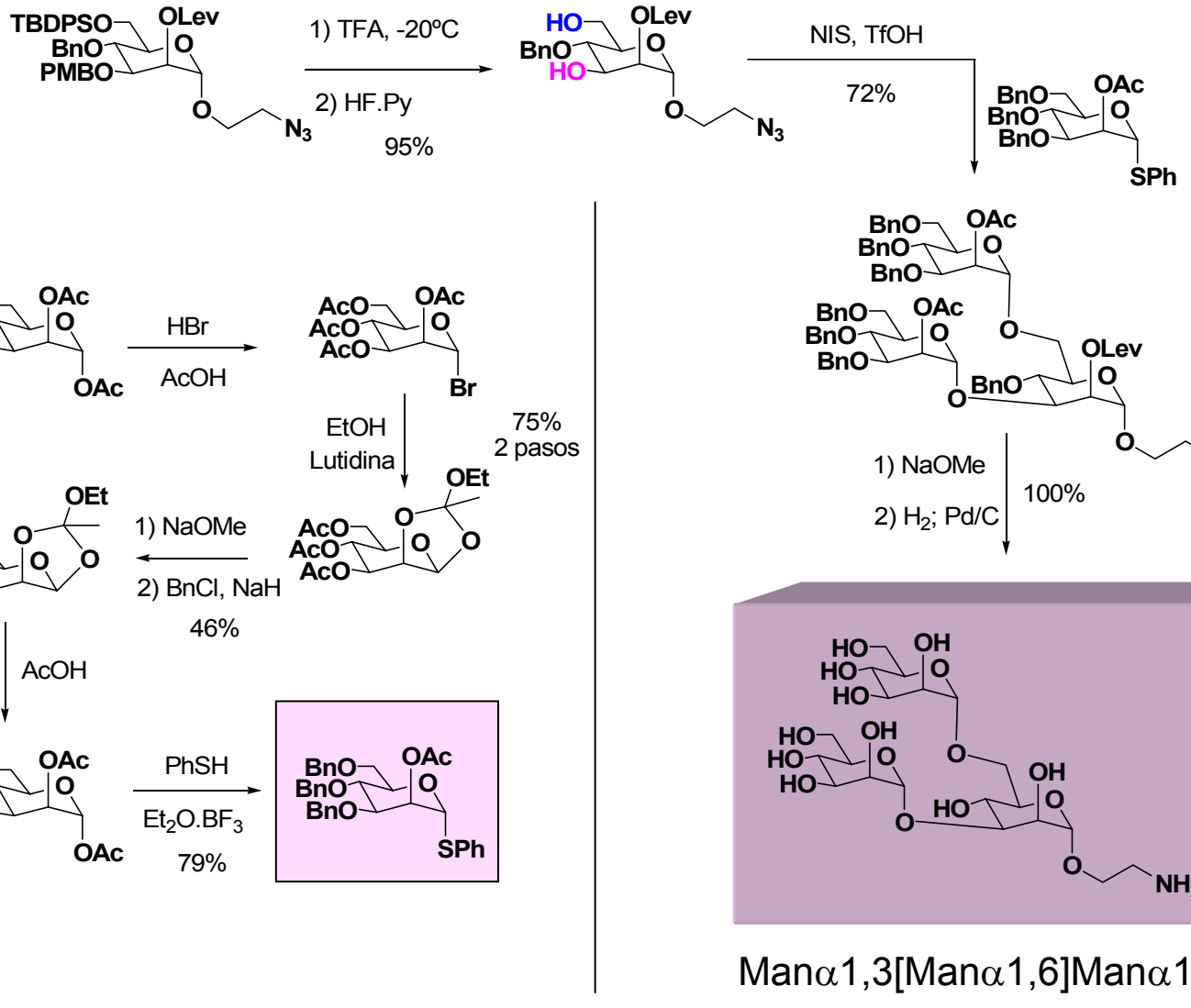
Manα1,3[Manα1,6]Man (yellow)

