cost
EUROPEAN COOPERATION IN SCIENCE & TECHNOLOGY

Funded by the European Union

COST ACTION CA 17140
NANO2CLINIC
CANCER NANOMEDICINE - FROM THE BENCH TO THE BEDSIDE

Carbosilane glycodendrimers for anticancer drug delivery

Designed for STSM 3rd Call

Final CA17140 Conference, October 25-26, 2022, Rome, Italy

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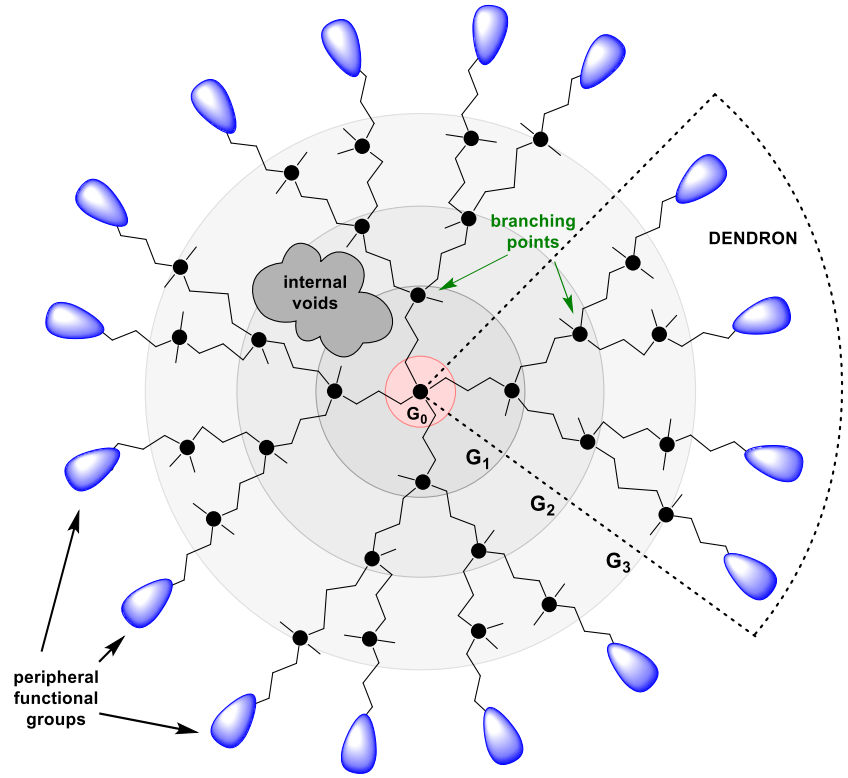
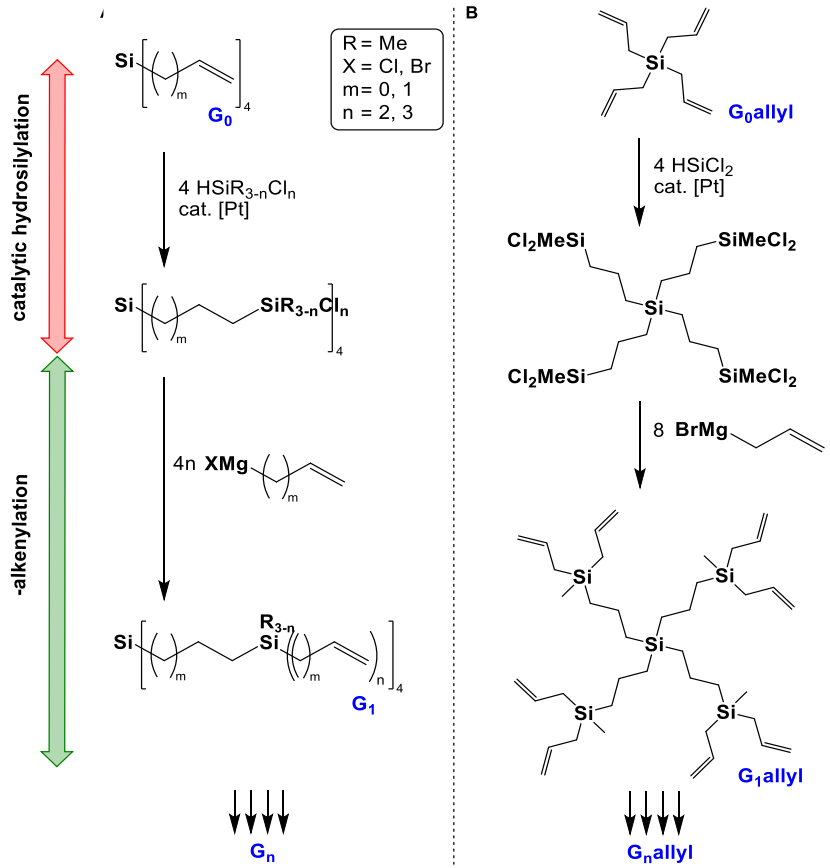
UNIVERSIDADE da MADEIRA
CQM CENTRO DE QUÍMICA DA MADEIRA

Functionalized Carbosilane Dendrimers

for bioapplications



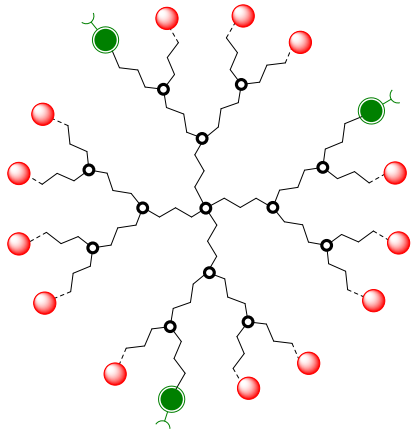


Macromolecules **1993,** *26* (5), 963-968
J. Chem. Soc. **1992,** (19), 1400-1401

