Chitosan-coated gold nanorods for the early detection of high-grade bladder cancer residual disease

Filippo Capancioni[1], Silvia Tortorella[1], Erica Locatelli[1], Emanuela Bua[1] Chiara Venegoni[2], Alessandra Caliendo[3], Irene Locatelli[2], Alessia Di Coste[2], Simona Camorani[3], Jithin Jose[4], Laura Cerchia[3], Massimo Alfano[2], Mauro Comes Franchini[1].

1) **Department of Industrial Chemistry "Toso Montanari", University of Bologna**, Via P. Gobetti 85, 40129 Bologna (Italy); 2) Division of Experimental Oncology/Unit of Urology; URI; IRCCS San Raffaele Scientific Institute, Milan, Italy; 3) Institute of Experimental Endocrinology and Oncology "Gaetano Salvatore", National Research Council, 80131, Naples, Italy;
4) FUJIFILM Visualsonics Inc., Amsterdam, The Netherlands.

INTRODUCTION

Thiolated chitosan (Chit-SH) is a biopolymer derived from chitin, with broad range of beneficial properties for biomedical applications[1], including the possibility of integration into nanosystems for the selective detection and treatment of tumors[2]. **Gold nanorods (GNRs)** are a class of biocompatible nanomaterials suitable for tumour detection and treatment, thanks to their tunable optical absorption in the so called «biological window [3]». The high affinity between gold and thiol groups of Chit-SH can be exploited for obtaining an active nanosystem for **tumour targeting**, but the poor reproducibility in the scale-up of their synthesis constitutes an obstacle to their medical application .

AIM OF THE PROJECT

1) **Synthesis of thiolated chitosan-coated GNRs (GNRs@Chit)**
- Critical process parameters study and optimization
- Scale-up from lab to pilot scale (30L)

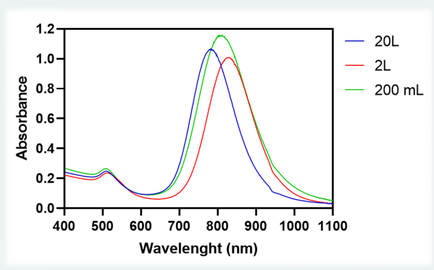
2) **GNRs@Chit functionalization with anti-integrin α5β1 aptamer (GNRs@Chit-Apt)**

3) **GNRs@Chit-Apt application in bladder cancer (BC) treatment**
- Photoacoustic imaging (PAI) for BC early detection (lesions <1mm)
- Preclinical tests

