7th scientific days of the CNRS network Molecular Imaging Agents - GDR AIM Joint meeting of GDR AIM & Mi2B

30.06.25 – 02.07.25

Nantes University

Booklet of Abstracts

PROGRAMME

MONDAY 30/06/25			
13h	**WELCOME COFFEE**		
14h-14h30	**OPENING ADDRESS**		
Sandrine HUCLIER (Chair of the conference) **Antoine GOULLET (Dean of Science of the Sciences Pole - Nantes University)** **Célia BONNET & Victor GONCALVES (coordinators of the GDR AIM)**			
Session 1 – Chair : Sandrine HUCLIER			
14h30-15h20	**PL01**	VUGTS Daniëlle	Translational ^{89}Zr-immuno-PET: from basic radiochemistry to drug imaging in early phase clinical trials
15h20-15h45	**C01**	PINEAU Julie	Antibody-based probes for targeted ^{64}Cu-PET imaging and ^{67}Cu-therapy
15h45-16h10	**C02**	CUSTODIO Camille	Evaluation of ligands for rational design of chelators for metallic astatine-211
16h10-16h40	**COFFEE break**		
Session 2– Chair : Célia BONNET			
16h40-17h05	**C03**	BODIO Ewen	Metal-substituted aza-BODIPYs: expanding the frontiers of NIR fluorophores for bimodal imaging
17h05-17h30	**C04**	SY Maryame	Mn-bispidine multimers as high-performance MRI contrast agents
17h30-17h55	**C05**	SALIEN Jelena	Comparison of NOTA-derivatives for nanobody PET imaging
18h-20h	**COCKTAIL DINNER – POSTER SESSION**		

TUESDAY 01/07/25			
Session 3 – Chair : Eléna ISHOW			
9h00-9h50	**PL02**	BONNET Sylvestre	Photochemical detection of metal ions and complexes for diagnostic and therapy
9h50-10h15	**C06**	DOAN Bich-Thuy	Multimodal MRI and optical imaging for the evaluation of theranostic magnetic complexes and nanocrystals in PDT-PTT phototherapy
10h15-10h40	**C07**	DA SILVA Isidro	Production of ^{165}Er
10h40-11h10	**COFFEE BREAK**		
Session 4 – Chair : Bich-Thuy DOAN			
11h10-11h35	**C08**	PEYROT Fabienne	Nanoformulated, innovative EPR probes for the evaluation of oxidative stress
11h35-12h00	**C09**	LEROUX Marion	Original fluorescent probes for use in microscopy on Caenorhabditis elegans
12h00-12h25	**C10**	CHARTIER Baptiste	New lanthanide complexes for in vivo luminescent biphotonic and MRI imaging
12h25-14h00	**LUNCH**		

TUESDAY 01/07/25			
Session 5 – Chair : Ali OUADI			
14h00-14h15	Brief overview of Mi2B network and Labex PRIME by Marie-Laure GALLIN-MARTEL & Denis DAUVERGNE		
14h15-15h05	**PL03**	FRELIN Anne-Marie	Dosimetry and biological effect studies in targeted radionuclide therapy @ IN2P3
15h05-15h30	**C11**	AMMOUR Luis	MAPSSIC, an implantable microprobe for beta+ neuroimaging of awake and freely moving rats
15h30-15h55	**C12**	OUADI Ali	Overview of IPHC – Strasbourg's activities in molecular imaging
16h00-16h45	**COFFEE BREAK – POSTER SESSION**		
16h45-17h30	**ROUND TABLE – Future of GDR**		
19h30	**GALA DINNER**		

WEDNESDAY 02/07/25			
Session 6 – Chair : Latifa RBAH-VIDAL			
9h00-9h25	**C13**	HADDAD Férid	Overview of radionuclide production at Arronax
9h25-9h50	**C14**	DEMARTINÉCOURT Théo	Daily elution, monitoring and performance assessment of radiochemically pure ^{44}Ti/^{44}Sc generator
9h50-10h15	**C15**	HERY Simon	PiB-derivative metal complexes for selective imaging of amyloid peptides
10h15-10h40	**C16**	COURSEYRE Marion	Bispidine radiotracers for ^{64}Cu/^{67}Cu theragnostics: proof of concept for multiple myeloma
10h40-11h10	**COFFEE BREAK**		
Session 7 – Chair : Sara LACERDA			
11h10-11h35	**C17**	CHEVALET Romain	Ruthenium speciation along the purification process of the daughter radionuclide
11h35-12h00	**C18**	MÉTIVIER Cassandra	CXCR4-targeted PET agents for solid tumors
12h00-12h20	**ORAL COMMUNICATION PRIZES – CLOSING ADDRESS**		
12h20-14h00	**LUNCH**		

LIST OF POSTERS

P01	ARIZTIA Julien	Thiol-reactive linkers: an alternative for radioimmunoconjugate development
P02	BANCHERI Louane	Preclinical evaluation of Lysyl oxidase-like 2 as a target for the nuclear imaging of idiopathic pulmonary fibrosis
P03	DIEBOLD Léa	Toward tumour theranostics : Hypoxia activation as a tool for therapy and diagnostic
P04	FAUDEMER Zélie	New PTAs for fluorine-18 radiolabelling
P05	HUCLIER Sandrine	Coupling DOTA-hexapeptide with Scandium-44/Copper-64 for early detection of brain amyloid deposits by PET imaging in Alzheimer's disease
P06	ISHOW Eléna	Mutimodal nanoassemblies for high-contrast diagnostics of cancer cells and bacteria
P07	SEQUIN Johanne	Evaluation of new imaging probes in the second transparency window (NIR-II) on the LIOPA platform
P08	SIRE Charline	^{18}F radiolabeling of new Zinc-responsive contrast agents
P09	TESTARD Clara	RNA-imaging: ^{11}C labelling of mRNA for PET imaging of new vaccines and therapies

Booklet of Abstracts

Plenary Lectures

Translational ^{89}Zr-immuno-PET: from basic radiochemistry to drug imaging in early phase clinical trials

Daniëlle VUGTS

Amsterdam University Medical Center, Amsterdam, The Netherlands

d.vugts@amsterdamumc.nl

^{89}Zr-immuno-PET has emerged as a powerful modality to visualize and quantify the *in vivo* distribution of biologicals such as monoclonal antibodies, antibody-drug conjugates and nanoparticles, informing on pharmacokinetics, tumor targeting and off-target uptake. Over the past ten years, new developments with regard to chelator chemistry, PET instrumentation and modelling have resulted in the use of ^{89}Zr-immuno-PET outside the oncology domain. In this presentation, the translational chain of ^{89}Zr-immuno-PET will be discussed and some translational examples presented.

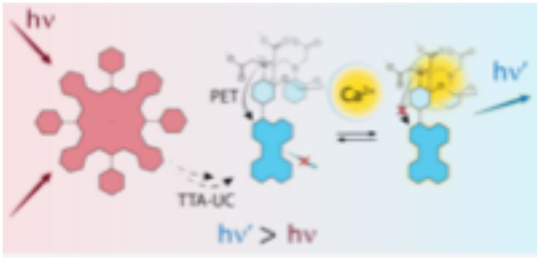
Photochemical detection of metal ions and complexes for diagnostic and therapy

Sylvestre Bonnet[1]

[1] *Leiden Institute of Chemistry, Leiden, The Netherlands*

bonnet@chem.leidenuniv.nl

Some metal ions such as Ca^{2+} are essential to health but difficult to observe in biological systems. One needs innovative photochemical methods to highlight their presence or quantify their concentration, which may reveal sickness. Other metal ions such as Ru^{2+} can be introduced in the body in the form of a prodrug for photoactivated chemotherapy. Following their fate and their excretion becomes essential to control the therapy and minimize the potential side effects of the treatment. In this presentation, I will present two stories. In the first one, Ca^{2+} ions are imaged via an upconversion system in the context of endothelial disfunction, a reversible, early form of vasculatory diseases. In the second story, I will describe how the first coordination sphere of ruthenium complexes influences their emission properties, and how histidines and methionine residues can be used to develop ruthenium-peptide conjugates for photoactivated chemotherapy that can be traced by emission imaging *in vivo*.



This work was supported by the European Union's Horizon 2020 Research and Innovation programme LOGICLAB under the Marie Sklodowska Curie grant agreement 813920.

References

[1] V. D. Andreeva, I. Regeni, T. Yang, A. Elmanova, M. Presselt, B. Dietzek-Ivansĭc,s. Bonnet, *J. Phys. Chem. Lett.* **2024**, *15*, 7430.

[2] Liyan Zhang,# Gangyin Zhao,# Trevor Dalrymple, Yurii Husiev, Hildert Bronkhorst, Gabriel Forn-Cuní, Bruno Lopes-Bastos, Ewa Snaar-Jagalska,* and Sylvestre Bonnet *ACS Central Science* **2024** *10*, 2294.

Dosimetry and biological effect studies in targeted radiotherapy @ IN2P3

Anne-Marie Frelin,[1,3] Rachel Delorme[1,3]

[1] *Grand accélérateur National d'Ions Lourds (GANIL), CEA/DRF-CNRS/IN2P3, Boulevard Henri Becquerel, 14076 Caen, France*
[2] *Univ. Grenoble Alpes, CNRS, Grenoble INP, LPSC-IN2P3, 38000 Grenoble, France*
[3] *on behalf of the GDR Mi2B*

anne-marie.frelin@ganil.fr

Many IN2P3 laboratories are involved in targeted radionuclide therapy (TRT), as evidenced by the recent structuring of IN2P3 Master Projects, with a focus on Targeted Alpha Therapy (TAT). In this modality, one of the teams' study topics concerns the optimization of treatments. This optimization must take into account complex dose distributions and biological effects mainly caused by large ranges of dose rate and dose deposition. These factors give rise to research projects that can be divided into dose and biological effect studies and address different objectives and different scales.

In radiotherapy, dose deposition is generally the first lever of action for treatment optimization and an important metric for treatment planning and evaluation. In TRT, dose distribution is closely linked to activity biodistribution and can vary significantly over space and time, as well as between patients. This makes personalized dosimetry desirable. However, temporal monitoring of biodistribution is a significant clinical challenge, complicating personalized dosimetry in TRT. Projects developing innovative gamma and Compton cameras are underway to address this issue and make personalized dosimetry achievable.

A second major challenge with high linear energy transfer particles is to take into account the multiscale biological effects, whether for treatment planning or evaluating outcomes. Radiobiology experiments are performed using different irradiation configurations, such as sealed and unsealed sources and accelerated beams, to study biological effects from the subcellular to the in vivo scale. Dosimetry methods and instruments are developed to adapt to different biological models and configurations of irradiation, providing accurate, reliable dose references that improve the reproducibility and comparability of experiments. These data will constrain cellular and multicellular biological effect models and identify the effects of the tumor environment. Ultimately, this will contribute to upgrading dose planning to treatment effect planning.

A brief overview of the various projects that contribute to these objectives will be provided during this presentation.

Booklet of Abstracts

Oral Communications

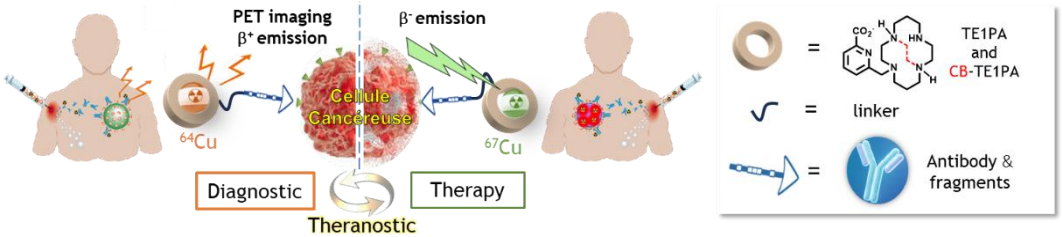
Antibody-based probes for targeted ^{64}Cu-PET imaging and ^{67}Cu-therapy

Julie Pineau[1,5], Dyhia Amrane[2,5], Patricia Le Saëc[1], Nicolas Joubert[2], Caroline Denevault-Sabourin[2], Nicolas Aubrey[3], Emmanuel Garcion[4], Michel Chérel[1], Nathalie Le Bris[5], Raphaël Tripier[5], Latifa Rbah-Vidal[1]

[1] *Nuclear Oncology team 2, CRCI2NA UMR INSERM 1307, Nantes University*
[2] *IMT team, EA 7501 GICC, University of Tours*
[3] *BioMAP team, ISP INRA UMR1282, University of Tours*
[4] *GLIAD team, CRCI2NA, UMR INSERM 1307, UMR CNRS 6075, University of Angers*
[5] *COSM team, UMR CNRS-UBO 6521, CEMCA, University of Brest*

julie.pineau@univ-nantes.fr

In oncology, early diagnosis and treatment adapted to a given pathology is essential for efficient care and survival of patients with limited side effects and reduced recurrence. In this context, the **TARGET'IN** project, involving the collaboration of five research group with complementary skills, propose to develop targeted diagnostic and therapeutic probes using antibodies as specific vector. Nuclear medicine allows to meet this need through the use of the matching radionuclide pair copper-64/copper-67. It is a single "all-in-one" theranostic object (therapy + diagnosis) able to *i)* diagnose tumors by PET imaging at their earliest stage thanks to specific targeting, carried out here with copper-64 (β^+, $t_{1/2}$=12.7 h) and *ii*) treat them by the same targeting process with copper-67 (β^-, $t_{1/2}$=62.0 h). This approach can be used for any type of cancer by changing the biological vector. In this consortium, we propose to work on breast cancer and glioblastoma to highlight this versatility and the contribution of the technique used. Until now, the stability of copper-based radiopharmaceuticals was the limiting factor in their use, particularly with the famous chelating agent DOTA. For this, ten years ago, the partner of Brest has developed chelating agents based on cyclam monopicolinate, known as TE1PA and CB-TE1PA, capable of forming a particularly stable and inert copper complex. [1,2] Our current work consists of finding the best combination [copper complex – linker – biological vector] against breast cancer or glioblastoma, using ^{64}Cu-immuno-PET imaging and envisaging ^{67}Cu-targeted radiotherapy. This presentation will focus on the chelating agents design and syntheses, antibody bioconjugation, ^{64}Cu-radiolabeling and *in vitro* studies.