Orthogonally protected mannose



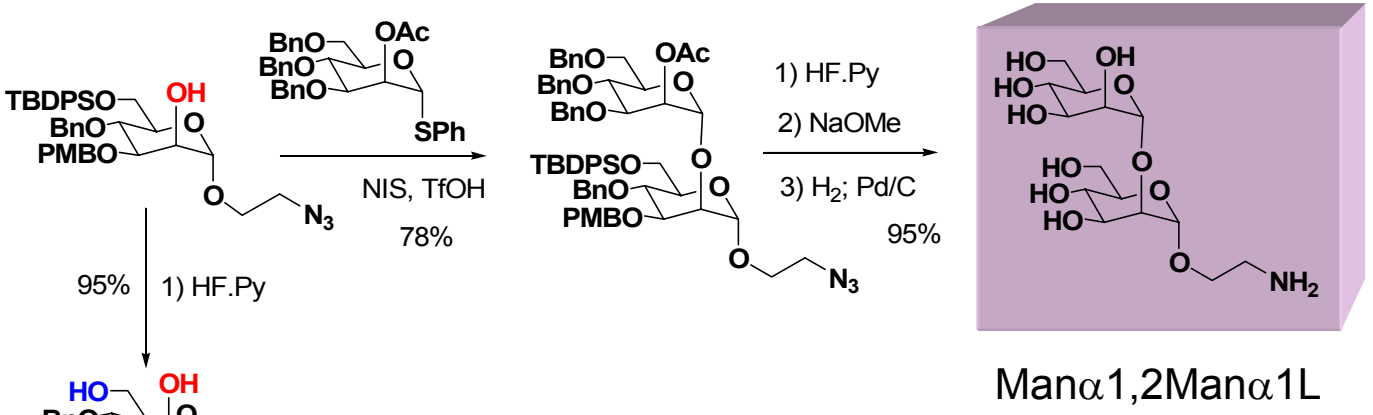
Tetrahedron Lett., **2006**, *47*, 2475-2478

