



صناعة الأدوية في الجزائر

Séminaire « L'industrie pharmaceutique en Algérie »



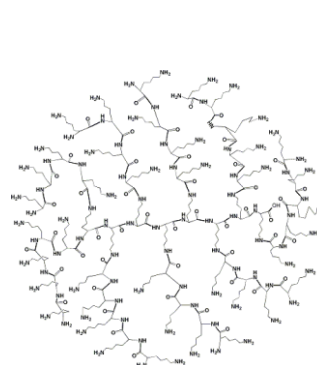
**Recherche et développement du laboratoire Colcom en
nanoparticules poly-lysines biocompatibles appliquées à la
biologie et la médecine**

Our Company



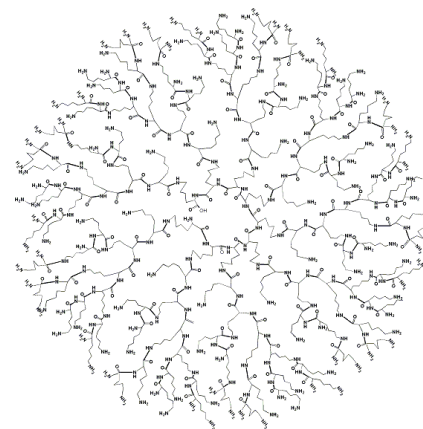
TECHNOLOGY & KNOW-HOW

Nanotechnologies
Dendri-Graft of Lysines (DGLs)



G2

50 lysines – 4,5 nm



G3

120 lysines – 7 nm

Activities

Research reagents provider

Contract research organization: Services & Customization of DGLs

→ More than 70 publications (www.colcom.eu), 6 patent applications

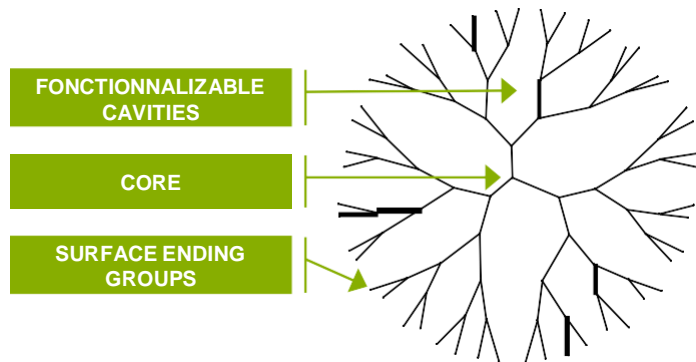
References, Customers and Partnerships



Structure & Synthesis

Etymology:

- *dendri* : « tree-like structure »
- *meros* : « part of »



Structure:

- 3D nanomolecule
- core and internal cavities (host molecule)
- successive « tree-like » layers (or generations)
- multivalency: surface end-groups (customization)

Properties:

1) Physicochemical (membrane interaction, cellular uptake,...):

Depending on the size, global charge, hydrophobic/hydrophilic balance,...

→ ALL CUSTOMISABLE

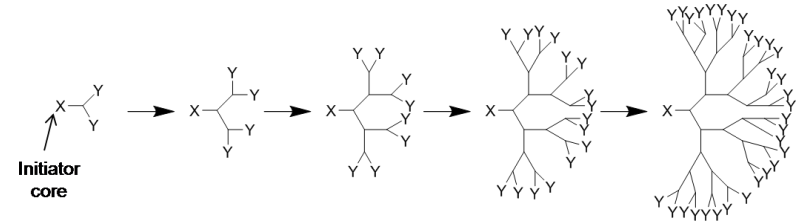
2) Biological:

Depending on the choice of monomer unit and functional end-groups

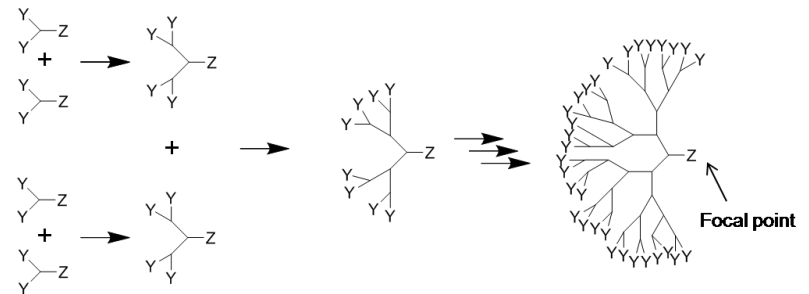
→ 2 KEY POINTS

Synthesis:

- Divergent (extension from the core)



- Convergent



Once upon a time... Dendrimers

1980 1985 1990 1995 2000 2005 2010 2015 2020

Birth

Resistance
Controversy

Acceptance
Recognition

PoC in Animals
Human Clinical Trials

Market

1978 1984
1st synthesis (Vögtle) 1st article PAMAM

polydispersity?
crosslinked particles?
no relevant properties?
scale up?

1992
1st company

1999
1st Intern.
Conference

2003
1st FDA IND
(Starpharma)

2004
1st clinical
trial

2006
Acquisition
DNT/Starpharma

2019
1st launching
(VivaGel® BV)



1st patent



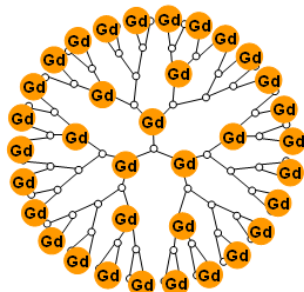
Donald A.
Tomalia



Examples of 1st targeted applications

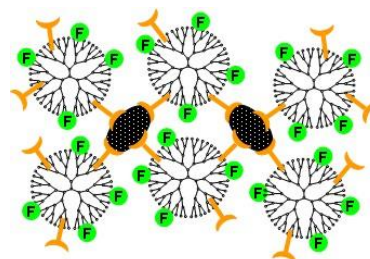
IRM CONTRAST
AGENT

Gadomer-17
(dendrimer of lysine)



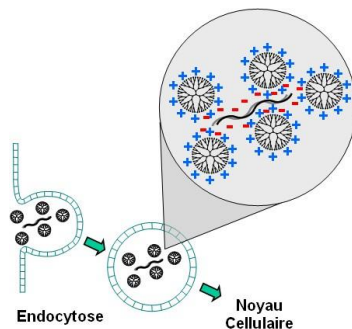
BIOLOGIC AGENT
DETECTION

Alert Ticket



GENETIC TRANSFECTION
AGENT

SuperFect®
(PAMAM)

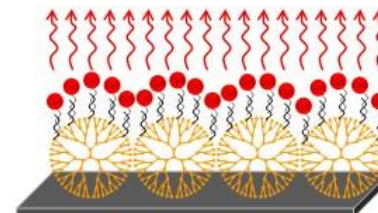


HEART ATTACK
DIAGNOSTIC AGENT

Stratus CS
(PAMAM G5)

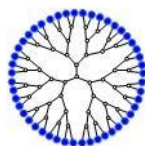
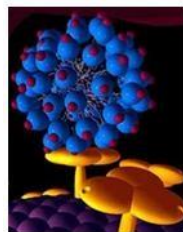
DADE BEHRING

