



NanoSpain
Conf  2017
March 07-10, 2017 — San Sebastian (Spain)

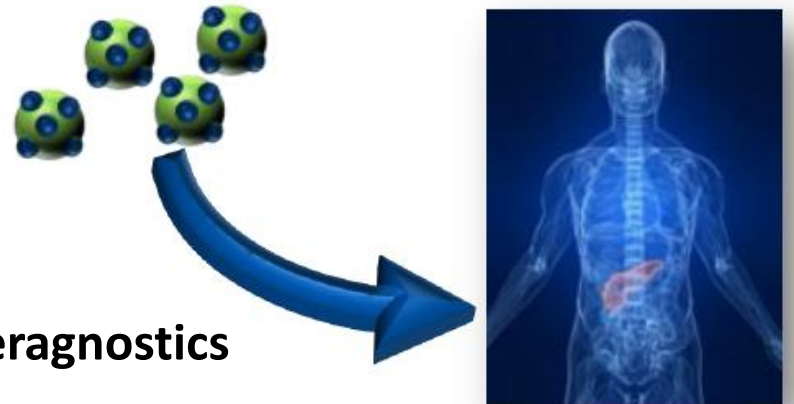
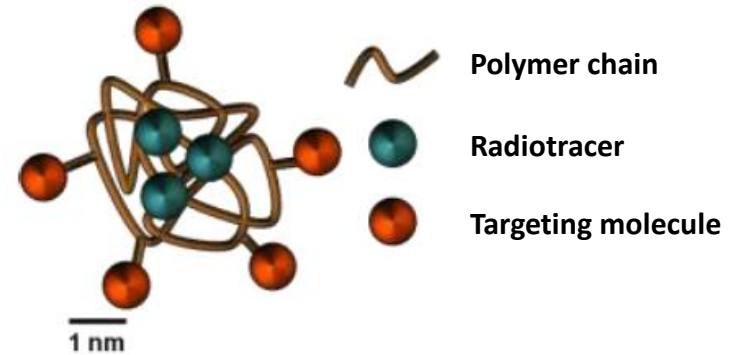
IK4  CIDETEC
Research Alliance

Poly(methacrylic acid)-based single-chain
polymer nanoparticles for targeting and
imaging pancreatic tumors *in vivo*

Marco Marradi

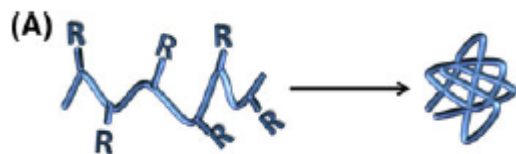
Pancreatic cancer

- 5th cause of death from cancer in Europe in 2012
- estimated 78,000 deaths (6.2% of total)
- low 5-year survival rate (around 5%)
- Late detection and 85% unresectable
- Limitation of current imaging systems for early detection, accurate staging and post-therapy monitoring

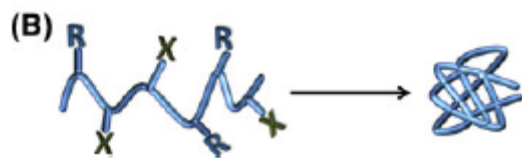


Nanotechnology-based approaches for new theragnostics

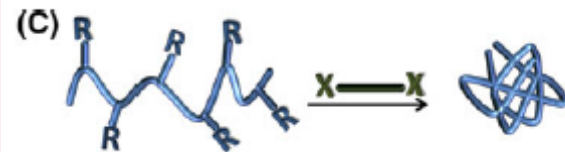
FUNCTIONAL POLYMER NANOPARTICLES



Intrachain homocoupling



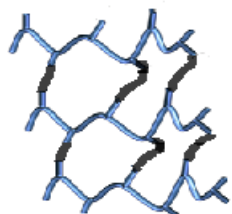
Intrachain heterocoupling



Crosslinker-induced intrachain collapse

Drawbacks:

- **Ultra-dilute** reaction conditions
(no viable for **large scale synthesis**)
to avoid inter-molecular cross-linking

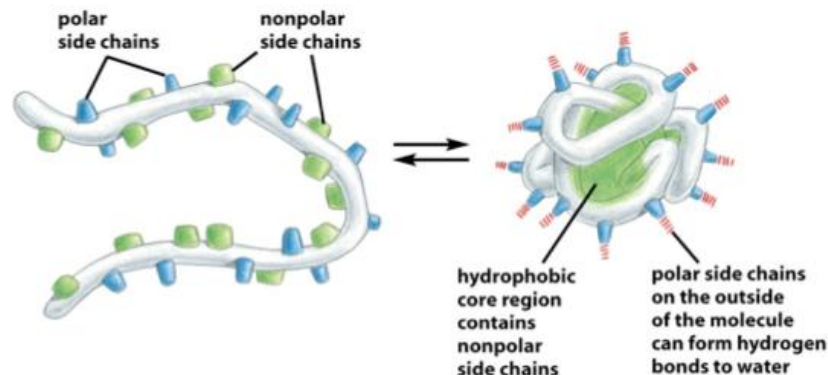


Inter-crosslinking

- **High temperatures**
- Use of **organic solvents**
- Use of **metal catalysts**