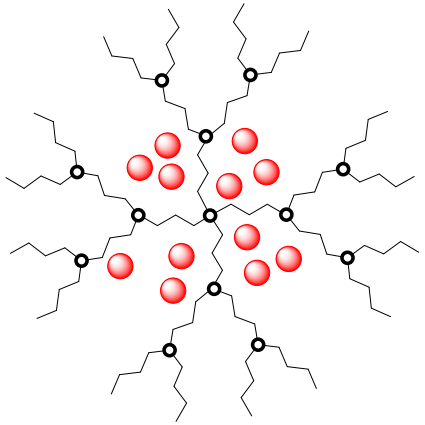
Functionalized Carbosilane Dendrimers

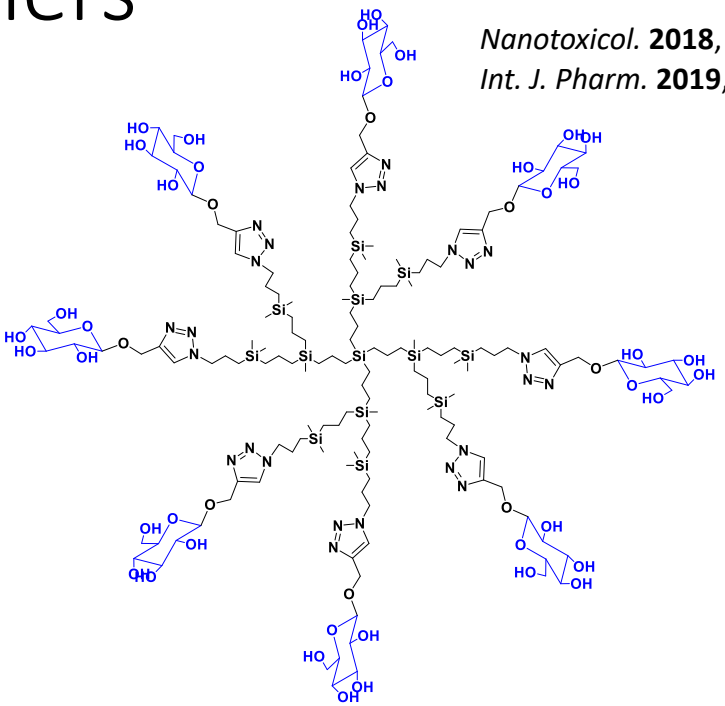
Vectors of therapeutic cargo

covalent conjugation



encapsulation



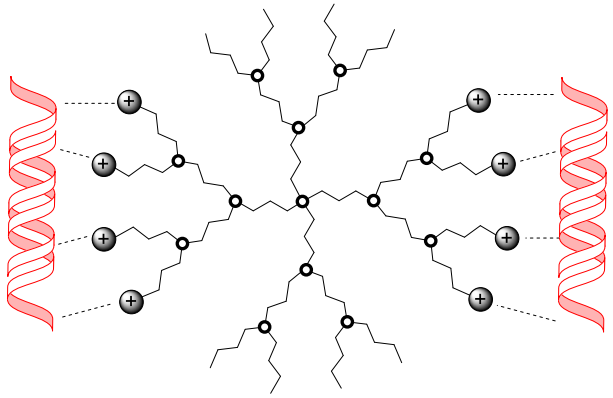


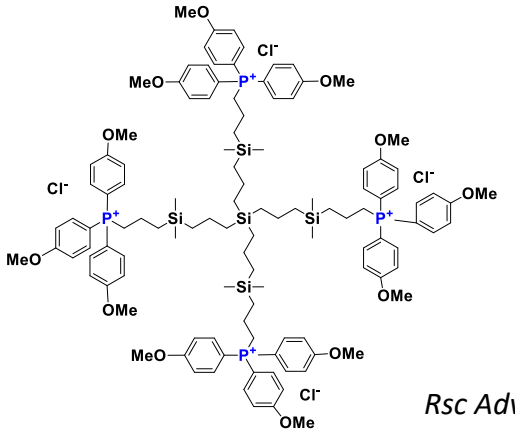
Nanotoxicol. **2018**, 12 (8), 797-818
Int. J. Pharm. **2019**, 579, 119138

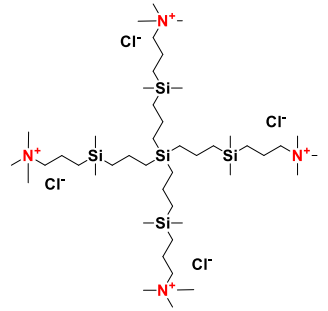
therapeutic compounds

targeting molecules, fluorescent labels

electrostatic complexation



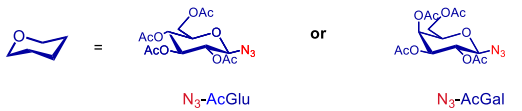


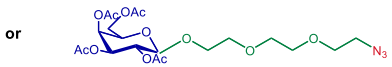


Rsc Advances **2017,** 7 (30), 18724-44
Int. J. Pharm. **2019,** 562, 51-65
*Phys.Chem.Chem.Phys.***2018**, 20(21),53-64

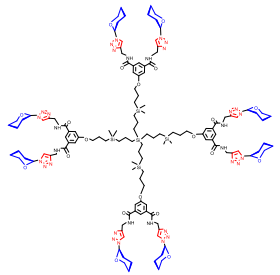
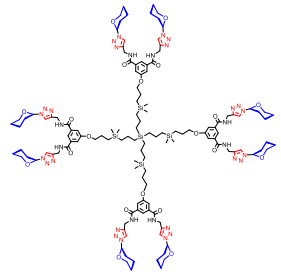
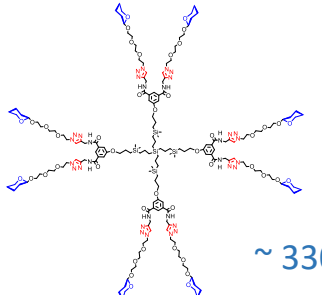
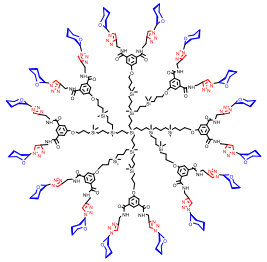
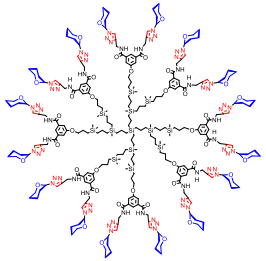
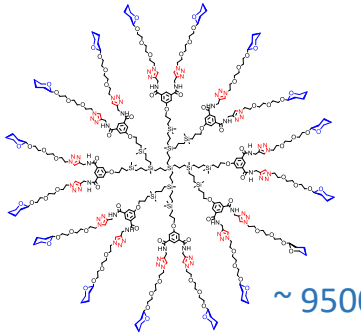
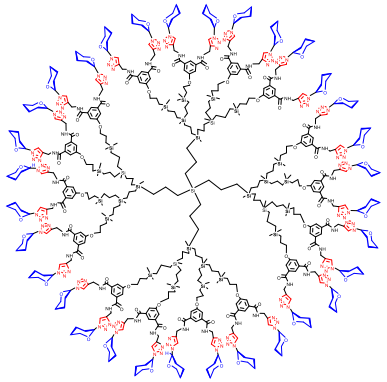
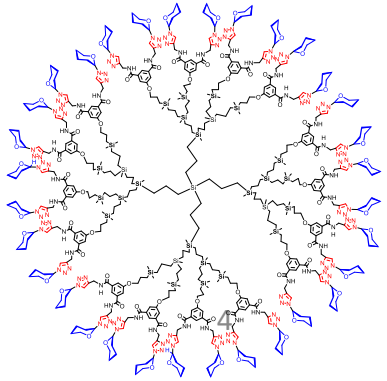
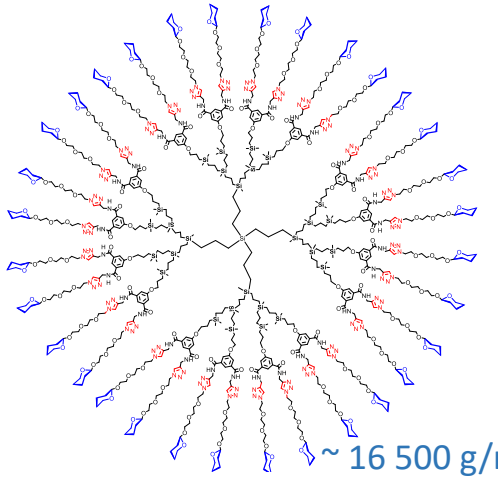
Carbosilane glycodendrimer library