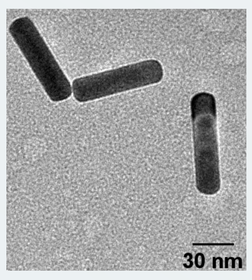
SYNTHESIS AND CHARACTERIZATION

STEP 1a - GNRs@CTAB

NIR-VIS

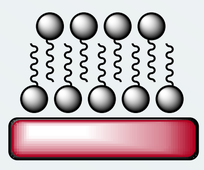


TEM



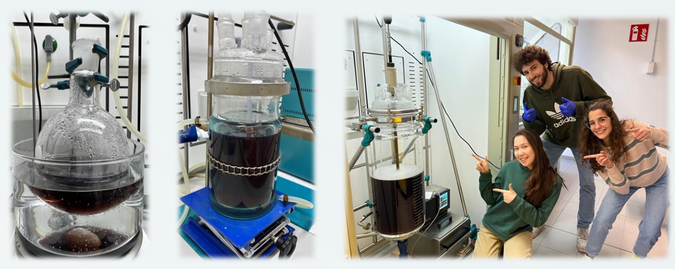
Gold precursor salts

Seed-mediated growth method



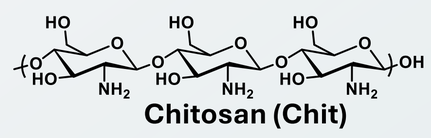
GNRs@CTAB

Scaled-up synthesis

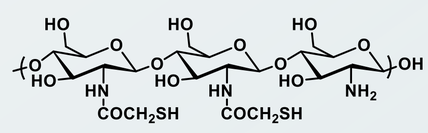


200 mL *2 L* *20 L*

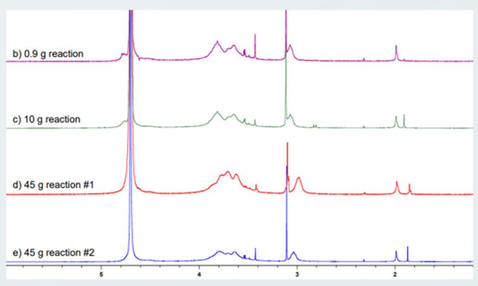
STEP 1b – Chit-SH



Synthetic modification with TGA



NMR



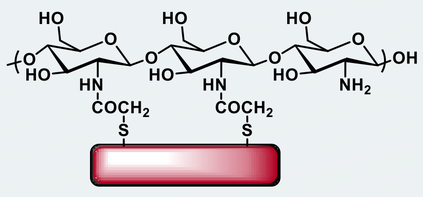
Scaled-up synthesis

2 L (40x scale-up)

Scaled-up dialysis

STEP 2 - GNRs@Chit

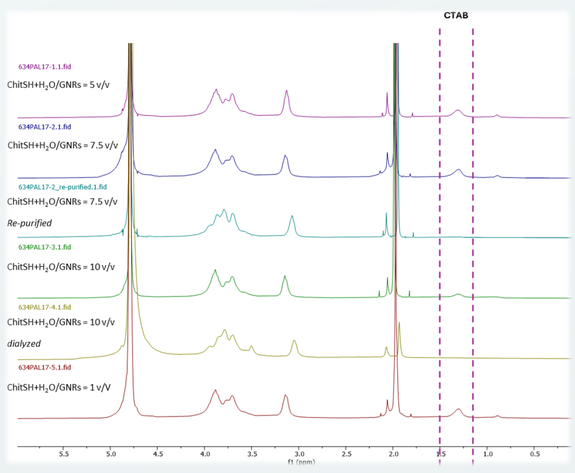
*GNRs coating with **Chit-SH***



GNRs@Chit

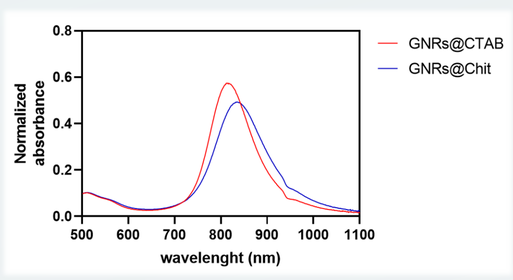
GNRs@Chit after basic precipitation

NMR



GNRs@Chit NMR spectra after purification

NIR-VIS

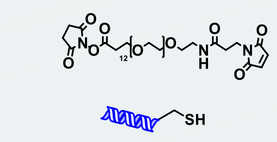


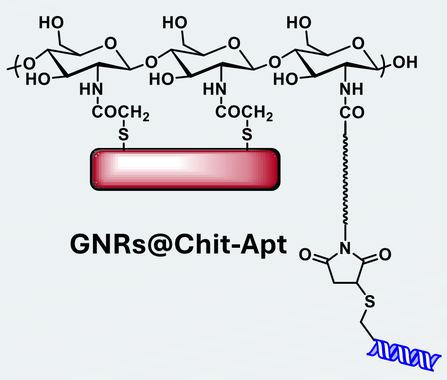
Centrifuged GNRs@Chit

STEP 3 - GNRs@Chit-Apt

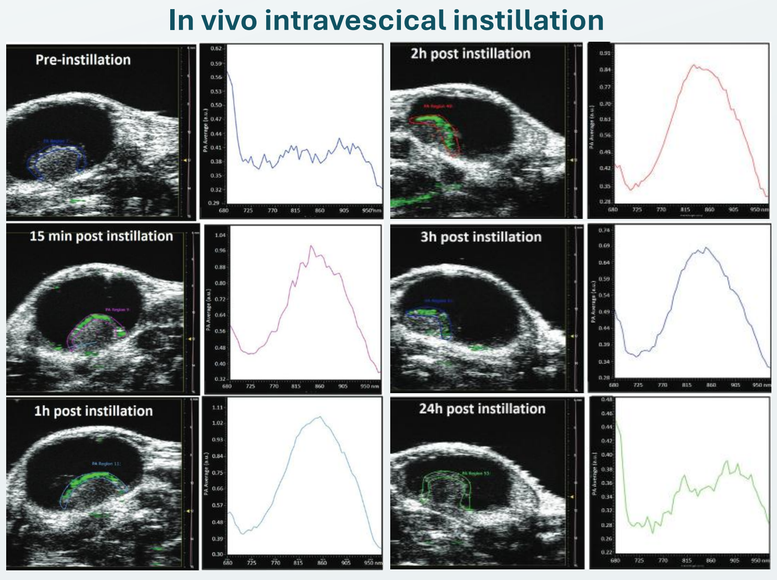
*1) Conjugation of **NHS-PEG$_{12}$-maleimide***

*2) Conjugation of **Apt***

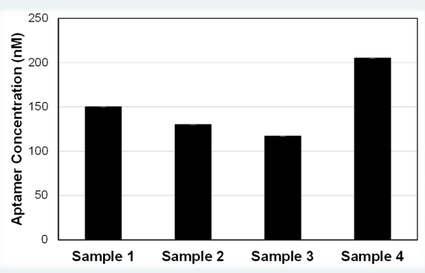




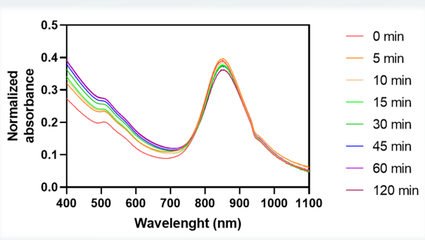
In vivo intravescical instillation



Apt concentration



Stability in human urine



CONCLUSIONS

- ✓ **Critical process parameters optimized** for each reactive step
