平成 29 年度
放射線安全規制研究戦略的推進事業費
短寿命α核種等の RI 利用における
合理的な放射線安全管理のあり方に関する研究

事業報告書

平成３０年３月

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研究の概要

本研究は，医療において放射性同位元素を用いた診断・治療が発展し続けている現在において，短寿命α核種等を用いた内用療法を含む放射線医療の更なる発展を目指した国内における研究開発が，科学的知見に基づく合理的な安全管理の下に促進されるために，放射線防護を関連法令や指針の上でどのように確保し，将来の国民医療の向上につなげるかについて，放射線業務に従事する者及び公衆の防護の最適化の観点から検討を進めた。これまで，厚生労働科学研究費補助金研究（地域医療基盤開発推進研究事業）「医療における放射線防護と関連法令の整備に関する研究（H26-医療-一般-019）」など長年に渡り高度な放射線診療に対応した放射線防護を推進するための研究を進められ，既にその成果の一部が医療法等の関係法令に取り入れられ，放射線診療の発展と放射線防護の整備に寄与している。例えば，α核種である Ra-223 は治療薬として承認されて広く利用され，臨床上の適用拡大に向けた取り組みも進められている。更に At-211, Ac-225 などの他の短寿命α核種，α核種以外にも Lu-177, Cu-67 などの短寿命核種について，放射線治療に関する基礎研究及び臨床研究が進められ，国内外で急速に利用が高まりつつある。一方で，前臨床・臨床研究等の研究開発におけるこのような核種の利用は，我が国においては，放射性同位元素等による放射線障害の防止に関する法律（昭和 32 年法律第 167 号。以下，「障害防止法」という。）の規制下で取り扱われることとなるが，同法及び関係法令におけるこのような将来の医療利用が期待される短寿命α核種に対する規制は，長寿命α核種を想定したものであり，短寿命α核種に適用すると過剰な管理となり，使用及び管理に伴う作業の非効率化につながりかねない。安全の確保を前提とする一方で，科学的知見に基づく合理的な規制が導入されなければ，国の目指す放射線療法の更なる充実に支障を来し，世界の医薬品開発競争にも後れをとることが懸念される。このような課題を解決することは極めて重要であり，本研究では平成 29 年度から 2 年間の計画で国内外における医療用又は医療用として期待される短寿命α核種等の研究開発と安全管理，短寿命α核種等の利用の将来ニーズについて調査し，実態に則した規制のあり方について検討を進めることとした。

１． 研究目的及び研究方法

近年，RI 内用療法の有効性を高める研究が近年精力的に行われ，短寿命α核種が治療薬の研究・開発に必須となっており，世界各地の先進的な研究施設において，短寿命α核種の臨床応用に向けた研究が進められている[1][2]。しかしながら，このような将来の医療利用が期待される短寿命α核種の利用はこれまでは，利用実態がほとんどなかった。その証拠にアイソトープ等流通統計によると，主な非密封 RI 供給量に示された 36 核種に α 核種は含まれず，全体の流通量の 10,036,880MBq に対して，リスト外のその他核種全体量 194 MBq の一部に過ぎない（2016 年度）[3]。このようにこれまで利用の想定がなかったこともあり，障害防止法及び関係法令における，実態を踏まえた規制については十分に検討されているとは言い難く，従来α核種に対する規制はその核種毒性[1)]（Radionuclide toxicity），半減期による，内部被ばくした際のα線の人体への影響を考慮した極めて厳しいものであり，この規制がそのまま短寿命核種に適用されることは，実態にそぐわない過剰な管理となり，使用及び管理に伴う作業の非効率化につながりかねない。安全の確保を前提とする一方で，科学的知見に基づく合理的な規制が導入されなければ，国の目指す放射線療法の更なる充実が，実態にそぐわない規制により支障が来され，世界の医薬品開発競争にも後れをとることも懸念される。このような背景の下，平成 29 年度は医療用又は医療用として期待される短寿命α核種等の利用における規制，安全管理に関して，我が国の現在の規制における課題の解決に向けた，以下の研究を行った。

[1)] IAEA Technical Report Series No.15-A Basic Toxicity Classification of Radionuclides (1963) [4]による分類

1.1 国内外の実態調査及び国内規制における課題の検討

(1) 国内主要施設における実態調査及び国内規制における課題の検討

国内における実態調査は，当該核種の利用状況や利用における放射線防護措置に関して調査するため，短寿命α核種等を実際に利用して研究に取り組んでいる先進的な施設である量子科学技術研究開発機構放射線医学総合研究所（以下，「放射線医学総合研究所」という。），

福島県立医科大学，国立医薬品食品衛生研究所において，利用実態を踏まえた規制の妥当性の観点からそれぞれの施設において以下の調査を実施するとともに短寿命α核種等の RI 利用に際して許可申請時に必要となる空気中濃度などの，我が国の規制下で求められる評価について検討し，実態との乖離などのような点について改善が図られるべきか，そのために科学的視点で明らかにすべきことは何か等を 1.2 の研究連絡会議において議論した。

○放射線医学総合研究所では,At-211 にて標識したカテコールアミン類似物質である At-211 MABG の合成を開発するなど放射性薬剤の基礎的検討がなされているので，α核種の新規薬剤開発における規制を中心に検討した。

○福島県立医科大学では，α核種 RI 内用療法の臨床応用を計画しているので，α 核種の臨床研究における規制を中心に検討した。

○国立医薬品食品衛生研究所では，放射性医薬品等の規制科学の立場で，短寿命α核種を含む RI を利用している。使用者の立場で規制のあり方について検討する他，医薬品研究開発から承認への全プロセスに精通した立場で，規制のあり方について検討を進めた。

(2) 諸外国における実態調査

諸外国においても実際にα核種を利用して医療における研究開発に取り組んでいる高度な施設は限られている。そこで本研究では，α核種の利用状況や利用における放射線防護措置及び規制の実態を調査するため，α核種の臨床応用の研究開発に世界的な業績を持つ以下の２施設を訪問し，施設責任者を含めた研究者との対面での質疑応答，施設・設備の詳細な確認によって，利用実態を踏まえた規制の妥当性の観点からそれぞれの施設において調査を実施した。また，本研究に関連する施設として空気中濃度モニタリング手法やその品質保証に関連した知見をもつ IRSN エアロゾル計量物理学研究所におけるα核種の利用状況や利用における放射線防護措置及びモニタリングの実態の調査を実施した。

○ヨーテボリ大学は，α核種の製造及びα核種の新規薬剤の開発を実施しており，また研究者が本主任研究者とともに ICRP[2] の RI 内用療法の Publication において α 核種を中心に関わっていることから国際的なα核種の使用，廃棄，技術基準，規制等について調査検討を行った。

[2] 国際放射線防護委員会（International Commission on Radiological Protection の略）

○ARRONAX 研究所は，これまで α 核種及び β 核種を含めた RI 内用療法の臨床応用に長年

に渡って先駆的研究を実施しており，本主任研究者が留学した研究施設（フランス保健医学研究所 U211）の後継的研究施設でもあるところから，α 核種の臨床研究における規制を中心にして，欧州諸国における RI 内用療法の規制を含めて，調査研究を行った。

○IRSN エアロゾル計量物理学研究所は，放射性エアロゾルモニタリングの計量に係わる専門機関であり，内部被ばく評価において特に重要なα核種を利用するほか，フランスにおける取扱い施設におけるモニタリング及びその品質保証に関する知見を有する。本機関では，取扱い実態及びフランスにおけるモニタリング手法とその品質保証に関する調査を主に実施した。

1.2　研究連絡会議の設置及び開催等

本研究を的確かつ円滑に推進することを目的として，本研究に携わる研究者等による研究連絡会議を 2 回（9 月及び 1 月）開催した。会議には主任研究者，研究協力者，研究参加者，プログラムオフィサー及びプログラムオフィサー補佐が参加した。本会議においては，1.1 の具体的内容を議論し，研究成果を本報告書にとりまとめた。

2.　研究結果

2.1　国内主要施設における実態調査及び国内規制における課題の検討

国内における実態調査は，短寿命α核種等を実際に利用して研究に取り組んでいる先進的な施設である放射線医学総合研究所，福島県立医科大学，国立医薬品食品衛生研究所において実施し，次のことが明らかとなった。

・国内外において，表１に示すような短寿命α核種の臨床応用に向けた研究が進められていることがわかった[5][6]。
・At-211 の臨床応用に向けた前臨床研究において必要とされる量について検討し，およそ 222 MBq（6 mCi）程度を想定すべきであることがわかった。
・臨床応用の場合，米国デューク大学にて行われた At-211 標識抗体（At-211-81C6）による神経細胞芽腫治療に関する治験（Phase I, II）によると 71 - 347 MBq （1.9 - 9.4 mCi）／ヒトの投与量[7]の想定が必要であることがわかった。

表 1　臨床利用可能なアルファ線放出核種の例 *

α核種	系列	半減期	Eα	親核種 **	子孫核種	最終安定核	放出粒子数	族分類
At-211	4n+3	7.2 h	5.98, 7.59 MeV	[Bi-209] Rn-211 (14.6 h)	Po-211, Bi-207	Pb-207	1α	ハロゲン
Bi-212	4n	61 min	6.21 MeV	Pb-212 (10.6 h)	Po-212+	Pb-208	1α1β	窒素族
Bi-213	4n+1	46 min	8.54 MeV (Po-213)	Ac-225 (10.0 d)	Po-213+	Bi-209	1α2β	窒素族
Ra-223	4n+3	11.4 d	5.98 MeV	Th-227 (18.7 d)	Rn-219+	Pb-207	4α2β	アルカリ土類金属
Ac-225	4n+1	10.0 d	5.94 MeV	[Ra-226 (1600y)] Th-229 (7880y)	Fr-221+	Bi-209	4α2β	アクチノイド
Th-227	4n+3	18.7 d	6.14 MeV	Ac-227	Ra-223+	Pb-207	5α2β	アクチノイド
Tb-149	――	4.2 h	4.08 MeV	[Gd-152, Eu-151]	Eu-145+	Nd-145	0.2α2β	ランタノイド

*細野眞, Isotope News (2013)[1]を一部改変

** [ ] 内は加速器で製造する場合に利用するターゲット元素を示す

・このような臨床応用が期待される短寿命α核種の利用に際しては，種々の係数（排気／空気中濃度限度等）が，表2の例に示すとおり，他の短寿命核種と比較して 1 ～ 2 桁厳しい評価となっているため，その他 RI の使用量を相当圧迫する要因となっている他，これまでのように一律に過剰に安全側な値を採用すると，全てのファクタについて安全係数が大きいため，規制基準値の厳しいα核種では，短寿命であるにもかかわらず全体として実態にそぐわない相当保守的な評価となりうることが見込まれた。このような課題ついて改善が図られるべきか，そのために必要な科学的知見については，海外における事例も参考に，今後典型的なモデルの例を用いた事例研究を行うことで明らかにすべきであると考えられた。また，表3に示す通り，上記の種々の係数の下となっている実効線量率定数について ^{226}Ra（半減期：1600 年）と ^{223}Ra（半減期：11.43 日）とを比較すると，両核種の半減期は大きく異なるにも関わらす，定数は同じオーダであり，根拠となっている実効線量率定数そのものの正当性についても確認することが望ましいと考えられた。

・放射性医薬品の開発承認申請の前臨床においては，動物実験が必須になるが，特に空気中濃度の評価に際しては，動物へ投与された RI は，投与以降，その飛散率を 1 として扱われるため，一般的な施設では小規模の代謝実験程度しか現実的に扱えず，より実態を踏まえた科学的知見に基づく評価が必要であることが示唆された。

表２　医療応用に関連する核種等の空気中濃度限度の例

核種	空気中濃度限度(Bq/cm^3)	空気中又は排気中濃度限度(Bq/cm^3)
^{211}At	8×10^{-4}	7×10^{-6}
^{223}Ra	4×10^{-6}	2×10^{-8}
^{99m}Tc	1×10^{0}	9×10^{-3}
^{131}I	2×10^{-3}	1×10^{-5}

表 3　実効線量率定数の例

COMMITTED EFFECTIVE DOSE PER UNIT INTAKE e(g) VIA INHALATION AND INGESTION (IAEA BSS)
^{223}Ra　5.7×10^{-3}（inhalation）1.0×10^{-4}（ingestion） ^{226}Ra　2.2×10^{-3}（inhalation）2.8×10^{-4}（ingestion）
Limits of radioactivity concentration for the air in controlled areas
^{223}Ra　4×10^{-6} ^{226}Ra　9×10^{-6}
Limits of radioactivity concentration for discharged air from controlled areas
^{223}Ra　2×10^{-8} ^{226}Ra　4×10^{-8}
Limits of radioactivity concentration for discharged water from controlled areas
^{223}Ra　5×10^{-3} ^{226}Ra　2×10^{-3}