Every minute of every day™



BIOCIDE

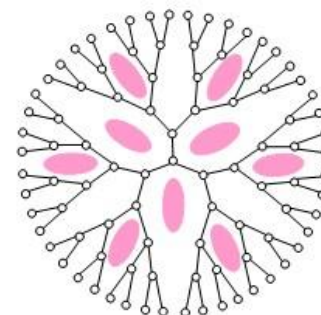
VivaGel
(dendrimere of lysine)



← Surface HIV

DRUG DELIVERY

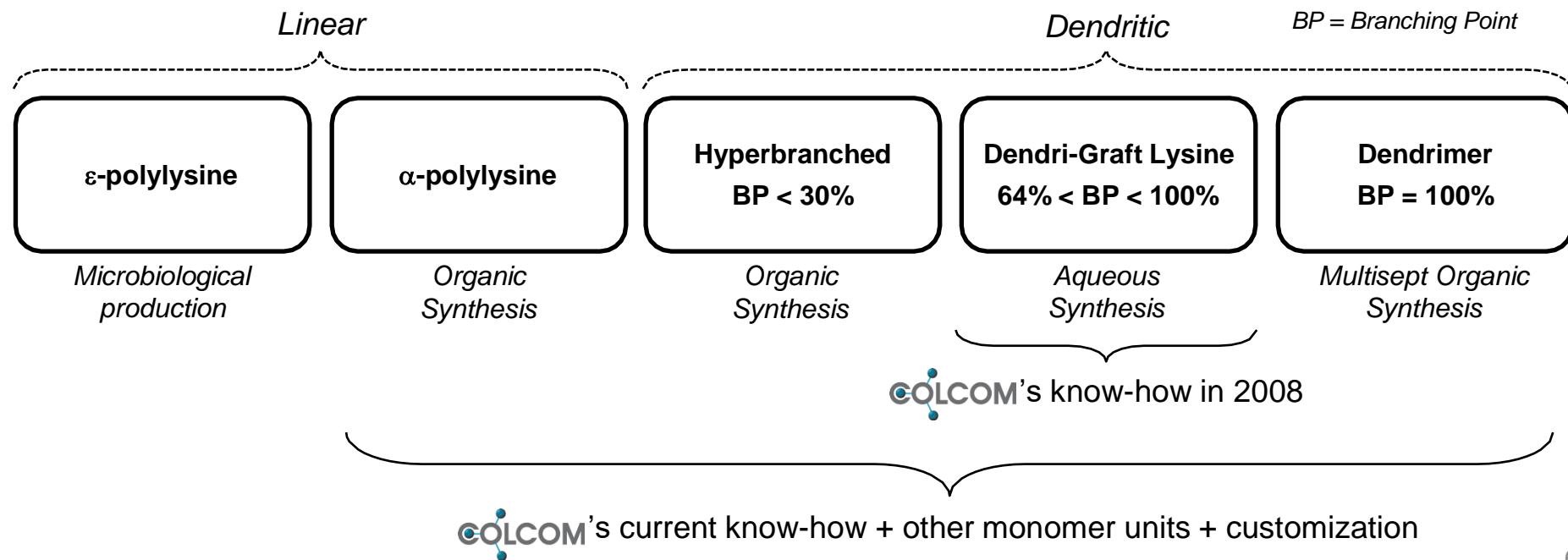
Under development



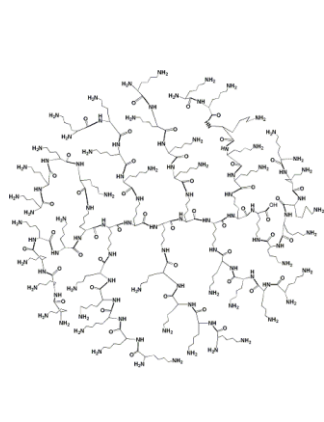
Polymer & Poly-L-Lysines overview

Four Major Classes of Macromolecular Architectures:

Nomenclature applied to Poly-lysines:

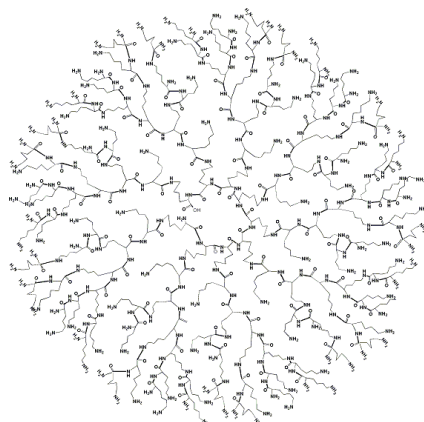


Dendri Graft of Lysines (DGLs)



G2

50 lysines – 4,5 nm



G3

120 lysines – 7 nm

“Dendron” in Greek mean “Tree”

Dendri Graft of Lysines : “Tree-like” polymer of Lysines

Generation	G1	G2	G3	G4	G5
Structure	Linear	DGL	DGL	DGL	DGL
Molecular Mass (kDa)	1.5	8.6	22.0	65.0	170.0
Diameter (nm)	2	4.5	7	11	16
Number of groups	8	50	120	360	1000

DGL
COLCOM

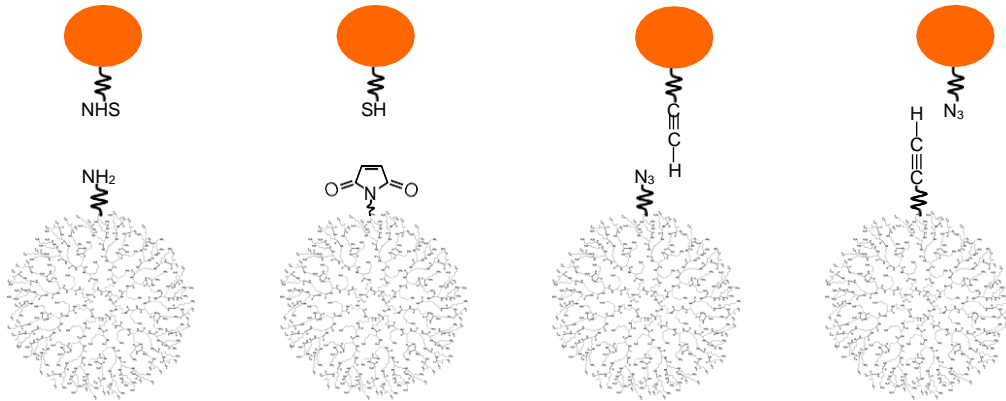
Other
Dendrimers + **DGL**
COLCOM

PROPERTIES	ADVANTAGES	BENEFITS	ADDED VALUE
100% L-Lysine Structure flexibility Exponential synthesis	Water solubility Safe by design Sizes & Price	Biocompatible Improve solubility Decrease toxicity	
Multivalence 80% of free volume 3D structure	Customization Active targeting Encapsulation	Reduce side effects Enhance half-life & therapeutic effect	

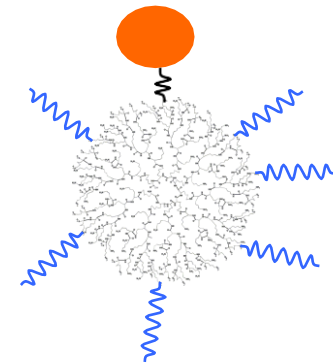
DGLs customization

Depending on the specifications, DGL customization (size, charge, linker, ligand,...)