- ✓ 3 generations
- ✓ 3 series differing in sugar moiety



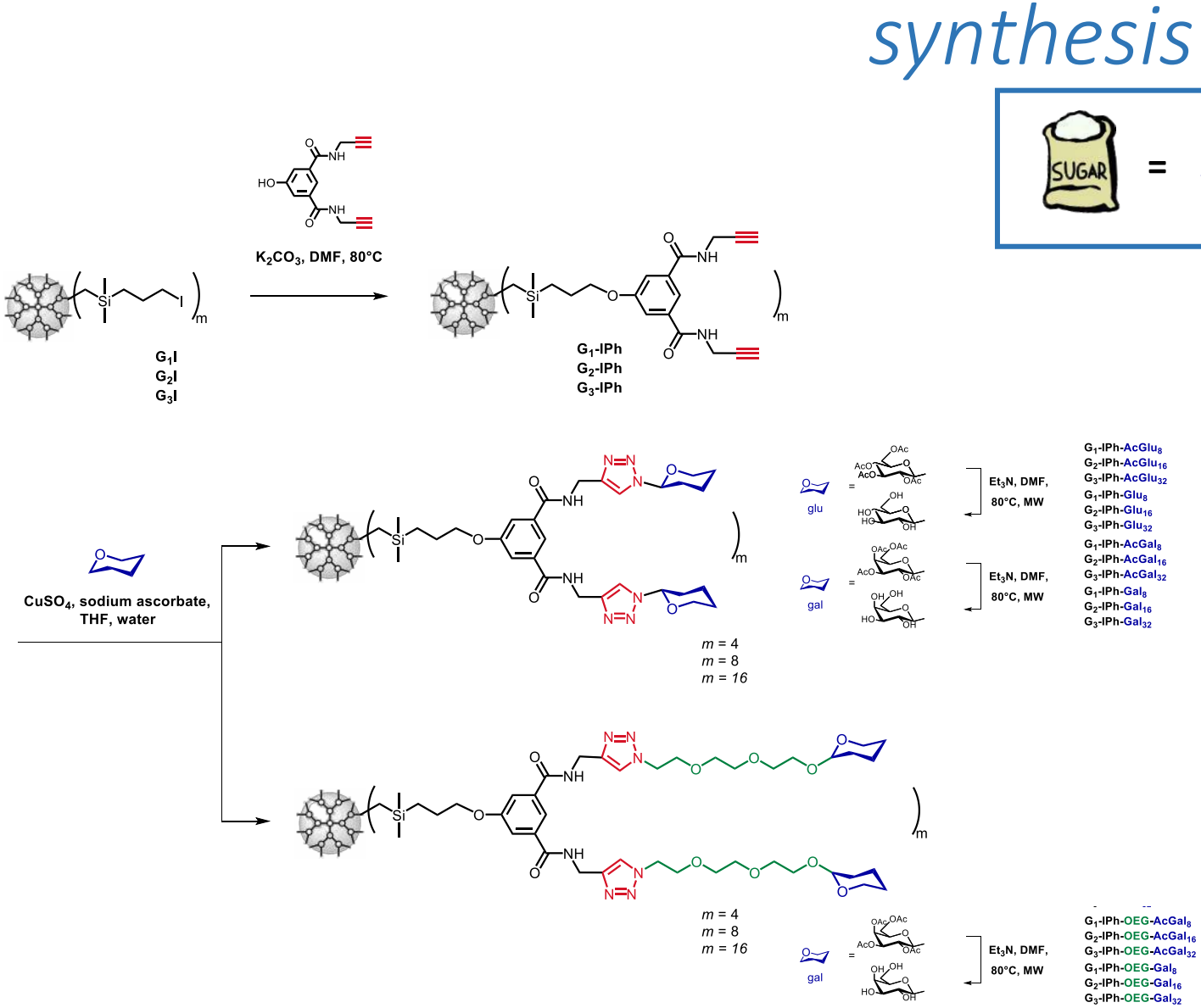


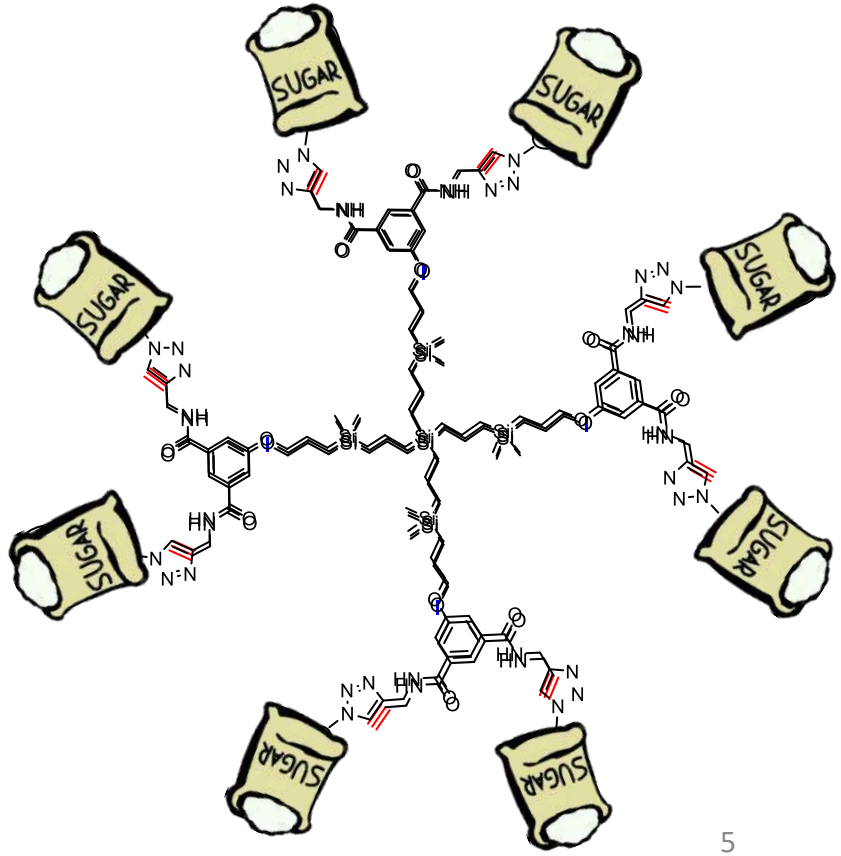
18 N_3-OEG-AcGal

	GLU series	GAL series	OEG-GAL series
G1			 ~ 3300 g/mol
G2			 ~ 9500 g/mol
G3			 ~ 16 500 g/mol

Carbosilane glycodendrimers

synthesis





Carbosilane glycodendrimer library

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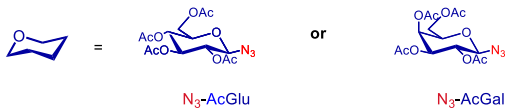