以上のことから，国内における規制に関連して検討すべき項目は以下のとおりと考えられる。

・空気中濃度限度，空気中／排気濃度限度が，実際の線量評価に及ぼす影響とその妥当性

・実効線量率定数の妥当性

・飛散率の妥当性（(例) 動物実験における飛散率：1，等）

2.2　諸外国における実態調査

本研究では，スウェーデン１施設及びフランス２施設を訪問調査した。

ヨーテボリ大学は，短寿命α核種の製造及びα核種の新規薬剤の開発を実施している。本調査においては，国際的な臨床応用に向けた短寿命α核種の利用の現状，今後の利用が期待される核種等の研究開発ニーズの現状について調査した。また，関連する施設を訪問し，利用の実態について調査を進めた。更に，訪問先研究者が本主任研究者とともにICRPのRI内用療法のPublicationにおいてα核種を中心に関わっていることから国際的な短寿命α核種の使用，廃棄，技術基準，規制等について意見交換を行った。訪問先において，我が国おける標的 α 線治療（TAT[3]）の現状を紹介し，今後の TAT 発展のための課題などについて議論した。

ARRONAX研究所は，短寿命α核種であるAt-211などの製造及びα核種の新規薬剤の開発のための前臨床研究を実施している。同施設の施設見学を行うとともに，短寿命α核種等の使用，廃棄，技術基準，規制等について調査した。更に，我が国おける標的TATの現状を紹介し，今後のTAT発展のための課題などについて議論した。

空気モニタリング装置の試験のための放射性エアロゾル暴露場を保有しているIRSN-Aerosol Physics and Metrology Laboratoryでは，RI使用施設における空気モニタリングとその品質保証に関して調査した。

以上の海外調査によって，次のことが明らかとなった。

・スウェーデンでは，90年代半ばから短寿命α核種の研究が進められ，当初は長半減期核種と同様に核種毒性について規制当局は懸念していたが，短寿命であり放射線防護の観点から影響は無視できるレベルであることを説明し，物理的半減期などの科学的知見に基づいて，合理的に安全管理が行われていることが分かった。

・フランスにおいても，施設の運用にあたっては，施設と行政の間で合理的な管理について議論し，短寿命α核種の利用によって得られる医学的利益に対して，その利用によるリスクのレベルが許容範囲内であれば，合理的な規制の下で安全を確保しつつ利用されていることが確認できた。

・各国においてDIS[4]は国際的に共通認識である年10μSvの基準に基づいて運用されていた。ただし，本手法の運用が合理的かは，保管設備の規模や核種の分別の容易性など各施設によって異なり，全ての施設が本基準に基づく措置をとっているわけではないことも分かっ

た。

・非密封 RI 施設における RI 利用では，リスク評価に基づく規制の運用と，安全確保のためのモニタリングを基本とした考え方に基づき実運用されていることが確認できた。

・運用にあたっては，IAEA BSS No. GSR Part 3[5]に基づき欧州各国に先駆けて使用核種の特性に応じた合理的な規制（Graded approach など）が導入されていた。以下に，その具体例を示す。

(1) 各国施設では高度な専門知識を有する放射線防護を専門とする物理士の下に安全管理体制が構築され，このような専門家が規制当局への対応も行っている。

(2) ARRONAX 研究所では，入退室は IC カードにより，各室への入室許可はこのカードで管理され，放射線業務従事者が全ての室に入室できるわけではなく，個別に制限し，入室許可は，放射線業務従事者の各作業に関する教育訓練に基づき与えられている。

(3) 近年，我が国においても水晶体等価線量評価に関して ICRP Pub103 の取り入れが検討されているが，ARRONAX 研究所では，規制要求されていないものの作業者の防護メガネ及び線量計の装着を独自に実施している。

これらは IAEA BSS No. GSR Part 3 の以下の要求事項などに基づくものと考えられる。

①Requirement 6: Graded approach

The application of the requirements of these Standards in planned exposure situations shall be commensurate with the characteristics of the practice or the source within a practice, and with the likelihood and magnitude of exposures.

研究の内容や核種の性質に応じた合理的な Graded approach で施設を運用し研究を実施

②Requirement 11: Optimization of protection and safety

The government or the regulatory body shall establish and enforce requirements for the optimization of protection and safety, and registrants and licensees shall ensure that protection and safety is optimized.

高度な専門家が構築した管理体制による防護の最適化

③Requirement 15: Prevention and mitigation of accidents

Registrants and licensees shall apply good engineering practice and shall take all practicable measures to prevent accidents and to mitigate the consequences of those

accidents that do occur.

モニター，安全装置，安全設備，（鍵）などによる事故防止，万が一の場合の汚染や被ばくの抑止

④Requirement 26: Information, instruction and training

Employers, registrants and licensees shall provide workers with adequate information, instruction and training for protection and safety.

教育訓練の実施，およびその受講に応じた利用許可の付与など

[3] α線内用療法（Targeted Alpha Therapy の略）

[4] DIS　Decay in Storage の略。許可を受けた RI が比較的半減期の短い核種で汚染された廃棄物を，減衰させることを目的に一定期間適切に保管すること。

[5] Radiation Protection and Safety of Radiation Sources: International Basic SafetyStandards General Safety Requirements Part 3 （放射線防護と放射線源の安全：国際基本安全基準）

３．まとめ

本研究では，実際にα核種を利用して研究開発を実施している高度な施設（国内：放射線医学総合研究所，福島県立医科大学，国立医薬品食品衛生研究所，海外：ヨーテボリ大学，ARRONAX 研究所，IRSN-Aerosol Physics and Metrology Laboratory）を対象に施設責任者を含めた研究者との質疑応答，施設設備の確認によって調査を実施し，短寿命α核種等の利用に対する合理的規制，安全管理等について研究を進めた。本研究によって臨床応用に向けた短寿命α核種を用いた研究が国内外で行われ，我が国においても将来に渡って利用ニーズがあることが分かった。一方で，本研究における国内調査は，短寿命α核種の製造能力を有する 2 施設及び研究利用施設 1 施設の調査にとどまっており，本研究で抽出された課題が国内における利用ニーズを完全に網羅したものとは言い難く，更に広範囲の調査を実施し，具体的な規制見直しのニーズについて取りまとめるとともに，その効果について検討する必要があることが認識された。海外調査においては，短寿命α核種等を用いた新しい手法の開発促進おいては，IAEA BSS No. GSR Part 3 の考え方に基づく合理的な規制の運用がなさることが重要であり，それが実際に可能であることが確認できた。また，短寿命核種の利用に際し

て，各国においてDISが実運用されていることが確認できた。例えば，フランスにおいては，Code de l'environnement 環境法（Art.L 542.1-1）において，code de la santé publique （公衆衛生法）に規定される活動（医療など）に係る，非常に半減期の短い核種の放射性廃棄物は，放射性廃棄物としての特別な認可ない部門で扱えるよう，十分に減衰させる，という趣旨の規定があり，very short lived-waste として 100days 未満が適用可能となっていることがわかった[8]。

なお，各施設が短寿命α核種の利用にあたって合理的な安全管理を行う上で，実際にどのような評価を行っているか，具体的に規制当局が要求している事項は何かなどのより詳細事項については，具体的に規制に生かす視点から，調査の成果を具体的に示すために，今後調査研究の必要があることが認識された。

謝辞

本研究は，原子力規制庁平成 29 年度放射線対策委託費（放射線安全規制研究戦略的推進事業費）の支援のもと実施した。ここに謝意を表する。

平成29年度放射線安全規制研究戦略的推進事業費（短寿命 α 核種等のRI利用における合理的な放射線安全管理のあり方に関する研究）事業

委員名簿

平成３０年３月現在（敬称略）

	氏名	所属
主任研究者	細野　眞	近畿大学　医学部　教授
研究協力者	織内　昇	福島県立医科大学先端臨床研究センター　ふくしま国際医療科学センター　先端臨床研究センター教授／副センター長
〃	右近　直之	福島県立医科大学先端臨床研究センター　ふくしま国際医療科学センター　先端臨床研究センター助教
〃	永津　弘太郎	国立研究開発法人量子科学技術研究開発機構　標識薬剤開発部　放射性核種製造チーム　主任研究員
研究参加者	伊藤　哲夫	近畿大学　原子力研究所　所長
〃	山西　弘城	近畿大学　原子力研究所　教授
〃	松田　外志朗	近畿大学　原子力研究所　准教授
〃	山田　崇裕	近畿大学　原子力研究所　准教授
外部有識者	蜂須賀　暁子	国立医薬品食品衛生研究所　生化学部　第一室長
プログラムオフィサー	中村　吉秀	公益社団法人日本アイソトープ協会　シニアアドバイザー

参考文献

[1] 細野眞 （2013） α線内用療法の現状と展望，Isotope News, No.711, 2-7

[2] Dekempeneer, et al.（2016）Targeted alpha therapy using short-lived alpha-particles and the promise of nanobodies as targeting vehicle

[3] 日本アイソトープ協会 （2017） アイソトープ等流通統計 2017

[4] IAEA Technical Report Series No.15-A Basic Toxicity Classification of Radionuclides (1963)

[5] Sophie Poty, et al. Alpha Emitters for Radiotherapy: Basic Radiochemistry to Clinical Studies – Part 2. Journal of Nuclear Medicine, published on March 1, 2018 as doi:10.2967/jnumed.117.204651 (2018)

[6] George Sgouros et al. Radiobiology and Dosimetry of a-Particle Emitters for Targeted Radionuclide Therapy*. THE JOURNAL OF NUCLEAR MEDICINE・Vol. 51・No. 2・February 2010

[7] Guérard F, Gestin JF, Brechbiel MW. Production of [^{211}At]-astatinated radiophamaceuticals and applications in targeted a-particle therapy. Cancer Biotherapy and Radiopharmaceuticals （2013） DOI: 10.1089/cbr.2012.1292

[8] Radioactive waste management. IRSN Thematic series

主な研究成果

- Hosono M. Radiation protection in therapy with radiopharmaceuticals. BER2018 International Workshop on Biological Effects of Radiation, March 21, 2018, Osaka, Japan.（invited lecture）
- Hosono M. Individualized treatment planning in radionuclide therapy, Symposium:Radiological protection in nuclear medicine for personalized care. World Federation of Nuclear Medicine and Biology 2018, April 22, 2018, Melbourne, Australia.（invited lecture）

参考資料

・1.　第一回研究連絡会議　議事

・2.　第二回研究連絡会議　議事

・3.　Targeted Alpha Therapy Group, University of Gothenburg, Sweden

・4.　ARRONAX 研究所

・5.　Aerosol Physics and Metrology Laboratory　(LPMA)

・6.　量子科学技術研究開発機構　放射線医学総合研究所　施設設備・研究紹介

　　福島県立医科大学　施設設備・研究紹介

・7.　欧州及び我が国における主な短寿命α核種製造・利用研究拠点

参考資料 1

平成 29 年度放射線対策委託費（放射線安全規制研究戦略的推進事業費）
規制等整備・ 運用領域
短寿命α核種等の RI 利用における合理的な放射線安全管理のあり方に関する研究
（細野班）
第 1 回研究連絡会議概要

日　時：平成 29年 9月5 日（火）13時30分 - 16時
場　所：新大阪丸ビル本館 503 号室
　　大阪市東淀川区東中島 1-18-5
出席者：
（主任研究者）細野　眞
（研究協力者）右近直之（織内 昇　代理）、永津弘太郎、伊藤哲夫、山西弘城、松田外志朗、
　　山田崇裕
（外部有識者）蜂須賀暁子
（プログラムオフィサ）中村吉秀
（オブザーバ）原子力規制庁 長官官房　放射線規制部門　制度係長　吉岡正勝

（出席 10 名　順不同、敬称略）

議　事：
1.　細野班研究計画について
2.　我が国における短寿命α核種等の RI 利用の規制における課題について
3. 平成 29 年度研究実施について
（1）海外調査
（2）国内調査
4. 今後の予定

配布資料：
資料 1：平成２９年度放射線安全規制研究戦略的推進事業費（短寿命α核種等の RI 利用における合理的な放射線安全管理のあり方に関する研究）事業 計画書
資料 2：我が国における短寿命α核種等の RI に係る規制の現状と課題について
資料 3：ARRONAX 研究所概要
資料 4：TAT グループ概要
資料 5：Aerosols physics & metrology laboratory, IRSN 概要

以上

参考資料 2

平成 29 年度放射線対策委託費（放射線安全規制研究戦略的推進事業費）
規制等整備・ 運用領域
短寿命α核種等の RI 利用における合理的な放射線安全管理のあり方に関する研究
（細野班）
第２回研究連絡会議概要