Advantages:

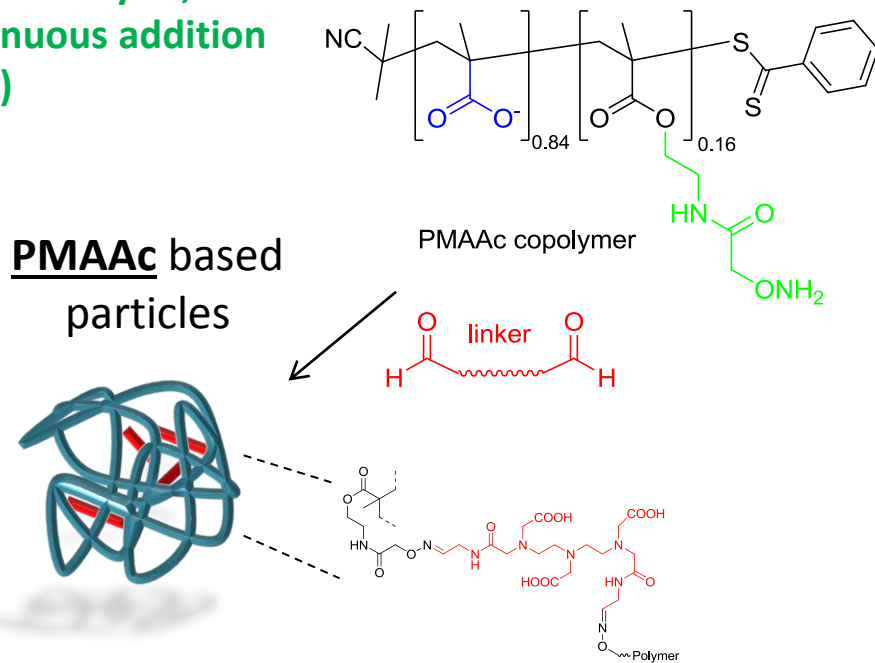
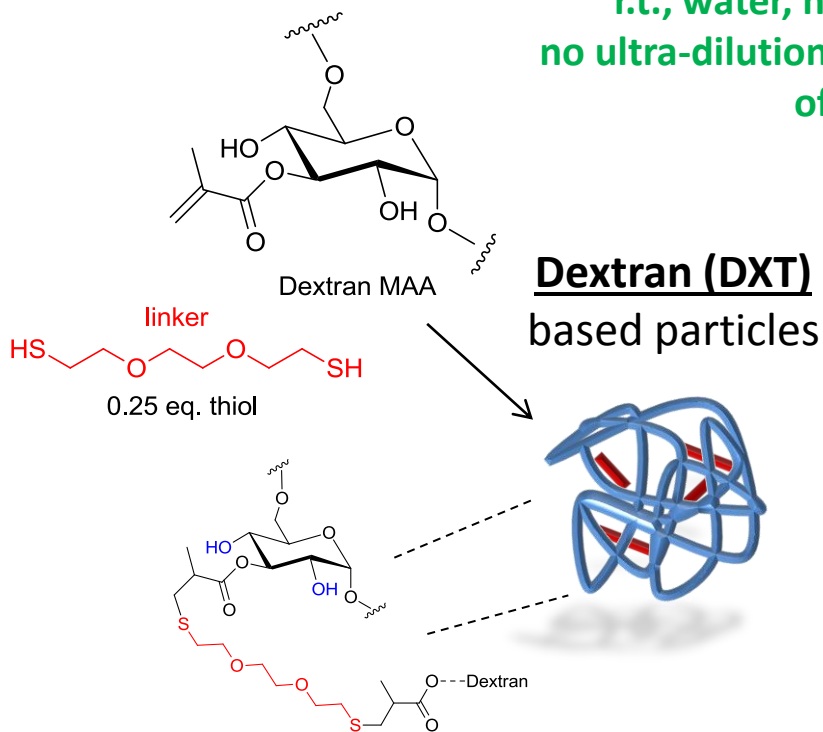
- Vast choice of **precursors** (biocompatible, polyfunctionalizable, ...)
- Control in size (from 200nm down to 5nm)
- Mimicking the protein folding