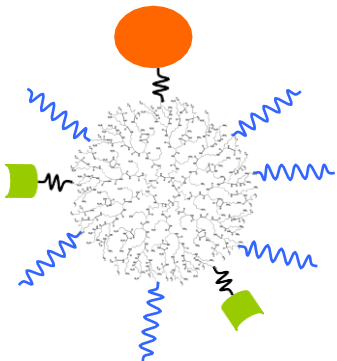
1) Load with drugs, peptide sequences, ligands



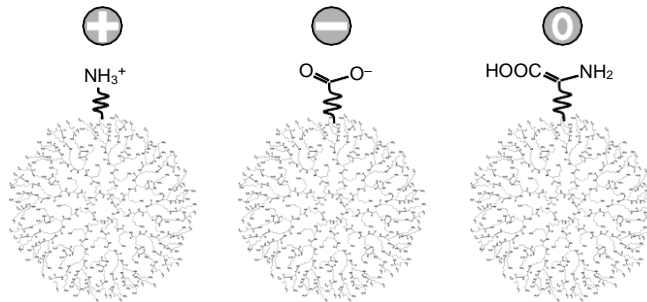
2) Enhance solubility



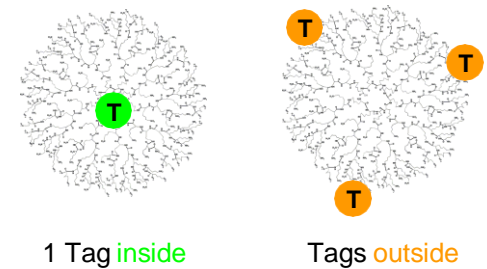
3) Target or by-pass



4) Adapt the global charge

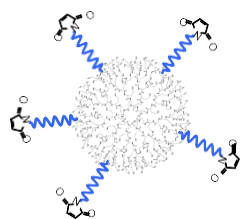


5) Follow & Localize

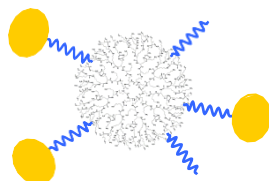


DGLs customization

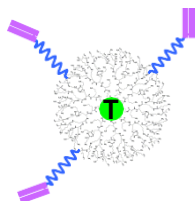
PEG-Maleimide



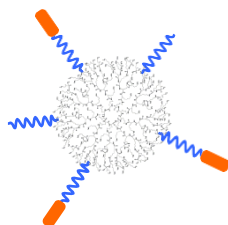
PEG-Drug



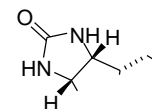
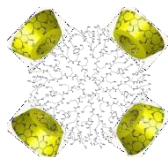
Tag inside +
Fab fragment



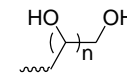
PEG-Peptide



Cyclodextrin



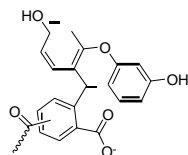
Biotine



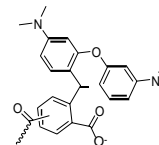
Polyols



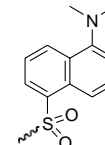
Peptides



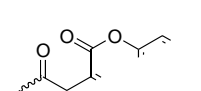
Fluoresceine



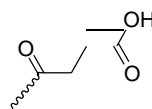
Rhodamine



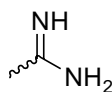
Dansyle



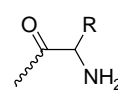
Coumarine



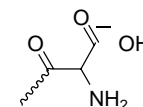
Acide succinique



Guanydines



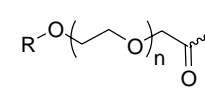
Acides aminés



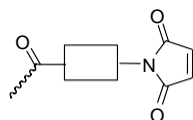
Aspartique



Alkyles



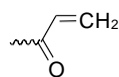
PEGs



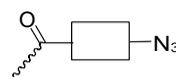
Maléimides



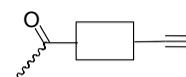
Cl-acétyles



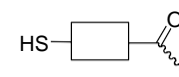
Acryloyles



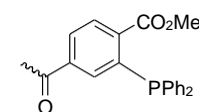
Azotures



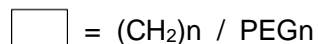
Acétyléniques



Thiols



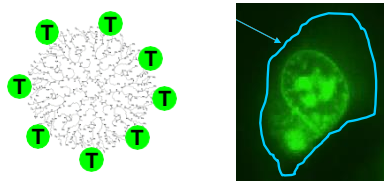
Phosphines



Proof of concept for biological app.

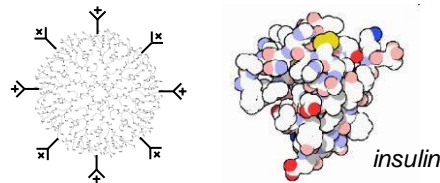
More than 70 publications (www.colcom.eu)

Membrane Penetration



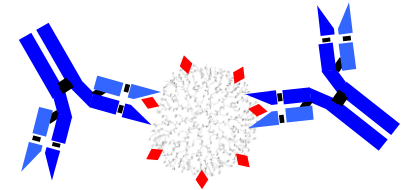
Tsogas et al., *Biomacromolecules*, 2007

Drug Delivery



Sideratou et al, *J. Colloid Interf. Sci*, 2010

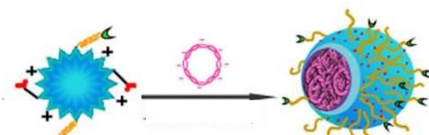
In Vivo Antibody Production



Romestand et al., *Biomacromolecules*, 2010

Targeted Drug Delivery

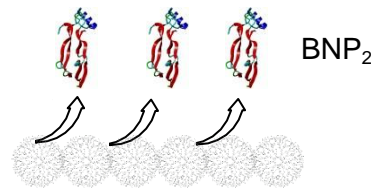
(chemotherapy and gene therapy effects)



Drug Carrier and Transfection Agent *In vivo*

Liu et al., *Biomaterials*, 2012

Growth factors controlled release



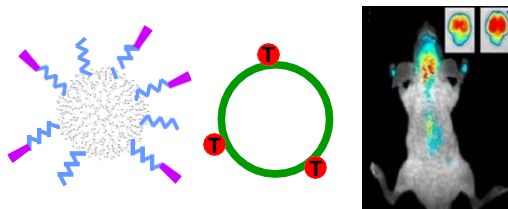
Mendoza et al, *ACS Nano*, 2011

MRI Contrast Agent



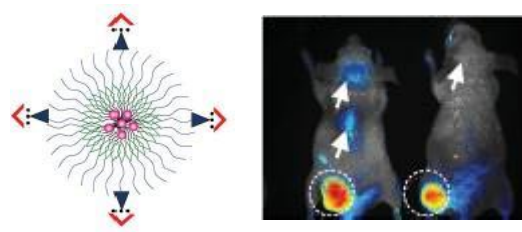
Ogawa et al., *Bioconjugate Chem.*, 2010