日　時：平成 30 年 1 月 31 日（火）13 時 00-15 時 30 分
場　所：近畿大学　東京センター大会議室
〒103-0028　東京都中央区八重洲1 丁目8 番 16号 新槇町ビル13 階
出席者：
（主任研究者）細野 眞
（研究協力者）織内 昇、永津弘太郎、右近直之、伊藤哲夫、松田外志朗、山田崇裕
（外部有識者）蜂須賀暁子
（プログラムオフィサ）中村吉秀
（オブザーバ）原子力規制庁 長官官房　放射線規制部門　制度係長　吉岡正勝

（出席 10 名　順不同、敬称略）

議　事：
1.　海外調査報告
2.　今後の短半減期 α 核種の利用ニーズと規制緩和が望まれる点について
（1）基礎研究、前臨床関連
（2）臨床研究関連
（3）臨床応用に向けた RI 研究利用全般
3.　短半減期 α 核種の利用に際しての規制のあり方についての総合討論
4. 次年度研究計画について
5.　その他

（会議資料）
資料 1　海外調査報告（案）スウェーデン
資料 2　海外調査報告（案）フランス
資料 3　短寿命α核種等の RI 利用における放射線安全管理のあり方に関する研究（細野班）報告書（QST 永津氏）
資料 4　短半減期α核種の利用に際して、規制緩和してもらいたい・今後の短半減期α核種の利用ニーズについて（福島県立医大右近氏）
資料 5　非密封 RI 利用について（国立衛研蜂須賀氏）
（参考資料）
参考資料 1-astatinated radiopharm and cancer therapy
参考資料 2-Ohshima Sudo KY MABG EJNMMI2018
参考資料 3-各国における放射性廃棄物規制除外（クリアランス）の動向（11-03-04-05）- ATOMICA -
参考資料 4-軽水炉の使用済燃料（04-07-01-02）- ATOMICA -

UNIVERSITY OF GOTHENBURG

Targeted Alpha Therapy Group

Targeted Alpha Therapy Group

The Targeted Alpha Therapy Group is the result of collaborations among departments at the University of Gothenburg, Sahlgrenska University Hospital, and Chalmers University of Technology as well as different foreign centers such as the PET & Cyclotron Unit at Rigshospitalet in Copenhagen, the Memorial Sloan-Kettering Cancer Center in New York, and the Institute for Transuranium Elements in Karlsruhe, Germany.

The coordination of all research activities in the group is though perfomed by people working at different departments at the Sahlgrenska Academy, University of Gothenburg.

The main goal is to develop strategies for the treatment of disseminated cancer using alpha-particle emitters as the leading actor. The research areas covered by the Targeted Alpha Therapy Group include studies of the chemistry related to the labeling of radionuclides to different ligands, studies of pharmacokinetics and different aspects of radiation physics such as dosimetry, and clinical studies in which the developed treatment strategies are evaluated, such as in our recently published phase I study.

Targeted Alpha Therapy Group

Research

The goal with the research is to develop strategies for the treatment of disseminated cancer using alpha-particle emitters as the leading actor.

The Targeted Alpha Therapy Group (TAT Group) started its research activities along with the publication of 2 articles in 1998. Since then, 60 articles have been published/accepted in peer-reviewed scientific journals and 8 PhD thesis have been presented, all relating to the work within the group.

Staff working in or in close collaboration with the TAT Group has different backgrounds varying from nuclear or radiation physics, chemistry, medicine and molecular or microbiology.

The cooperation within the group is characterized by a frequent and continuous exchange of ideas and planning of experiments, among the whole or parts of the group.

Targeted Alpha Therapy Group

Main Group Research

The main research areas in the TAT Group range from studies of the labeling chemistry of radionuclides to different ligands, studies of pharmacokinetics and different aspects of radiation physics such as dosimetry, to clinical studies in which treatment strategies are evaluated, such as in our recently completed phase I study.

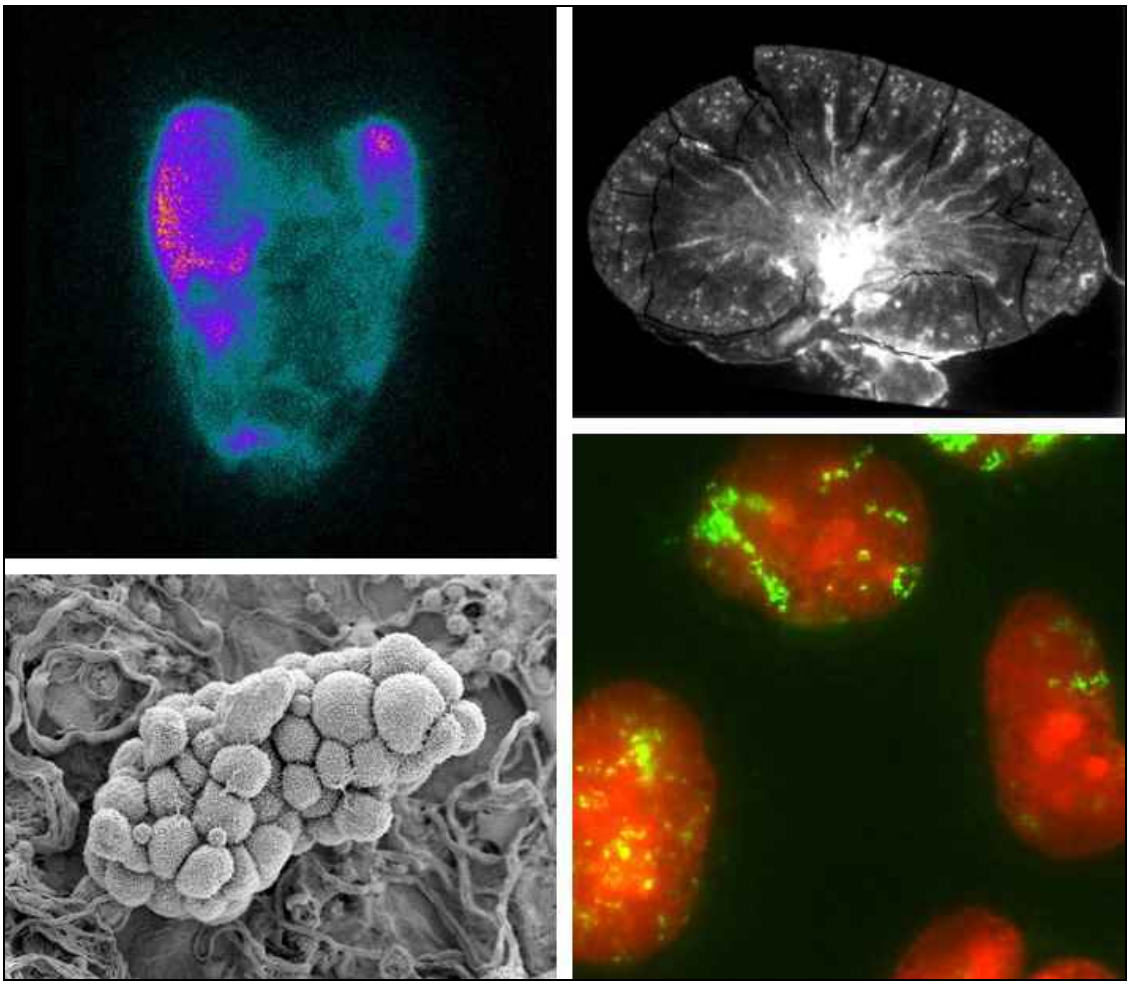

Fig. 1. ***Upper left***: Anteroposterior gamma-camera scan of the abdominal and thoracic area after infusion of 211At-MX35 F(ab´)2 in a patient in the phase I study. ***Upper right***: Distribution of 211At-IgG in mouse kidney 2 h after i.v. injection visualized in a 20 µm cryosection using the Alpha Camera. ***Lower left***: Scanning electron microscopy image showing a small tumor loosely adhered to the peritoneum in a mice. ***Lower right***: Double-strand breaks, visualized as H2AX-foci, in cells irradiated with 1 Gy by alpha particles. Particle passages across the whole nucleus are seen in some cells.

Targeted Alpha Therapy Group

Radiobiology

Cellular Radiobiology of Alpha Particles

DNA-damage and cellular consequences

Initial investigations on in vitro radiobiology effects of alpha particles from ^{211}At were conducted by Stig Palm and collaborators, revealing a severe inhibition of growth in two tumour cell lines, and a RBE of 5 and 12, respectively (Palm et al, 2000a and Palm et al, 2000b). The team now working on cellular effects of irradiation with alpha particles is headed by Associated Professor Kecke Elmroth together with Karin Magnander, Kristina Claesson and other colleagues.

Background

The main focus in our current cellular investigations of high-LET effects is determination of the Relative Biological Effectiveness (RBE) for alpha particles under different conditions. The purpose of studying radiobiology of alpha particles is to increase our knowledge of how damage is inflicted and how cells deal with it in order to improve targeted radiotherapy. It has been generally accepted that double-strand breaks are critical lesions, involved in chromosomal aberrations and decreased survival.

Recently, another type of complex damage, the clustered lesion, has been considered important as well. Clustered lesions are defined as 2 or more lesions induced within 10-20 base pairs and may, if repaired at all, challenge the different repair systems despite the fact that the lesions per se are quite simple (single-strand breaks, base damages or abasic sites). It has been suggested that high-LET radiation induces more clustered lesions compared with low-LET but this has not yet been proved experimentally. Although the induction yield seems to decrease with LET the repair may still be severely compromised.

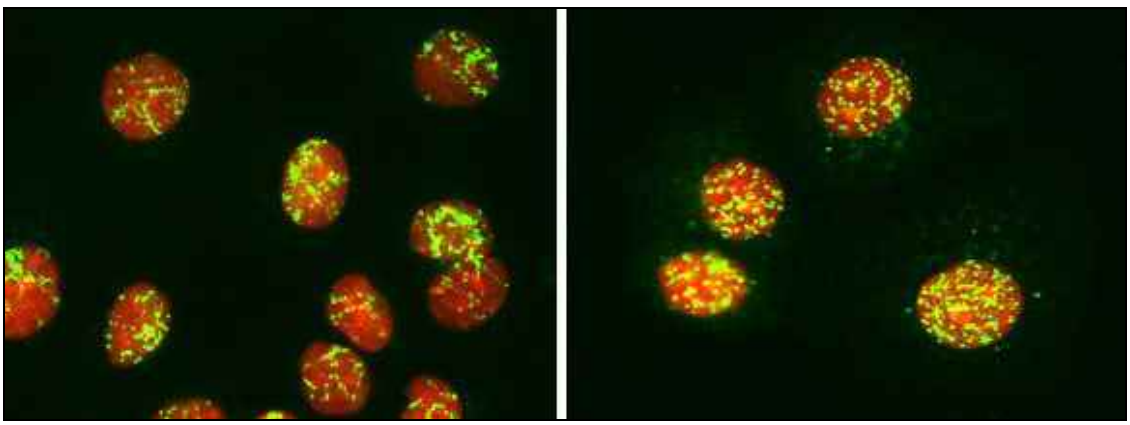

Fig. 1. *Double-strand breaks (green) in irradiated cell nuclei (red). Breaks are visualized by labeling of gamma-H2AX foci, emerging within minutes at the site of DNA breaks. Left: cells*

irradiated with alpha particles from ^{211}At. Right: cells irradiated with X-rays. ***Click image for higher resolution****.*

Alpha particles result in high RBE for induction of double-strand breaks

Recent in vitro investigations focus on induction and repair of complex DNA lesions, i.e. double-strand breaks and clustered lesions induced by alpha particles. Using pulsed field electrophoresis and fragment analysis, which more correctly quantifies non-randomly induced double-strand breaks, we have shown that the induction yield is almost three times higher for alpha particles from ^{211}At than for low-LET radiations at the same absorbed dose (Claesson et al, 2007).

Breaks do not appear randomly in the cell nucleus. When analyzing the DNA fragment distribution resulting from the breaks, an excess of small DNA fragments and a depletion of larger fragments is evident. This is expected due to the inhomogeneous deposit of energy along the radiation track across chromatin fibres (Fig. 1, left). From this work it is also evident that protection against alpha particles by soluble intrinsic scavengers is not as important as for low-LET radiation.

RBE in different cell cycle phases

In studies completed during the spring of 2009, we have further investigated the role of cell cycle position for induction of complex DNA damage. In synchronized fibroblast cells, RBE for both double-strand breaks and clustered DNA lesions in different cell cycle phases was determined. While clustered DNA lesions do not increase with LET or dose, double-strand breaks certainly do, resulting in a RBE close to 3 for all phases. When analyzing the resulting clonogenic survival, the RBE increases dramatically to 8-9 for all phases. The exception is cells in mitosis, showing a more pronounced effect but a lower RBE for both induction and survival.