References

[1] A.-S. Navarro, T. Le Bihan, P. Le Saëc, N. L. Bris, C. Bailly, C. Saï-Maurel, M. Bourgeois, M. Chérel, R. Tripier, A. Faivre-Chauvet, *Bioconjugate Chem.* **2019**, *30*, 2393–2403.

[2] J. Pineau, L. M. P. Lima, M. M. L. Roy, S. Marionneau-Lambot, M. Cordier, P. L. Saëc, J. R. Zeevaart, C. H. S. Driver, A. Faivre-Chauvet, N. L. Bris, R. Tripier, *Chem. Commun.* **2023**, *59*, 888–891.

Evaluation of ligands for rational design of chelators for metallic astatine-211

Camille Custodio,[1] Samuel Mador,[2] Pierre Rouanet,[2] Ferid Haddad,[3] Nicolas Galland,[2] Gilles Montavon,[4] Lu Liu,[5] François Guérard[1]

[1]CRCI2NA, Nantes, France *[2]CEISAM, Nantes, France*
[3]ARRONAX, Saint-Herblain, France *[4]Subatech, Nantes, France*
[5]IPHC, Strasbourg, France

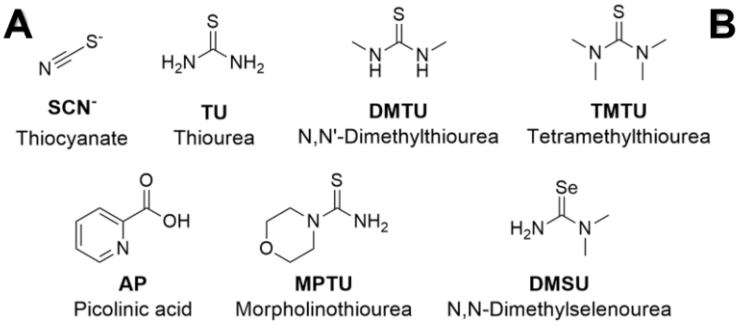
camille.custodio@etu.univ-nantes.fr

Objectives: Despite being a promising candidate for targeted alpha therapy, ^{211}At applications have been limited by the in vivo instability of compounds labeled by formation of carbon-astatine aryl bonds.[1] Recent advances have focused on the radionuclide's halogen reactivity, leaving room for development of labeling strategies using its metallic properties.[2] In this work, we evaluated interactions between the metallic species At^+ and soft Lewis bases, as groundwork for the rational design of chelating agents for At(I).

Methods: A series of small monodentate ligands (Figure A) were studied by liquid/liquid extraction in a biphasic system (toluene/0.1 M $HClO_4$).[3-4] Changes of distribution of ^{211}At activity after equilibration in presence of increasing ligand concentration were indicative of interactions between astatine and the ligand. Fitting of the experimental data allowed to determine both the stoichiometry and stability constant of the formed complexes. Extracted log β values were compared to results obtained from ligand exchange DFT calculations.

Key findings: Stability constants demonstrated stronger interactions of astatine with thiourea-based ligands compared to anionic ligands. Preliminary results on a selenious ligand, N,N-dimethylselenourea, showed significant improvement of the stability constant of the formed complexes. (Table B) Further investigation is ongoing to confirm the superiority of selenious over sulfur-based ligands. Highest affinity ligands will be selected to form preorganized structures as the next step for rational design of chelators for metallic ^{211}At.

A



B

Ligand	Logβ_1	Logβ_2
TU	5.6 ± 0.3	9.0 ± 1.6
DMTU	5.8 ± 0.5	9.5 ± 1.5
TMTU	6.0 ± 0.5	9.3 ± 0.6
MPTU	6.2 ± 1.4	10.8 ± 2.1
DMSU	7.0 ± 0.3	12.4 ± 0.12

References

[1] Vanermen, M. et al. *EJNMMI Radiopharm. Chem*. **2024**, *9*, 69.

[2] Guérard, F et al. *Acc. Chem. Res*. **2021**, *54*, 3264.

[3] Liu, L. et al. *Inorg. Chem*. **2022**, *61*, 13462.

[4] Champion, J. et al. *Inorg. Chim. Acta.* **2009**, *362*, 2654.

Metal-substituted aza-BODIPYs: expanding the frontiers of NIR fluorophores for bimodal imaging

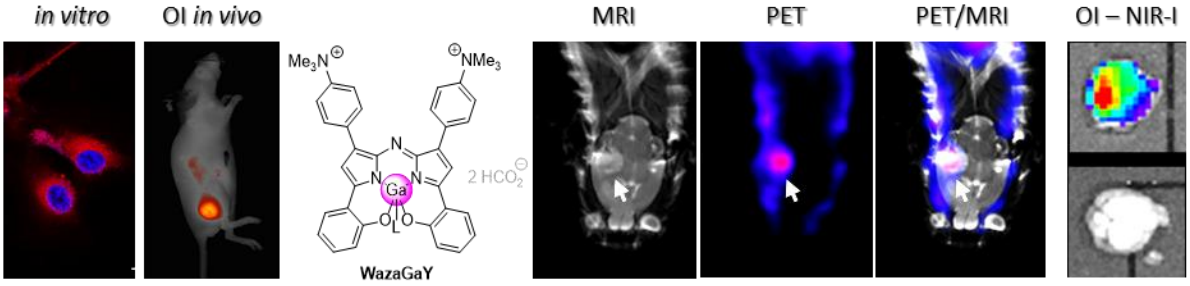
Charlotte Cave,[1] Mohamed Bendellaa,[2] Mathieu Moreau,[1] Pierre-Simon Bellaye,[3] Annika Sickinger,[4] Ludmilla Verrieux,[5] Shaymaa Al Shehimy,[4] Bertrand Collin,[3] Pierre Le Gendre,[1] Arnaud Fihey,[5] Benoit Busser,[2,6] Christophe Bucher,[4] Boris Le Guennic,[5] Olivier Maury,[4] Lucie Sancey,[2] Ewen Bodio[1,6,7*]

[1] ICMUB, Université de Bourgogne, CNRS UMR 6302, F-21000 Dijon, France.
[2] Univ. Grenoble Alpes, INSERM U 1209, CNRS UMR 5309, IAB, 38000 Grenoble, France.
[3] Service de médecine nucléaire, CGFL, Dijon Cedex 21079, France.
[4] Univ Lyon, ENS de Lyon, CNRS UMR 5182, Laboratoire de Chimie, F-69342 Lyon, France.
[5] Univ Rennes, CNRS, ISCR, UMR 6226, 35000 Rennes, France.
[6]Institut Universitaire de France Institut Universitaire de France (IUF) 75005 Paris, France
[7]Nantes Université, CNRS, CEISAM, UMR 6230, F-44000 Nantes, France

ewen.bodio@univ-nantes.fr

Aza-boron-dipyrromethenes (aza-BODIPYs) have emerged as a class of highly promising fluorophores, comparable to rhodamines and cyanines, due to their ease of synthesis, high stability, and strong fluorescence in the near-infrared (NIR-I, 700–900 nm). These properties make them ideal candidates for *in vivo* fluorescence imaging and surgical guidance. Some of our developed aza-BODIPYs extend emission into the NIR-II window (1000–1700 nm),[1] enhancing imaging resolution.

While structural modifications have focused on tuning the substituents and conjugation of aza-BODIPYs, little attention has been given to the boron center itself. In this work, we explore the impact of replacing boron with metal centers, leading to the synthesis and characterization of novel aza-Metal-DIPY complexes. [2] Special emphasis will be placed on a gallium derivative that has been water-solubilized and designed as a bimodal probe for both NIR fluorescence and PET imaging. We will present its biodistribution in tumor-bearing mice (U87MG, IGROV1, A375), its application in fluorescence-guided surgery, [3] and its potential for PET imaging through radiolabeling with [$^{6\ 8}$ Ga]. These results highlight the promise of metal-substituted aza-BODIPYs for advanced bioimaging applications.



References

[1] A. Godard *et al. Bioconjugate Chem.* **2020**, 31, 1088.

[2] A. Godard *et al. Inorg. Chem.*, **2023**, 62, 5067.

[3] M. Bendellaa *et al. J. Med. Chem.* **2024**, *67*, 18, 16635.

Mn-bispidine multimers as high-performance MRI contrast agents

Maryame Sy[1], Cyrille Charpentier[1], Agnès Pallier[2], Éva Tóth[2], Loïc J. Charbonnière[1] and Aline M. Nonat[1]

[1] *Equipe de Synthèse pour l'Analyse (SynPA), Institut Pluridisciplinaire Hubert Curien (IPHC), UMR 7178, CNRS/Université de Strasbourg, ECPM, France*
[2] *Centre de Biophysique Moléculaire, CNRS UPR 4301/Université d'Orléans, Rue Charles Sadron, F-45071 Orléans, France.*

maryame.sy@unistra.fr

In this study, we aim to develop Mn(II)-based contrast agents for MRI. Previous work has shown that bispidine ligand are capable of coordinating Mn(II) forming kinetically inert, monohydrated complexes with high relaxivity values (r_1 = 3.51 to 4.89 $mM^{-1}s^{-1}$ at 60 MHz, 25 °C).[1] To further increase the relaxivity and compete with the new generation of gadolinium-based contrast agent,[2] we synthesized dimeric, trimeric and tetrameric Mn(II)-bispidine complexes based on MnL_1 (Figure A) which exhibited the best properties (r_1 = 4.14 $mM^{-1}s^{-1}$, $t_{1/2}$ > 140 days in the presence of 50 equivalents of Zn(II) at 37 °C). [1c] The relaxivities of these complexes were determined (Figure B) and the biodistribution of the two trimeric complexes were studied (Figure C). Both complexes demonstrated a rapid renal clearance and a low liver accumulation. Finally, a highly rigid tetrameric complex was designed and showed a high relaxivity of 7.62 $mM^{-1}s^{-1}$ at 37 °C and 60 MHz (Figure B) [3]. This complex was selected as the lead candidate for further developments.

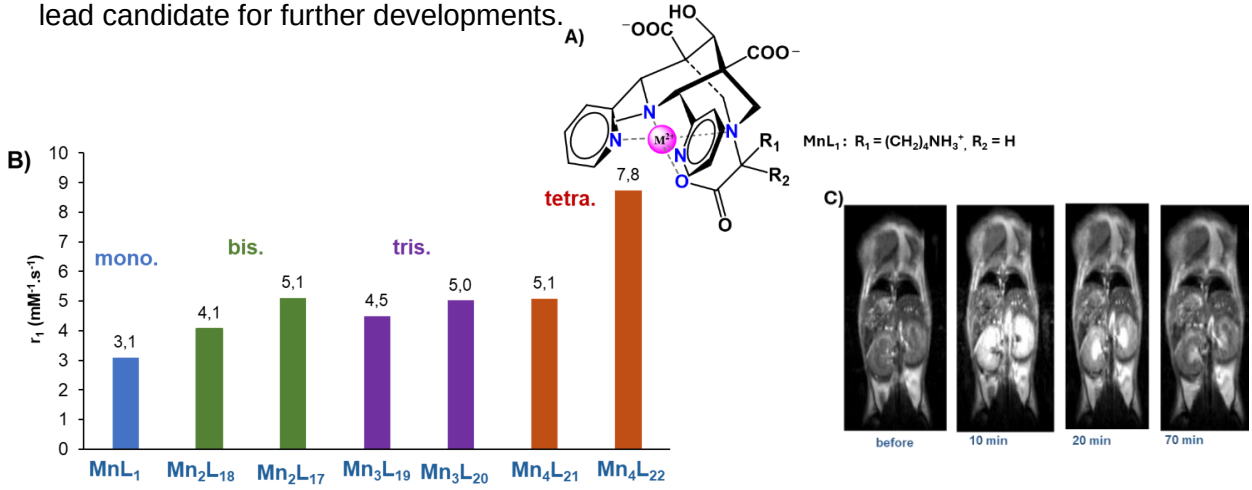


Figure: A) General structure of Mn-bispidine complexes. B) Relaxivities of the complexes at 60 MHz and 37 °C. C) *In vivo T1-weighted MR images before (9.4 T) injection of a trimeric Mn complex and at 10, 20 min and 70 min post-injection (0.06 mmol kg^{-1}).*

References

[1] *a*) D. Ndiaye, M. Sy, W. Thor, *Chem. Eur. J.* **2023**, *29*, e202301880; b) M. Sy, D. Ndiaye, I. Da Silva, *Inorg. Chem.* **2022**, *61*, 13421; c) D. Ndiaye, M. Sy, A. Pallier, *Angew. Chem. Int. Ed.* **2020**, *59*, 11958; d) L. Charbonnière, M. Sy, E. Jakab Toth, WO2023126336A1.