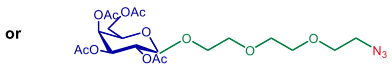

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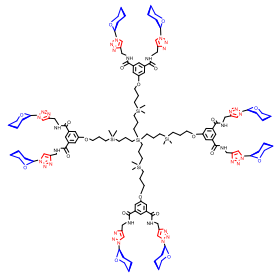
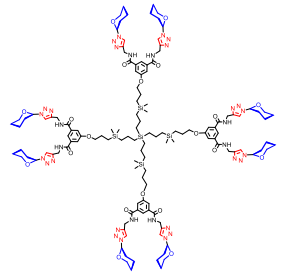
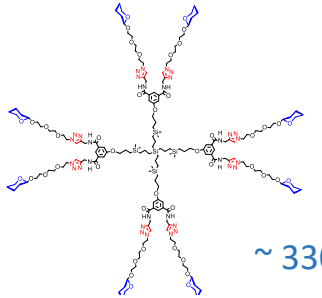
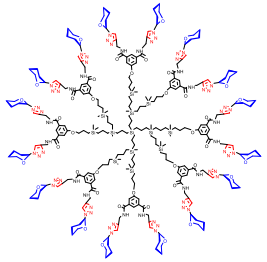
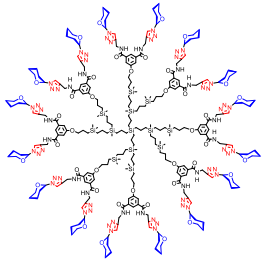
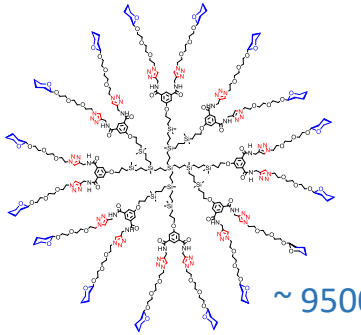
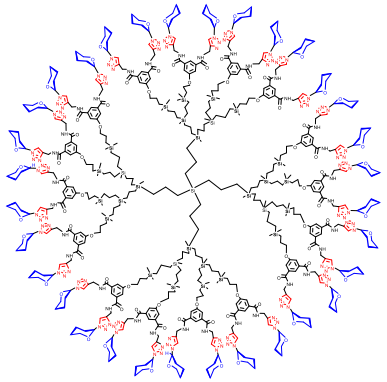
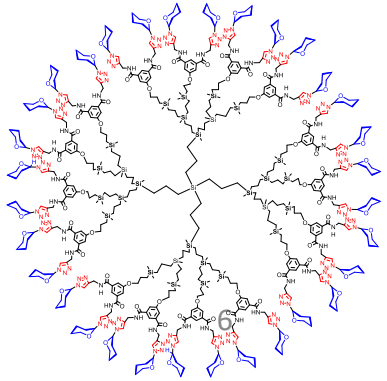
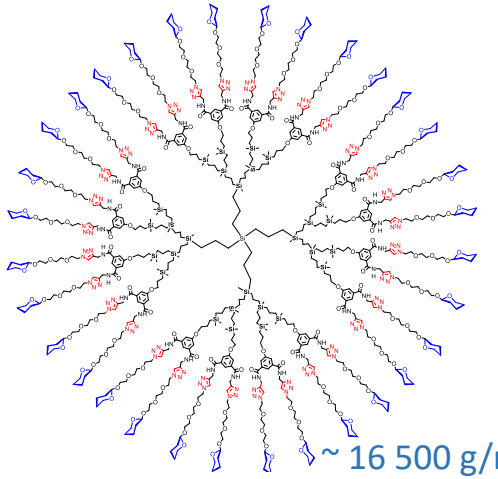
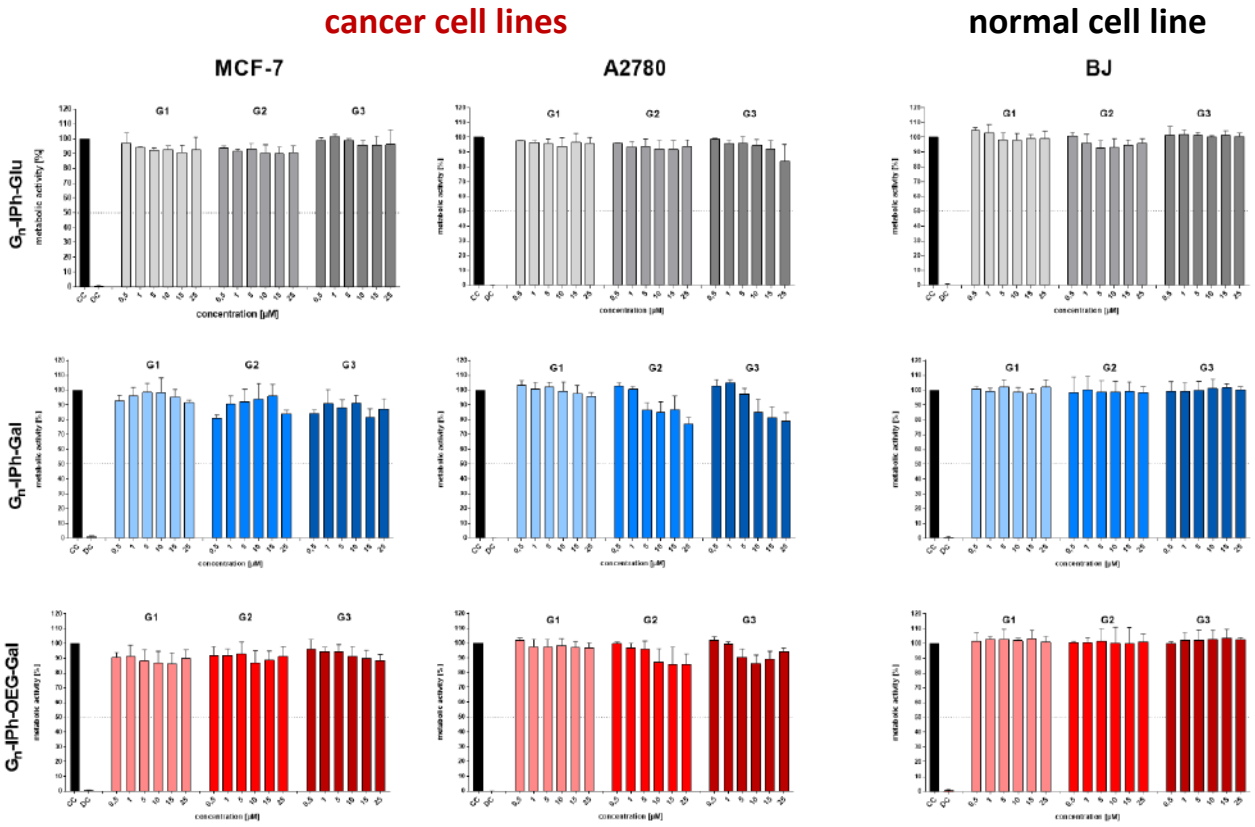
G1			 ~ 3300 g/mol
G2			 ~ 9500 g/mol
G3			 ~ 16 500 g/mol

Carbosilane glycodendrimers
cytotoxicity



In vitro cell viability at different concentrations of G_1-G_3 DDMs of the series G_n-IPh-Glu, G_n-IPh-Gal, and G_n-IPh-OEG-Gal. Cell viability values in 0.5–25 µM concentration range. CC = cell control, DC = death control.