CNS Targeting



Liu et al., *Biomaterials*, 2010






















Tumor Targeting



Han et al., *SMALL*, 2013

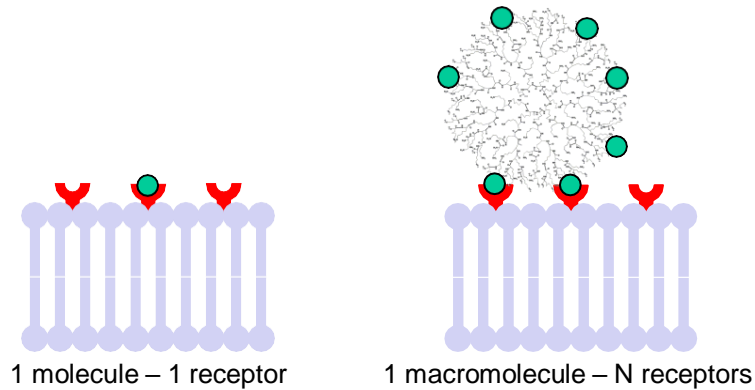


Other topics & projects

PROJECT	TOPIC	OUTCOMES	PARTNERS and/or FUNDING
DHERMIC	Tissue Engineering (skin regeneration and modeling)	Patent (WO2017194761A2)	  
BIOCHIP	Detection of extraterrestrial life biomarkers at low concentrations	Patent (WO2009112430A1)	  
SACAD'EAU	Water analysis and decontamination	Patents (WO2014135593A1, WO2017005754A1)	  
COSMETIC	Confidential	Patent Clinical trial	Confidential
IMDENDRIM	In situ anti-cancer agent	Patent (WO2015104589A1) Clinical trial	
PCI NANO	Chemotherapies delivery system	On-going project	    
CAPSECURE	Selective delivery of hydrophobic drugs in GI oncology	On-going project	
SPINEFLEX	Treatment of vertebral fractures	On-going project	    

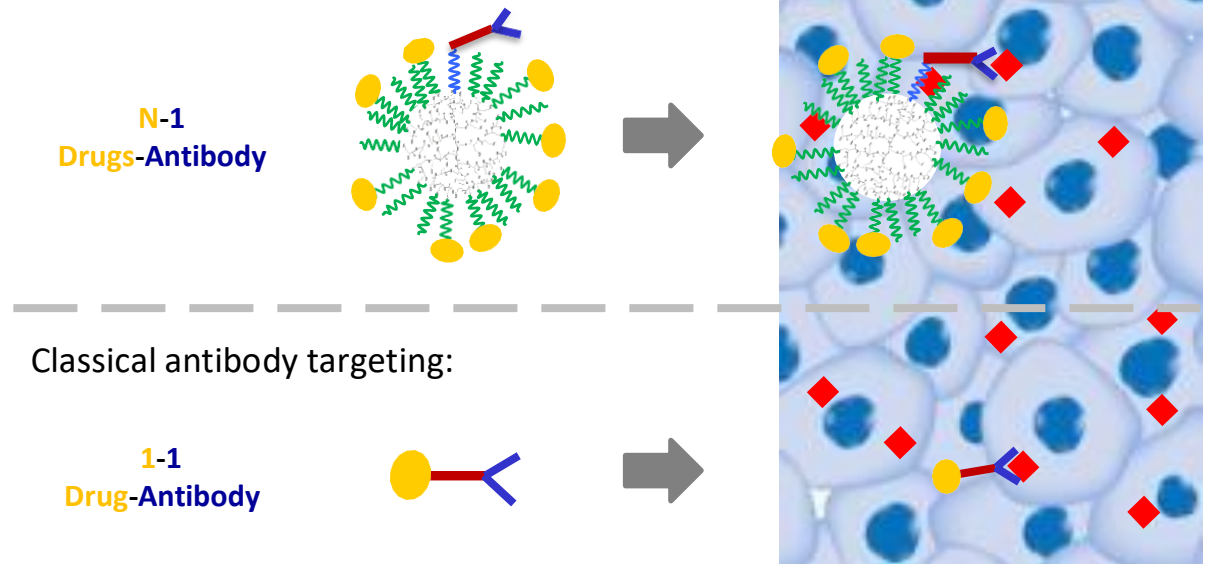
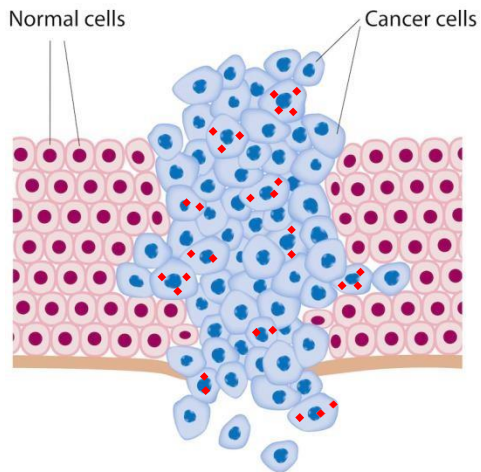
Strategies for targeting therapy

1) Passive delivery system:

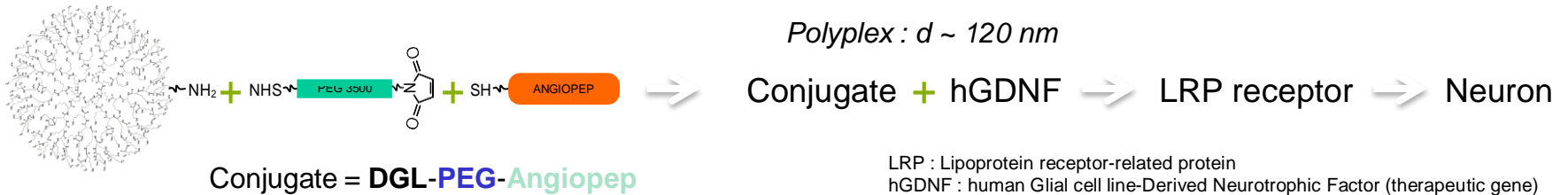


2) Active delivery system:

Cancer cells express targetable antigen (◆)



Central Nervous System gene delivery with an Angiopeptide-conjugate



	rotenone	hGDNF	Days	0	...	15	...	25	...	35	37	39	41	43	45
Blank control	O	O	Group 1 (oil)			BT		BT		BT					BT,I,H
Negative control	X	O	Group 2 (DPA/GFP)			BT		BT		BT					BT,I,H
	X	X	Group 3 (DPA/hGDNF)			BT		BT		BT					BT,I,H
	X	X	Group 4 (DPA/hGDNF)			BT		BT		BT					BT,I,H
	X	X	Group 5 (DPA/hGDNF)			BT		BT		BT					BT,I,H
	X	X	Group 6 (DPA/hGDNF)			BT		BT		BT					BT,I,H

n = 15

Behavioral Tests:

Treatment with hGDNF markedly increase number of line crossing and decrease inactive time

Tyrosine Hydroxylase Immunohistochemistry:

Increasing the injection induce apparent recovery of dopaminergic neurons

Multiple dosing administrations could be applied in long-term gene therapy of chronic Parkinson's Disease.