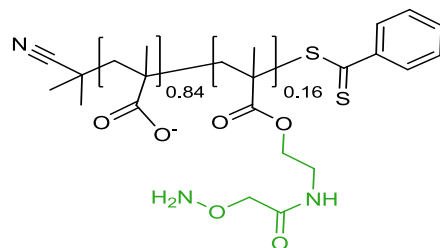


A process for preparing water-dispersible SCPNs: **versatility**

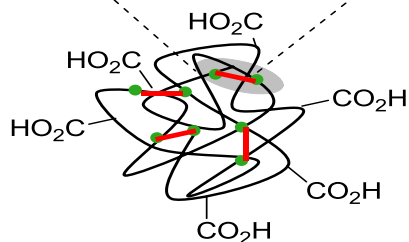
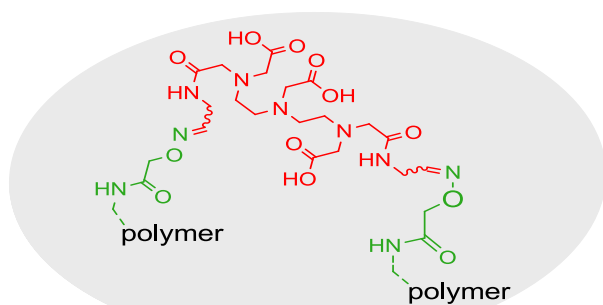
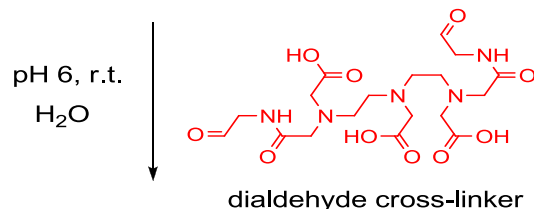
Wide range of polymer particles have been obtained based on natural and synthetic polymers.

**r.t., water, no metal catalysts,
no ultra-dilution (continuous addition
of linker)**

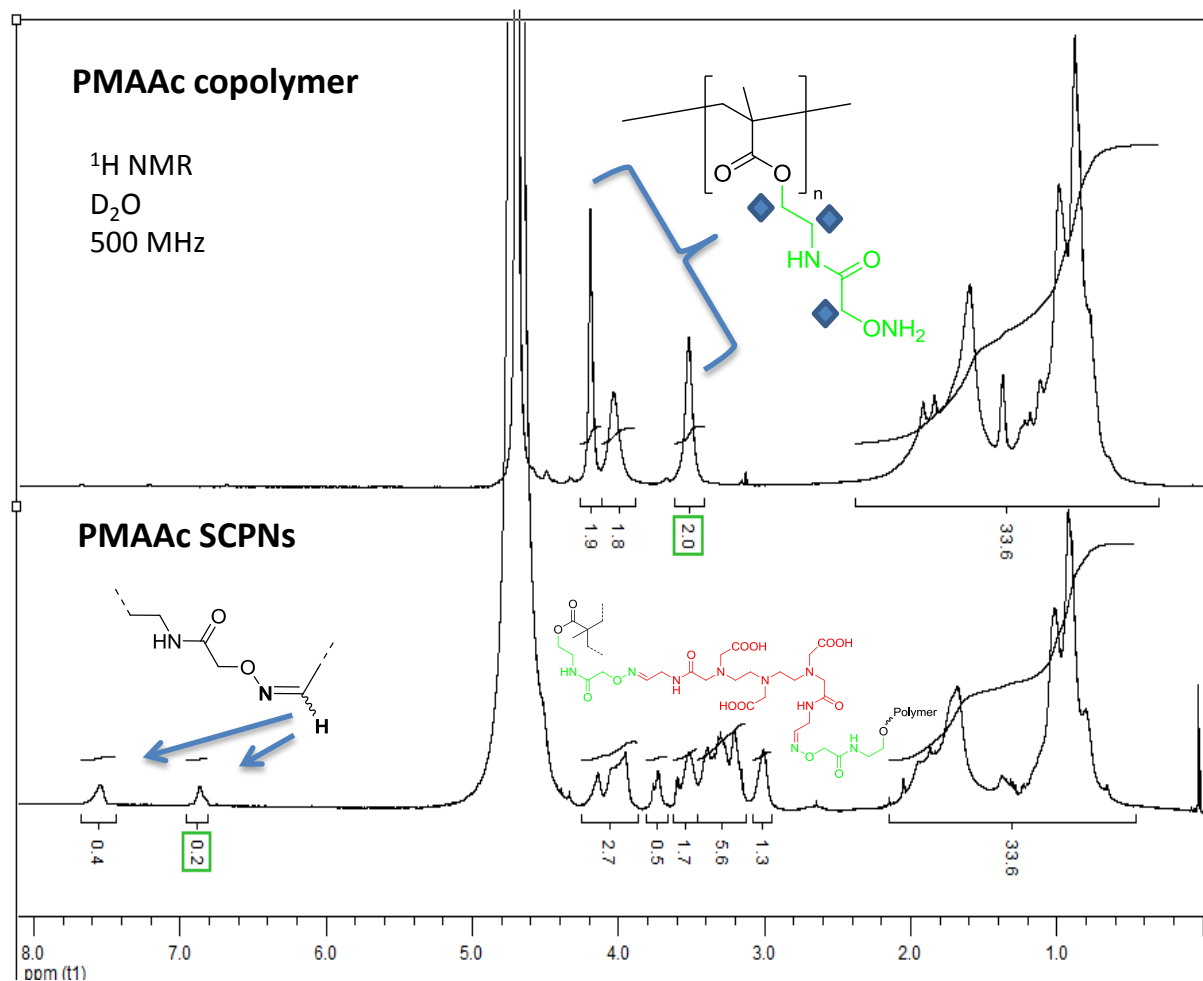
Marradi et al. *J. Mater. Chem. B*, 2017, 5, 1143Marradi et al. *Biomacromolecules*, 2016, 17, 3213