Ongoing projects

Future experiments include studies on the importance of chromatin structure, survival of cells with different intrinsic radiosensitivity and cell cycle arrest responses in cells irradiated with alpha particles. Another important project just initiated is evaluation of cellular toxicity and biodosimetry after in vivo exposure to alpha particles using visualization of double-strand breaks in individual cells by immunohistochemistry directed against gamma-H2AX foci (Fig. 1). One advantage using this method for quantification of double-strand breaks is that effects after clinically relevant doses can be investigated.

References

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2. Palm S., Andersson H., Bäck T., Claesson I., Delle U., Hultborn R., Jacobsson L., Köpf I. and Lindegren S. In vitro effects of free ^{211}At, ^{211}At-albumin and ^{211}At-monoclonal antibody compared to external photon irradiation on two human cancer cell lines. Anticancer Research 20, 1005-1012, 2000a.
3. Palm S., Andersson H., Bäck T., Claesson I., Delle U., Hultborn R., Jacobsson L., Köpf I. and Lindegren S. Cell growth kinetics of the human cell line Colo-205 irradiated with photons and astatine-211 alpha-particles. Anticancer Research 20, 1807-1812, 2000b.

Targeted Alpha Therapy Group

Staff

The persons associated with the Targeted Alpha Therapy Group has different background varying from nuclear or radiation physics, chemistry and medicine to molecular or microbiology.

Some of the people also work within other projects but the co-operation within the group is always characterized by a frequent and continuous exchange of ideas and planning of experiments. In the lists to the left you will find the regular staff, the close collaborators and persons just occasionally appearing as co-workers or co-authors to the group.

Targeted Alpha Therapy Group

Collaborators

Already from the beginning of the research activities performed by the Targeted Alpha Therapy Group there have been a number of different collaborations.

The most important ones are shown in the list to the left. Some of the collaborations have been continuously ongoing for a long period of years, *e.g.* the PET & Cyclotron Unit at Rigshospitalet in Copenhagen and the Department of Nuclear Chemistry at Chalmers University of Technology in Gothenburg, while others are just recently established, *e.g.* The Institute for Transuranium Elements (ITU) in Karlsruhe, Germany.

Targeted Alpha Therapy Group

Publications

Since the Targeted Alpha Therapy Group started its research activities with the publication of 2 papers 1998 there have been 60 papers published/accepted in peer-reviewed scientific journals and books, 7 PhD theses has been presented and 16 MSc theses has been completed.

A number of conferences have also been attended at which both oral and poster presentations have been made. Also, some articles in daily papers and other popular science newspapers have been published.

Please make a choice in the list to the left to view all the articles, PhD theses, MSc theses, or popular science articles related to the Targeted Alpha Therapy Group.

Targeted Alpha Therapy Group

Chemistry

The standard production of ^{211}At is by irradiating stable bismuth with helium ions (alpha-particles) using cyclotron irradiation, and date back to the discovery of the nuclide in 1940 by Corson et al (1).

Although no cyclotron is available in Gothenburg collaborations with the Department of Physics Oslo University, Norway, Forschungszentrum Dresden-Rossendorf, Germany and Cyclotron and PET Unit, Rigshospitalet, Copenhagen, Denmark has provided us with ^{211}At on a regular basis since 1994. Early on much of the work was focused on the procedures, distillation and chemistry to obtain ^{211}At and ^{211}At-labeled antibodies.

In 2001 a novel procedure on distillation and work up of ^{211}At from irradiated Bi-targets was developed (2). With this method the nuclide is rapidly converted into a chemical useful form for subsequent labeling chemistry. The labeling chemistry has been based on the method first developed by Zalutsky and co-workers (3), see Figure 1 below.

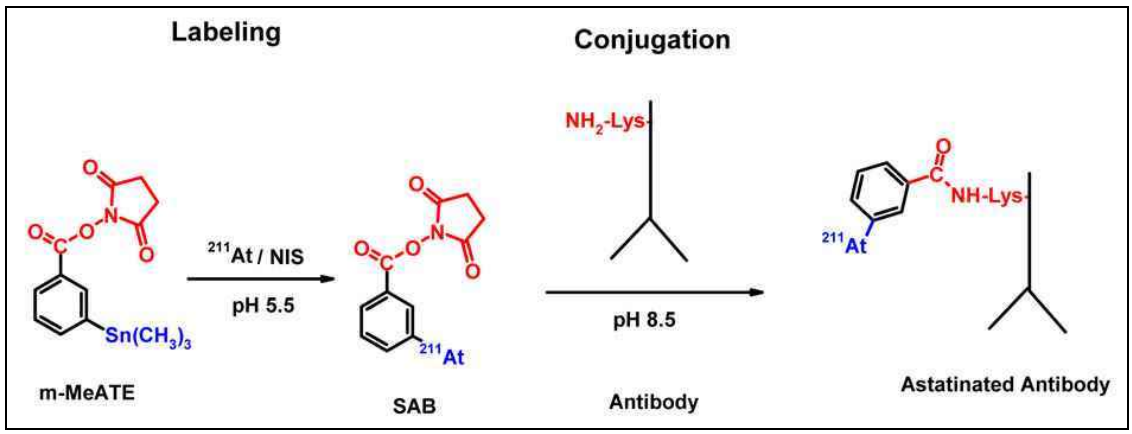

Fig. 1. *Labeling of N-succinimidyl 3-(trimethylstannyl)benzoate followed by conjugation of labeled reagent to antibody.* ***Click image for higher resolution****.*

With this method labeled antibodies which have been sufficient for preclinical evaluations have been produced. However, we encountered problems when taken the research into a clinical phase I study on patients with recurrent ovarian carcinoma. The levels of activity planned for the study were difficult to reach by the conventional procedure for labeling the antibody. Therefore an extensive effort was put on developing the chemistry, and in 2008 a new chemical route that substantially improves the labeling efficacy in astatination of antibodies was developed (4), see Figure 2. In this way the levels of activity required to continue into a phase II clinical study now is possible to reach.

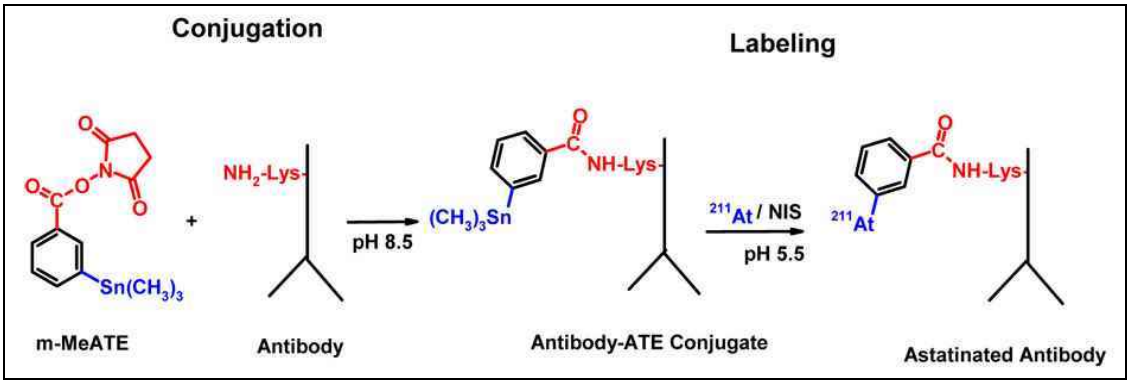

Fig. 2. *Conjugation of antibody with the labeling reagent N-succinimidyl 3-(trimethylstannyl)benzoate (m-MeATE) followed by labeling of immunoconjugate with ^{211}At.* ***Click image for higher resolution****.*

In addition to astatine we have access to another interesting alpha -particle emitting radionuclide, Bismuth-213. It is available through a close collaboration with the Institute of Transuran Elements (ITU) Karlsruhe, Germany. They provide us with ^{225}Ac/^{213}Bi generators from which we can elute ^{213}Bi in a pure chemical form for subsequent labeling to antibodies. The labeling is performed via bifunctional chelates. Several different chelates for antibody are commercially available. Generator protocols and protocols for labeling that result in very good radiochemical yields have been developed at ITU.

The half-life of ^{211}At (7.2 h) and ^{213}Bi (46 min) is generally too short for conventional radioimmunotherapy except for a few special applications such as blood-born or intracavitary cancer treatments, e.g. intraperitoneal (i.p.) and intrathecal (i.t.) treatments (5-7). This is due to the relatively slow in vivo distribution and slow clearance rates of radiolabelled antibodies. Most of the injected radioactivity will therefore decay before reaching its target.

In order to circumvent the unfavourable pharmacokinetics of radiolabeled antibodies, different ways of improving the distribution of the radioactivity have been suggested, employing various pretargeting techniques (8-10). With this type of technique modified antibodies are administered for pre-binding to the tumor antigens. A sufficient time is introduced, to allow non-bound antibodies to be cleared from the circulating system, or a clearing agent is administered to enhance the clearance rate before injecting the labeled effector molecule. The effector molecule recognizes a tag on the antibody and due to the small size of the effector molecule as compared to labeled antibodies, it will localize the target more rapidly and the non-bound fraction will be cleared more efficiently, thus increasing tumor uptake and lowering the dose to normal tissue.

A successful pretargeting protocol will improve the tumor-to-normal tissue absorbed dose ratio for all types of applications involving antibodies for tumor targeting together with radionuclides with short half-lives, e.g. ^{213}Bi and ^{211}At. In this project a pretargeting strategy including the pretargeting molecule avidin/strepavidin conjugated antibody and an effector-molecule based on biotinylated, labeled and charge modified polylysine is investigated, see Figure 3 below.

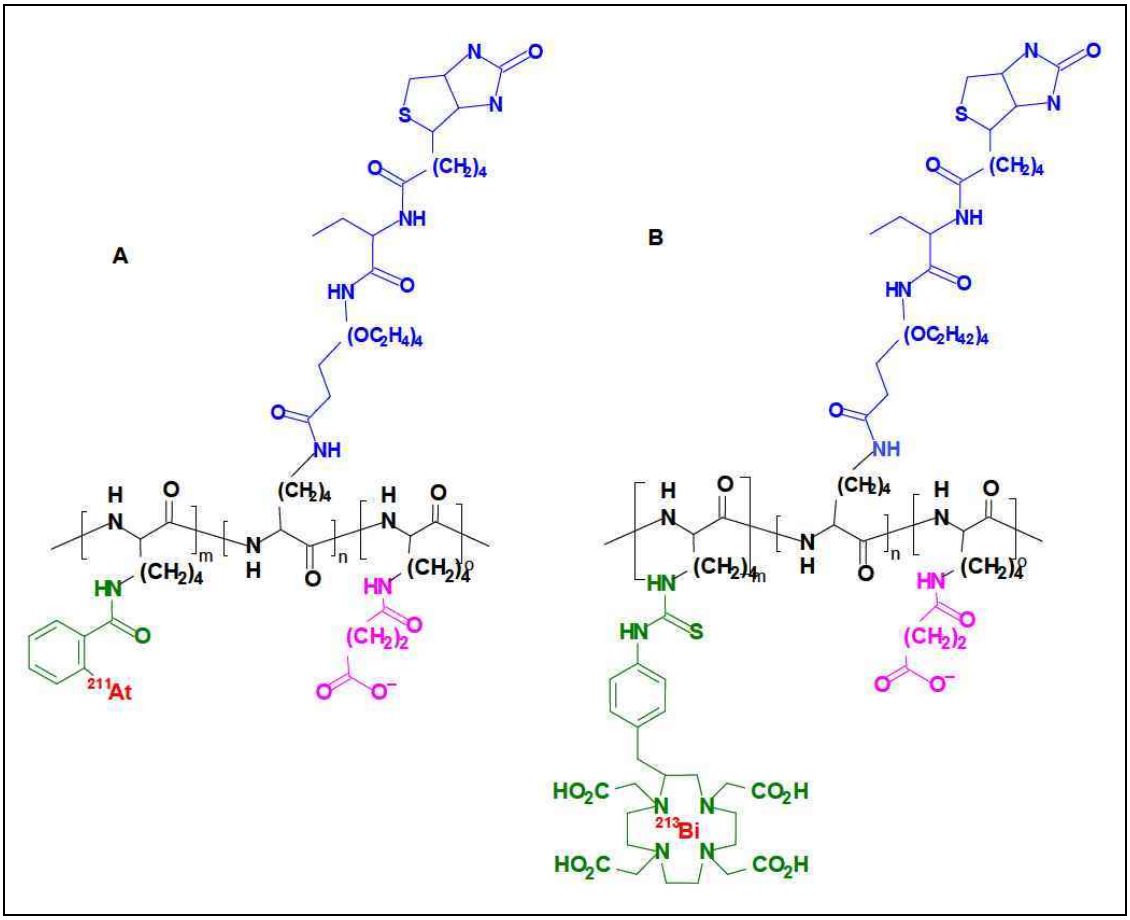

Fig. 3. *Schematic structures of effector molecules.* ***A:*** *Labeled with ^{211}At via N-succinimidyl 3-(trimethylstannyl)benzoate.* ***B:*** *labeled with ^{213}Bi via the kelate DOTA. Different parts of the molecule are presented as, BLUE: biotin residue; GREEN/RED: radiometal-kelate- or radiohalogen-reagent residue; PURPLE: succinic acid residue following charge modification.* ***Click image for higher resolution****.*

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Targeted Alpha Therapy Group

Clinical Studies

All research performed by the Targeted Alpha Therapy Group, irrespective of weather it concerns basic radiobiological, physical or chemical issues, has the final goal of becoming beneficial for the patient. The last 10 years of research by the group has resulted in a phase I study being completed (Journal of Nuclear Medicine 2009;50:1153–1160).