[2] a) J. Lohrke, M. Berger, T. Frenzel, *Invest. Radiol.* **2022**, *57*, 629 ; b) I. Maimouni, C. Henoumont, M.-C. De Goltstein, *Invest. Radiol.* **2025**, *60*, 234.

[3] A. Nonat, L. Charbonnière, M. Sy, WO2023126336A.

Comparison of NOTA-derivatives for nanobody PET imaging

Jelena Saliën,[1,2] Sam Floru,[1] Michiel Van Hove,[1] Tessa De Pauw,[1] Romane Vizier,[2] Jessica Bridoux,[1] Victor Goncalves,[2] Franck Denat,[2] Sophie Hernot[1]

[1] *Vrije Universiteit Brussel (VUB), Brussels Health Campus/Faculty of Medicine and Pharmacy, Molecular Imaging and Therapy, Laarbeeklaan 103, 1090 Brussels, Belgium*

[2] *Univ. Bourgogne Europe, Institut de Chimie Moléculaire de l'Université de Bourgogne (ICMUB), UMR CNRS 6302, 9 Avenue Alain Savary, 21078 Dijon, France*

Jelena.Salien@vub.be

To this day, [^{18}F]FDG is the clinical standard for cancer diagnosis using PET imaging.[1] This tracer can highlight metabolically active tissues such as cancer cells, but also inflammation and fibrotic tissue. Since radiochemotherapy often results in the latter, a tracer able to specifically image cancerous tissue for treatment follow-up would be an added clinical value.

In this study, a Nanobody™ (Nb) targeting CEACAM5 was modified with NOTA-chelators enabling complexation with gallium-68, a positron-emitting radionuclide whose half-life perfectly matches the fast biodistribution of Nbs. Carcinoembryonic antigen (CEACAM5) is an unchallenged molecular marker overexpressed in > 80% of colorectal cancers, and therefore an interesting target. To investigate the influence of the bioconjugation handle on the pharmacokinetic properties of these targeted PET-tracers, four different NOTA-derivatives containing the same macrocyclic chelator and different reactive functions for bioconjugation were compared in a preclinical study (Figure 1A). *In vitro*, the tracers' quality controls, labelling with gallium-68, stability and affinity for the target were assessed. Afterwards, their biodistribution was evaluated *via in vivo* PET/CT imaging in a subcutaneous mouse model.

In conclusion, we synthesised and preclinically evaluated four Nb-tracers carrying different NOTA-derivatives. Despite their high structural similarity, differences in

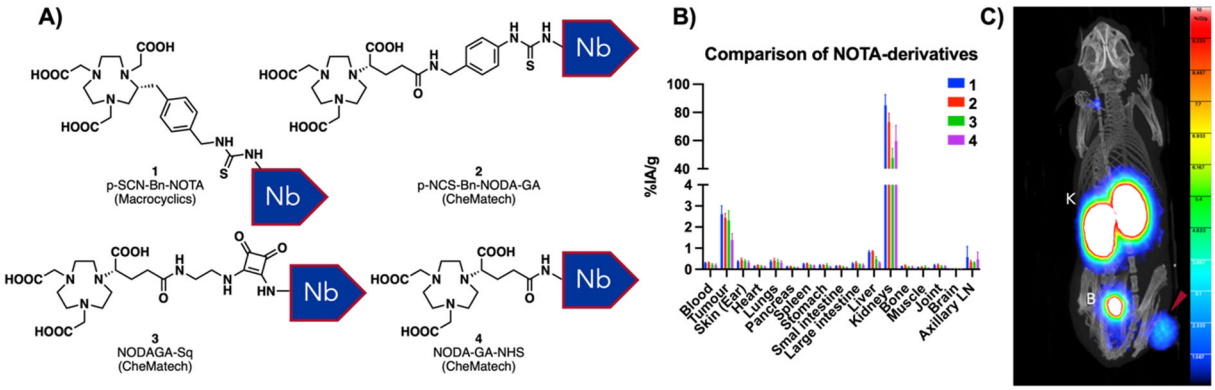


Figure 1 A) Structures of four Nb-based PET tracers bearing different NOTA-derivatives; B) *ex vivo* biodistribution of all tracers showing tumour uptake and renal clearance; C) PET/CT image of tracer **3**, tumour indicated in red

[1] J. Fletcher, B. Djulbegovic, H. Soares, *J. Med. Chem.* **2008**, *49 (3)*, 480-508

Multimodal MRI and optical imagings for the evaluation of theranostic magnetic complexes and nanocrystals in PDT-PTT phototherapy.

Y. Wang[1,4], S. Boumati[1], J. Gateau[5], C. Ménager[3], A. Sour[6], V. Heitz[6], C. Richard[2], P. Mesdom[4], J. Seguin[2], N. Mignet[2], G. Gasser[4], Y. Corvis[2]*, BT. Doan[1]*

[1] SEISAD, ICLEHS, ENSCP ChimieParisTech, Université PSL, CNRS, Paris
[2] VICT, UTCBS, Université Paris Cité, CNRS, Inserm, Paris
[3] PHENIX, Sorbonne Université, Paris
[4] ICB, ICLEHS, ENSCP, ChimieParisTech, Université PSL, CNRS, Paris
[5] LIB, Sorbonne Université, CNRS, INSERM, Paris
[6] UNISTRA, Université de Strasbourg
Contact: bich-thuy.doan@chimieparistech.psl.eu

Introduction : Among biomedical imaging methods, MRI is an advantageous technology for its 3D properties and contrast in vivo. Optical imaging is a complementary modality that enables biodistribution studies at microscopic cellular level as well as at macroscopic whole-body scale. The overall aim of the project is to develop and evaluate a new therapeutic strategy using in vitro to in vivo theranostic formulations of molecular and nanoparticles which can be activated by light for photodynamic synergestic PDT and/or photothermal PTT therapy, and guided by MRI and optical imaging for dynamic drug monitoring.
Building on previous work in theranostics [1-3], we aim to present multimodal MRI and optical bioimaging methods enabling to develop new formulations of light-activated photoactive antitumor drugs in magnetic nanoparticles or molecules.
A first example of photoactive molecules and a second example of nanocrystals for MRI imaging and NIR optical imaging will be presented.
Methods: We propose to evaluate first magnetic and fluorescent porphyrins and second fluorescent and magnetic IO- porphyrin PS as nanocrystals suspension in aqueous environment [2] by relaxivities studies at 7T. Then, the physicochemical, phototherapeutic and bioimaging features of these formulations have been evaluated in vitro to in vivo [2-4].
Results: We have carried out the synthesis and physicochemical characterization of functionalized Gd porphyrins and thermosensitive nanocrystals of porphyrin PSs for PDT, with IO nanoparticles for PTT and MRI. Fluorescent and magnetic porphyrins, and monodispersed and stable formulations NPs (150 nm) were obtained. For NPs, we have developed light-induced hyperthermia experiments, including evaluation of ferromagnetic nanocrystals able to induce hyperthermia for enhanced bioavailability of photosensitisers leading to PDT and PTT. We have evaluated their MRI and optical imaging efficiency by measuring relaxivity (r_2 = 210 $mM^{-1}.s^{-1}$) and fluorescent properties, respectively, as well as 89 to 38% photo conversion efficiency.

Discussion/Conclusion: We have proposed bimodal imaging methods in vitro to in vivo to conceive or evaluate innovative bimodal theranostics for *in vitro* evaluation of molecular magnetic PS complexes and NCs PS/ IO nanoparticles to generate PDT-PTT therapies. The magnetic PS complexes and NCs are promising nanotheranostic triggerable scaffolds with efficient MRI and/or fluorescent features for image guided therapy. Preclinical developments with subsequent *in vitro* and *in vivo* therapeutic formulations combined with toxicology testing will open a possible translation to the clinic.

References

[1] Venditto VJ. et al. *Adv. Drug Deliv. Rev.* **2013**, 65, 80-8.
[2] Castillo Henríquez L, … A, Mignet N, Corvis Y. Small. **2024**, 20(25):e2306054.
[3] Boumati S…. Doan BT. ACS Appl Bio Mater. **2023,** 6(11):4791-4804.
[4] Thébault CJ, … Mignet N., Menager C., Doan BT. J. *Control. Release* **2020**, 322, 137.

Production of ^{165}Er

P.A.Ellison,[1] J.W. Engle,[1] D. Baux,[2] T. Stora,[3] C. Duchemin,[3] M. Jensen,[4]

E. Nigron,[5] A.Guertin,[5] F. Haddad,[5] I.A. Da Silva[2]

[1] Department of Medical Physics, Madison, USA
[2] CEMHTI ,CNRS, Orléans, France
3 CERN Medicis, Genève, Switzerland
4 DTU-NUTECH, Roskilde, Denmark
5 Arronax/Subatech, Nantes, France

Idasilva45160@cnrs-orleans.fr

^{165}Er has a half-life of 10.36 h sufficient for production (irradiation, separation, radiolabeling, and injection). It emits low-energy X-rays (47 - 55 keV) useful for preclinical bimodal MRI/SPECT imaging. Moreover, it has a higher number of Auger emissions (AE) (a lower X-ray fluorescence yield, no conversion electrons (CE) or gamma rays). This lanthanide is a good candidate for Auger therapy dosimetry studies

For its production, various process have been investigated in collaboration with other laboratories : irradiation of naturally monoisotopic holmium targets produces ^{165}Er using 6 - 16 MeV protons via ^{165}Ho(*p,n*)^{165}Er [1] using a biomedical cyclotron (Madison, WI, USA) or 10 – 20 MeV deuteron irradiation via ^{165}Ho(*d,2n*)^{165}Er[2] with a research cyclotron (Orleans, FR). But owing to the short half-life of ^{165}Er (10.36h), the need of logistics of transport and its required skills (radiochemistry) and separation time (4 - 5h), research laboratories without on-site cyclotron and radiochemical expertise in separation exclude the use of this AE emitter. But ^{165}Er (10.36h) by decay of ^{165}Tm (30.1h) could reduce these drawbacks. For that, ^{165}Tm will be produced using ^{165}Ho(α,x)^{165}Tm with a 35 – 70 MeV alpha particles (Nantes, FR) or by spallation (CERN, CH) [3]. It's the concept of the ^{165}Tm/^{165}Er generator [4] developed at time in our laboratory.

All methods will be presented and compared.

References

[1] a) I. Da Silva, TR. Johnson , JC. Mixdorf et al.. *Molecules*. **2021**, *26(24),*1. b) GJ. Beyer, SK. Zeisler, DW Becker, *Radiochim. Acta.* **2004**, *92(4-6)*, 219. c) F. Tárkányi, A. Hermanne, S. Takács, et al., *Nucl Instrum Methods Phys Res, Sect B,* **2008**, *266(15)*, 3346.

[2] a) J. Vaudon, L. Frealle, G. Audiger, et al., *Instruments*. **2018**, *2(3)*, b) F. Tárkányi, A. Hermanne, S. Takács, et al., *Nucl Instrum Methods Phys Res, Sect B*. **2008**, *266(16),*3529., c) A. Hermanne, R. Adam-Rebeles, F. Tarkanyi, et al., *Nucl Instrum Methods Phys Res, Sect B,* **2013**, *311*, 102.

[3] a) DE. Fiaccabrino, P. Kunz, V. Radchenko, *Nucl Med Biol*, 2021, *94-95*, 81, b) C. Duchemin, TE. Cocolios, K. Dockx, et al., *Appl Radiat Isot*., **2021,** *178*., 109983, c) Y. Blumenfeld, T. Nilsson, P. Van Duppen., *Phys Scr*. **2013**, *152*.

[4] Z. Baranyai, G. Tircsó, F. Rösch., *Eur J Inorg Chem*., **2020,** *(1)*,36.