- ✓ **Assessed reproducibility** for GNRs LLSPR in the biological window (770 nm<λ<850 nm)
- ✓ **Scaled-up synthesis** (7L of GNRS@Chit-Apt)
- ✓ Succesfull **preclinical applications** (imaging+therapy)

NEXT PERSPECTIVES

- **GMP production** of GNRs@Chit-Apt
- **Clinical tests on humans**

BIBLIOGRAPHY

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(2) Alchera, E.; Monieri, M.; Maturi, M.; Locatelli, I.; Locatelli, E.; Tortorella, S.; Sacchi, A.; Corti, A.; Nebuloni, M.; Lucianò, R.; Pederzoli, F.; Montorsi, F.; Salonia, A.; Meyer, S.; Jose, J.; Giustetto, P.; Franchini, M. C.; Curnis, F.; Alfano, M. Early Diagnosis of Bladder Cancer by Photoacoustic Imaging of Tumor-Targeted Gold Nanorods. *Photoacoustics* **2022**, *28*, 100400. https://doi.org/10.1016/j.pacs.2022.100400.
(3) Venegoni, C.; Tortorella, S.; Caliendo, A.; Locatelli, I.; Coste, A. D.; Locatelli, E.; Capancioni, F.; Bua, E.; Camorani, S.; Salonia, A.; Montorsi, F.; Jose, J.; Moschini, M.; Cerchia, L.; Franchini, M. C.; Alfano, M. Urine-Stable Aptamer-Conjugated Gold Nanorods for the Early Detection of High-Grade Bladder Cancer Residual Disease. *Adv Healthcare Materials* **2025**, *14* (10), 2403314. https://doi.org/10.1002/adhm.202403314.

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