PMAAc copolymer



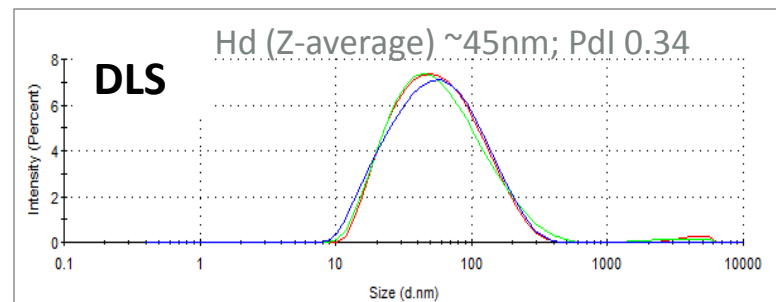
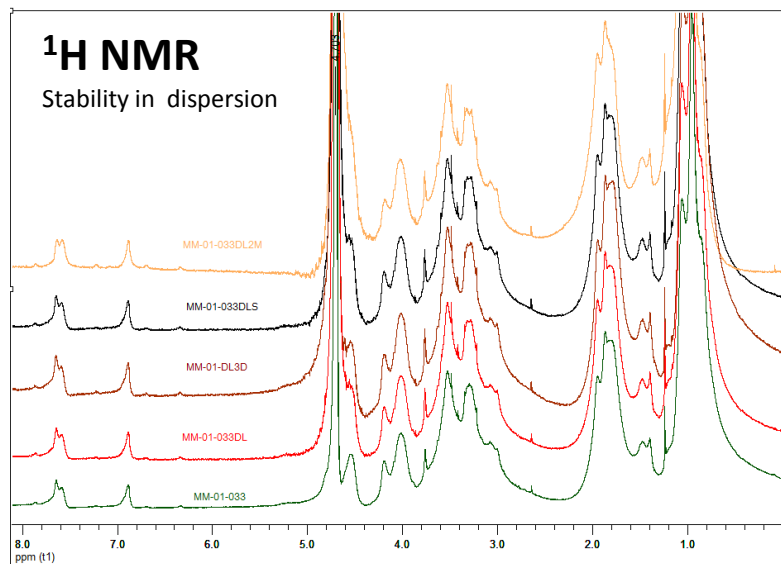
PMAAc SCPN



The *O*-alkyloximes are stable to mild reducing agents
2.2 < pH < 3.7 PRECIPITATION

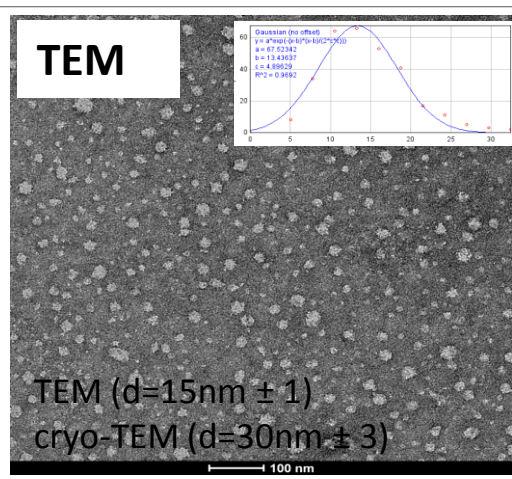
^1H NMR

Stability in dispersion

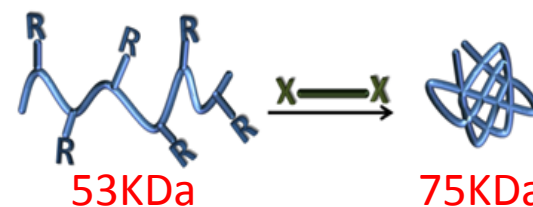
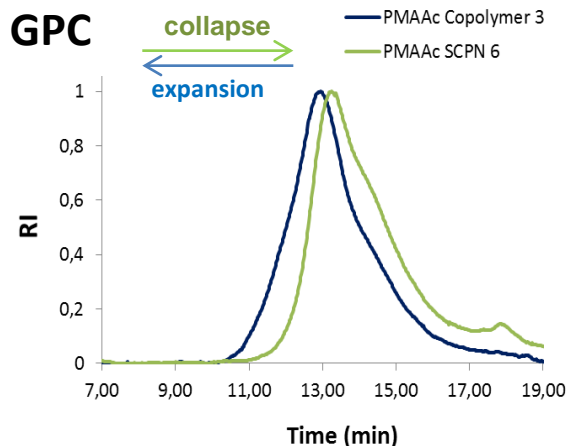


Z-pot (NaCl 1mM, pH 7): -40 mV

TEM



GPC



Sample (2 mg/mL, H ₂ O) filtered (0.2 μm)	Diffusion Coeff. (D)
Precursor-polymer	408 ± 4
Single chain-nanoparticle	474 ± 5