Below follows a short presentation of the methods, results, and conclusion from that study. Questions regardning this study should be addressed to Professor Ragnar Hultborn.

The phase I study:

The alpha-particle emitter ^{211}At labeled to a monoclonal antibody (mAb) has proven safe and effective in treating microscopic ovarian cancer in the abdominal cavity of mice. Beginning in 2005, women in complete clinical remission following second-line chemotherapy for recurrent ovarian carcinoma were enrolled in the study. The aim was to determine the relevant pharmacokinetics for assessing absorbed dose to normal tissues and investigating the toxicity.

Methods: Nine patients underwent laparoscopy 2–5 d before the therapy; a peritoneal catheter was inserted and the abdominal cavity was inspected to exclude the presence of macroscopic tumor growth or major adhesions. Peritoneal scintigraphy was done using 99mTc-LyoMAA to study the fluid distribution in the abdominal cavity. Approximately 500 MBq of ^{211}At was labeled to 0.7 mg of MX35 F(ab')2 using the reagent N-succinimidyl 3 (trimethylstannyl) benzoate. Fifty MBq (3 patients) or 100–200 MBq (6 patients) in 1–2 L of Extraneal was infused via the peritoneal catheter. Gamma camera whole-body scans were acquired at 1, 6, 12, and 20 h (occasionally up to 48 h) after infusion and a single-photon emission computed tomography (SPECT) scan was acquired at 3–6 h. Samples of blood, urine, and peritoneal fluid were collected at 1–48 h. Haematology as well as renal and thyroid function were followed for a median of 23 months.

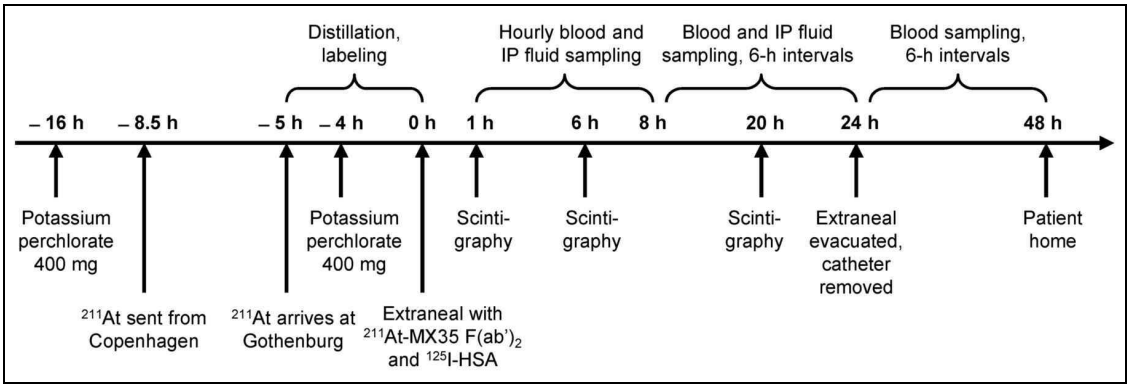


Fig 1. *Schematic overview of logistics of therapeutic procedures. IP = intraperitoneal.* ***Click image for higher resolution****.*

Results: Pharmacokinetics and dosimetric results were related to the initial activity concentration (IC) of the infused solution. The decay-corrected activity concentration decreased with time in the peritoneal fluid to 50% IC at 24 h, increased in serum to 6% IC after 30 h, and increased in the thyroid to 127 ± 63% IC at 20 h without blocking and less than 20% IC with blocking. No other organ uptakes could be detected. The cumulative urinary excretion was 40 kBq/(MBq/L) at 24h. The estimated absorbed dose to the peritoneum was 15.6 ± 1.0 mGy/(MBq/L), to red bone marrow was 0.14 ± 0.04 mGy/(MBq/L), to the urinary bladder wall was 0.77 ± 0.19 mGy/(MBq/L), to the unblocked thyroid was 24.7 ± 11.1 mGy/(MBq/L), and to the blocked thyroid was 1.4 ± 1.6 mGy/(MBq/L) (mean ± 1 SD). No adverse effects were observed either subjectively or in laboratory parameters.

Conclusion: This study indicates that by intraperitoneal administration of ^{211}At-MX35 F(ab')2 it is possible to achieve therapeutic absorbed doses in microscopic tumor clusters without significant toxicity.

Radiation Physics

In preclinical studies, as well as in the clinical phase I study recently published, there are always different aspects of radiation physics that has to be regarded.

These aspects range from the radiation safety during handling of various radioactive sources, calculation of microdosimetric properties regarding irradiation of single cells, dosimetry and the determination of the absorbed dose to tumors, bone marrow or other normal tissues, estimate of the relative biological effectiveness (RBE) for various end-points, estimation of the maximum tolerated absorbed dose or activity for various treatment situations, and considerations regarding tumor cure probability for different treatments and therefore irradiation situations.

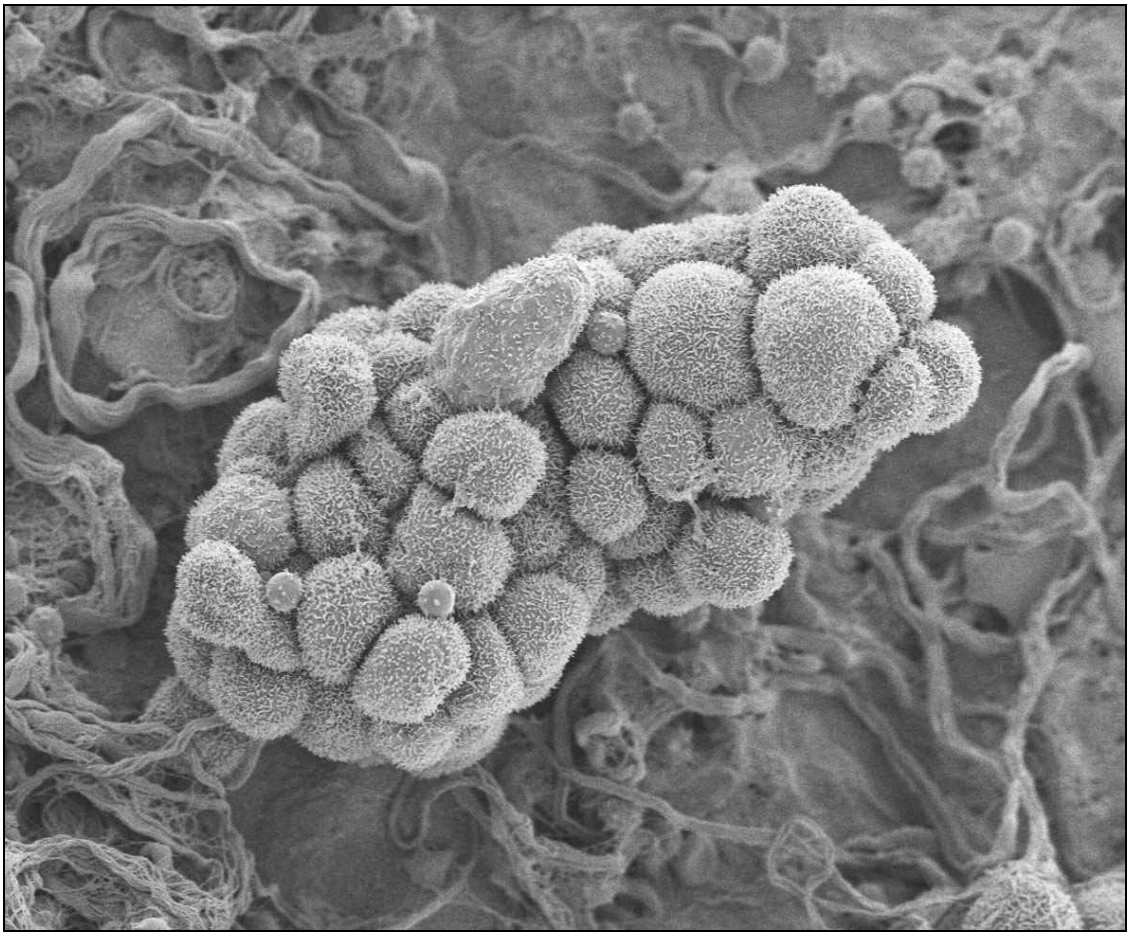


Fig. 1. *The scanning electron microscopy image illustrates microvilli-covered ovarian cancer cells forming a small tumor on thr peritoneum. A network of fibrin, forming individual filaments and more coarse bundles, partly covers the tumor and the peritoneal surface. In the work in J Nucl Med 2006;47:1342–1350 calculations were done in order to estimate the*

absorbed dose to differently sized tumors. ***Click image for higher resolution****.*

Some examples of studies in which different aspects of radiation physics have been considered are presented below.

In the work in Med Phys 2004;31:218-225 a microdosimetric analysis of ^{211}At irradiation of cancer cells was done. A custom-built computer program based on the Monte Carlo method was used to simulate the irradiation. The results show that ^{211}Po atoms, created on a cell surface by the decay of ^{211}At atoms, will diffuse from the cell during its life-span. The increasing distance to the cell nucleus will drastically decrease the probability of the emitted alpha particle to hit the cell nucleus. The conclusion was that for dispersed cells, the diffusion of ^{211}Po atoms will reduce the total absorbed dose from cell-bound ^{211}At by a factor of 2.

In the work in J Nucl Med 2005;46:464–471 the myelotoxicity and the RBE for alpha-particles emitted from ^{211}At was investigated in a pre-clinical study. An RBE of 3.4 ± 0.6 and 5.0 ± 0.9 was found when comparison was made with electrons emitted from ^{99m}Tc or generated by gamma rays emitted from an external ^{60}Co source. The end-point parameter was degree of myelosuppression.

J Nucl Med 2005;46:464–471

Finally, in the work in J Nucl Med 2005;46:2061–2067 the aim was to evaluate the RBE of ^{211}At compared with that of ^{60}Co gamma-irradiation. The endpoint was growth inhibition (GI) of subcutaneous xenografts. The Balb/c received an intravenous injection of ^{211}At-labeled monoclonal antibody MX35 F(ab')2 at different levels of radioactivity (0.33, 0.65, and 0.90 MBq). To calculate the mean absorbed dose to tumor, a separate biodistribution study established the uptake of ^{211}At in tumors and organs at different times after injection. External irradiation of the tumors was performed with ^{60}Co. The biodistribution study showed the uptake of the immunoconjugates by the tumor to be 14% after 7 h. At 40 h, the ratio of uptake in tumors to uptake in blood reached a maximum value of 6.2. The administered activities of ^{211}At corresponded to absorbed doses in tumors of 1.35, 2.65, and 3.70 Gy. The value (mean ± SEM) for D37 was 1.59 ± 0.08 Gy. Tumor growth after ^{60}Co external irradiation showed a value for D37 of 7.65 ± 1.0 Gy. The corresponding RBE of ^{211}At irradiation was 4.8 ± 0.7.

Imaging

Different imaging techniques is repeatedly used in the research and gives important additional information regarding for example the distribution of radioactivity in tissues. Below is listed the imaging techniques use by the Targeted Alpha Therapy Group.

Alpha Camera Imaging

This newly developed technique by Tom Bäck makes it possible to image and pin-point the location of the alpha particles emitted by for example ^{211}At and ^{213}Bi in *in vivo* samples, with a resolution of ~20 µm.