Mannose oligosaccharides



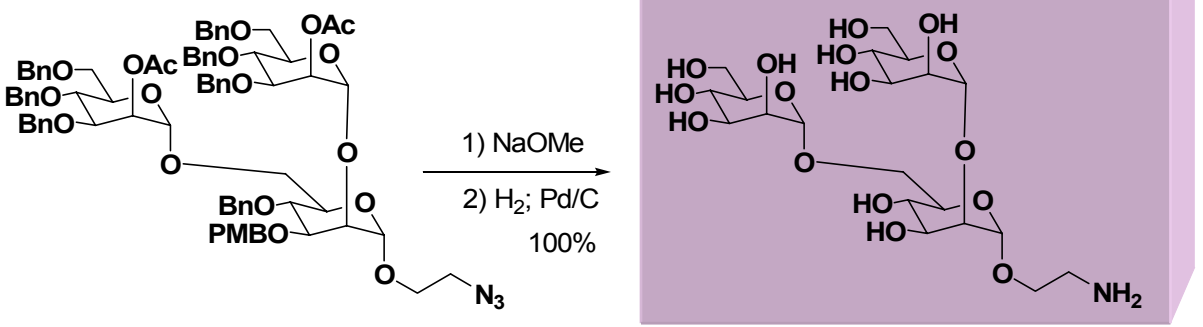
Manα1,3[Manα1,6]Manα1L

Mannose oligosaccharides II



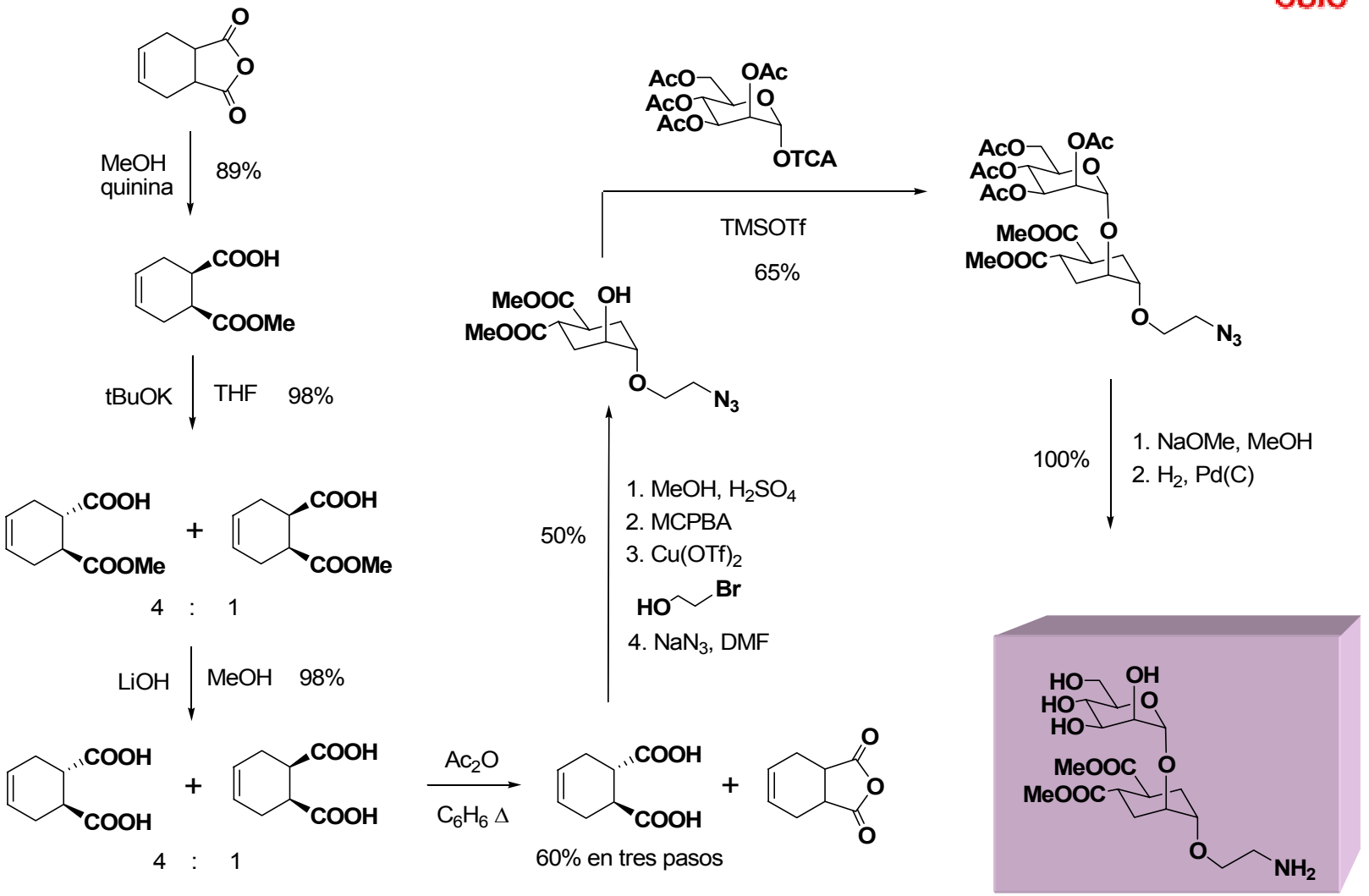
Manα1,2Manα1L

Manα1,2[Manα1,6]Manα1L



Mannose mimics

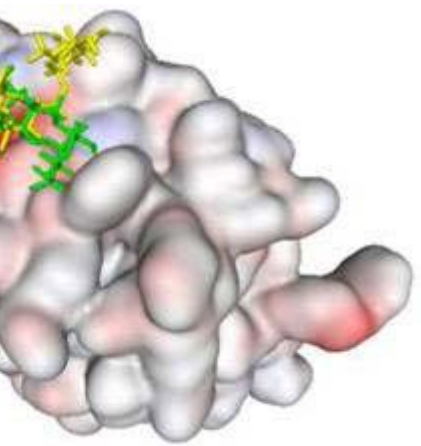
Prof. Anna Bernardi, University of Milan



Tetrahedron Asymm., **1999**, *10*, 3403-3407

Antiviral activity of monovalent sugars

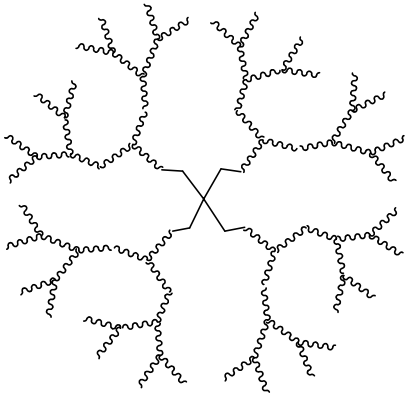
Qualitative activity using a model of Ebola cis infection



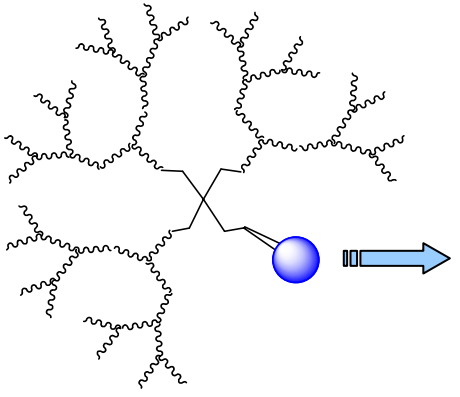
Perspectives

Applications in Nanomedicine

- **Selective Vectors in gene therapy**
- **New generation of Vaccines**
- **Selective drug delivery systems**