Viscosizer - Proof of collapse

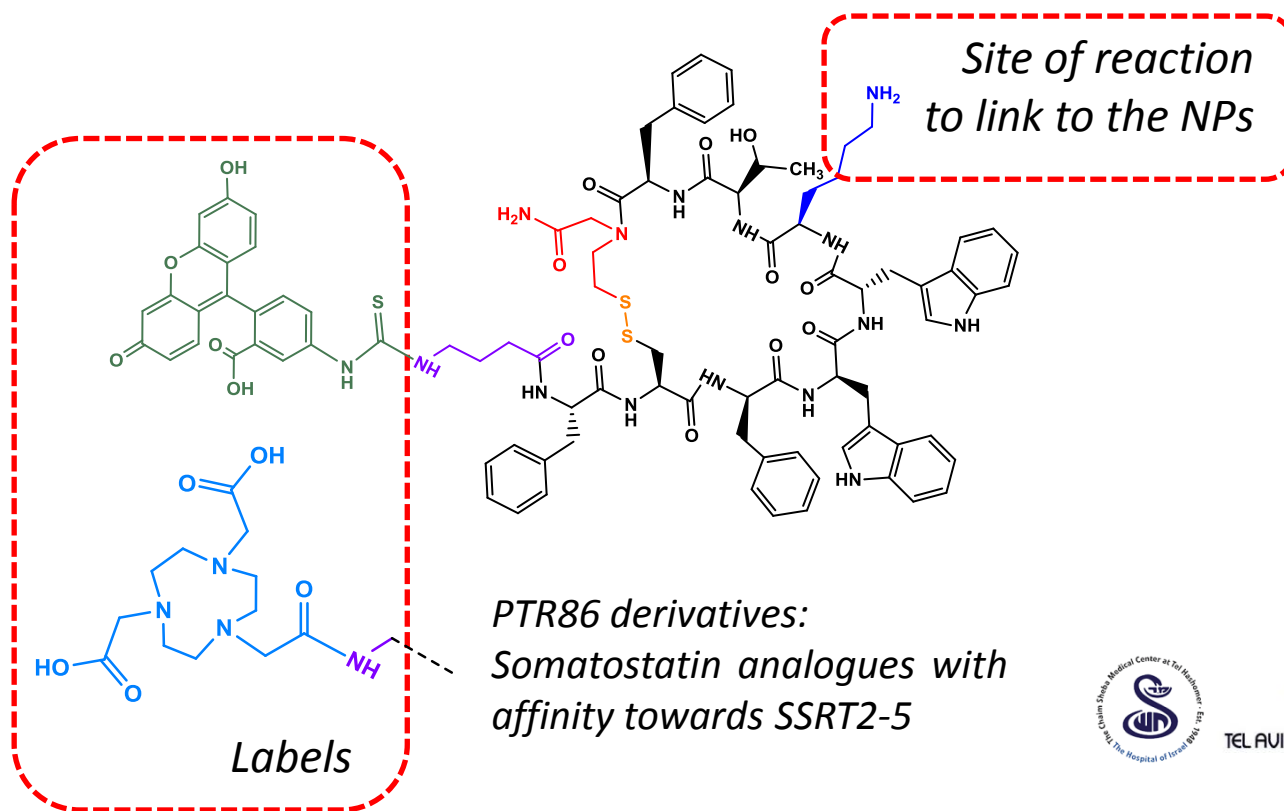
Indication of collapse?

Somatostatin analogue (peptide PTR86)

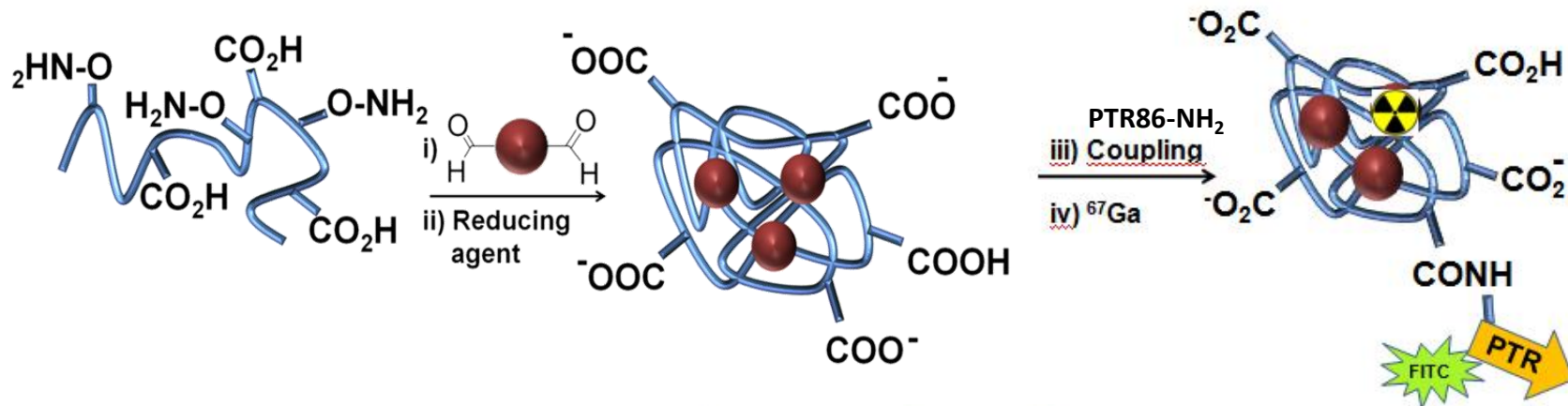
Somatostatin receptors are over-expressed in a variety of malignant tumors.