Nanoformulated, innovative EPR probes for the evaluation of oxidative stress

Aleksandra Rančić[1], Nikola Babić[1], Samuel Ajayi[2], Guillaume Force[1], Rabah Gahoual[2], Claire Mader[3], Sonia Lajnef[1], Caroline Roques[2], Fabienne Peyrot[1,4]

[1] *Université Paris Cité, CNRS, Laboratoire de Chimie et de Biochimie Pharmacologiques et Toxicologiques, Paris*
[2] *Université Paris Cité, CNRS, Inserm, Unité des Technologies Chimiques et Biologiques pour la Santé, Paris*
[3] *Université Paris Cité, BioMedTech Facilities, Paris*
[4] *Sorbonne Université, INSPE de Paris, Paris*

caroline.roques@u-paris.fr ; fabienne.peyrot@u-paris.fr

Introduction: Oxidative stress is associated with numerous chronic and acute conditions with high societal impact but there is a lack of non-invasive methods for its *in vivo* detection. Electron Paramagnetic Resonance (EPR) is considered the gold standard by the American Heart Association for measuring redox physiology. However, the availability of EPR probes constitutes a major limitation to its development. We recently designed innovative, protected hydroxylamines as redox-sensitive probes along with tailored nanovectors.

Methods: Masked hydroxylamines were synthesized from corresponding nitroxides by introducing an innovative, esterase-sensitive protecting group. To encapsulate the hydrophobic probes, the optimal oil phase and surfactants were determined and nanoemulsions were prepared using microfluidization. Characterization of nanoemulsions was carried out using DLS, cryo-TEM and HPLC-UV. The proof of concept of oxidative stress detection was obtained by EPR *in vitro* on glioblastoma cells. The cytotoxicity of probe-loaded nanoemulsions was evaluated on endothelial cells. The *in vivo* toxicity, biodistribution and EPR signal of a protected hydroxylamine probe as well as those of a nitroxide analogue were assessed in healthy mice.

Results: Prototype probes were synthesized on a gram scale. Optimized, probe-loaded nanoemulsions obtained displayed a hydrodynamic diameter around 140 nm, with a homogenous distribution and were stable at room temperature for up to 3 months. Moreover, EPR probes submitted to microfluidization (a high energy process of homogenization) were not degraded through the process. The encapsulation efficiency and drug loading of the probe were estimated at 99% and 33% respectively. *In vitro*, when the prototype probe was incubated with U87 cells along with a redox-cycling hydroxyquinone, we observed a dose-dependent EPR signal increase relative to control in treated cells. Preliminary *in vivo* experiments conducted on healthy mice did not show any acute toxicity of the nanoformulated probes for both the nitroxide and the protected hydroxylamine. Moreover, the stability of both types of probes was retained *in vivo*, thus allowing for successful EPR detection (spectroscopy and imaging) of the nitroxide probe.

Conclusion: Innovative EPR probes were successfully loaded in tailored nanoemulsions. High concentrations of probes could be safely delivered to endothelial cells *in vitro*. *In vivo*, nanoemulsions appear as best suited to deliver amounts of EPR probes compatible with imaging.

Original fluorescent probes for use in microscopy on *Caenorhabditis elegans*

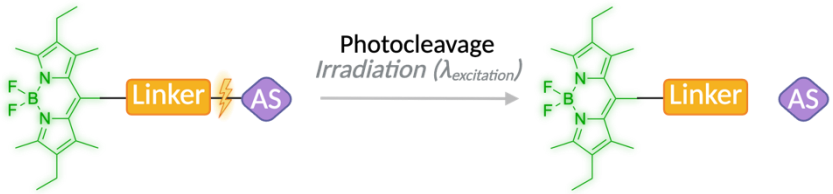
Marion Leroux,[1] Muriel Bonnet,[2] Philippe Veisseire,[2] Stéphanie Bornes,[2] Pierre Chalard,[3] Clément Michelin[3]

[1]*Université Bourgogne Europe, ICMUB, UMR CNRS 6302, 21000 Dijon, France*
[2]*Université Clermont Auvergne, INRAE, VetAgro Sup, UMRF, 15000 Aurillac, France*
[3]*Université Clermont Auvergne, UMR CNRS 6296, Clermont Auvergne INP, ICCF, 63000 Clermont-Ferrand, France*

Marion.leroux@u-bourgogne.fr

Following an active substance within an organism is a major stake. Indeed, this allows, among other things, to facilitate the discovery of new targets of interest. To do this, fluorescence microscopy is a particularly well-suited tool.

The aim of this research work is to develop a novel fluorescent labelling technique to follow the fate and the impact of a molecule on a microorganism *in vivo*, in the intestine of the nematode *Caenorhabditis elegans*.[1] After ingestion, the vector's path can be followed thanks to the nematode's transparency at the bodipy's fluorescence wavelength.[2] Then, after an irradiation at another specific wavelength, the active substance is released,[3] allowing the study of its fate and impact *in vivo*.



Scheme 1: Schematic depiction of a fluorescent probe of interest

This approach will allow the following of the effect of a drug *in vivo* and could be used as an innovative screening process. For this purpose, several fluorescent probes were designed and synthetized to be coupled to an active substance or a microorganism of interest via a potentially cleavable link. Preliminary *in vitro* and *in vivo* studies were carried out to determine the cleavage kinetics, toxicity of the probes and irradiation wavelength.

References

[1] C. Poupet, C. Chassard, A. Nivoliez, S. Bornes, *Front. Nutr.* **2020**, *7*, 135.
[2] L. Vellanki, S. Ritambhara, R. Mangalampalli, *Reports in O. Chem.* **2016**, 1-24.
[3] L. Josa-Culleré, A. Llebaria, *ChemPhotoChem.* **2021**, *5*, 296.

New lanthanide complexes for in vivo luminescent biphotonic and MRI imaging

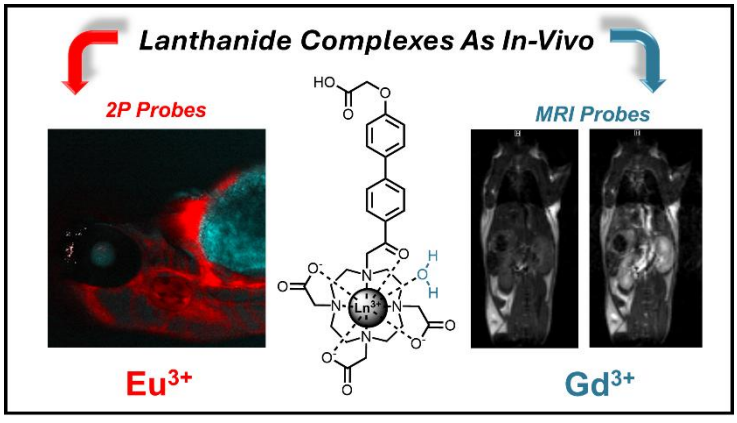
Baptiste Chartier,[1,2] Dina Akl,[3] Guillaume Micouin,[3] Akos Banyasz,[3] Olivier Maury,[3] Véronique Frachet-Martel,[4] Alexei Grichine,[4] Luke Marchetti,[5] Célia Bonnet,[5] Lamiaa Ali,[6] Magali Gary-Bobo,[6] Didier Boturyn,[2] Olivier Sénèque[1]

[1]Univ. Grenoble Alpes, CNRS, CEA, IRIG, LCBM (UMR5249), Grenoble, France ;
[2]Univ. Grenoble Alpes, CNRS, DCM (UMR5250), Grenoble, France;
[3]Univ. Lyon, ENS de Lyon, CNRS, Laboratoire de Chimie (UMR5182), Lyon, France;
[4]Univ. Grenoble Alpes, INSERM, CNRS, IAB (U1209), Grenoble, France ;
[5]Centre de Biophysique Moléculaire, Orléans, France ;
[6]IBMM, Univ Montpellier, CNRS, ENSCM, Montpellier, France

baptiste.chartier@cea.fr

In recent decades, lanthanide(III) complexes have been widely used in medicine as MRI and nuclear imaging probes. These elements have similar coordination properties, making it possible to use a single complex for several applications. Despite their fantastic luminescent properties compared with conventional fluorescent probes (long luminescence lifetime, large stokes shift, fine emission band at fixed wavelengths from visible to near infrared, high photostability, low tendency to aggregation), they have not been widely explored for these applications. Indeed, these complexes require an antenna that absorbs and transfers its energy to the excited state of the lanthanide. They often require high-energy excitation (in the UV, <370nm), out of the optical transparency window of the biological samples and also damaging for cells. The two-photon absorption provide a solution to this issue: molecules are excited by the simultaneous absorption of two photons of half the energy, shifting the excitation in the near infrared, which is the appropriate range for a deeper and more accurate imaging.

Most luminescent lanthanide complexes optimized for microscopy imaging are based on a ligand that saturates the lanthanide coordination sphere, in order to suppress some non-radiative de-excitation due to water molecules. Consequently, their Gd(III) analogue cannot be used as an MRI contrast agent, as this technique requires exchangeable water molecules in the Gd(III) coordination sphere. Here we describe a new family of lanthanide complex that combines a DO3A macrocycle with a fourth substituted acetophenone that serves an antenna for 2P absorption. This system shows excellent 2P luminescence properties with Eu(III) despite a coordinating water molecule and good MRI properties with Gd(III) thereby opening the way to 2P luminescence/MRI bimodal imaging. This presentation presents the synthesis, the optical and magnetic properties of theses complexes as well as their in-vivo imaging applications in zebrafish embryos and mice respectively.



MAPSSIC, an implantable microprobe for β^+ neuroimaging of awake and freely moving rats

S. El ketara [a,b], F. Agnese [d], L. Ammour [c], J. Baudot [d], S. Bouvard [e], O. Clausse [d], M. Dupont [f], F. Gensolen [f], M. Goffe [d], M. Kachel [d], J. Laurence [f], T. Weicherding [f], C. Wabnitz [d], C. Morel [f], P. Pangaud [f], L. Zimmer [d], P. Lanièce [a,b], M.-A. Verdier [a,b]

a Université Paris-Saclay, CNRS/IN2P3, IJCLab, 91405 Orsay, France

b Université Paris Cité, IJCLab, 91405 Orsay France

c Nantes Université, CHU Nantes, F-44000 Nantes, France

d Université de Strasbourg, CNRS, IPHC UMR 7178, F 67000 Strasbourg, France

e Université Claude Bernard Lyon 1, CERMEP-Imagerie du vivant, CNRS, INSERM, Hospices Civils de Lyon, Lyon, France

f Aix Marseille Univ, CNRS/IN2P3, CPPM

email presenting author: luis.ammour@chu-nantes.fr

The MAPSSIC project addresses a major challenge in preclinical neuroscience: correlating molecular neuroimaging with behavior in awake and freely moving animals. Traditional preclinical imaging methods rely heavily on anesthesia, which alters brain function and limits the relevance of behavioral studies. To overcome this, the project is developing a miniaturized implantable microprobe based on CMOS Monolithic Active Pixel Sensor (MAPS) technology. This probe is designed to be used in awake rats after injection of β+ radiotracers commonly employed in positron emission tomography (PET), such as [^{11}C]Raclopride.

Unlike standard micro-PET systems that detect coincident γ-rays from positron annihilation, the MAPSSIC probe detects short-range positrons directly from its implanted location, enabling precise, local brain measurements. The device comprises two back-to-back MAPS sensors (14,700 µm × 700 µm × 200 µm), each with 128 × 16 binary pixels of 30 µm × 50 µm. Its 25 µm sensitive layer offers strong positron sensitivity while remaining transparent to γ-rays, and its total thickness of 400 µm ensures durability without damaging surrounding brain tissue.

Monte Carlo simulations on a rat brain phantom were conducted using time-activity curves (TACs) derived from dynamic micro-PET scans with [^{11}C]Raclopride. Additional simulations introduced controlled reductions in binding potential (BPND) in the striatum, from 5% to 30%, to test the probe's capacity for detecting changes. Results showed that over 93% of the signal was localized within 2 mm of the probe, and using a region of interest (ROI) centered on the striatum captured more than 92% of its signal. Kinetic modeling of striatal TACs demonstrated the probe's ability to accurately quantify BPND, though corrections were needed to offset cerebellar overestimation caused by partial volume effects.

The simulated BPND changes were detected with less than 4% error, confirming the probe's reliability. Moreover, the 2048-pixel matrix enabled preliminary image segmentation, successfully identifying specific brain structures and quantifying BPND variations. These features make the device promising for longitudinal and comparative behavioral neuroimaging.

The MAPSSIC project is advancing rapidly. Newly produced probes have passed initial physical tests and are now slated for biological validation in rodents, with performance to be compared against micro-PET, the current gold standard in preclinical molecular imaging.