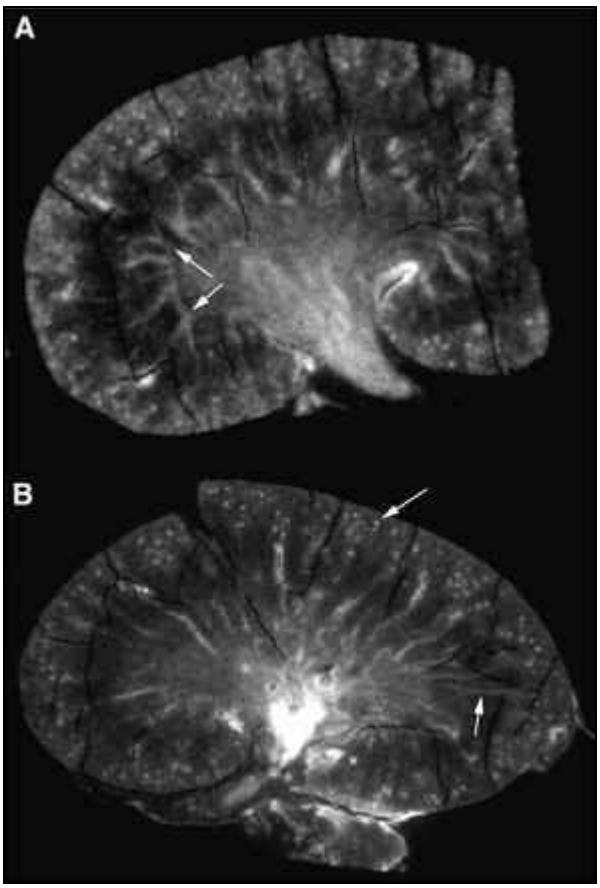


Fig. 1. *Cryosections of Balb/c nu/nu kidneys imaged with the Alpha Camera at 30 min (A) and 2 h (B) after intravenous injection of ^{211}At-IgG trastuzumab. White arrows indicate vascular branches (A) and medullary rays and glomeruli (B).* ***Click image for higher resolution****. J Nucl Med, 2010;51 (10):1616-23.*

Fluorescence Microscopy

This technique has been used when in radiobiological studies when investigationg the appearence of double-strand breaks in irradiated cell nuclei. Breaks are visualized by labeling of gamma-H2AX foci, emerging within minutes at the site of DNA breaks.

Light Microscopy

This technique is used on a regular basis when for example investigating the presence of microscopic tumors in hematoxylin/eosin coloured tissue samples in our therapy studies.

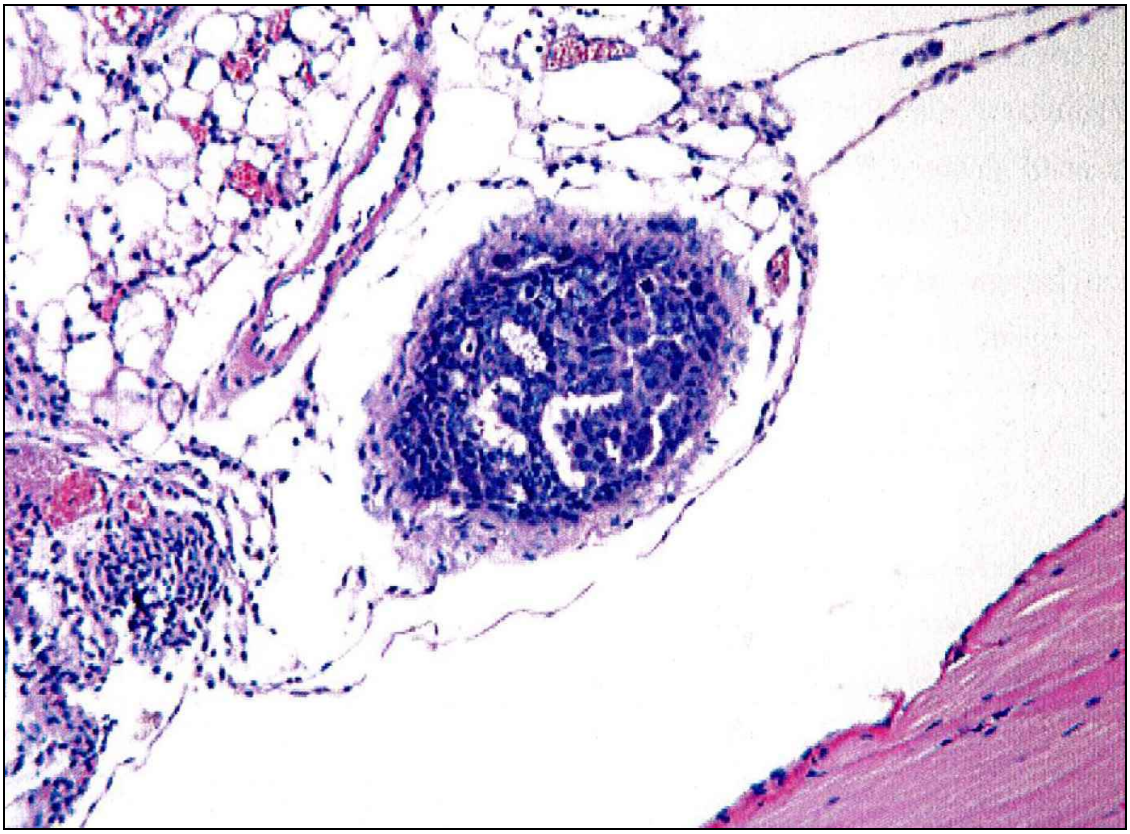


Fig. 2. *Light microscopy image of micrometastasis 2 weeks after intraperitoneal inoculation of ovarian tumor cells (OVCAR-3) in vivo (Balb/c nu/nu). Original magnification x 10.* ***Click image for higher resolution****. Image from Håkan Andersson's PhD thesis in 2000.*

Scanning Electron Microscopy (SEM)

This technique has been used in some studies when for example investigating the relationship between the estimated absorbed dose to differently sized small tumors and the therapeutic efficacy.

Transmission Electron Microscopy (TEM)

This technique has been used occasionally when investigating how the ovarian tumor cells for example are attached to the peritoneal lining during studies of the therapetic efficacy.

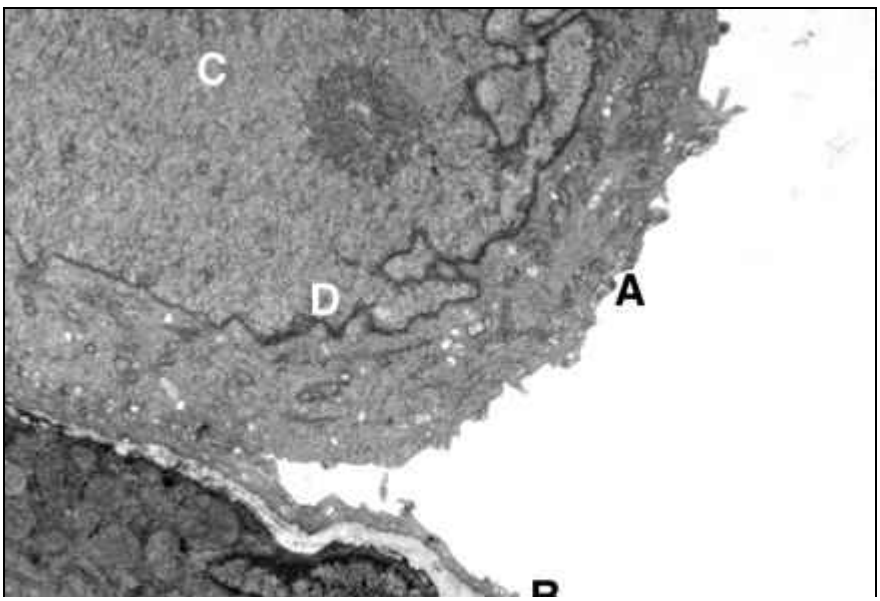


Fig. 3. *TEM image of a tumor cell covered with microvilli (A) adhered to the mesothelial cell layer (B) on peritoneum. The nucleus can also be seen (C), together with its envelope (D). The specimen shown was taken from upper left quadrant of*

abdominal wall from Balb/c nu/nu. ***Click image for higher resolution****. J Nucl Med, 2006;47(8):1238-40.*

Scintigraphy/CT/SPECT-CT

Various standard nuclear-medicine imaging techniqes have been utilized in for example the phase I study to image the localization of radioactivity in the body.

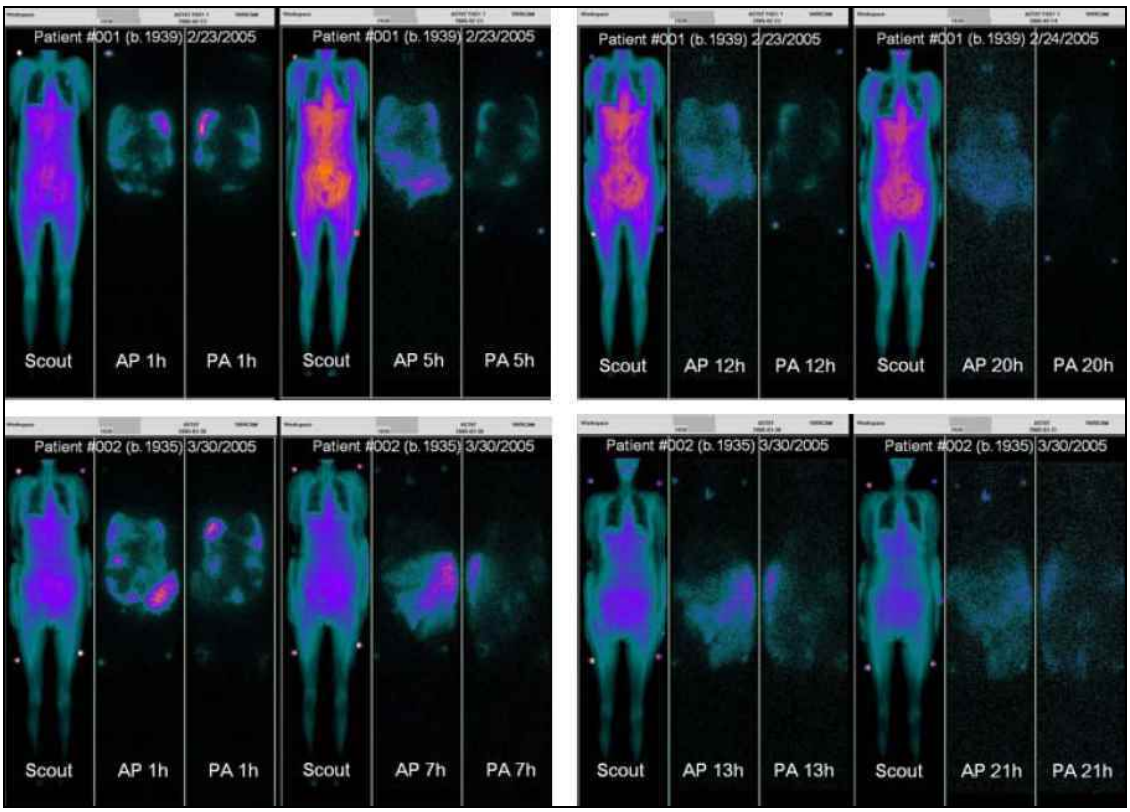


Fig 1. *Gamma-camera images of 2 patients in our phase I study on intraperitoneal infusion of ^{211}At-MX-35 F(ab')2 for ovarian carcinoma. Images show anterior-posterior (AP) and posterior-anterior (PA) views of patients up to 24 h postinfusion.* ***Click image for higher resolution****.*

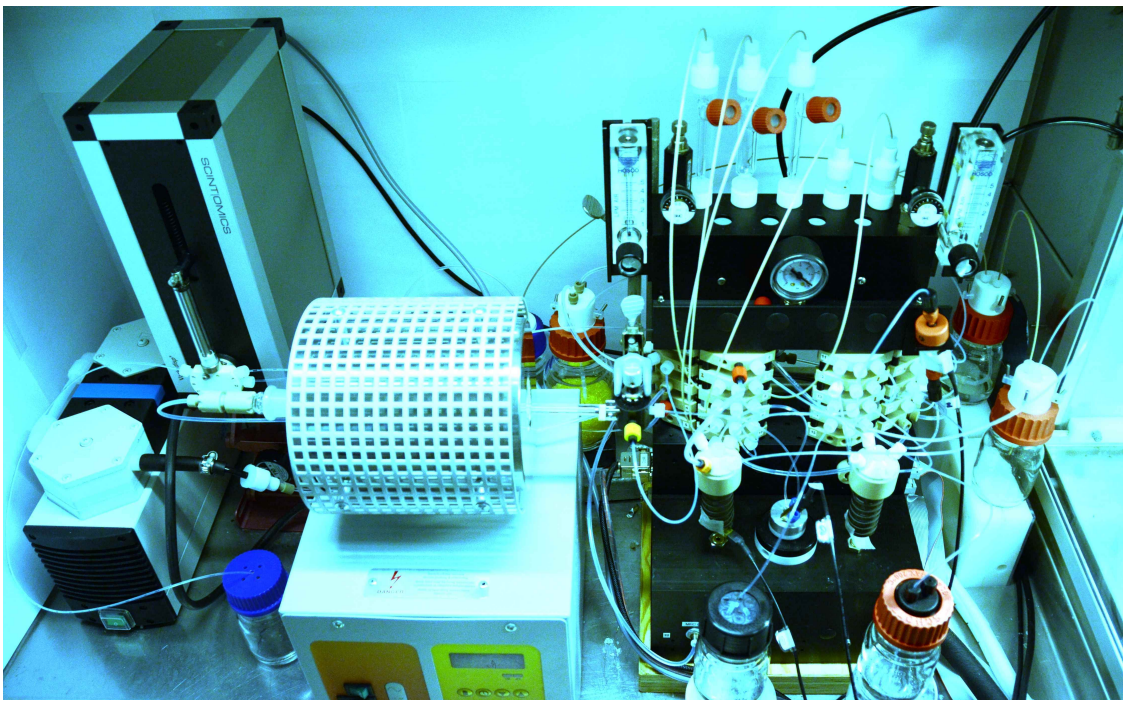
Targeted Alpha Therapy Group

Automation

After more than 20 years of experience in working with the α-emitting radionuclide Astatine-211 (At-211) the chemistry branch of the TAT-group has begun developing an automatic system for production of At-211-radiopharmaceuticals.