FITC-PTR86

PTR86-NOTA



Functionalization: Targeted and radiolabeled PMAAc-SCPNs



Quantification of PTR86 loading
by fluorescence spectroscopy

Fluorescent imaging
with targeted peptides

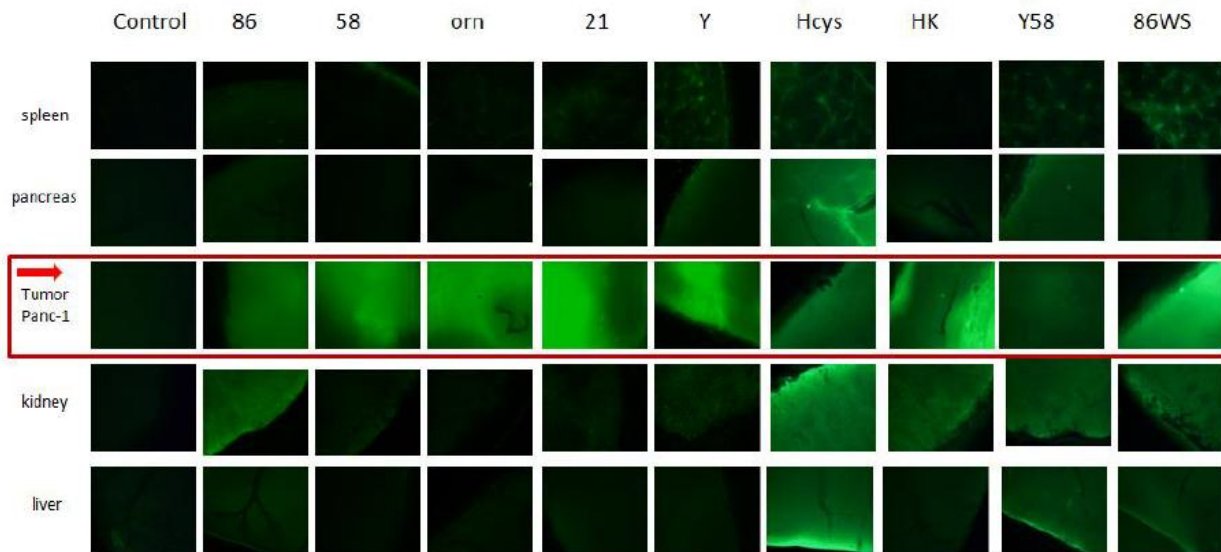
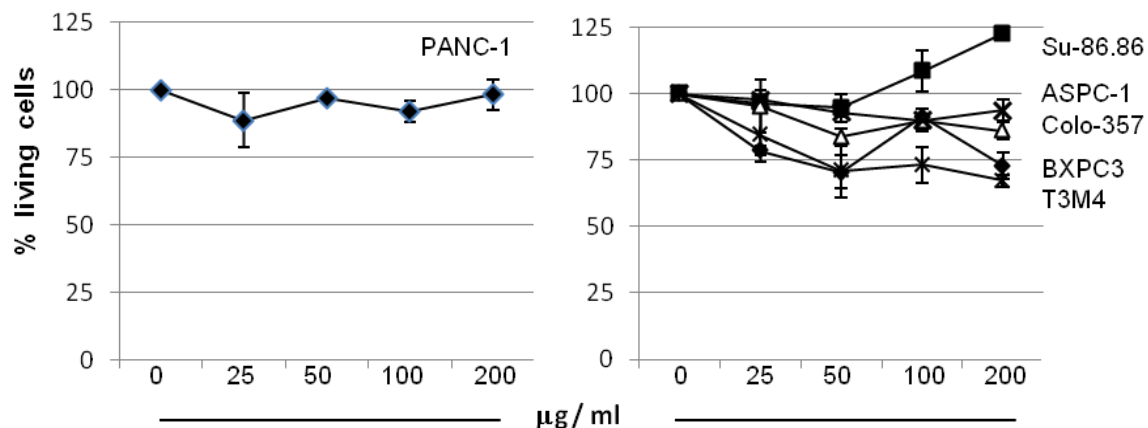


Figure 5.5. Signal intensity in the different organs (namely, spleen, pancreas, tumour, kidneys and liver) for the different SSTR ligands assayed. PTR-58 was selected for subsequent *in vivo* investigations using nuclear imaging.