Overview of IPHC - Strasbourg's activities in molecular imaging

Ali OUADI[1]

[1]*Institut pluridisciplinaire Hubert-Curien (IPHC) - UMR 7178, 23 rue du Loess, 67200 Strasbourg, France*

ali.ouadi@iphc.cnrs.fr

Overview of radionuclide production at Arronax

F. Haddad [1,2] on behalf of GIP Arronax team

[1] *GIP Arronax, Saint Herblain, France*
[2] *Subatech, Nantes, France*

haddad@subatech.in2p3.fr

Radionuclides are used in different fields of medicine from oncology, neurology to cardiology, for either diagnostic or therapy. In most cases, radionuclides must be coupled to a carrier molecule to target the cells of interest. Many radionuclides may be of medical interest due to their emitted radiations (beta / alpha emitters, Auger emitters) and/or their half-lives that have to be adapted to the carrier molecule's transit time and to the pathology.
Over the last years, two breakthroughs occurred in nuclear medicine:

- The theranostic approach [1] that combines imaging information and therapeutic use of radionuclides. This new paradigm shows great promises especially because it may allow personalizing the treatment to each patient. The diagnosis test done prior to the treatment allows to determine patient response and to determine the needed injected dose for the therapeutic agent. After treatment, the imaging agent can be used to follow the patient response to the injected radiopharmaceutical. Finally, this approach allows a better control of the targeting and increases the benefit/toxicity ratio as useless treatments on patients with no response to the diagnosis test are avoided.
- The use of alpha emitters in some patients shows very promising results [2].

Radionuclides are traditionally produced in (research) reactors, in cyclotrons accelerating protons or are, in some cases, obtained from radionuclide generators. The rapidly growing demand for theranostic applications has stimulated researchers to explore alternative approaches for effectively producing radionuclides. Among them, several have been established or are in development, such as photonuclear reactions, nuclear reactions induced by charged particles other than proton, extraction of progeny from very long-lived sources, spallation reactions employing high-energy accelerators possibly combined with mass separation techniques.
Arronax possesses a 70 MeV multiparticle high energy high intensity cyclotron that allows to produce non-conventional radionuclides for the scientific community and the industry.

The purpose of this presentation is to present our facility, including our radiopharmacy in which we make radiopharmaceuticals for clinical trials, and our pipeline of radionuclides.

References

[1] SC. Srivastava, JPM 2013; 47(1)31-46
[2] C. Kratochwil et al, J Nucl Med. 2016 Dec;57(12):1941-1944.

Daily elutions, monitoring and performance assessment of radiochemically pure $^{44}Ti/^{44}Sc$ generator

Théo Demartinécourt,[1] Ferid Haddad,[1,2] Sandrine Huclier-Markai[1,2]

[1] GIP ARRONAX, 1 rue Arronax, F-44817 Nantes Cedex 3, France
[2] SUBATECH, 4 rue Alfred Kastler, BP 20722, 44307 Nantes Cedex 3, France

demartinecourt@arronax-nantes.fr

Introduction: Recent progresses on the chemical separation of ^{44}Ti from Sc target material allows the establishment and the use of a $^{44}Ti/^{44}Sc$ generator. Evaluating performances of the generator requires a long-term reliability analysis, ^{44}Ti breakthrough rates monitoring and resin stability studies. Furthermore, Radchenko et al [1] described a prototypical generator design with “forward” and “reverse” flow elution mode capabilities which minimize ^{44}Ti Breakthrough[2]. This work evaluates the performance of the alternate flow elution mode versus direct elution one with the aim of providing a high specific ^{44}Sc activity on a daily basis for radiolabelling uses while limiting ^{44}Ti breakthrough.

Methods: ^{44}Ti aliquot from the pre-purified target is eluted through a DOWEX 1X8 column. Then the eluate is evaporated to dryness and recovered in HCl medium. The recovered solution is loaded onto a pre-conditioned ZR-resin column. Both $^{44}Ti/^{44}Sc$ alternate and direct flow elution mode generator are daily eluted in HCl at different concentrations. To perform radiolabelling studies, an additional DGA® resin column can be used. ^{44}Sc activities are measured with a High Purity Germanium (HPGe) gamma spectrometer and ^{44}Ti release is monitored. Finally, performances are evaluated.

Results: The medium change induced by the DOWEX 1X8 column elution allows a better control of the solution acidity and then, a better knowledge of radionuclides interactions. Five 150 kBq $^{44}Ti/^{44}Sc$ generator have been set-up so far. Daily elutions show a reproducible 150 kBq ^{44}Sc elution without any ^{44}Ti breakthrough. Nevertheless, ^{44}Ti breakthrough is observed after 23 - 24 elutions. For radiolabelling studies, using a DGA® column is mandatory to decrease the acidity of ^{44}Sc solution to be biologically suitable. Unfortunately, the use of this column imply a 33% activity loss. Nevertheless, by decreasing HCl concentration, the longevity of the generator is higher and a direct use of ^{44}Sc for radiolabelling studies is possible.

Conclusion: The performances of $^{44}Ti/^{44}Sc$ generator were evaluated. It can provide constant and high specific ^{44}Sc activity on a daily basis without observing ^{44}Ti Breakthrough until a high number of elution. Even with a lower acidity, DGA® purification process of the eluted ^{44}Sc fraction is still necessary to optimize radiolabelling processes. Promising perspectives of this generator are thus foreseen for its daily use imaging facilities with a higher ^{44}Sc activity.

References

[1] V. Radchenko et al. / J. Chromatogr. A 1477 (2016) 39–46
2] D.V. Filosofov, N.S. Loktionova, F. Rosch, Radiochim. Acta 98 (2010) 149–156.

PiB-derivative metal complexes for selective Imaging of amyloid peptides

S. Hery,[1] S.Majdoub,[1] I. Relich,[2] A. Brison,[2] S. Lacerda,[1] J.F. Morfin,[1] C. Hureau,[2] E. Jakab Toth,[1]

[1] *Centre de Biophysique Moléculaire, CNRS, Orléans, France*
[2] *Laboratoire de Chimie de Coordination, CNRS, Toulouse, France*

simon.hery@cnrs-orleans.fr

Aggregation of misfolded peptides is a hallmark of increasingly prevalent diseases, including type 2 diabetes mellitus (T2DM) and Alzheimer's disease (AD), implying amyloid forms of amylin and Aβ-, respectively.[1] The AD brain is characterized by neuronal loss leading to brain atrophy and by the presence of amyloid plaques and neurofibrillary tangles. On the other hand, the accumulation of amylin (islet amyloid polypeptide, IAPP) that is co-secreted with insulin and deposited in the Langerhans Islets, is observed in >90% of T2DM patients and contributes to cell toxicity. [2] Increasing evidence suggests a link between T2DM and AD, likely mediated by fibril "cross-seeding" processes when one amyloid protein promotes aggregation of another.

In this context, selective visualization of amylin- and Aβ-amyloids becomes important.[3] Despite the high number of imaging probes reported for amyloid detection, so far none has shown selectivity for amylin- vs. Aβ-amyloids.[6-7] Here we report the development and characterization of new imaging probes for the selective detection of amylin.

After multi-step synthesis of different imaging probe candidates, we have recently identified two chelates, bearing two PiB units in cis (GdLcis) or trans position on a cyclen (GdLtrans), with selectivity for amylin- vs. Aβ-fibrils. Dissociation constants (K_d) measured by SPR are 2-3 orders of magnitude higher for amylin- than for Aβ-fibrils. When in vitro peptide aggregation is monitored with ThT competition assays, the effect of the compounds is different for Aβ and amylin: GdLcis and GdLtrans readily replace ThT from preformed amylin-fibrils (more than control complexes bearing one PiB unit), but not from Aβ-fibrils. These complexes are the first examples with several orders of magnitude higher affinity for amylin fibrils than for Aβ-fibrils. The reasons for this selectivity are not yet identified. Novel complexes are currently investigated in order to gain insight into the structural parameters and charge effects that might be responsible for selectivity.

References

[1] Y. Zhang et al, *Chin. J. Chem. Eng.*, **2021**, *30*, 225.

[2] a) V.L. Villemagne et al, *Lancet Neurol.*, **2013**, *12(4)*, 357. b) D. Milardi et al, *Chem. Rev.*, **2021**, *121(3)*, 1845.

[3] a) M. E. Oskarsson, et al, *Am. J. Pathol.,* **2015**, *185(3)*, 834. b) J. Fawver et al, *Curr. Alzheimer Res.,* **2014**, *11(10)*, 928.

[4] a) K. Saito et al, *Nucl. Med. Biol.*, **2022**, *106*, 72. b) M. Yoshimura et al, *Bioconjug. Chem.*, **2016**, *27(6)*, 1532.

Bispidine Radiotracers for $^{64}Cu/^{67}Cu$ Theragnostics: Proof of Concept for Multiple Myeloma

Manon Courseyre,[1,2] Adeline Gasser,[2] Stéphane Erb,[1] Oscar Hernandez-Alba,[1] David Brasse,[1] Ali Ouadi,[1] Olena Varuk,[3] Alain Roussel,[3] Patricia Le Saec,[4] Michel Chérel,[4] Yannick Guilloux,[4] Alexandre Detappe,[2] Aline Nonat,[1]

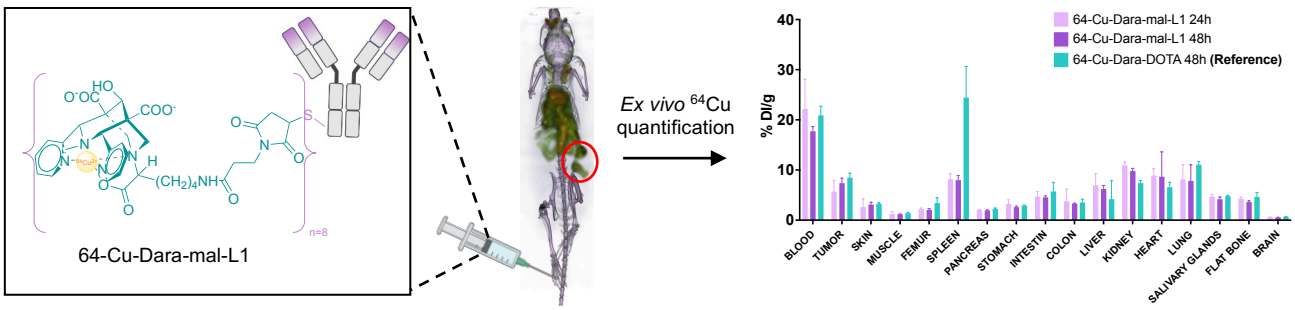
[1] Institut Pluridisciplinaire Hubert Curien (IPHC), Strasbourg, France
[2] Institut de Cancérologie Strasbourg-Europe (ICANS), Strasbourg, France
[3] Laboratoire d'Ingénierie des Systèmes Macromoléculaires (LISM), Marseille, France
[4] Centre de Recherche en Cancérologie et Immunologie Nantes-Angers (CRCI²NA), Nantes, France

manon.courseyre@etu.unistra.fr

Multiple myeloma (MM) is the second most common blood cancer in the world and is incurable, making sensitive imaging, such as immuno-PET, important for detecting minimal residual disease (MRD). Also, the introduction of radiopharmaceutical therapy, in addition to other therapies, has improved patient care, hence showing an increase in median survival.[1]

This study presents a novel bispidine-based copper-64 and copper-67 radiotracer conjugated to daratumumab, a CD38-targeting monoclonal antibody, for specific immuno-PET imaging and potential β^- radioimmunotherapy of MM. Two conjugation strategies were employed to attach bifunctional bispidine ligand (L1), which had been identified previously as a promising ^{64}Cu PET tracer.[2] A site-specific conjugation to thiol groups was achieved, yielding a degree of labeling (DOL) of 8, as confirmed by SEC-MS. This conjugate maintained high binding affinity verified by receptors affinity studies and showed favorable radiolabeling efficiency, serum stability, and pharmacokinetics comparable to unmodified daratumumab. Both ^{64}Cu and ^{67}Cu radiolabeling were successfully achieved with high radiochemical yields (RCY) > 93%. PET imaging in a MM xenograft model confirmed targeted tumor accumulation, showing comparable results to the DOTA chelator with lower uptake in the spleen.

These findings support the clinical potential of bispidine tracers as theranostic radiopharmaceuticals, with ongoing work on nanobody-based tracers to further optimize imaging through improved pharmacokinetics.



References

[1] a) H. Ludwig, S. Novis Durie, A. Meckl, *The Oncologist.* **2020**, *25*, e1406; b) M. Minnix, V. Adhikarla, E. Caserta, *J. Nucl. Med*. **2021**, *62*, 795.

[2] a) A. Roux, R. Gillet, S. Huclier-Markai, *Org. Biomol. Chem*. **2017**, *15*, 1475; b) R. Gillet, A. Roux, J. Brandel, *Inorg. Chem*. **2017**, *56*, 11738.