DGL-based polyplex showed higher cellular uptake and gene expression compared to PAMAM-based counterpart.

DGL is a potential dendrimer in the field of gene therapy.

Study Case 2: CNS Gene Delivery

Yang Liu et coll. BIOMATERIALS ; 2010 ; 31 ; 5246-57
Department of Pharmaceutics, School of Pharmacy, Fudan University Shanghai, China

Delivery to the central nervous system through the blood-brain barrier with non-viral gene delivery systems

(Central Nervous system diseases : Alzheimer, Parkinson, Brain tumors)

Leptin = 146 amino-acid polypeptide
Secreted in the blood stream
Action specifically located in the brain



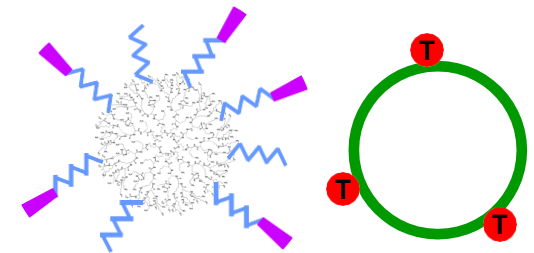
Leptin30 = Sequence of 30 amino-acid (position 61-90) observed to possess the highest brain/plasma ratio
Leptin30 as brain-targeting ligand

Study of Brain-Targeting efficiency of Nanoparticles :

DGL G3 + Polyethylene Glycol + Leptin30 / plasmid DNA

Nano size (141 ± 33 nm) and positively charged ($+1.15 \pm 0.47$ mV)

DGL G3 (NH_3^+)₈₀ (PEG)₂₇ (PEG-Leptin30)₁₆



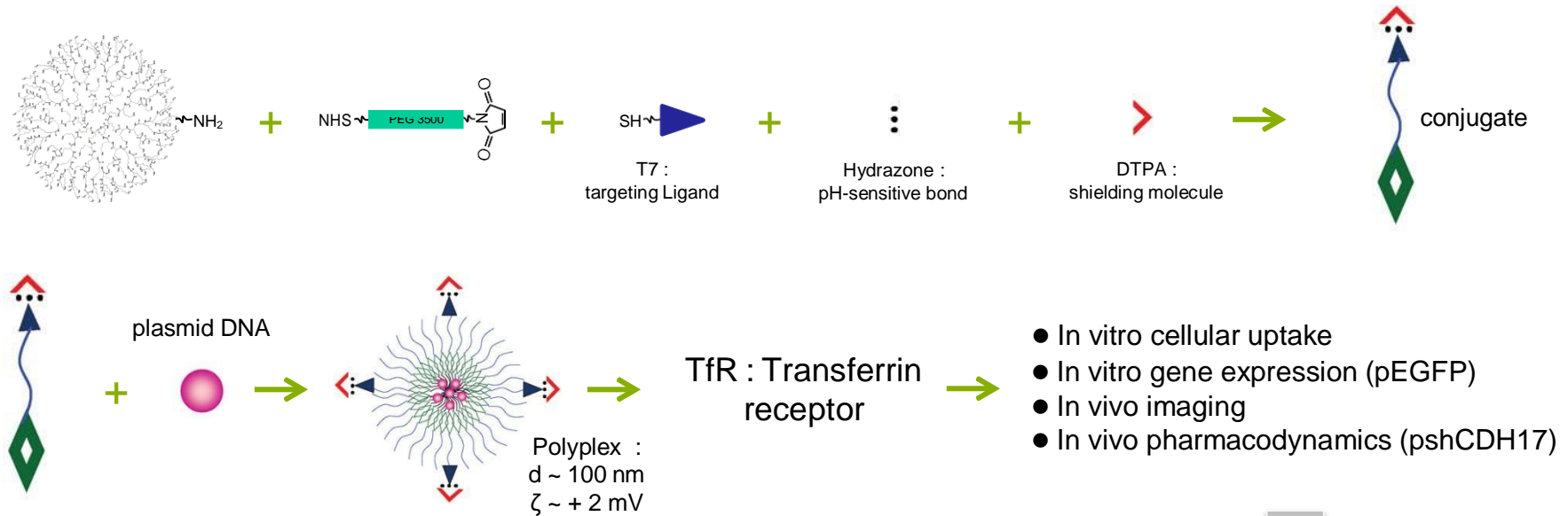
3 factors influence the cellular uptake : Surface charge / PEGylation / Brain targeting ligand
DGL : Better biocompatibility / Less toxic and biodegradable / Tissue selectivity

“DGL holds great promise as the non viral vector for brain-targeting gene delivery”

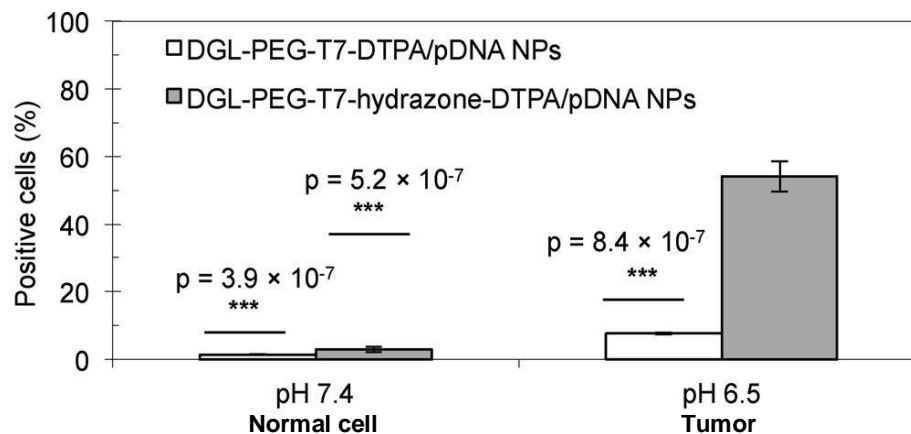
Study Case 3: Tumor Targeting

Liang Han et coll. SMALL ; 2013 ; 9(21) ; 3647–58
Shanghai Public Health Clinical Center Fudan University, Shanghai, China

Active/Inactive the receptor-mediated endocytosis on tumor/normal cells
Reducing the overall dose by concentrating the drug in the targeted tissue



Cellular Uptake



Reduce or even avoid the uptake by healthy cells.