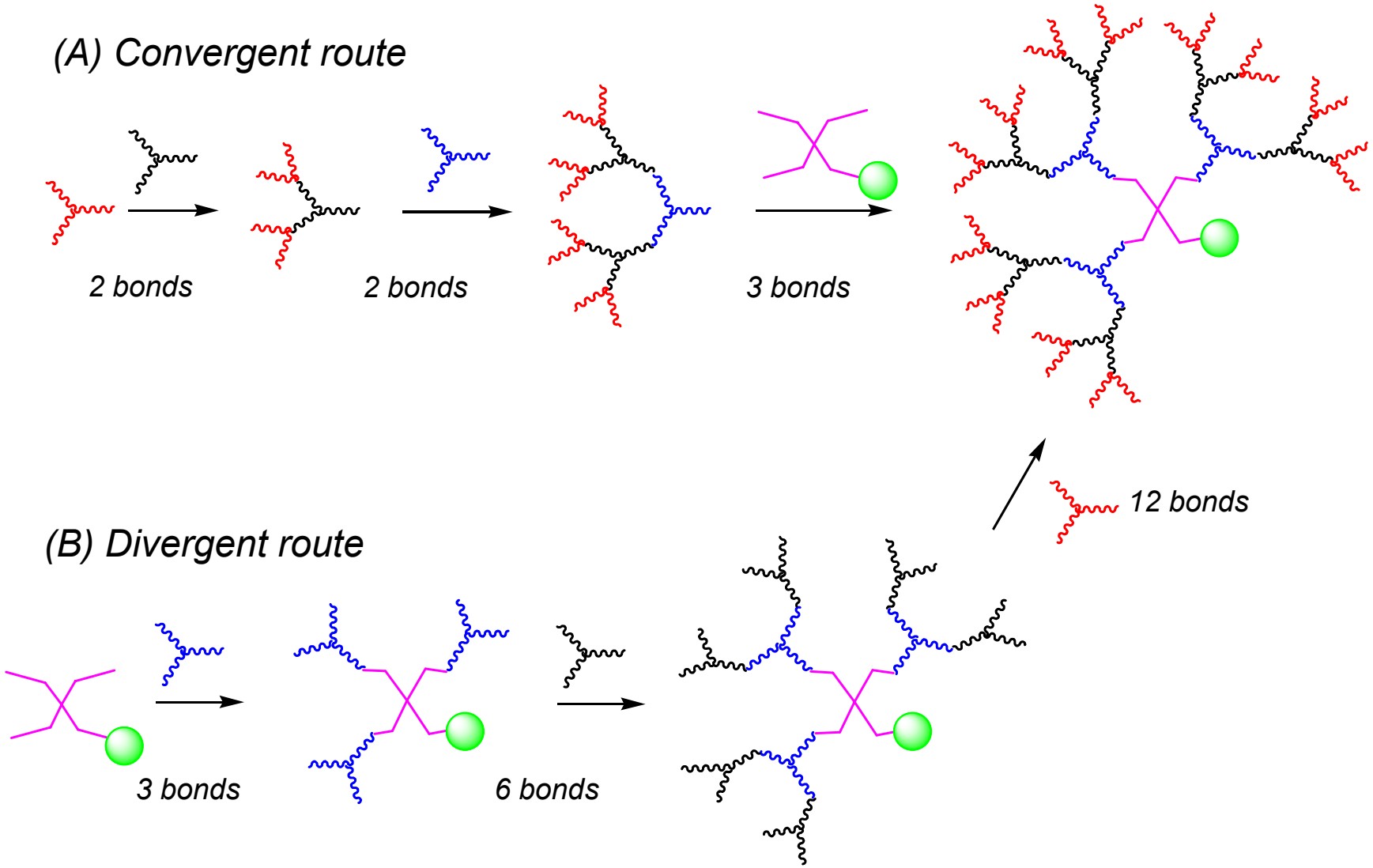
Multivalent system (Dendrimer)



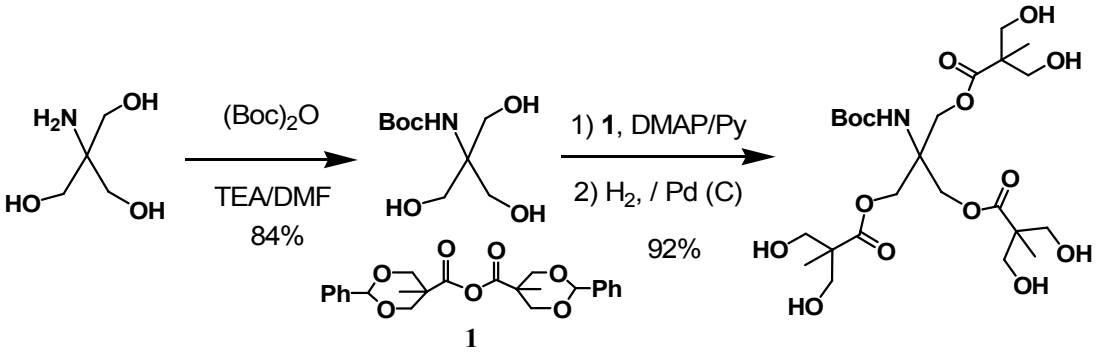
Fluorecense Tag
Biotine
Maleimide; peptides