Ruthenium speciation along the purification process of the daughter radionuclide

R. Chevalet[1], A.C. Bonraisin[1], C.Wagner[2], F. Haddad[1,3], S. Huclier-Markai[1,3 *]

[1]*GIP ARRONAX, 1 Rue Aronnax – CS 10112 – 44817 Saint-Herblain Cedex, France ;*
[2]*ORANO R&D, Le Prisme - 125 avenue de Paris – 92320 Châtillon Cedex France ;*
[3] *SUBATECH, 4 rue A. Kastler, BP 20722, 44307 Nantes cedex 3, France* ;
sandrine.huclier@subatech.in2p3.fr

Introduction : Radioactive isotopes of ruthenium are important elements related to the nuclear medicine field. Among them, ^{97}Ru isotope is intended to be useful for both diagnostic and therapeutic purposes due to its convenient physical properties ($T_{1/2}$ = 2.9 d, Eɣ= 215.7 keV (85.8%) and 324.5 keV (10.2%)). ^{97}Ru is produced through ^{nat}Mo(α,x)^{97}Ru. Ruthenium 97 must be purified after the target dissolution in HNO_3 and HF using a 3-steps process, involving 3 different anionic exchange resins and elutions in HCl solutions at different concentrations. The first objective is to understand the chemical behavior of ruthenium at every step of the purification process. Indeed, ruthenium speciation is quite complicated and must be understood and strictly controlled for reaching an optimal purification process. The second objective is to explore an alternative process using another anionic exchange resin that could be useful for tetravalent species of Ru.

Methods: Ion exchange experiments were conducted using three different anionic exchange resins: A, B and C. For each step, the speciation of ruthenium has been monitored by UV spectrometry in order to establish if ruthenium is eluted within +III or +IV redox state. In addition, the eluted fractions have been quantitatively measured by ICP-MS in order to quantify the retention of ruthenium onto the different columns along the process.

Results: The speciation of the cold ruthenium starting solution was strictly controlled and was identify as $[RuCl_6]^{2-}$. UV-visible analyses evidenced that Ru (IV) is totally retained on resin A whichever species of the starting solution, i.e. $[RuCl_6]^{2-}$ or $[Ru^{IV}(OH)_2Cl_4]^{2-}$. That is not in agreement with experimental observations during the “real” purification process with dissolved ^{97}Ru from irradiated Mo targets. Our assumption is that Ru (III) is formed in the target dissolution solution together with Ru (IV) and that Ru (III) is eluted onto the column with resin A. In order to check this hypothesis, a solution containing 100% of cold Ru (III) was prepared and eluted onto a column with resin A and shows that Ru (III) is eluted. By contrast, the elution onto a column with resin B, showed that Ru is totally eluted and within the same speciation than the starting solution, i.e. $[RuCl_6]^{2-}$. Finally, in the alternative process involving resin C, Ru (IV) is retained on the column whereas Ru (III) is eluted.

Conclusion: The speciation of ruthenium was determined and controlled at the different steps. However, the purification step involving resin A indicated that ruthenium (IV) in a chloride medium was retained on the column while Ru (III) was eluted. The hypothesis is that only Ru (III) is involved during the purification step. Another hypothesis is that Ru nitro-nitrate complexes, i.e. $Ru(NO)(NH_3)_4^{3+}$, are formed during the first step and are highly stable[1]. Investigations will be done to check this assumption.

(1) Boglaienko, D. Ruthenium Speciation and Distribution in the Environment: A Review. *Sci. Total Environ.* **2024**, *951*, 175629. https://doi.org/10.1016/j.scitotenv.2024.175629.

CXCR4-Targeted PET Agents for Solid Tumors

Cassandra Métivier[1,2], Patricia Le Saëc[1], Nour El Ayoubi[1,2,3], Férid Haddad[2,3], Sandrine Huclier[2,3], Latifa Rbah-Vidal[1]

[1] Nantes Université, Inserm UMR 1307, CNRS UMR 6075, Université d'Angers, CRCI²NA, 44000 Nantes
[2] SUBATECH: Laboratory of Subatomic Physics and related Technologies UMR 6457 - IMT Atlantique. Nantes University, 44307 Nantes, France
[3] GIP Arronax, 44800 Saint-Herblain, France

cassandra.metivier@univ-nantes.fr

Introduction: CXCR4, widely overexpressed in cancers, is a key target for imaging and therapy [1,2]. While [^{68}Ga]Ga-Pentixafor has shown strong potential in PET/CT imaging of hematologic cancers, its efficacy in solid tumors is limited by CXCR4 heterogeneity and poor accessibility [3,4]. Monoclonal antibodies, with high affinity and extended biological half-life, offer a promising alternative. This study compares two copper-64 radiolabelled anti-CXCR4 antibodies, one Fc-modified (mAb2), to [^{68}Ga]Ga-Pentixafor in solid tumor models. **Methods & Results:** CXCR4 expression was analysed by flow cytometry in three triple-negative breast cancer cell lines (MDA-MB-231, DU4475, MDA-MB-468) and a cervical carcinoma cell line (HeLa), revealing nanomolar affinity of the antibodies and positive membrane expression for DU4475 and HeLa cells, and negative for MDA-MB-231 and MDA-MB-468 cells. Comparative studies were conducted using two ^{64}Cu-labelled anti-CXCR4 antibodies conjugated to DOTA and [^{68}Ga]Ga-Pentixafor. In vitro analyses revealed the specific internalisation of [^{64}Cu]Cu-DOTA-mAb1 and [^{68}Ga]Ga-Pentixafor in all four cell lines. The membrane binding profiles of [^{64}Cu]Cu-DOTA-mAb1 were consistent with the flow cytometry data. PET/CT imaging in NOD-SCID mice bearing DU4475 and MDA-MB-231 tumors revealed distinct biodistribution: mAb1 accumulated in both tumors model, as well as in the spleen; meanwhile, mAb2 bound significantly to the DU4475 tumors, but less significantly and much more heterogeneously to the MDA-MB-231 tumors. [^{68}Ga]Ga-Pentixafor also exhibited higher uptake in DU4475 tumors, as for mAb2, while showing rapid elimination through the urinary tract shortly after injection, reducing the availability of the tracer to the tumor. **Conclusions:** CXCR4-targeting antibodies, especially Fc-modified mAb2, show promising results for PET/CT imaging of solid tumors. Further studies are needed to assess their uptake and compare them to [^{68}Ga]Ga-Pentixafor.

References

[1] M. Kircher, P. Herhaus, M. Schottelius, A. K. Buck, R. A. Werner, H.-J. Wester, U. Keller and C. Lapa, *Ann Nucl Med*, **2018**, 32, 503–511.

[2] A. K. Buck, S. E. Serfling, T. Lindner, H. Hänscheid, A. Schirbel, S. Hahner, M. Fassnacht, H. Einsele and R. A. Werner, *Eur J Nucl Med Mol Imaging*, **2022**, **49,** 4133–4144.

[3] C. Lapa, M. Schreder, A. Schirbel, S. Samnick, K. M. Kortüm, K. Herrmann, S. Kropf, H. Einsele, A. K. Buck, H.-J. Wester, S. Knop and K. Lückerath, *Theranostics*, **2017**, 7, 205–212.

[4] A. K. Buck, A. Haug, N. Dreher, A. Lambertini, T. Higuchi, C. Lapa, A. Weich, M. G. Pomper, H.-J. Wester, A. Zehndner, A. Schirbel, S. Samnick, M. Hacker, V. Pichler, S. Hahner, M. Fassnacht, H. Einsele, S. E. Serfling and R. A. Werner, *J Nucl Med*, **2022**, 63, 1687–1692.

Booklet of Abstracts

Poster presentations

Thiol-reactive linkers: an alternative for radioimmunoconjugate development

Julen Ariztia[1], Marion Leroux[1], Mathieu Moreau[1], Claire Bernhard[1], Delphine Vivier[1], Pierre-Simon Bellaye[2], Alexandre Cochet[1,2], Alexia Daoust[3], Franck Denat[1]

[1] *Université de Bourgogne, ICMUB UMR CNRS 6302, Dijon, 21000, France*
[2] *Centre Georges François Leclerc, IMATHERA, BioSanD, Dijon, 21000, France*
[3] *Oncodesign Precision Medicine, Dijon, 21000, France*

julen.ariztia-iribarren@u-bourgogne.fr

Bioconjugation between maleimide and thiol groups on proteins is a widely used strategy in the preparation of antibody-drug conjugates (ADCs). However, the resulting thiosuccinimide bond often lacks long-term stability, leading to retro-Michael deconjugation. To overcome this, new alternatives to maleimide-based conjugation are already employed in the ADC field.[1-3] Nevertheless, their potential for radioimmunoconjugates remains largely unexplored. In this study, we investigated the potential of three alternative thiol-reactive groups (maleimide, aryl-maleimide and benzoyl acrylate) for radioimmunoconjugate development.

Each thiol-reactive group was successfully coupled to DFO with good yields. The resulting compounds were then conjugated to a small cysteine-tagged protein (~20 kDa). Among the three compounds, DFO aryl-maleimide exhibited the highest reactivity, achieving full conversion in under 60 minutes with 10 equivalents.

To monitor the formation of hydrolyzed maleimide (more stable towards deconjugation) a hydrolysis study was performed. At pH 8, aryl-maleimide-DFO underwent near-complete hydrolysis (95%) within 6 hours, compared to 36% for maleimide-DFO. Retro-Michael deconjugation was assessed by incubating the three conjugates with a large excess of glutathione. After 14 days, aryl-maleimide-DFO showed greater resistance to deconjugation, with less than 2% deconjugation observed, compared to the other two DFO conjugates (> 50%). Finally, the conjugates were radiolabeled with ^{89}Zr, achieving good radiochemical yields.

This study highlights the improved reactivity and stability of aryl-maleimide conjugates, which exhibit promising resistance to deconjugation and effective radiolabeling performance.

References

[1] Szijj, P. A. *et al. Drug Discov. Today Technol.* **2018**, *30*, 27–34

[2] Ravasco, J. M. J. M. *et al. Chem. – Eur. J.* **2019**, *25* (1), 43–59.

[3] (a) Bernardim, B. *et al. Nat. Commun.* **2016**, *7* (1), 13128 (b) Bernardim, B. *et al. Nat. Protoc.* **2019**, *14* (1), 86–99.

Preclinical evaluation of Lysyl Oxidase-like 2 as a target for the nuclear imaging of idiopathic pulmonary fibrosis

Louane Bancheri[1], Mathieu Moreau[1], Pierre-Simon Bellaye[2], Romane Vizier[1], Victor Goncalves[1]

[1]ICMUB, Dijon, France
[2]CGFL, Dijon, France
louane.bancheri@ube.fr

Introduction:

Idiopathic pulmonary fibrosis (IPF) is a chronic, irreversible disease marked by excess ECM deposition, destroying lung architecture.[1] LOXL2, an enzyme involved in fibrogenesis, catalyzes lysine deamination in collagen, promoting cross-linking. This study aimed to develop a LOXL2-targeting antibody-based radiotracer for in vivo PET/CT imaging of pulmonary fibrosis.

Methods:

AB0023 (Biointron), a murine anti-LOXL2 antibody, was site-specifically conjugated to DFO via microbial transglutaminase.[2] Lung fibrosis was induced in mice (n = 3-4/group) via intratracheal administration of bleomycin (2 mg/kg) 14 days prior to the imaging study. The immunoconjugate was labeled with zirconium-89. ([89Zr]Zr-DFO-AB0023) was injected intravenously (25 µg/mouse, 5 MBq). Four groups (control, fibrotic, blocking, isotype control) were imaged at 24–168 h post-injection and ex-vivo biodistribution was studied by gamma counting.

Results and Discussion:

DFO conjugation yielded a DOL of 1.3. DFO-AB0023 retained high affinity for murine LOXL2. Radiometallation (30 min, 37 °C) led to a RCY of 96%, at a specific activity of 135 MBq/mg. PET/CT and biodistribution data showed higher lung uptake in fibrotic mice (45.2 ± 5.46 %ID/g) vs. healthy mice (8.91 ± 1.48 %ID/g) at 168 h. The blocking group and the isotype control group showed a reduced uptake in fibrotic lungs (26.5 ± 7.06 %ID/g and 20.3 ± 10.0 %ID/g respectively), demonstrating [89Zr]Zr-DFO-AB0023 specificity.

Conclusion:

[89Zr]Zr-DFO-AB0023 enables specific PET/CT imaging of fibrotic lung areas, showing promise as a non-invasive IPF diagnostic tool.

Acknowledgments:

This work was supported by Cancéropôle EST (LOXSTIC project) and ANR-24-CE18-7483 (SUPRALABEL project).

References:

[1] Martinez, F. J. et al. Idiopathic pulmonary fibrosis. Nat. Rev. Dis. Primer 3, 17074 (2017).

[2] Barry-Hamilton, V. et al. Allosteric inhibition of lysyl oxidase–like-2 impedes the development of a pathologic microenvironment. Nat. Med. 16, 1009–1017 (2010).