Masked T7 could not recognize the TfR and mediate endocytosis

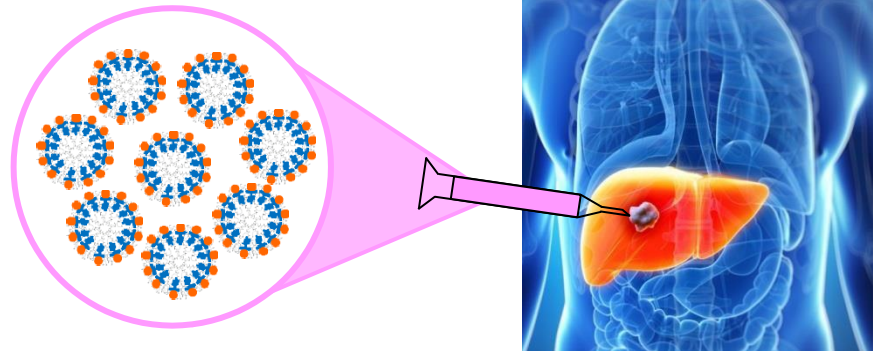
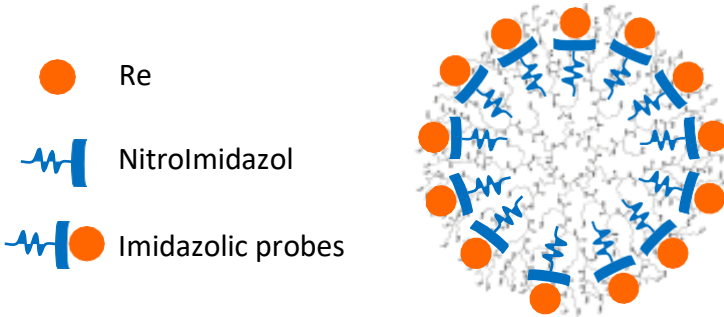
Study Case 4: Imdendrim

Treatment of liver metastases, inoperable and not responding to conventional treatment.

Word patent (WO2015104589A1): In situ anti-cancer agent derived from [188re]rhenium nitro-imidazole ligand loaded poly-l-lysine dendrimer administrated by direct CT guided stereotactic intrahepatic injection.

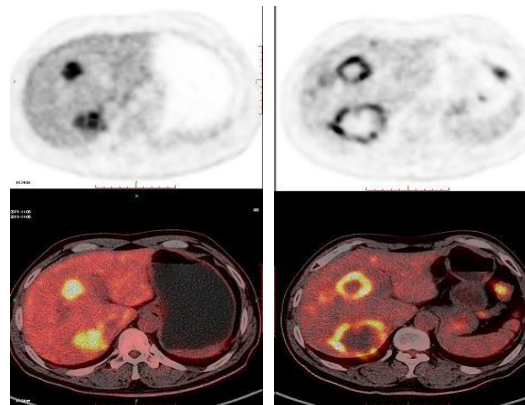
In situ Injection

Complex dendrimer rhenium



Imdendrim remains in site of injection
24hs post injection, avoiding systemic effect

1st Human trial with DGL



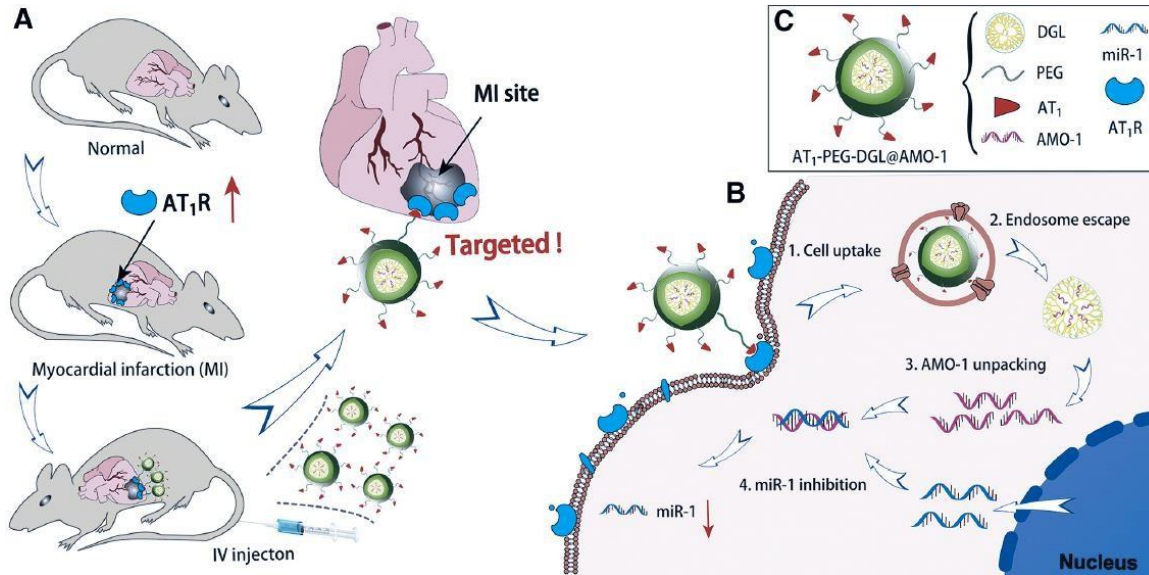
DOI: 10.1200/JCO.2018.36.15_suppl.e15569
Journal of Clinical Oncology 36, no. 15 suppl



Study Case 5: Myocardial Infarction

NANOMEDECINE: Nanotechnology, Biology, and Medicine ; 2018 ; 14 ; 619-631
Shanghai Jiao Tong University, Shanghai, China College of Engineering & Applied
Science, University of Cincinnati, Cincinnati, OH, USA.

Nanovector anchored with AT1 targeting peptide and μ RNA inhibitor to attenuate cardiomyocyte apoptosis.



miR-1: promotes apoptosis of cardiomyocytes
(down-regulates Bcl-2 and PKC ϵ proteins)

AMO-1: Anti-miR-1 antisense oligonucleotide

AT1R (Angiotensin II type 1 receptor): over-expressed

Polyplex
DGL G3 – PEG – AT₁ / AMO-1
($d = 200 \pm 16.5$ nm, $\zeta = +4.06 \pm 0.34$ mV)

Potential target for early MI therapy by down-regulation of miR-1 expression

Figure 1. Schematic illustrations of AT₁-PEG-DGL@AMO-1 complex formation and delivery.

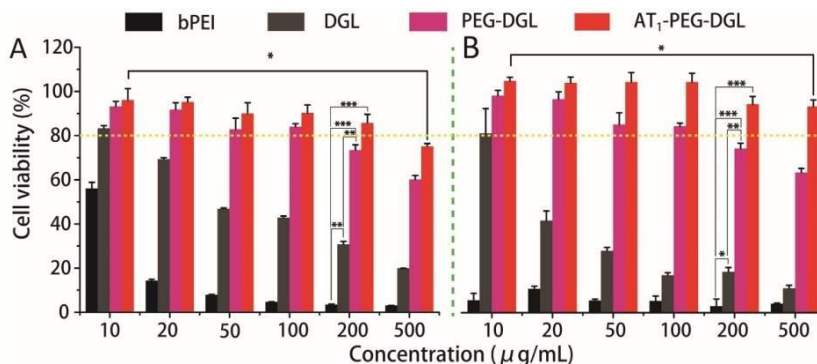


Figure 3 (Suppl. Mat.). Cytotoxicity examination of bPEI, DGL, PEG-DGL and AT₁-PEG-DGL under different concentrations. (A) H9C2 cells; (B) primary myocardial cells.