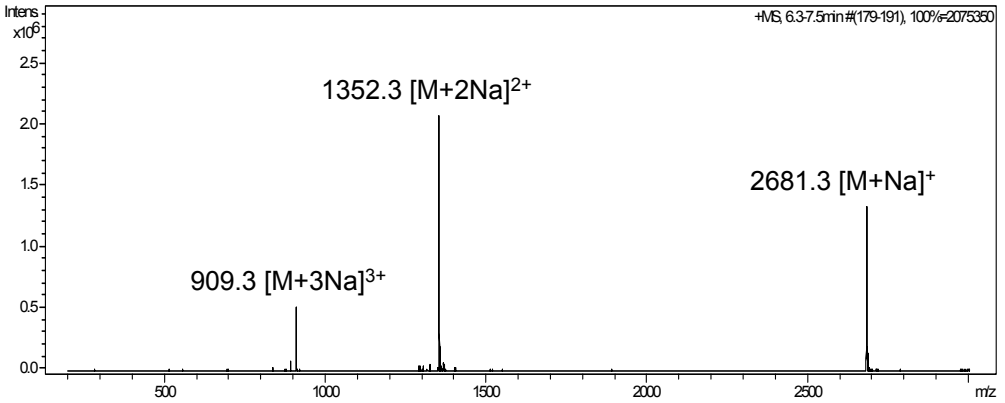
Multivalent system (Dendron)