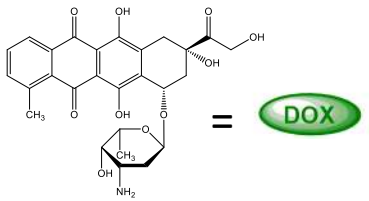
resazurin assay *(0.5 – 25* µM)

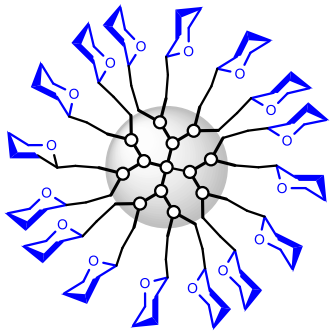
MCF	human breast cancer (adenocarcinoma)
A2780	human ovarian carcinoma
BJ	human fibroblast

- ✓ IC_{50} >> 25 µM
- ✓ Biocompatibility across dendrimer generation, series, and cell lines

Carbosilane glycodendrimers

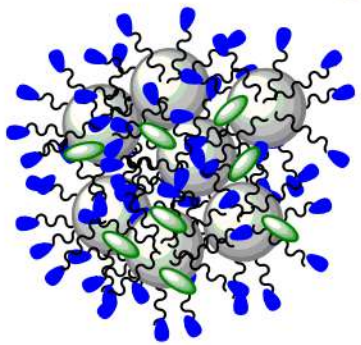
DOX encapsulation



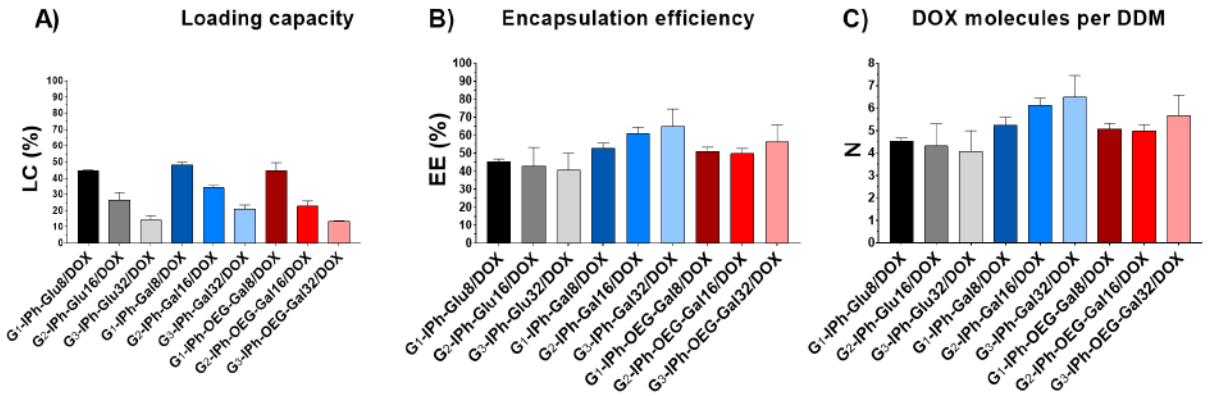


10 molar excess

DOX



glyco-DDM/DOX clusters



Encapsulation characteristics of glyco-DDM/DOX complexes. A) loading capacity (LC), LC = $W_e/W_c \times 100$, where W_e is the weight of encapsulated DOX, and W_c is the weight of DOX/DDM complex; B) encapsulation efficiency (EE), EE = $W_e/W_t \times 100$, where W_e is the weight of encapsulated DOX, and W_t is a total DOX weight; C) amount of DOX molecules per DDM (N), N = $n(DOX_{encaps.})/n(DDM)$, where $n(DOX_{encaps})$ is a molar quantity of encapsulated DOX and n(DDM) is a molar quantity of the DDM.

UV-Vis determination

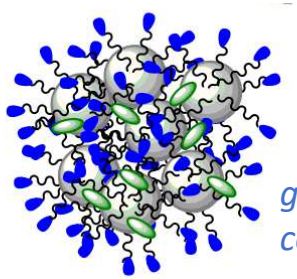
- ✓ 41 -65 % of DOX (4-6 DOX molecules) was encapsulated across generations and series
- ✓ Loading capacity „favors“ lower generations

BUT

glyco-DDM/DOX clusters

Biomacromolecules **2022**, 23 (1), p. 276–290

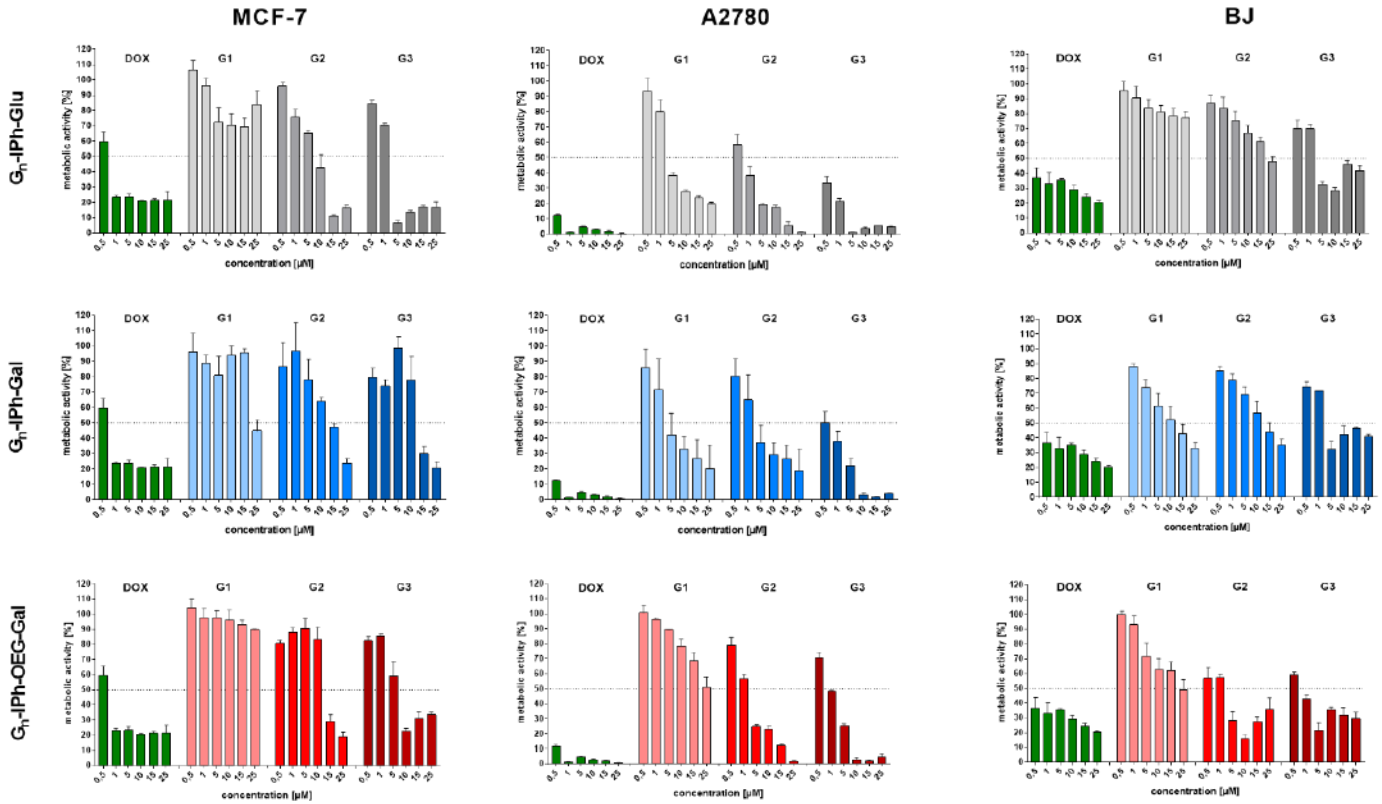
+ computational modeling
+ DLS measurements