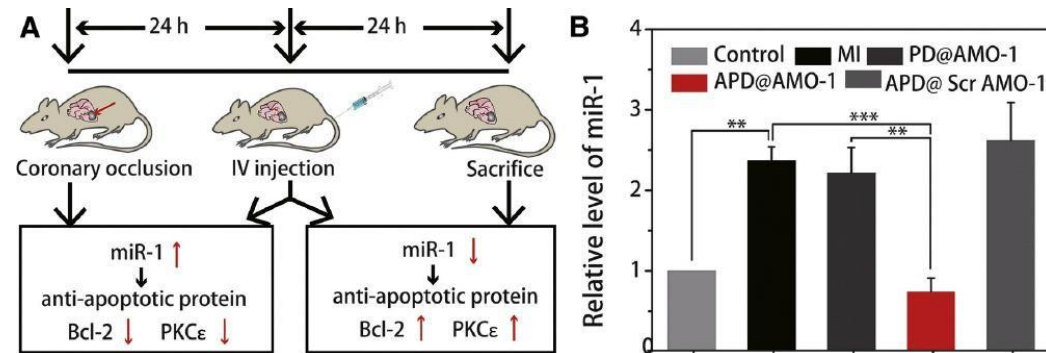


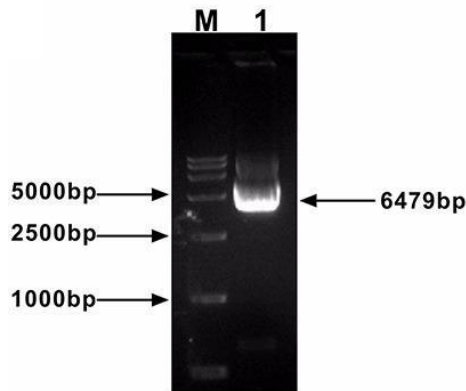
Figure 6. Effects of AT₁-PEG-DGL@AMO-1 on miR-1 level in the heart 1 day after administration. (A) Study design. (B) MiR-1 quantification through real-time RT-PCR. PD: PEG-DGL; APD: AT₁-PEG-DGL; Scr AMO-1: Scramble AMO-1.

Study Case 6: DNA vaccine

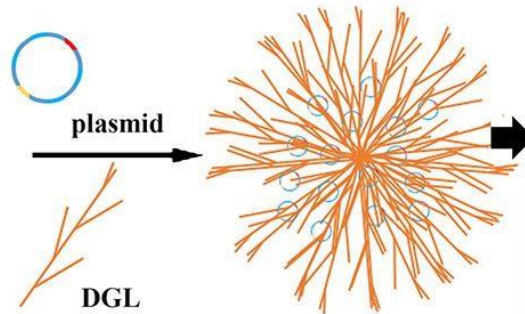
Kai Zhao et coll. Nanomedicine: Nanotechnology, Biology and Medicine
Volume 27, July 2020, 102209
Laboratory of Chemical Engineering Process and Technology, Heilongjiang University,
Harbin, China and Department of Physiological Sciences, College of Veterinary Medicine,
Oklahoma State University, Stillwater, OK, USA

Dendrigrraft poly-L-lysines delivery of DNA vaccine effectively enhances the immunogenic responses against H9N2 avian influenza virus infection in chickens

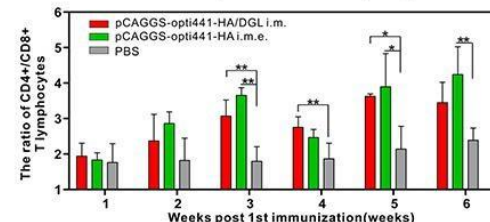
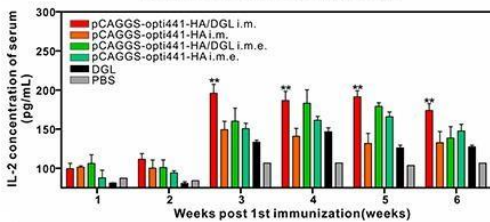
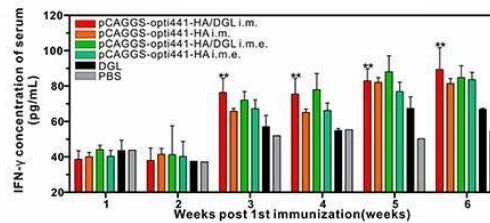
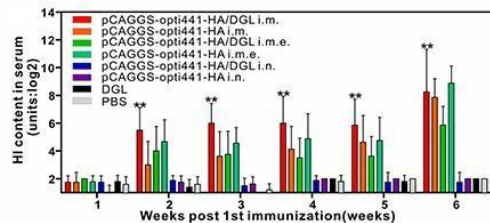
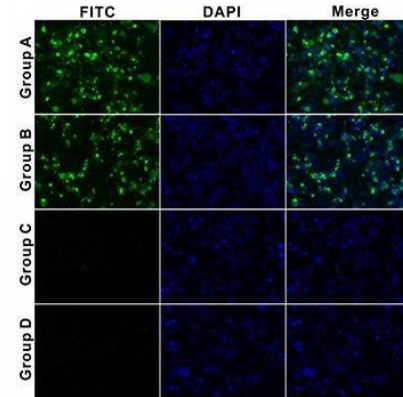
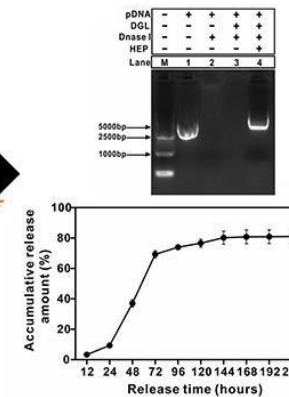
Construction of plasmid