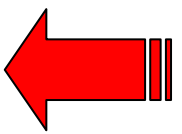
Synthetic route for 3G dendrons

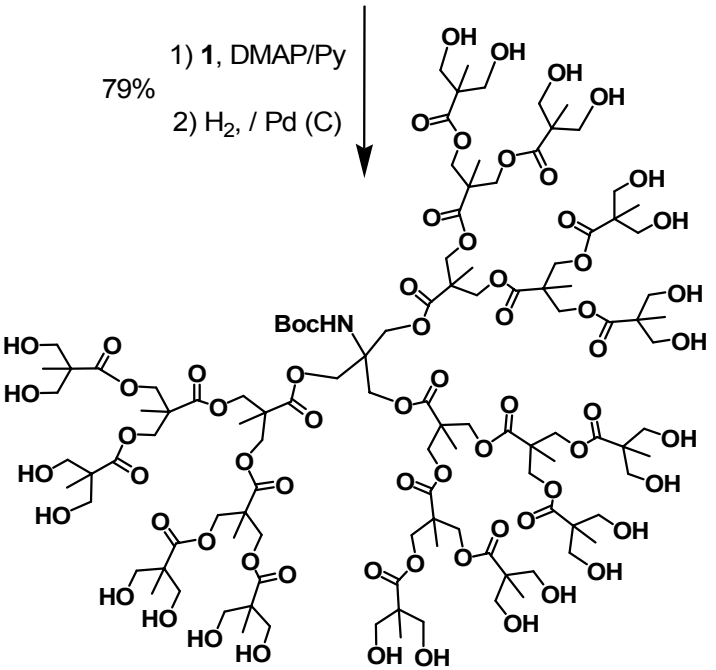


Divergent Route (B) for 3G dendron