glyco-DDM/DOX complexes

Glyco-DDM/DOX complexes *cytotoxicity*

cancer cell lines | **normal cell line**



resazurin assay *(0.5 – 25 μM)*

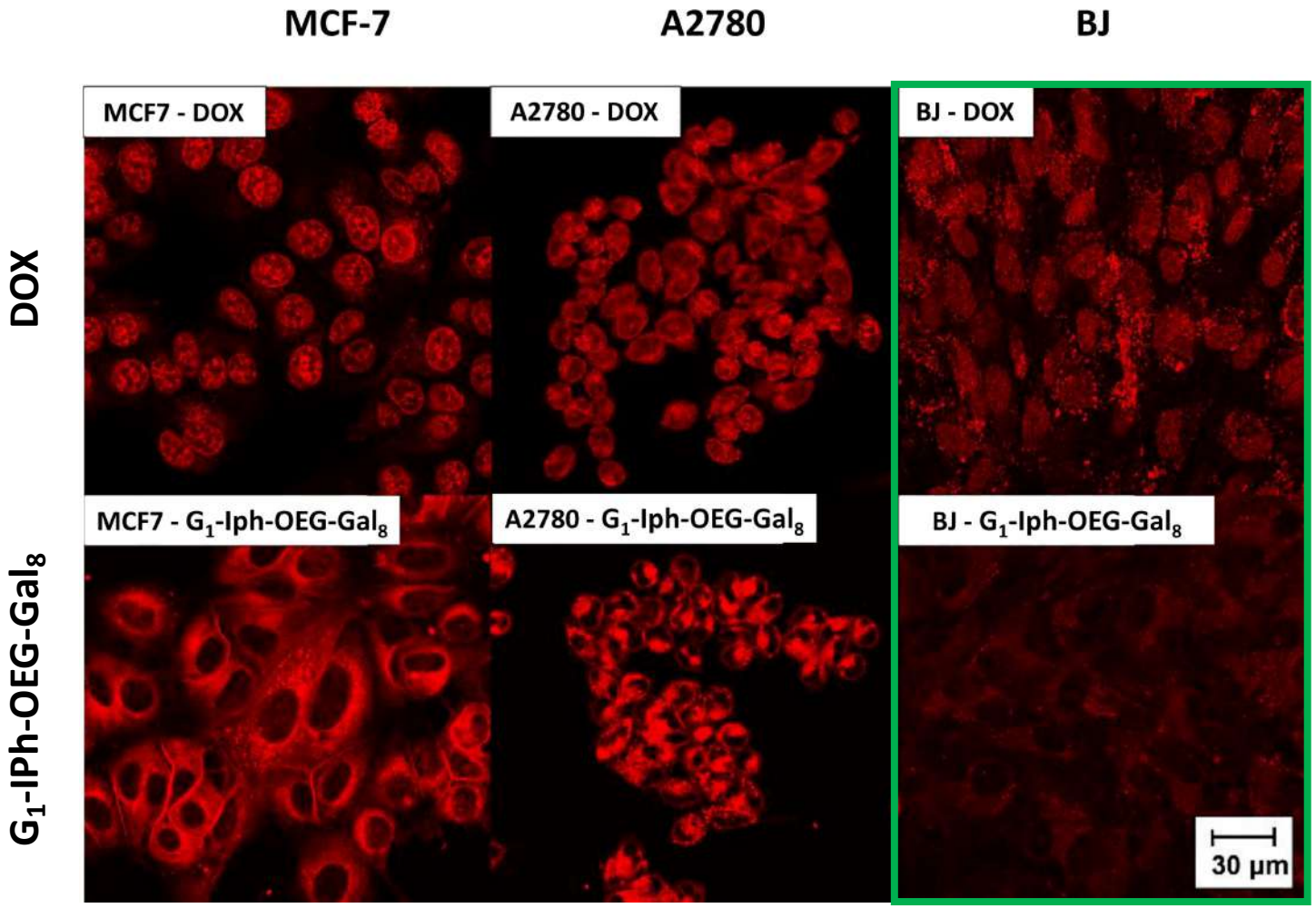
MCF	human breast cancer (adenocarcinoma)
A2780	human ovarian carcinoma
BJ	human fibroblast

- ✓ Cytotoxicity increases with generation
- ✓ Negligable influence of the DDM series
- ✓ elevated toxicity towards A2780 compared to MCF-7 (IC_{50} 5-6 times lower)

In vitro cell viability of cancer (MCF-7, A2780) and normal (BJ) cell lines after treatment with free DOX and glyco-DDM/DOX complexes (G_n-IPh-Glu/DOX, G_n-IPh-Gal/DOX, and G_n-IPh-OEG-Gal/DOX) at different concentrations of encapsulated DOX (0.5 – 25 μM range). Untreated MCF-7, A2780, and BJ cells were used as a control.

IC_{50} (μM)										
	DOX	G_n-IPh-Glu/DOX			G_n-IPh-Gal/DOX			G_n-IPh-OEG-Gal/DOX		
		G_1	G_2	G_3	G_1	G_2	G_3	G_1	G_2	G_3
MCF-7	0.7±0.1	24.9±3.5	5.8±0.8	2.8±0.2	19.8±3.1	13.2±1.7	4.3±0.7	>25.00*	5.3±0.8	4.8±0.6
A2780	0.1±0.0	4.0±0.3	0.7±0.1	0.3±0.0	4.1±0.7	3.1±0.5	0.6±0.1	31.6±2.0	1.7±0.1	1.1±0.1
BJ	0.4±0.1	58.3±7.9	20.5±2.1	2.3±0.3	9.7±1.1	11.7±1.1	2.2±0.2	20.1±1.8	1.2±0.2	0.8±0.1