Although manual methods of synthesizing At-211 radiopharmaceuticals can be feasible, they would require expert skills and result in only small scale production. This is why future clinical advancements with At-211 will rely on the possibility of automatic production, similar to the development in PET radiochemistry. An automated system is not only useful for standardizing radiopharmaceutical preparations and increasing radiation safety but also reduces the risk of human error.

Our method is based on the combination of dry distillation to isolate astatine-211 in a chemical useful form and a synthesis module for producing radiopharmaceuticals. These components have been merged into a complete automatic process platform where the unique hardware is controlled by a single computer software.



To increase radiation safety, the system is adapted to fit inside a glovebox or a small lead-shielded, hot-cell to minimize exposure to the volatile, radioactive astatine. This compact platform is very versatile as it can accommodate any type of target used for the cyclotron

The Arronax cyclotron characteristics

ARRONAX is an isochronous cyclotron with 4 high hill sectors. The overall size of the cyclotron is of the order of 4m. The working RF frequency is 30.45 MHz with an RF cavity composed of two dees at a voltage of 65 kV.
Table 1 summarises the characteristics of the beam for the four types of particles.

Table 1: ARRONAX cyclotron characteristics

Extracted Particles	Energy range (MeV)	Highest possible current (µAe)	most common current range (µAe)
H+	35 - 70	375 x 2	0.05 - 100 x 2
He2+	70	70	0.07 - 0.1
HH+	35	50	0.1 - 1
D+	15 - 35	50	0.05 - 1.2

The cyclotron has the capacity to send simultaneously proton beams in both opposite beamlines. It is regularly used in this configuration.

For more information, please have a look at http://accelconf.web.cern.ch/AccelConf/IPAC2011/papers/WEPS069.PDF [http://accelconf.web.cern.ch/AccelConf/IPAC2011/papers/WEPS069.PDF]

Laboratories & apparatus

In the controlled area of ARRONAX several laboratories allow the handling of radioactive isotopes safely:

- 2 radiochemistry laboratories (ZCE1 and ZCE2) each containing two fume hoods and glove box.
- 1 cell culture laboratory microscope with a time lapse, a CO2 incubator, a centrifuge, a biological safety cabinet (PSM) and a fume hood
- 1 radiolabeling laboratory equiped with a hot cell and a phospho-imager cyclone.
- 1 metrology room containing three gamma spectrometers with their lead shielding, two alpha spectrometrer,a liquid scintillator, a gas detector alpha / beta total.
- 2 target preparation room containing two electroplating system, a binocular.
- 1 control room containing three quality HPLC which can be coupled to a radioactivity detector or IT-TOF, an UV-visible spectrometer, a GC-MS, an ICP-AES, an electromobility device.
- 1 GMP production lab containing 3 shielded cells.
- 1 sterile GMP production lab containing 5 ht cells equipede with an ionisation chamber.

Access to these laboratories and the materials is possible for external users. To do this, you must make a request to plandemanip@arronax-nantes.fr [mailto:plandemanip@arronax-nantes.fr].

Radionuclide production

As from the beginning of the 2000s, a technological revolution dramatically modified the prospects of nuclear medicine.

This technological revolution was the introduction of PET imaging with 18FDG in the routine practice of nuclear oncology. At the same time a substantial progress has been achieved in radionuclide therapy development as well, especially in radioimmunotherapy and radiopeptide therapy. All these developments open large prospects both in diagnostic imaging and radionuclide therapy with the availability of a lot of carrier molecules which are currently evaluated in preclinical and clinical studies. Beyond oncology, new innovative radiopharmaceuticals are expected to be validated in the coming years, in cardiology and neurology.

In this context, some new needs show up for original and innovative positron, beta- and alpha-emitting radionuclides.

1- PET imaging

For PET imaging, fluorine-18 is undoubtedly the radionuclide of choice, due to its favourable radiophysical characteristics. A lot of new carrier candidates, including FLT, F-MISO, FES, F-choline and F-DOPA, have been clinically evaluated and some of them could be approvedjavascript:toggle_preview();
Mode prévisualisation for a routine use in the coming years. However, the short physical half life (110 minutes) of fluorine-18 requires its production in a cyclotron located at a short distance of each user centre. That's why there is more and more interest for positron-emitting radionuclides with short half-lives but which can be produced in a generator and especially for **[*gallium-68*]** (physical half-life: 68 minutes) for which the father is germanium-68 (with a long half-life of 271 days). Such a generator 68Ge/68Ga has the great advantage to be used for a few months in a nuclear medicine department but germanium-68 needs to be produced in a cyclotron with a high intensity due to its low production yield.

Moreover, fluorinated molecules have a small size and consequently fast kinetics after intravenous injection, which is compatible with the relative short physical half life of fluorine-18. However, for larger carrier molecules, such as antibodies or more generally immunoconstructs, blood kinetics is much slower and maximal tumor accretion is observed relatively late, some days after intravenous injection. This time interval is not compatible with the 110 minutes half-life of fluorine-18. For this new imaging application named immuno-PET, new radionuclides with longer half-lives are needed. **[*Iodine-124*]** is a positron-emitting radionuclide with a physical half-life of 4.2 days which favorably fits with the blood kinetics of antibodies for immuno-PET imaging.

[*Copper-64*] (half-life: 12.7 hours) is another positron-emitting radionuclide of great interest which is also considered for routine production.

Another clinical application which needs some radionuclides with half-lives longer than that of fluorine-18, even for small molecules with fast blood kinetics, is the pre-therapeutic dosimetric calculation. For this application, the innovative approach consists in taking into consideration some pairs of positron- and beta-emitting radionuclides.

Given the present clinical routine use of iodine-131 and yttrium-90 for the labeling of immunoconstructs and peptides, the favorite pairs of radionuclides are iodine-124/iodine-131 and yttrium-86/yttrium-90. However, for the latter pair, a high energy gamma ray emitted at a high rate by yttrium-86 is a real drawback for the routine use of this radionuclide.

Another highly requested pair of radionuclides is copper-64/copper-67 due to the favorable characteristics of both radionuclides.

In cardiology, thallium-201 and technetium-99m MIBI (Cardiolite®) radiopharmaceuticals have been used in clinical practice for some decades, for the diagnosis of myocardial ischemia. However the low energy of the gamma rays emitted by these radionuclides requires an attenuation correction to be introduced which has some limitations. These limitations result in a relative high percentage of false positive results which can lead to some useless invasive coronarography procedures.

[*Rubidium-82*] is a positron-emitting radionuclide which behaves like thallium-201 and is taken-up by the myocardial muscle. The high energy (511 keV) annihilation photons allow to achieve a reliable attenuation correction. Consequently it has been clearly shown that the diagnostic specificity of rubidium-82 imaging is significantly higher than that of thallium-201 or technetium-99m MIBI SPECT imaging. Rubidium-82 has a very short physical half-life (75 sec) and is produced, in a generator, by decay of strontium-82 which has a 25.5 day physical half-life. This very short half-life of rubidium-82 allows to perform both rest and stress imaging tests in less than 30 minutes as compared with a few hours for thallium-201 or technetium-99m MIBI SPECT imaging.

Strontium-82/rubidium-82 generators have been used in the US for more than a decade but currently, the production capability of high activity of strontium-82 is seriously limited in the production centers. ARRONAX cyclotron, with a high energy/high intensity of proton beam will allow to produce up to 600 generators a year.

2- Radionuclide therapy

The three currently used radionuclides for therapy are iodine-131, yttrium-90 and lutetium-177. They cover a range of beta energy which fits well with the range of small tumor sizes which are appropriate for this treatment modality. However iodine-131 emits a relatively high percentage of high energy gamma rays which requires some medical staff radiation safety constraints including some confining of patients in shielded rooms for a few days. These constraints seriously limit the number of patients who could have benefit of radionuclide therapy. Moreover yttrium-90, a high energy beta-emitter, is taken up by bone/bone marrow after release from its chelator coupled to the carrier molecule resulting in bone marrow irradiation which limits the injected activity. Additionally yttrium-90 does not emit gamma rays for pre-therapeutic imaging and yttrium-86 has too high energy gamma rays for routine imaging.

A radionuclide with favorable radiophysical and biological characteristics is **[*copper-67*]** (physical half-life: 61.5 hours) which has been preclinically and clinically evaluated for more than 2 decades. As compared with iodine-131 and yttrium-90, copper-67 has shown the highest therapeutic index in a few clinical studies. However its industrial production has been, up to now, limited by the lack of high energy (70 MeV), high intensity (a few hundreds of microamps) cyclotrons necessary for the production of high activities for clinical studies. ARRONAX cyclotron will be able to produce such high activities.

Finally alpha-emitting radionuclides are being more and more considered for their use in alpha-therapy because of their high LET (Linear Energy Transfer) which gives a high killing effect especially for small clusters of malignant cells. A few alpha-emitting radionuclides are available, including astatine-211, lead-212/bismuth-212 and actinium-225/bismuth-213. ARRONAX cyclotron will produce **[*astatine-211*]** (physical half-life: 7.2 hours) for preclinical and clinical alpha-therapy studies.

Radionuclides priduced byARRONAX

Radionuclide	Target	Nuclear reaction	Cross section (mbarns)	Needed Energy (MeV)
^{64}Cu	Ni	$^{64}Ni(p,n)$	≈ 675	15
^{68}Ge	Ga	$^{69}Ga(p,2n)$	≈ 550	
^{124}I	Te	$^{124}Te(p,n)$	≈ 590	15
^{82}r	RbCl	$^{nat}Rb(p,4n)$	≈ 98	
^{67}Cu	ZnO	$^{68}Zn(p,2p)$	≈ 10	70

Radiobiology

The interaction of ionizing radiation with living matter changes its intrinsic structure (breaking of chemical bonds, recombination ...) differently depending onseveral factors:
- The particle involved
- The linear energy transfer
- The density of energy deposition in the medium
- The total deposited dose.

The cyclotron ARRONAX delivers several types of particles: alpha, deuteron and proton with energies up to 68 MeV.
Dose rates vary between 0.01 and 10 MGy /s allowing us to study and compile data over a wide range of linear energy transfer (LET) and density of energy deposition for each incident ion. These features are unique and complementary to those of other accelerators in France and around the world.

The AX Hall of ARRONAX cyclotron is a room dedicated to experiments. It has several unique features:
- The ability to irradiate using a horizontal beam or a vertical beam
- The possibility, in the vertical beam configuration, to perform on-line time lapse microscopy.
- The possibility to pulse the alpha particle beam (variable inter-pulse duration from 330 ns up to 5 s)

Inside the facility, a cell culture laboratory is present equipped with a PSM, a centrifuge, an incubator, a -80 ° C freezer and various fridges and freezers. A time lapse microscope with controlled atmosphere is available in this room.

Developments in progress:
Innovative tools for dosimetry are being developed:
- The calibration of the optical density of radiosensitive films (including Gafchromic EBT2) as a function of the dose deposited by alpha particles and deuterons will be studied.

- A set of collimators and degraders is being developed in order to obtain spatially uniform beam with minimum energy dispersion.

- A new method for measuring the on-line dose will be implemented based on the measurement of bremsstrahlung emission from the irradiated medium.

- Implementation of time-lapse microscopy in beam.

Teams involved:
Development of radiobiology at ARRONAX is performed by a group of researchers from several laboratories (ARRONAX, CRCNA, ICO and Subatech).

Cross section measurements

Production of radionuclides

The radionuclides that are used for medical applications are generally produced artificially. They are obtained by sending projectiles on targets formed from stable elements in nature.

The choice of the projectile used depends on the decay mode of the radionuclide that is to be produced. For beta + emitting radionuclides, charged particles such as protons will be used, produced by an accelerator. For emitting radionuclides beta-, neutrons, produced in a reactor, are used. For alpha-emitting radionuclides, neutrons from reactors will also be used. Thus we see that accelerators and nuclear reactors are complementary with regard to the production of radionuclides.

The quantity of radionuclides produced during the irradiation of a target is proportional to the number of target nuclei and the flow of the projectile used. The coefficient of proportionality, which contains all the physics of the interaction, is called production cross section. It depends on the energy of the projectile.