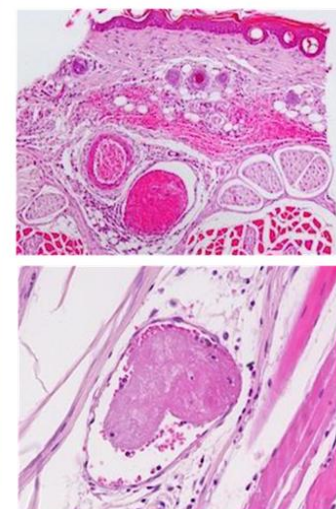
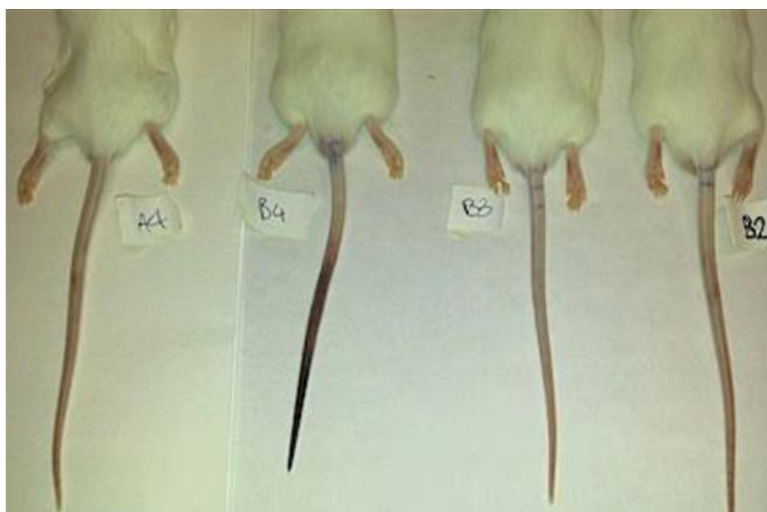
In vitro cytotoxicity to assess the safety of SCPN

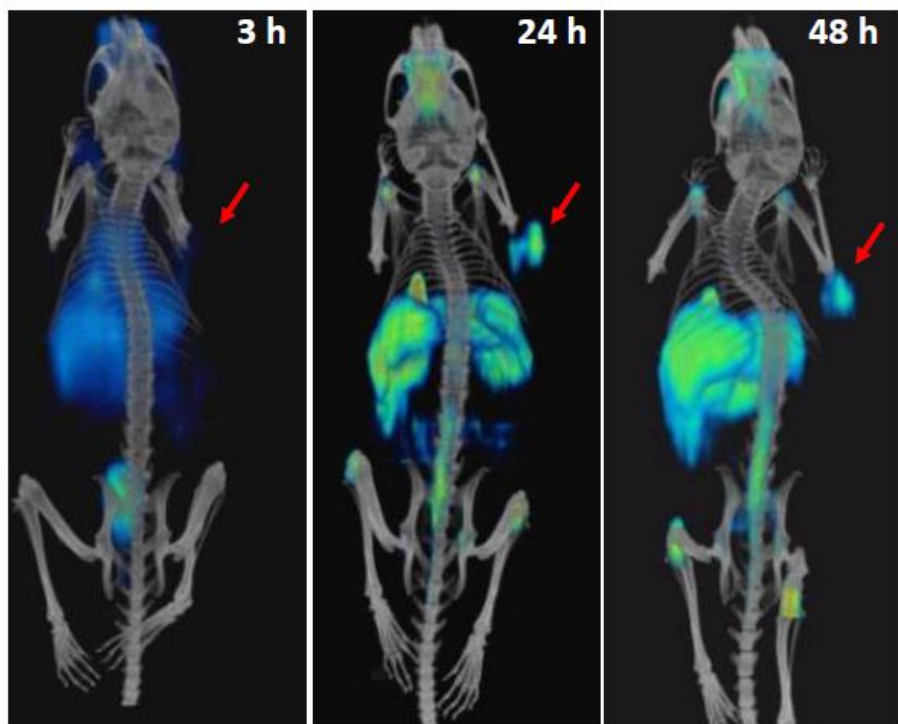
Toxicity was tested in 6 different pancreatic adenocarcinoma cell lines.