Glyco-DDM/DOX complexes
intracellular internalization



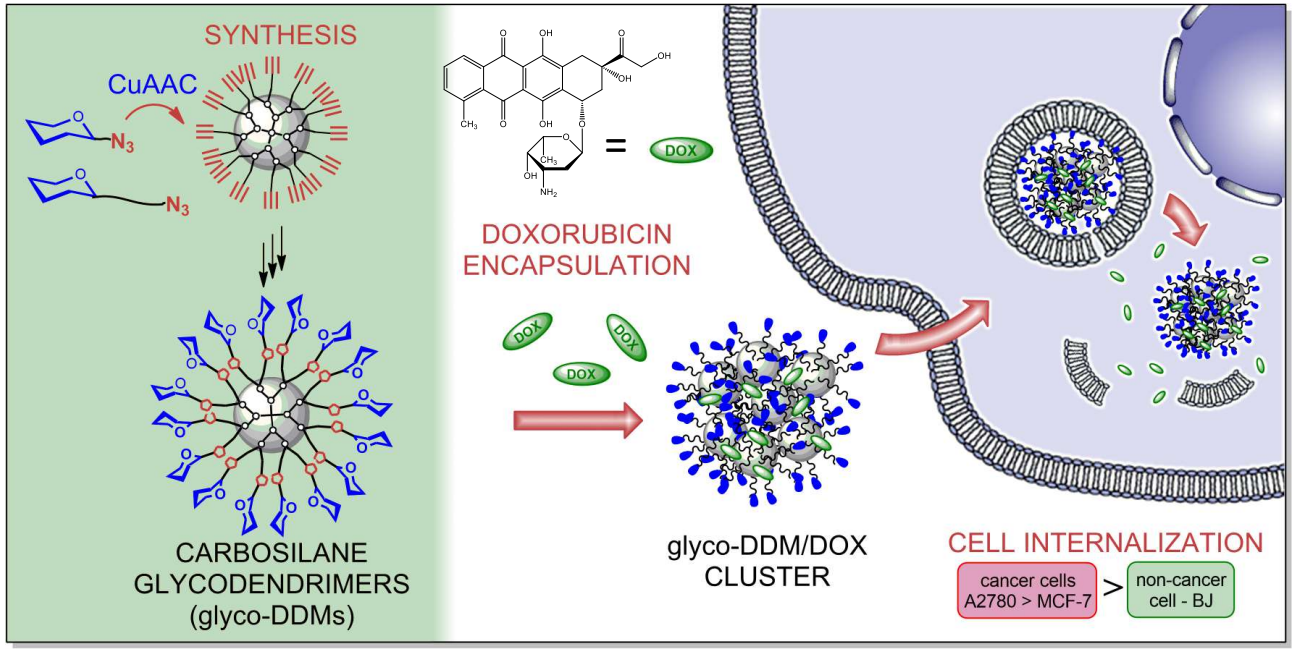
Confocal laser scanning fluorescence microscopy images of cellular internalization of DOX and G1-IPh-OEG-Gal8/DOX complexes in the three cell lines MCF7, A2780, and BJ after 90 min incubation.

Confocal fluorescence microscopy

- ✓ DOX accumulates in nucleus while glycoDDM/DOX complexes stay in cytoplasm
- ✓ **BJ** did not uptake glycoDDM/DOX complexes as easily as the cancer cell lines **MCF7** and **A2780**.

Carbosilane glycodendrimers
for anticancer drug delivery

STSM goals



- ✓ *Biocompatible carbosilane glycodendrimers*
- ✓ *cytotoxicity assay of DDM/DOX complexes*
- ✓ *Efficient DOX delivery to cancer cells*


BioMACROMOLECULES

pubs.acs.org/Biomac Article

Carbosilane Glycodendrimers for Anticancer Drug Delivery: Synthetic Route, Characterization, and Biological Effect of Glycodendrimer–Doxorubicin Complexes

Monika Müllerová, Dina Maciel, Nádia Nunes, Dominika Wrobel, Marcel Stofik, Lucie Červenková Šťastná, Alena Krupková, Petra Cuřínová, Kateřina Nováková, Matěj Božík, Marek Malý, Jan Malý, João Rodrigues,* and Tomáš Strašák*

Cite This: *Biomacromolecules* 2022, 23, 276–290 Read Online

- ✓ Size and Zeta potentials of the complexes
- ✓ DOX release profiles in physiological and acidic environment
- ✓ computer modeling

Acknowledgements

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INSTITUTE OF CHEMICAL PROCESS FUNDAMENTALS OF THE ASCR

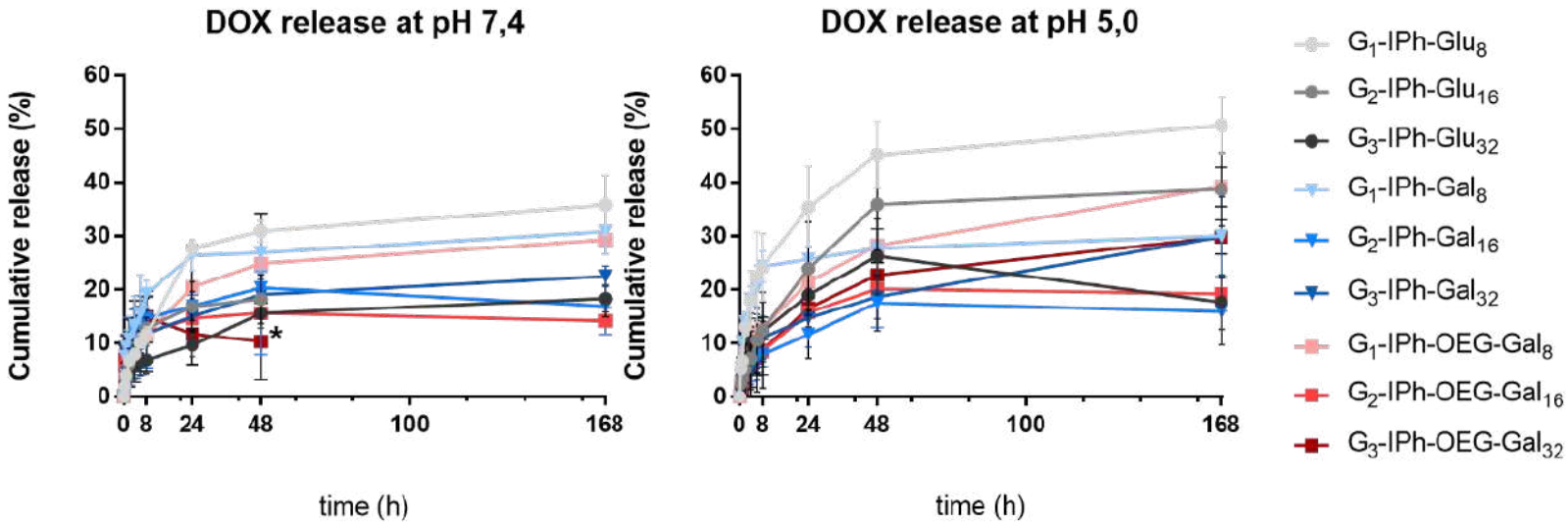
Tomáš Strašák
Lucie Č. Šťastná
Petra Cuřínová
Alena Krupková

SATA webinar (February/March 2023)

The power of collaboration in science:
Novel dendritic DDS and their prospects in anticancer therapy
by
Dr. Monika Müllerová
ICPF of the Czech Academy of Science
and MSc. Piotr Tarach
Department of General Biophysics of the University of Lodz

We acknowledge COST Action CA 17140 "Cancer Nanomedicine from the Bench to the Bedside" supported by COST (European Cooperation in Science and Technology).