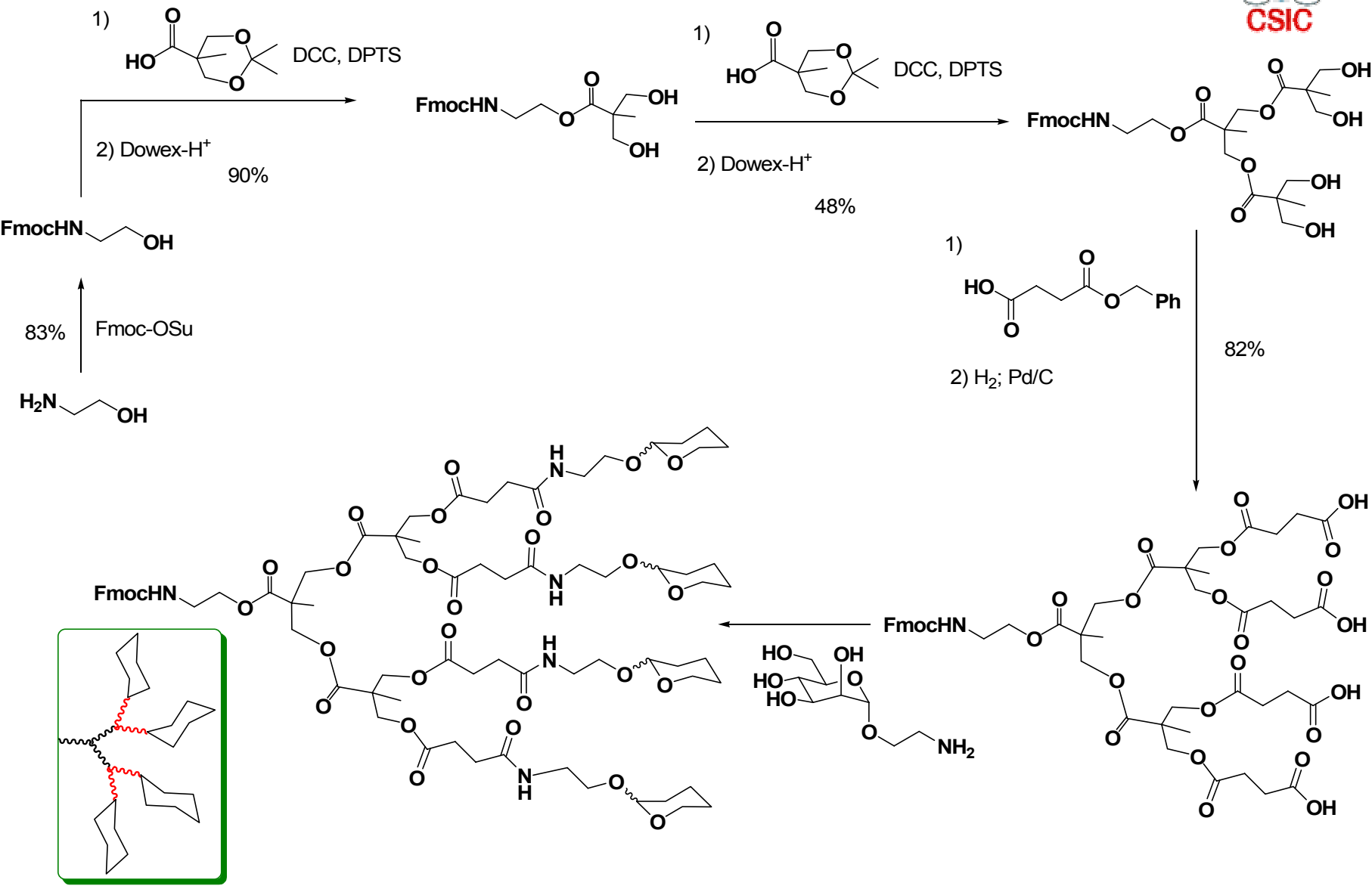




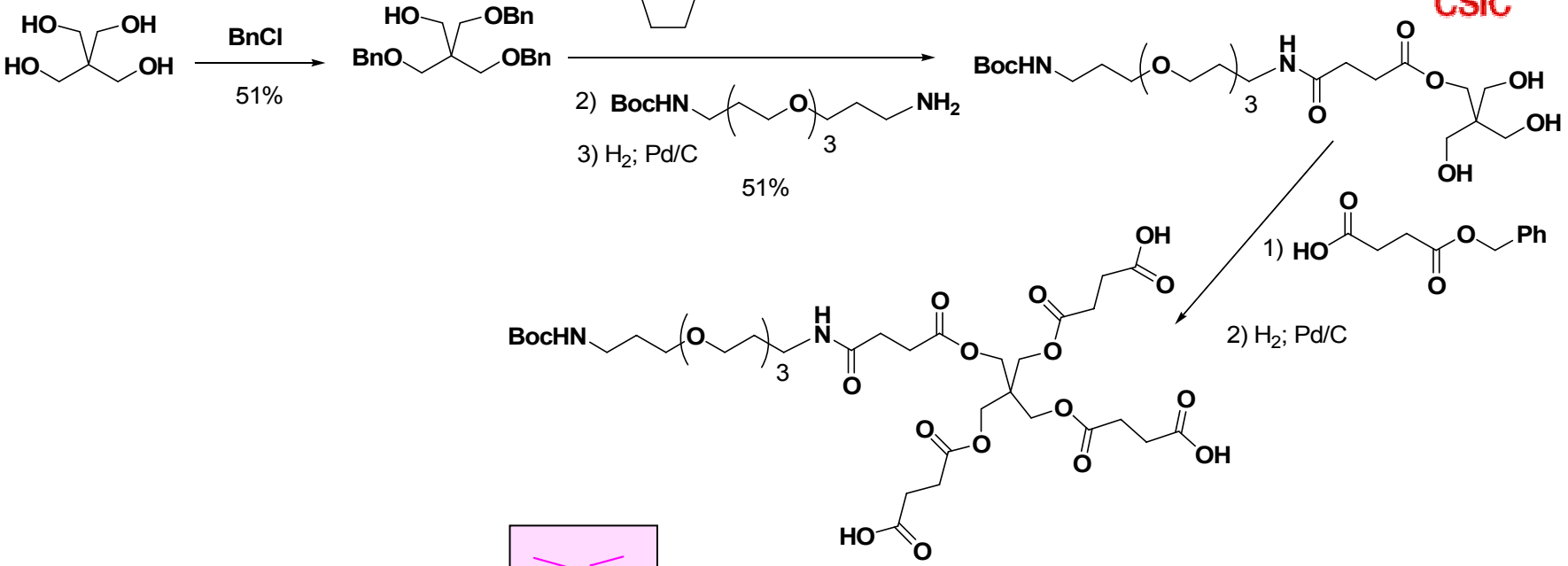


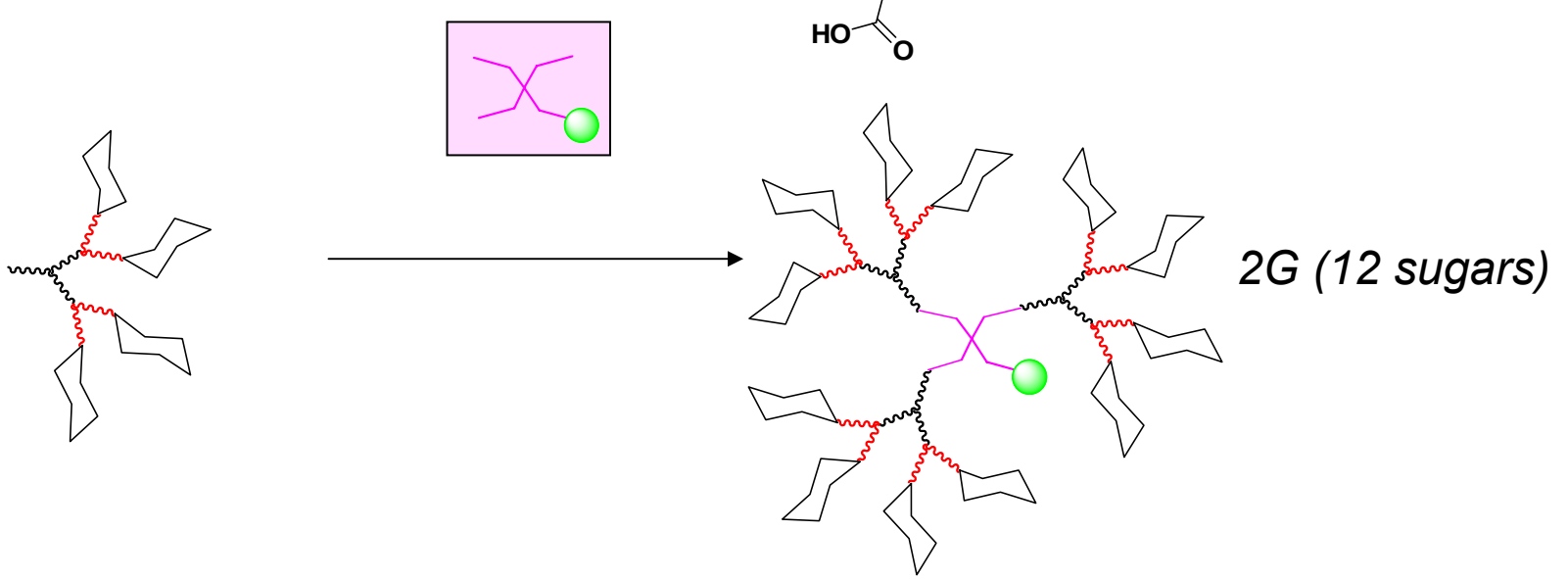


Convergent route (A)