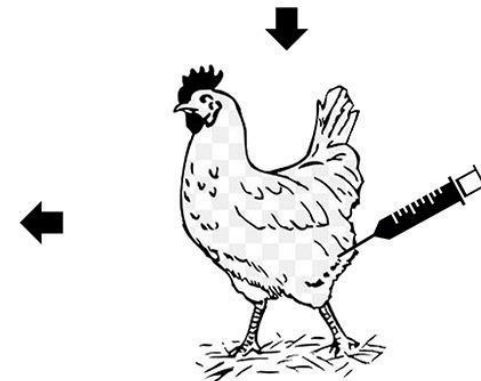
Synthesis of the pCAGGS-opti441-HA/DGL



Characterization

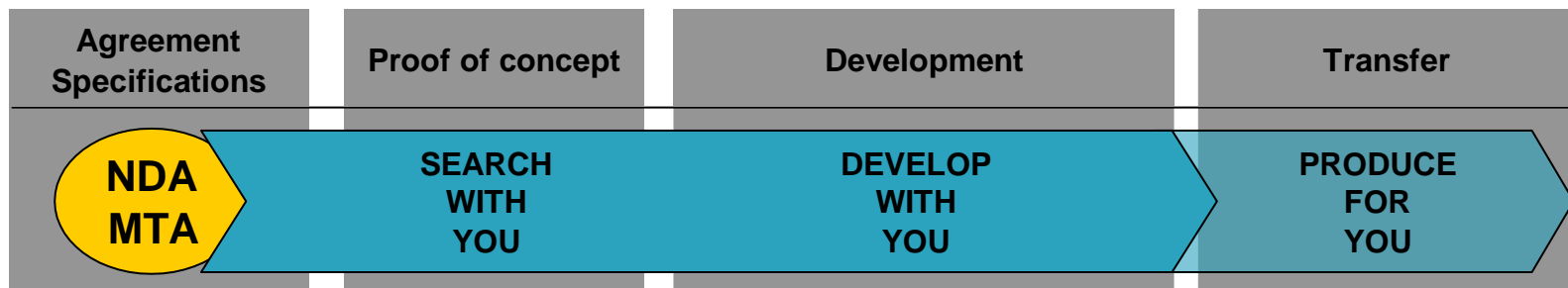
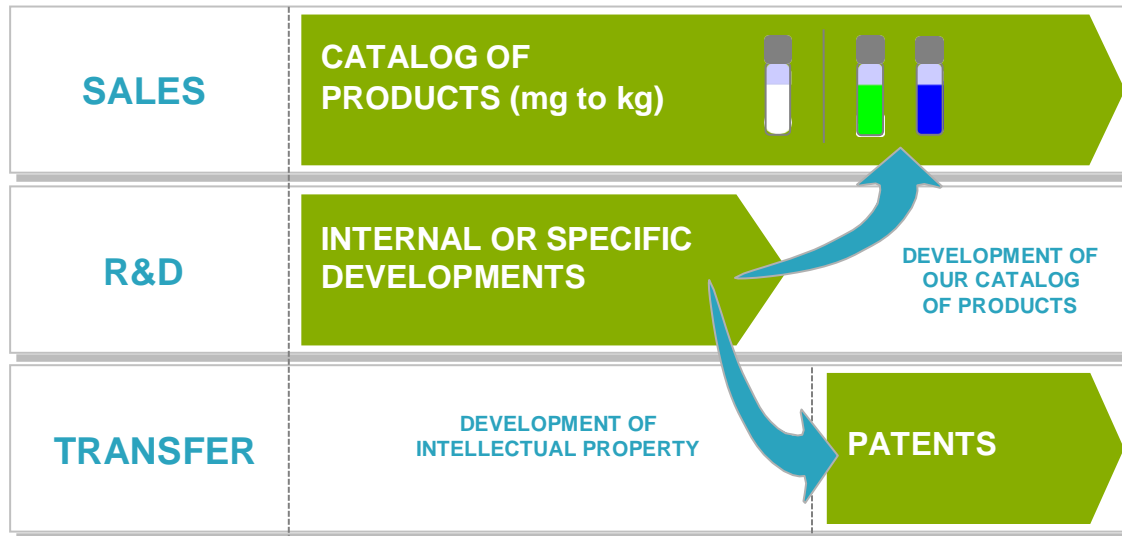


Results of the Immunization

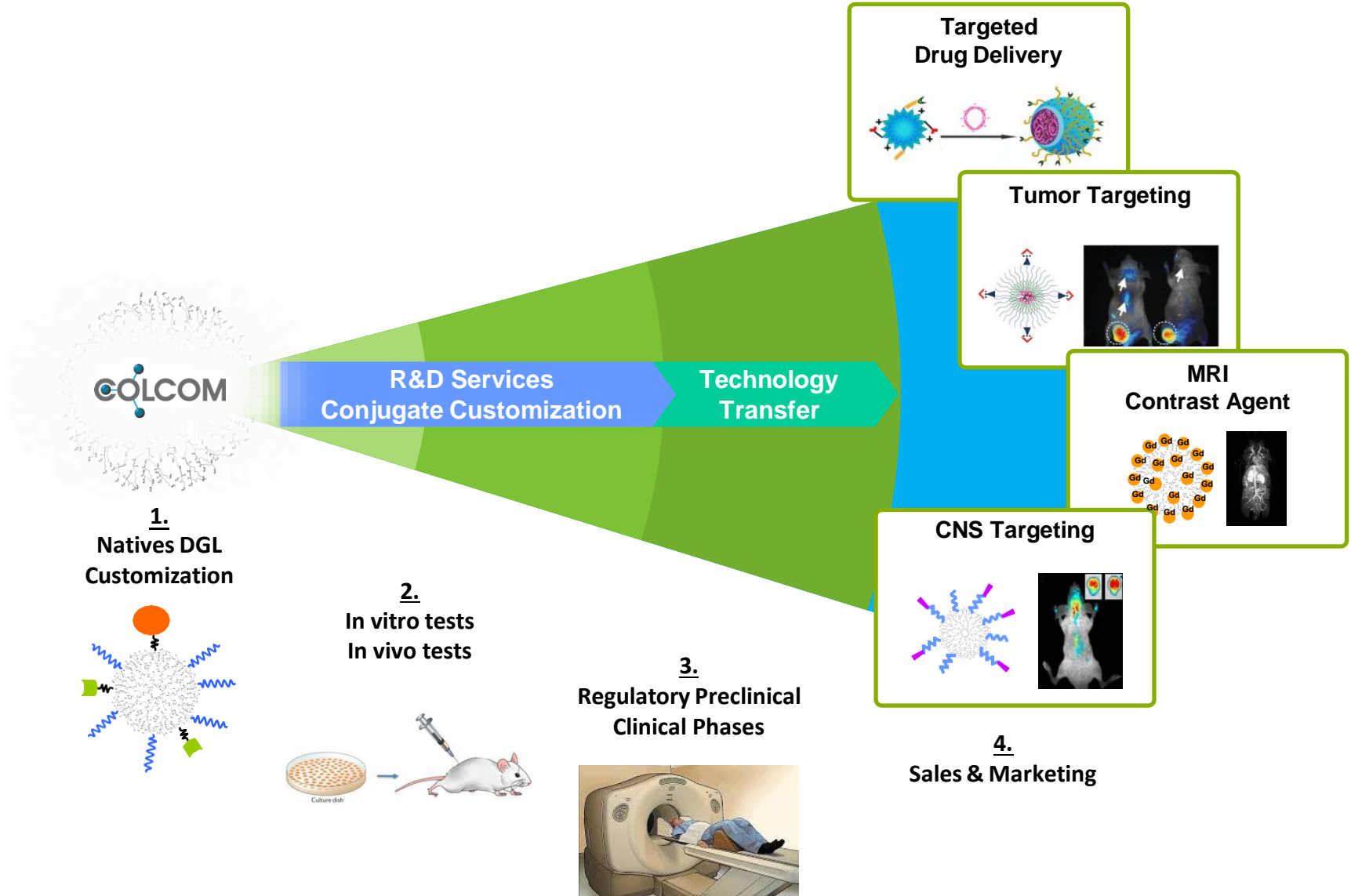


Intramuscular injection

Business Model



Colcom's business model





شكرا ا لنتباهكم

Thank you for your attention