Cumulative release study



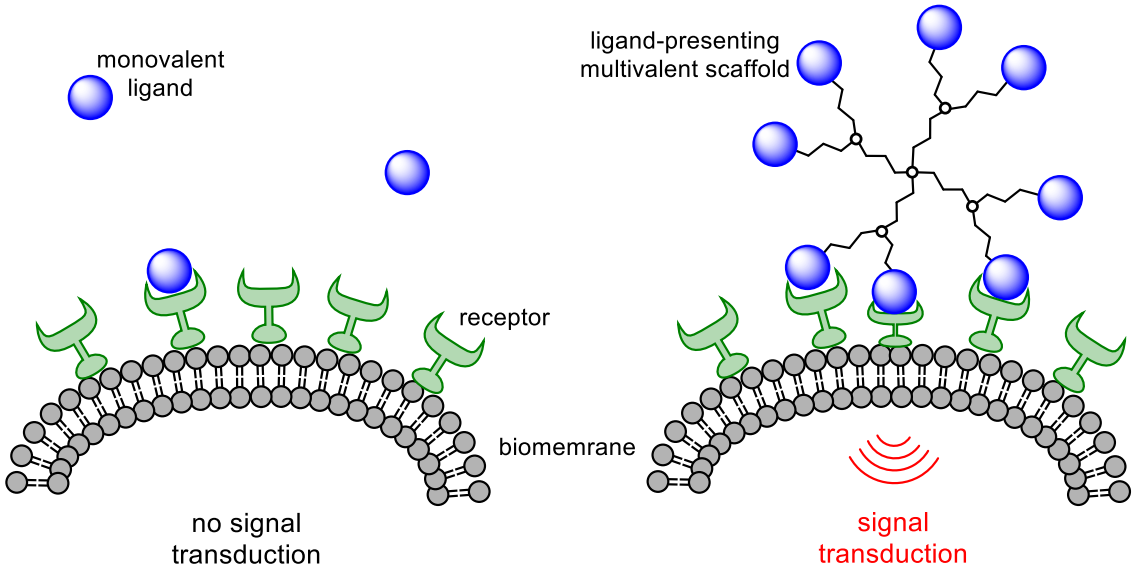
- *In vitro* DOX release from glyco-DDM/DOX complexes in PBS solutions at pH of 7.4 (left) and 5.0 (right). *The CR peaked in less than 48 hours.

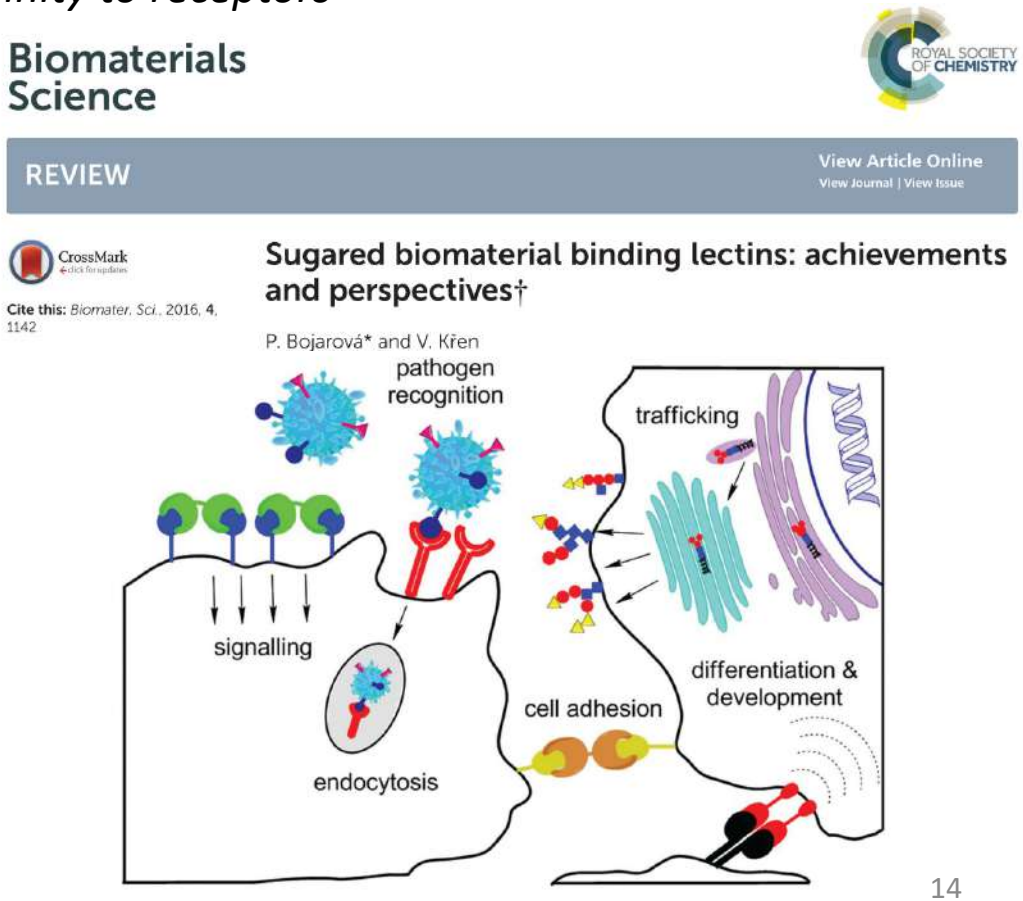
Carbosilane glycodendrimers
as drug delivery systems

Why 

?

- ✓ *Specific ligand – protein (saccharide – lectin) recognition promotes a plethora of essential biological functions*
- ✓ *Multivalent presentation of ligands amplifies the binding affinity to receptors*





Hydrodynamic diameters (d_h) and Zeta potential (ζ) of glyco-DDMs glyco-DDM/DOX complexes.

Compound	d_h [nm] in water		d_h [nm] in PBS		ξ-values [mV]	
	DDM	DDM/DOX	DDM	DDM/DOX	DDM	DDM/DOX
G_1-IPh-Glu_8	8 ± 1	48 ± 11	7 ± 1	67 ± 6	2.9 ± 0.6	-17.7 ± 5.5
G_2-IPh-Glu_{16}	14 ± 2	45 ± 3	16 ± 2	82 ± 31	-16.0 ± 1.2	-4.2 ± 0.1
G_3-IPh-Glu_{32}	17 ± 3	350 ± 10	18 ± 6	320 ± 31	-1.2 ± 0.9	-9.5 ± 0.2
G_1-IPh-Gal_8	6 ± 1	9 ± 1	6 ± 1	11 ± 3	-0.3 ± 0.7	-7.4 ± 0.7
G_2-IPh-Gal_{16}	14 ± 3	14 ± 3	14 ± 1	11 ± 2	7.2 ± 0.9	-23.7 ± 4.2
G_3-IPh-Gal_{32}	17 ± 2	16 ± 2	8 ± 2	16 ± 1	-2.3 ± 1.5	1.1 ± 0.1
G_1-IPh-OEG-Gal_8	5 ± 2	86 ± 12	6 ± 2	86 ± 9	-5.3 ± 1.5	-9.3 ± 0.7
G_2-IPh-OEG-Gal_{16}	6 ± 1	71 ± 6	6 ± 1	70 ± 13	-4.5 ± 1.0	-1.6 ± 0.9
G_3-IPh-OEG-Gal_{32}	7 ± 1	5 ± 2	20 ± 2	6 ± 2	-7.5 ± 2.1	-1.4 ± 1.7

IC50 values of the DDM/DOX complexes.

IC_{50} (μM)										
	DOX	G_n-IPh-Glu/DOX			G_n-IPh-Gal/DOX			G_n-IPh-OEG-Gal/DOX		
		G_1	G_2	G_3	G_1	G_2	G_3	G_1	G_2	G_3
MCF-7	0.7±0.1	24.9±3.5	5.8±0.8	2.8±0.2	19.8±3.1	13.2±1.7	4.3±0.7	>25.00*	5.3±0.8	4.8±0.6
A2780	0.1±0.0	4.0±0.3	0.7±0.1	0.3±0.0	4.1±0.7	3.1±0.5	0.6±0.1	31.6±2.0	1.7±0.1	1.1±0.1
BJ	0.4±0.1	58.3±7.9	20.5±2.1	2.3±0.3	9.7±1.1	11.7±1.1	2.2±0.2	20.1±1.8	1.2±0.2	0.8±0.1