Toward tumour therasnostics : Hypoxia activation as a tool for therapy and diagnostic

Léa Diebold[1-2], Célia Bonnet[1], Rob Elmes[2]

[1] *CBM Orléans, UPR4301, CNRS, Orléans, France*
[2] *Chemistry Department, Maynooth University, Maynooth, Ireland*

lea.diebold@mu.ie

Hypoxia, a diminished availability of molecular oxygen in bodily tissues, has long been a therapeutic target for cancer research given its major role in tumour growth and resistance to therapy.[1] Despite an explosion of information on hypoxia and its role in tumour development, there are still major questions to be addressed if the long-standing goal of exploiting tumour hypoxia in diagnostics and therapy are to be realised.[2,3] Effective approaches to reliably detect and image hypoxic areas within tumours are therefore critically required.[4]

Our approach is to achieving 'Smart Theranostics' by the synthesis of activatable magnetic resonance imaging (MRI) agents. It relies on a series of reductive enzymes upregulated at the site of hypoxic stress that provide an activation pathway of the agent predominantly available at a site of hypoxia. This goal will be achieve through an enzymatically responsive self-immolative scaffold[5] able to release MRI contrast agent and therapeutic drug. The final goal is to assess the hypoxic state of individual tumours in vivo with the high resolution afforded by MRI while simultaneously providing therapeutic effect (Figure 1).

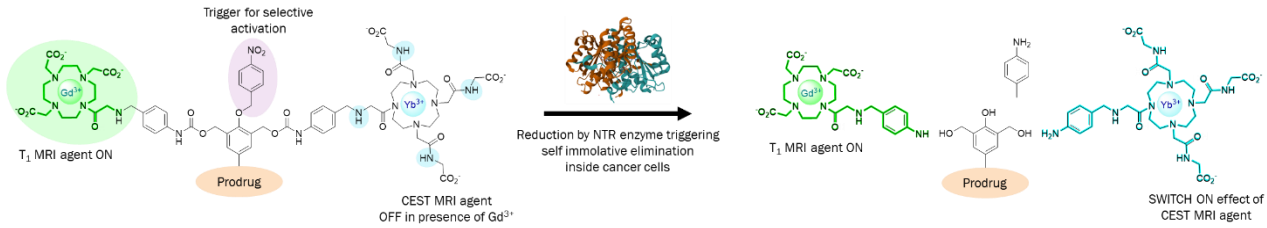


Figure 1. Concept of the theranostic agent

This presentation will focus on the synthesis of a symmetrical compound comporting one lanthanide specie, and preliminary data on the enzymatic cleavage and physico-chemistry measures taken of the released MRI agents.

(1) Zu, Y.; Wang, Z.; Yao, H.; Yan, L. Oxygen-Generating Biocatalytic Nanomaterials for Tumor Hypoxia Relief in Cancer Radiotherapy. *Journal of Materials Chemistry B* **2023**, *11* (14), 3071–3088. https://doi.org/10.1039/D2TB02751H.
(2) Brown, J. M.; Wilson, W. R. Exploiting Tumour Hypoxia in Cancer Treatment. *Nat Rev Cancer* **2004**, *4* (6), 437–447. https://doi.org/10.1038/nrc1367.
(3) Zhuang, Y.; Liu, K.; He, Q.; Gu, X.; Jiang, C.; Wu, J. Hypoxia Signaling in Cancer: Implications for Therapeutic Interventions. *MedComm (2020)* **2023**, *4* (1), e203. https://doi.org/10.1002/mco2.203.
(4) Do, Q. N.; Ratnakar, J. S.; Kovács, Z.; Sherry, A. D. Redox- and Hypoxia-Responsive MRI Contrast Agents. *ChemMedChem* **2014**, *9* (6), 1116–1129. https://doi.org/10.1002/cmdc.201402034.
(5) Erez, R.; Shabat, D. The Azaquinone-Methide Elimination: Comparison Study of 1,6- and 1,4-Eliminations under Physiological Conditions. *Org. Biomol. Chem.* **2008**, *6* (15), 2669–2672. https://doi.org/10.1039/B808198K.

New PTAs for Fluorine-18 Radiolabelling

Zélie Faudemer[1], Rémi Pelletier[1], Bertrand Kuhnast[1], Simon Specklin[1]

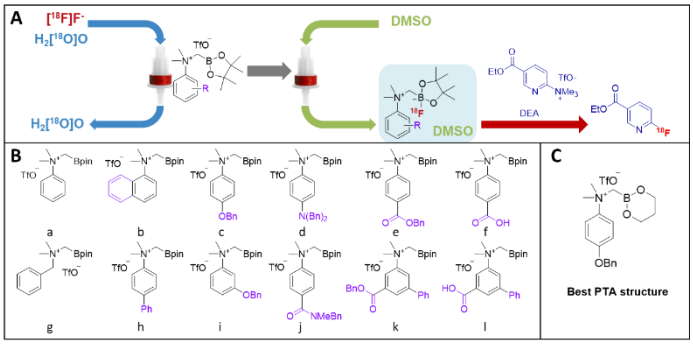
[1] *Université Paris-Saclay, Inserm, CNRS, CEA, Laboratoire d'Imagerie Biomédicale Multimodale Paris-Saclay (BioMaps), Orsay, France.*

zelie.faudemer@cea.fr

PET imaging has become an indispensable diagnostic method, with fluorine-18 labelled tracers being the most commonly used in clinical settings. The activation of the cyclotron-produced [^{18}F]fluoride is a central issue in fluorine-18 radiolabelling.[1] It is highly unreactive in water and has to be dehydrated in the presence of a phase transfer catalyst to restore its reactivity and solubility in an organic medium. The azeotropic drying step, which is time-consuming, is systematic in the radiosynthesis process of tracers. For these reasons, several alternative approaches have been proposed to shorten the radiolabelling processes [2,3]. The aim of this work is to develop original phase transfer agent (PTA) exploiting the boron-fluorine affinity to avoid the azeotropic drying step. (**Fig. 1A**)

A family of 12 PTAs, based on a dimethyl-ammoniomethylen-boronic ester, was successfully obtained. (**Fig. 1B**) Some of them showed promising results in the trapping-elution test (trapping > 95% and elution > 45%), especially those substituted with a benzyl ether (**c**), N,N-dibenzyl group (**d**), or an ester (**e**). Then, the radiofluorination using these PTAs was evaluated on a model pyridine. The best conversion was obtained for the PTA substituted with a benzyl ether (47 %). This structure was optimized by studying the impact of 4 different boronic esters. Conversions superior to 95% were obtained (85% with K[^{18}F]F·K222) by using dioxaborinane rather than pinacolborane. (**Fig. 1C**) This optimized PTA was then applied to several precursors like diaryliodonium salts, boronic esters or arylstannanes. The Cu-mediated fluorinations of arylstannane precursors gave the best results, prompting us to radiolabel more complex structures such as Dolutegravir (DTG) (radiochemical conversion of 73%).

This new class of easily accessible PTAs allowed the radiolabelling of several compounds, with different leaving groups and with comparable to better yields than with the traditional method using K[^{18}F]F·K222. Based on these promising results, this method is being adapted and automated.



References

[1] Haveman, L. Y. F. et al., *EJNMMI radiopharm. chem.* **2023**, *8*, 28.

[2] Perrio, C. *et al.*, *Chem. Commun.* **2016**, *53*, 340.

[3] Pees, A. *et al.*, *Chem. Commun.* **2018**, *54*, 10179.

Coupling DOTA-hexapeptide with Scandium-44/Copper-64 for early detection of brain amyloid deposits by PET imaging in Alzheimer's disease

Cyann Larralde,[1,2], Théo Demartinecourt,[2,3] Cyrille Garnier,[1] Sandrine Huclier-Markai[2,3]

[1] Univ Rennes, Inserm, EHESP, Irset, UMR_S 1085, Rennes, France
[2] Laboratoire Subatech, UMR 6457, IMT Atlantique /CNRS-IN2P3/Nantes Université, Nantes, France
[3] GIP ARRONAX, Saint-Herblain, France

sandrine.huclier@subatech.in2p3.fr

Alzheimer's disease is one of the major public health issues of the 21st century. This neurodegenerative pathology results in amyloid plaques and tangles caused by the abnormal auto-association of Tau protein, named Paired Helical Filaments and Straight Filaments. This protein is expressed in 6 isoforms, composed of 325 to 441 amino acids. The sequence $_{306}$VQIVYK$_{311}$ called PHF6, present in Tau protein, plays a key role in Tau self-association and that is often considered as the nucleation center of filaments. The aim is to develop theranostic molecules that could be used in Alzheimer's disease and Tauopathies. These molecules will be coupled to PET radionuclides (i.e. ^{44}Sc and ^{64}Cu), their stability was evaluated in biological solutions and as probes for an early detection of amyloid deposits in brain by PET imaging.
The chosen peptides are composed of natural "L" amino acids and have the same composition than Tau PHF6 peptide, but with different sequence. Previous results allow to select 6 "leader" hexapeptides with the qualities required to be used as therapeutics1. Two of them, named 4tp- and 6tp, present the highest inhibition of PHF6 fibrillation. 4tp- and 6tp peptides were coupled to DOTA chelate and then radiolabeled with Scandium-44/Copper-64 PET radionuclides in sodium acetate (0,25 mol L^{-1}; pH 4.5). The resulting radiolabeled compounds were further evaluated by challenging studies in in vitro biological solutions as human serum, phosphate buffered saline (PBS) and hydroxyapatite by measuring the radioactivity of Sc-44/Cu-64 at different incubation times at 37°C with autoradiographic system. These complexes were assessed by in vivo biodistribution and imaged by PET on pathological hTau.P301S and wild-type mouse model by measuring Cu-64 radioactivity at different times after the injection.

Radiolabeling of 4tp and 6tp-DOTA is optimum at pH 4.5 at 70°C after 30 minutes of contact, reaching a radiochemical yield of 100 % for both radionuclides (^{44}Sc and ^{64}Cu). The [^{44}Sc]Sc-4tp/6tp-DOTA complexes were stable in human serum and PBS up to 4hrs; whereas [^{64}Cu]Cu-4tp/6tp-DOTA complexes were stable up to 48hrs. In presence of hydroxyapatite, the [^{44}Sc]Sc-4tp/6tp-DOTA complexes showed a small Sc release. Preliminary PET images of transgenic mice carrying hTau.P301S pathological brain lesions showed a whole-body biodistribution with a fast urine clearance.

First studies showed encouraging results about the radiolabeling between 4tp/6tp-DOTA and Sc-44/Cu-64, the stability of [^{64}Cu]Cu-4tp/6tp-DOTA and [^{44}Sc]Sc-4tp/6tp-DOTA and the biodistribution in pathological hTau.P301S and wild-type mice in PET imaging. But cellular uptake and further animal studies are required to better asses the in vitro and in vivo effects of 4tp and 6tp-DOTA complexes, and notably to further evaluate their potential to become theranostic probes for Alzheimer's disease.

Multimodal nanoassemblies for high-contrast diagnostics of cancer cells and bacteria

Houda Benhelli-Mokrani,[1] Fabrice Fleury,[1] Christophe Blanquart,[2] Nicolas Stéphant,[3] Steven Nedellec,[4] Philippe Hulin,[4] Eléna Ishow[5]

[1] US2B UMR CNRS 6286, Nantes Université, Nantes, France
[2] CRCI²NA INSERM U1307, Nantes Université, Nantes, France
[3] IMN UMR CNRS 6502, Nantes Université, Nantes, France
[4] MicroPICELL, SFR Bonamy, Nantes Université, Nantes, France
[5] CEISAM UMR CNRS 6230, Nantes Université, Nantes, France

elena.ishow@univ-nantes.fr

Whereas inorganic nanoparticles and inert polymers have rapidly been considered as potential contrast agents and drug delivery matrices respectively, functional organic nanoparticles have appeared on the stage only recently. In particular, fluorescent organic nanoparticles (FONs), made of self-assembled small molecules are stirring considerable interest for their biodegradability, colloidal stability in physiological media, and finally easily tunable emission energy. After a brief survey of the main emissive materials commonly used as optical labels, we want to show that FONs represent very attractive systems for high-contrast optical bioimaging of various cells (cancer, bacteria, stem cells) and drug vectorization as well. Fine structural modulation of the constitutive FON units enables the introduction of complementary superparamagnetic iron oxide units, which generates photostable core-shell nanoarchitectures endowed with magnetic resonance imaging (MRI) and hyperthermia properties (Figure 1).[1] Emphasis will be put on the benefits of working with assembled nanoparticles over isolated ones in terms of imaging contrast,[2] biorecognition[3] and drug release efficiency.[4]