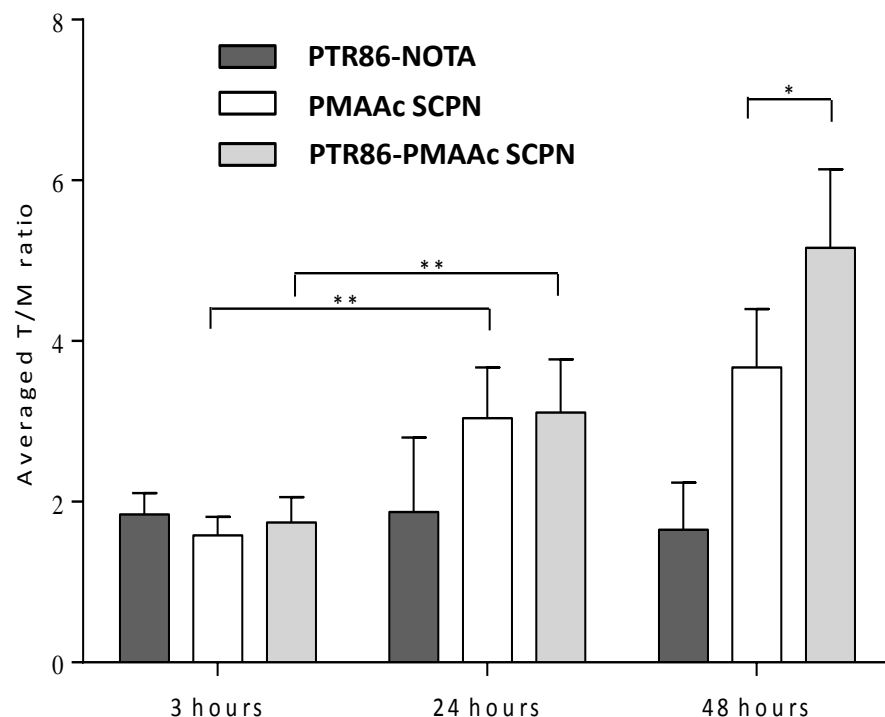
In vivo toxicity studies in animals

Dose: i.v. (10 ml/kg physiological saline) of PMAAc SCPNs (12,5 mg/kg and 100 mg/Kg) and sacrificed 24 hours after treatment. One (B4) out of five mice treated with 12,5 mg/kg had dark and necrotic tail.





SPECT-CT images of mice bearing subcutaneous human pancreatic ductal adenocarcinoma (PANC-1)
Dose: i.v. injection of 1 mg/Kg of SCPNs



Accumulation of the SCPNs in the tumor expressed as **tumor-to-muscle ratio**

- A new **synthetic route** has been established for the synthesis of PMAAc-SCPNs in **water**.
- Simultaneous incorporation of the **targeting** peptide PTR86 (receptor specificity) and the radionuclide ^{67}Ga for SPECT **imaging** was achieved.
- The results obtained in this study indicate higher retention of the SCPNs in the tumor when these were decorated with the PTR86 peptide, leading to higher tumor-to-muscle ratios as determined from *in vivo* images.

Collaborators:



Biomaterials Unit (CIDETEC)

Funding:



ETORTEK
biomaGUNE'15

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POSTER n°7

NanoPilot EU project: A Pilot plant for the production of Polymer based Nanopharmaceuticals in Compliance with GMP

www.nanopilot.eu



A Pilot Plant for the Production of Polymer-based Nanopharmaceuticals in Compliance with GMP

INTRODUCTION

What is NanoPilot?

NanoPilot is an EU funded (H2020) research project that brings together the expertise of 9 partners to set-up a pilot plant operating under GMP for the production of nanopharmaceuticals.



NanoPilot aims to accelerate the development of nanomedicine, currently in its infancy within the pharmaceutical sector, bringing into operation a flexible and adaptable pilot plant for the production of small GMP batches suitable for clinical trials.

AN INDUSTRIAL NEED TO COVER

Why NanoPilot?

The traditional business model of 'Big Pharma' has evolved to an Open Innovation (OI) model, in-licensing technology from academia or SMEs. Academia and innovative SMEs have become key players in the first stage of the development and proof-of-concept studies.



There is a high potential of innovation in SMEs/industries working in nanomedicine. However, in most cases clinical validation is still required and production in quantity and quality (GMP) needed remains a challenge. In this context, it is urgently needed to provide those SMEs with the tools that can help them to validate their technologies.

THE NANOPILOT CONSORTIUM



THE CONSORTIUM

9 partners have joined forces to guarantee the successful outcome of the proposed project.

2 INDUSTRIES
8. Syntex
9. Chemtrix

3 SMEs
5. Micronit
6. Meijon
7. Spinverse

4 RESEARCH GROUPS

1. IK4-CIDETEC
2. National University of Ireland, Galway
3. University of Santiago de Compostela
4. ADERA-UTZA

OBJECTIVES

What to expect from NanoPilot?

Three different nanopharmaceuticals will be produced during the project. The plant will integrate cutting-edge characterization techniques and microfluidics to control the production processes.

At the end of the project the pilot plant will be fully operating under GMP conditions, addressing some critical industrial needs:

- Production of small GMP batches for clinical trials.
- Flexibility and adaptability to prepare wide variety of nanopharmaceuticals.
- Specialization that guarantees the high quality of the products.

THREE DIFFERENT NANOPHARMACEUTICALS PRODUCTION



A RNAi-based nanomedicine for ocular pain treatment



A nanovaccine for HIV treatment



Hyaluronan particles for interstitial cystitis/painful bladder syndrome

CUTTING-EDGE CHARACTERIZATION TECHNIQUES AND MICROFLUIDICS INTEGRATION



Microreactors



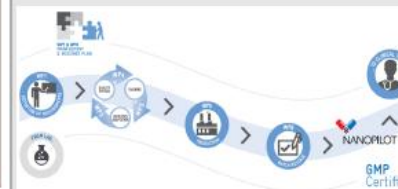
Microfluidics chip



Particle size fractionation in AAF

WORK PACKAGES

Project step by step



WP1: Definition of nanopharmaceuticals/design GMP production processes.

WP2: Adaptation of the facilities to a pilot plant working in compliance with GMP.

WP3: Training system implementation.

WP4: Quality system implementation.

WP5: Validation of GMP manufacturing processes and production.

WP6: Shipping and batch release.

WP7: Business and dissemination plan.

WP8: Management.





ESKERRIK ASKO
MUCHAS GRACIAS
THANK YOU VERY MUCH
MERCI BEAUCOUP
DANKESCHÖN