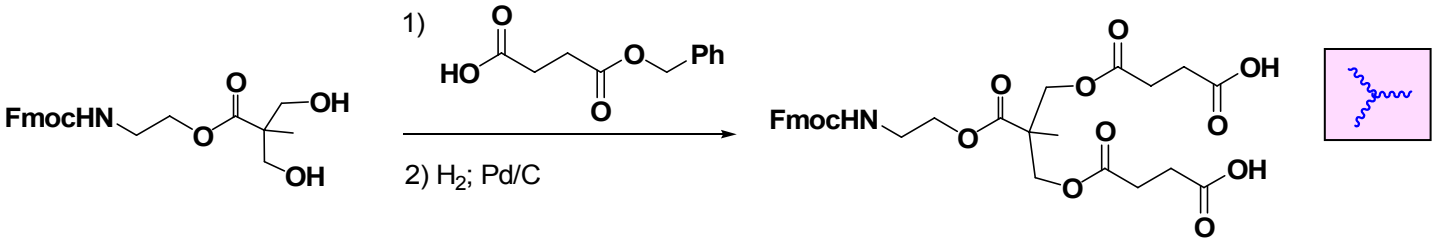
Convergent route (A) II



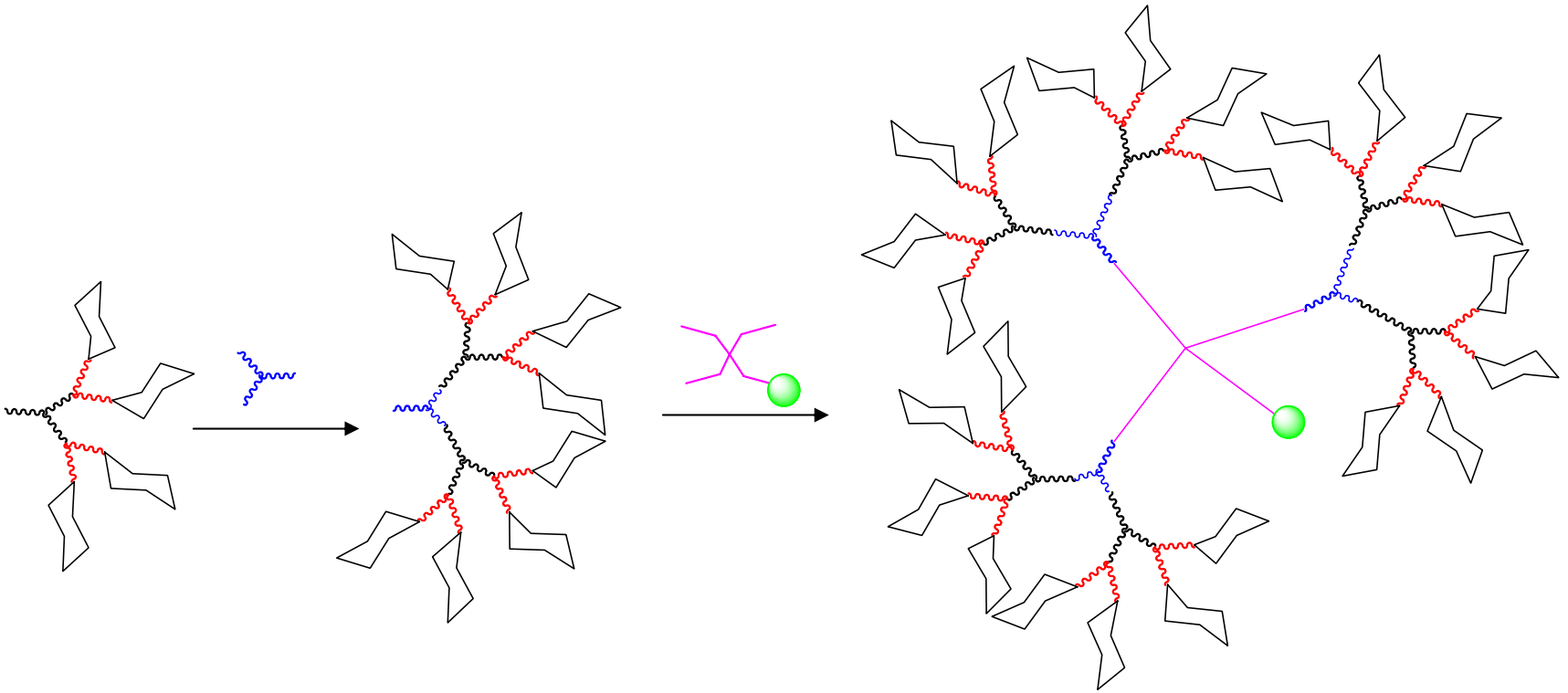


2G (12 sugars)

Dendron assembly



3G (24 sugars)



Acknowledgements

Grupo Carbohidratos, IIQ, CSIC, Sevilla

Dra. Eva Arce
José Juan Reina
Olivia Sánchez
Macarena Sánchez
Dr. Pedro M. Nieto

IQM, CSIC, Madrid

Dra. Nuria Campillo
Dr. Juan A. Páez

St Georges Hospital Medical School

Dr. Robin Shattock

Laboratorio de Microbiología Molecular Hospital 12 de Octubre, Madrid

Dr. Rafael Delgado
Fátima Lasala
Lorena Martínez

Milan University

Prof. Anna Bernardi
Sara Sattin

IBS, Grenoble

Dr. Franck Fieschi
Georges Tabarani

Funding

FIS; Programa Ramón y Cajal
MCyT; MEC; CSIC

The European Microbicides Project (EMPRO) EU FP6