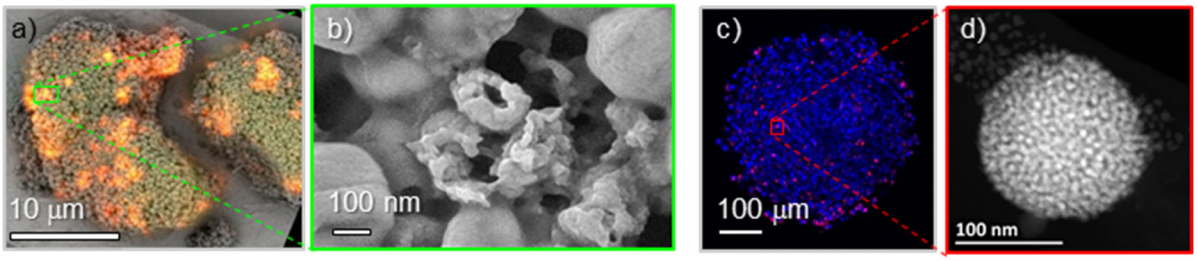


Figure 1. *a-b) Correlative light-electron microscopy of bacterial peptidoglycan layers interacting with FONs (zoom-out). c) Transparized multicellular cell spheroids as solid tumor models after internalization of magnetofluorescent nanoparticles (FONmag). d) TEM imaging of FONmag.*

References

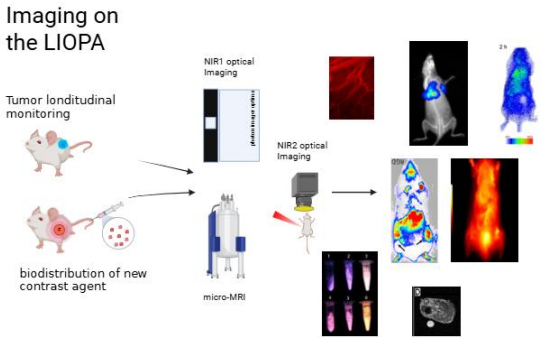
[1] a) A. Faucon et al., *J. Colloid Interface Sci.* **2016**, *479*, 139–149. B) T. Blondy et al., Nanoscale **2022,** *14,* 5884-5898.
[2] a) J. Boucard et al., *ACS Appl. Mater. Interfaces* **2019**, *11*, 32808-32814; *T. Briolay et al., Int. J. Nanomedicine* **2024**, *19*, 633-650.
[3] a) A. Faucon et al., *Nanoscale* **2017**, *9*, 18094–18106 ; b) J. Boucard et al., *ACS Omega* **2018,** *3*, 17392–17402.
[4] a) C. Linot et al., *ACS Appl. Mater. Interfaces* **2017**, *9*, 14242-14257 ; J. Boucard et al., *Small* **2018**, *14*, 1802307.

Evaluation of new imaging probes in the second transparency window (NIR-II) on the LIOPA platform

Thomas Naillon[1,2], Bruno Viana[2], Corinne Chanéac[1], Ghadir Kalot[3], Antoine Maruani[4], Bich-thuy Doan[5], Nathalie Mignet[3], Johanne Seguin[3]

[1]LCMCP, UMR7574 CNRS, Sorbonne Université, 4 Place Jussieu, 75005 Paris, France. [2]IRCP, UMR8247 CNRS, PSL Chimie ParisTech, 11 Rue Pierre et Marie Curie, 75005 Paris, France. [3]Université de Paris Cité, INSERM, CNRS, UTCBS, Faculté de Pharmacie, 4 avenue de l'Observatoire, 75006 Paris, France. [4]Université de Paris Cité, CNRS, Laboratoire de Chimie et de Biochimie Pharmacologiques et Toxicologiques, F-75006 Paris, France. [5]I-CLeHS UMR8060 CNRS, PSL Chimie ParisTech, 11 Rue Pierre et Marie Curie, 75005 Paris, France.

Johanne.seguin@u-paris.fr



The Laboratoire d'Imagerie Optique du Petit Animal (LIOPA) is recognized throughout the scientific community for its expertise in preclinical optical imaging in vivo. The platform is dedicated to bioluminescence and fluorescence imaging. Key applications include the biodistribution of innovative imaging agents, nanomedicine, and tumor growth monitoring to visualize therapeutic effects. Thanks to the development of InGaAs detectors, imaging in the second window of tissue transparency (NIR-II) has been possible for some years now. Here, we present the development of two imaging probes in this wavelength range: (i) silver sulfide (Ag_2S) and (ii) IRDye800-anti CD8.

(i) Ag_2S has been shown to be both effective and low in toxicity. It can be excited in the first transparency window of biological tissues (NIR-I) and emit radiation in the NIR-II window. This nanoparticle was initially developed for luminescence nanothermometry, but it can also be used as an imaging probe. The first biodistribution results on mice showed quick elimination in the bladder for Ag_2S@DTDTPA and in the liver for Ag_2S@PEG2000.

(ii) IRDye800CW, a clinically used near-infrared (NIR) dye, was coupled to either the full-length anti-CD8 antibody (FA) or the anti-CD8 Fab fragments using a site-specific approach based on disulfide rebridging. Both conjugated probes were administered to mice with subcutaneous CT26 tumors. In vivo, both probes stained the spleen, an organ rich in CD8 T lymphocytes, allowing for clear visualization at 5 and 2.5 hours post-injection, respectively. IRDye800-FA exhibited hepatobiliary and renal excretion, whereas IRDye800-Fab was primarily eliminated by the kidneys.

Conclusion: The LIOPA platform can validate new imaging probes. Significant optimization work is required to understand and improve the efficiency of Ag_2S. However, the fast renal elimination of Ag_2S@DTDTPA was interesting for tissue targeting. In another project, we proved the feasibility of imaging CD8 T lymphocytes in vivo in mice using NIR-II fluorescence imaging with IRDye800-anti CD8.

^{18}F radiolabeling of new Zinc-responsive contrast agents

Charline Sire,[1] Tom Di Santo,[2] Bertrand Kuhnast,[1] Célia Bonnet,[3] Olivier Sénèque,[2] Mylène Richard.[1]

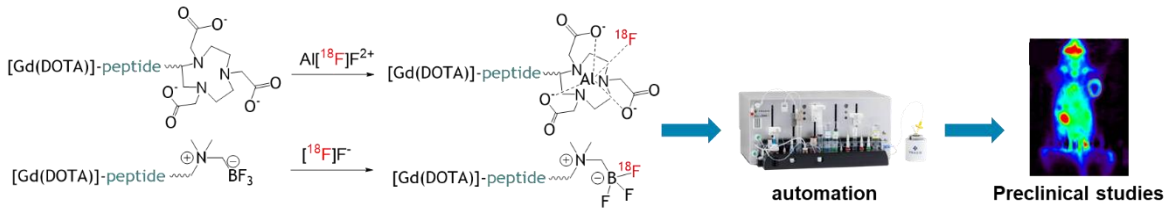
[1] *Université Paris-Saclay, CEA, CNRS, Inserm, BioMaps, Orsay, 91401, France*
[2] *Université Grenoble Alpes, CEA, CNRS, LCBM, Grenoble, 38054, France*
[3] *Université d'Orléans, CNRS, CBM, Orléans, 45071, France*

Charline.sire@cea.fr

Zinc is an essential element in the human body, as it plays a fundamental role in numerous biological processes. Dysregulation of zinc concentration can be associated to various pathologies, such as diabetes, neurodegenerative diseases, cancers… It is therefore crucial to develop methods for detecting zinc *in vivo*, in order to enable the early diagnosis of diseases that may arise from its dysregulation. For this purpose, Magnetic Resonance Imaging (MRI) constitutes a powerful non-invasive imaging technique that provides high-resolution images. Gadolinium-based contrast agents are commonly used to enhance the sensitivity of this technique due to their paramagnetic properties and can be designed to respond to specific biomarkers, such as Zn^{2+} concentration.[1]

Zn^{2+} responsive contrast agents based on zinc finger peptides, known for their high affinity and selectivity towards zinc ions, could be relevant probes in this context.[2] The MRI signal obtained depends not only on the local Zn^{2+} concentration but also on the *in vivo* concentration of the contrast agent. Consequently, accurate determination of the local *in vivo* concentration of the contrast agent is essential and can be achieved through biodistribution studies with PET imaging.

For this purpose, we opted for ^{18}F labeling with two strategies: the incorporation of a NOTA macrocycle capable of coordinating the Al[^{18}F]F synthon,[3] or the incorporation of a trifluoroborate moiety, which can be radiolabeled by ^{19}F/^{18}F isotopic exchange.[4] For these two methods, we have first carried out manual labeling on small scale to determine the optimal labeling conditions and checked the stability of the radiolabeled compounds. We are now focusing on the automation of the process in order to study the biodistribution of these original contrast agents in a diabetes mice model.



References

[1] C.S. Bonnet, *Coord. Chem. Rev.* **2018**, *369*, 91.
[2] M. Isaac *et al. Chem. Comm*. **2018**, *54*, 7350.
[3] Z. Liu *et al*. *Angew. Chem. Int. Ed*. **2014**, *53*, 11876.
[4] W.J. McBride *et al. J. Nucl. Med*. **2009**, *50*, 991.

RNA-imaging: ^{11}C labelling of mRNA for PET imaging of new vaccines and therapies

Clara Testard,[1,2] Mélanie Ethève-Quelquejeu,[2] Davide Audisio,[3] Laura Iannazzo,[2] Fabien Caillé[1]

[1] *Université Paris-Saclay, Inserm, CNRS, CEA, Laboratoire d'Imagerie Biomédicale Multimodale Paris Saclay (BioMaps), F-91401 Orsay, France*
[2] *Laboratoire de Chimie et Biochimie Pharmacologiques et Toxicologiques, UMR 8601, CNRS, Université Paris Cité, 75006, Paris, France*
[3] *Service de Chimie Bio-organique et de Marquage, CEA-DRF-JOLIOT-SCBM, Université Paris-Saclay, Gif sur Yvette, France*

clara.testard@cea.fr

mRNA-based therapies have been in constant expansion in the last years and ever since the first COVID-19 vaccines. In the course of their development, the study of mRNA biodistribution is paramount. In this area, PET-imaging of radiolabelled RNA could allow whole-body and direct visualization of its early spatio-temporal trafficking. The introduction of *β*+ emitters directly to oligonucleotides often require bulky chelators which may alter their structure and properties. On the contrary, smaller radionuclides such as ^{11}C are promising but challenging candidates for the labelling of RNAs. In this project, we aim to establish direct ^{11}C-labelling of oligonucleotides *via* a Staudinger/Aza-Wittig (SAW) tandem reaction[1]. To this end, a model N_3-modified dinucleotide was obtained using known phosphoramidite methodology[2]. Preliminary experiments are in progress to validate the efficacy of the SAW reaction in presence of water to introduce ^{11}C on the dinucleotide with suitable radiochemical conversion (RCC). Application to longer oligomers will confirm the validity of the approach, and allow the labelling of poly-A fragments for full mRNA imaging.

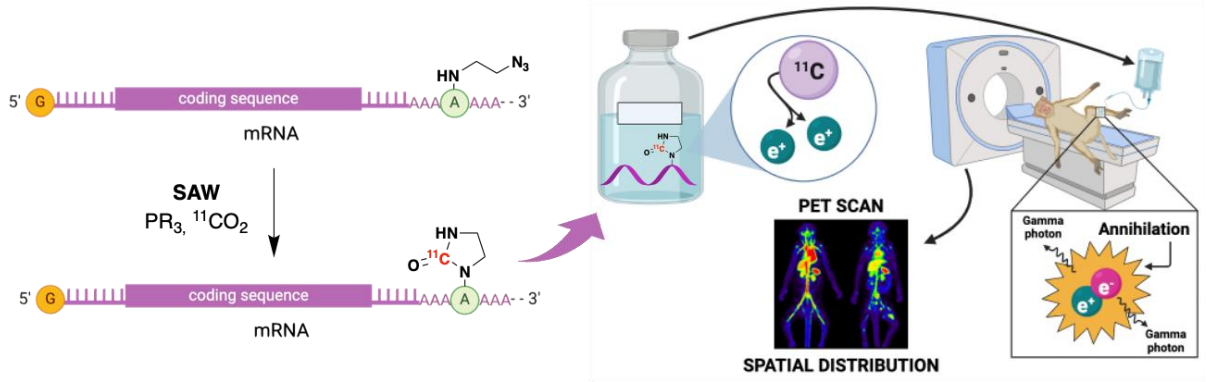


Figure 1. *^{11}C radiolabelling of RNA for PET-imaging*

References

[1] A. Del Vecchio, F. Caillé, A. Chevalier, O. Loreau, K. Horkka, C. Halldin, M. Schou, N. Camus, P. Kessler, B. Kuhnast, F. Taran, D. Audisio *Angew. Chem.* **2018**, *130*, 9892

[2] V. Meynier, L. Iannazzo, M. Catala, S. Oerum, E. Braud, C. Atdjian, P. Barraud, M. Fonvielle, C. Tisné, M. Ethève-Quelquejeu *Nucleic Acids Research*, **2022**, *50*, 10, 5793