To optimize the production of a radionuclide, we must determine the proper energy interval of our projectiles and take into account the fact that for a given projectile energy, several different nuclear reactions are possible. These other nuclear reactions will produce unwanted radioisotopes (contaminants) that must be removed later. It is therefore important during the optimization phase of irradiation parameters to see how we can avoid the production of these contaminants in order to simplify the subsequent work of radiochemical purification.

The production cross sections

This optimization work is done using the production cross sections of the different isotopes.

These cross sections are available, when they are known, in data bases (NNDC). For some reactions, cross sections are not known precisely and it is necessary to measure them again. To this end, a program of cross sections measurement is implemented at Arronax using the "stacked foils » technique.

Research partnerships

It is the policy of Arronax to propose and get involved in research partnerships. Purely academic or academia/industry partnerships are considered when their object is consistent with the original missions of Arronax.
Accordingly, Arronax is currently a partner of the Région Pays de la Loire NucSan (Nuclear Technologies for Health) research project, which, in addition to the Arronax GIP, includes 10 laboratories of Nantes and Angers, and of the "Investissements d'Avenir" laboratory of Excellence IRON (Innovative Radiopharmaceuticals for Oncology and Neurology), which brings together laboratories and clinical centers from Nantes, Angers, Tours, Caen, Toulouse, Orléans, Rennes and Strasbourg.
The ArronaxPlus equipment of excellence of the "Investissements d'Avenir" program another example of scientific and technological consortium, managed by Arronax with the goal of offering a coordinated group of technological platforms, from chemistry to clinical nuclear medicine, to help in the development of radiopharmaceuticals in all medical domains including oncology, cardiology and neurology.
Arronax is also a partner in two government subsidized academia/industry partnership: Theranean and QuantiCardi. These two projects are funded through the FUI (Fonds Unique Interministériel) managed by the Oséo agency:

- **Theranean** (Therapy through Neutron Activation using Nanoparticles) aims at developing a neutron activation device driven by a highly intense 70 MeV proton beam delivered by the Arronax cyclotron to activate holmium nano and microparticles for the treatment of cancer by brachytherapy. It is coordinated by the AAA company (Saint-Génis-Pouilly, France) and involves Subatech, the Nano-H S.A.S. company, the University Claude Bernard Lyon 1, the INSA-MATEIS (CNRS) laboratory, in collaboration with the Hospices Civils de Lyon.
- The objective of the **QuantiCardi project** is the preindustrial development of an integrated solution consisting in innovative components dedicated to the imaging of myocardic perfusion by Positron Emission Tomography (PET) to measure myocardic blood flow and the coronary reserve. A subnormal coronary reserve is symptomatic coronary insufficiency, disease responsible for 30 50% of death by cardiopathy in Europe. The project is headed by Lemer PAX company and involves Keosys, Subatech, IRCCyN-IVC and the CRCNA.

A large collaborative project, aiming at creating a national academic and industrial cluster in molecular radiotherapy, is being developed with Atlanpole Biotherapies in response to the "Projets Structurants des Pôles de Compétivité call for tender of the"Investissements d'Avenir".

history

After submission of a scientific project in 2001, the cyclotron Arronax reached full power on October 25, 2010 and has been operational since 2011.

2001: the scientific file written by Jacques Barbet, Jean-François Chatal, Jacques Martino and Yves Thomas is presented to scientific authorities and potential funders
May 3, 2002: after scientific expertises from CNRS, Inserm, CEA and Universities, the Ministry of Research formalizes a scientific evaluation of the submitted document
2002-2003: A study of technical and economic feasibility, co-funded by the Regional Council of Pays de la Loire and the State (Prefecture) and led by the University Hospital of Nantes, confirms the technical feasibility and more accurately assesses the costs of investment and operation
December 18, 2003: The project, led by the University of Nantes and supported by the Minister Francois Fillon and the President of the Regional Council of Pays de la Loire, Jean-Luc Harousseau, receives a favorable advice of the Interministerial Committee for the Planning and Development of the Territory (CIADT)
July 9, 2004: Jacques Auxiette, newly elected President of the Regional Council of Pays de la Loire, supports and decides to launch the project to be installed on the northern site of the Nantes University Hospital, on land made available by the hospital. The project ownership is delegated by the State to the Region Pays de la Loire during the building period.
End of 2004 : the financial closure of the estimated investment of € 36.9 million is provided with the following distribution
State: € 8.4 million
Region Pays de la Loire: 14.260 M €
European Funds: 7,490 M €
French
Poitou Charente Brittany Regional Council: 0.750 M €
Pays de la Loire Regional Council: 0,500 M €
General Council of Loire Atlantique: 2.00 M €
General Council of Maine et Loire: 0,300 M €
Nantes Métropole: 3.00 M €
Angers Loire Métropole: 0,200 M €

2005: definition of the equipments and award of public contracts
7 December 2006: laying of the first foundation stone
March 2008: delivery of the cyclotron, then assembly, tests and adjustments
November 7, 2008: Inauguration of the cyclotron and Arronax site in the presence of Prime Minister François Fillon, President of Region Jacques Auxiette, Deputy Mayor of Nantes Jean-Marc Ayrault, Mayor and Senator of Saint Herblain Jean-Charles Gautier

Inauguration le 07/11/2008

de gauche à droite:
J. C. Gautier, Sénateur-maire de St Herblain
J. Auxiette, Président du Conseil régional des Pays de la Loire
Y. Lecointe, Président de l'Université de Nantes
J. Martino, Directeur du GIP Arronax
F. Fillon, Premier Ministre

25 October 2010 :: the cyclotron reaches full power for 24 hours
2011: The cyclotron produces Strontium-82 in routine and R/D allows for the production of copper-64, Ge-68, Scandium-44 and radiolysis, radiobiology and physics experients.

The Arronax GIP

From a statutory point of view, Arronax is a « Groupement d'Intérêt Public » (GIP). Arronax is thus a public research institute with a private accounting. It is a kind of mixed economy system that allows it both to cooperate as equal with public research partners and to act as a company with respect to industrial customers (Siret: 13000411200012, DUNS: 295659028).

A GIP is a grouping of several public and private members, linked by a written agreement, approved by the State. Members of the Arronax GIP are the State and the Region Pays de la Loire, large national organizations of research, higher education and research institutions, hospitals. Members are gathered in an Administration Council which elects its president and vice president.

The Arronax GIP is constituted for a period of 25 years, renewable. First, it supports the operation of the cyclotron Arronax (Accelerator for Research in Radiochemistry and Oncology at Nantes AtlantiX), which is a large platform with an international Research and Development objective. The responsibility for management lies with the director of the GIP, who signs research contracts and service contracts with national or international, public or industrial partners.

Staff members working for the GIP are, researchers, engineers, technicians provided by its members, and employees of the GIP. Some counselors, and several members of the partnership laboratories cooperate regularly on the platform. Depending on the time period, the number of staff members is between 30 and 40.

Scientific Board members

The mission of the International Scientific Council of ARRONAX is to advise the management of the GIP in its strategic choices.

It meets once a year in the fall since 2004. It consists 14 members including 4 technical committee members:
■ **Professor Suresh Srivastava**, Professor of Radiology at the Medical School of the State University of New York at Stony Brook, director of Radionuclide and Research Division Radiopharmaceutical (R & RR) of the Medical Department at Brookhaven National Laboratory, Upton, New York (USA), President.
■ **Professor Marion de Jong**, Professor of Nuclear Biology, Erasmus MC, Rotterdam (Netherlands), Vice-President.
■ **Professor Patrick Bourguet** (Centre Eugène Marquis, Rennes), Professor of Biophysics and Nuclear Medicine, University of Rennes 1 (France).
■ **Professor Patrick Cozzone**, Professor of Biophysics at the Faculty of Medicine of Marseille, head of department at the Hospital La Timone, CHU Marseille, Chair of Biophysics at the Institut Universitaire de France, founder and directorof the Centre for Magnetic Resonance Biological and Medical CNRS (France).
■ **Professor Peter Ell**, Nuclear Medicine, Senior Investigator National Institute for Health Research (NIHR) and Professor Emeritus at University College London (UK).
■ **Professor Denis Guilloteau**, professor of pharmaceutical biophysics, Director of Inserm U619 "Dynamics and pathology of brain development," Director of Nuclear Medicine, CHU Bretonneau, Tours (France).
■ **Dr. Ulli Köster**, nuclear physicist, radioisotope production at the Institut Laue Langevin (Grenoble) and ISOLDE (CERN).
■ **Professor Jörg Kotzerke,** Professor of Nuclear Medicine, Director of the Department of Nuclear Medicine, University Hospital of Dresden (Germany).
■ **Dr. Bernard Laune**, Accelerators Physicist , Head of Mission for the Pole IN2P3 Accelerators CEA / CNRS, Technical Coordinator of the Accelerators Division of the Institute of Nuclear Physics of Orsay, Member of the National Programme for Research in Radiation Therapy (France).
■ **Professor Jacques Martino**, nuclear physicist, director of IN2P3 (CNRS), Paris (France).

Additional members from the Technical Committee:
■ **Professor Michel Chérel**, Professor of Biophysics, Pharmacist, Doctor of the University of Nantes, Centre for Research in Cancer Nantes-Angers, Nantes (France).
■ **Dr. Ferid Haddad**, Senior Lecturer at the University of Nantes, Deputy Director of GIP ARRONAX, Doctor of Physics, Subatech, Nantes (Nantes).
■ **Dr. Vincent Metivier**, Assistant Professor at the Ecole des Mines de Nantes, Doctor of Physics, Subatech, Nantes (France)
■ **Docteur Freddy Poirier**,Research ingineer at the CNRS, Doctor in accelerator physics, ARRONAX, Nantes(France).

The Arronax facility

The Arronax plant is installed in a 4000 m^2 building, located 1 rue Aronnax, Saint Herblain, in the suburbs of Nantes, near the north branch of the University Hospital. This building is built on a 10 000 m^2 plot of land in the Bio-Ouest Laënnec technology park.

The building is split into 3 parts:
• A conventional area (blue on plan) comprising offices, a conference room, the cyclotron control-command room, an electrical supply room and standard service areas (heating equipment, ventilation system, compressor, etc.).
• A controlled area (in yellow) in which all nuclear activities are performed. Here are the cyclotron, its reaction shields, its utilities, the various laboratories, storage areas for nuclear waste, target processing areas (hot cells, etc.)
• A technical space (in green), to allow for growth in activity, or for use by partners and R/D consortiums developed by Arronax.

Research units

Aerosol Physics and Metrology Laboratory (LPMA)

The Aerosol Physics and Metrology Laboratory (LPMA) has long been involved in developing aerosol science in France in conjunction with various academic and industrial partners. It is located at the CEA site in Saclay (Essonne, France). It is directed by François Gensdarmes.

Summary

Context and research themes

The LPMA carries out experimental research, studies and technical assessments related to the characterisation of source terms at facilities, in normal and accident situations.

This involves:

- basic research and studies, as well as applied studies, on aerosol physics and metrology, and on air contamination by natural sources, both radioactive and non-radioactive, and anthropogenic sources (formation, physical and chemical changes, and transfer, mainly via containment barriers);
- research and studies on the physics and chemistry of phenomena likely to occur during an accident situation at a nuclear facility;
- applying the results obtained to define, analyse and develop the related measurement devices, especially atmospheric samplers;
- testing devices used to measure radioactive contamination.

Research axes

Main areas

To fulfil its tasks, the LPMA develops knowledge in three main areas :

- Aerosol physics: sources (suspension, nucleation), changes in space and over time (condensation, evaporation, agglomeration, electrical charge) and aerosol deposition (deposition on surfaces, transfer via ducts).
- Aerosol metrology: measuring physical properties (aerodynamics, diffusion, electrical, aggregate morphology) and sampling methods (performance, use strategy).
- Purification of radioactive gases such as iodine and tritium.

Major research topics currently studied at LPMA

- Particle suspension by air flow or by the fall of potentially dispersible materials (powders, contaminated objects, liquids), suspension during dismantling operations.
- Characterisation of aerosols emitted during a fire situation and the suspension of contaminants emitted by materials subject to fire.
- Nanoparticles (characterising emissions, specific metrology, transfer and deposition, containment and filtration)
- Sampling aerosols for workstation monitoring.
- Radon, thoron and decay product metrology.
- Assessing the performance of air contamination monitors.
- Trapping radioactive iodine and tritium on activated carbon and zeolite filters.

Specialties and researchers

Services provided

- Assessment and characterisation of α,β et γ radioactive contamination measurement equipment.
- Verification and calibration of radon measurement instruments.
- Measurement and characterisation of particle releases at industrial facilities.

Researchers

François Gensdarmes, head of laboratory
Sylvain Bondiguel, technician
Sylvain Fauvel, engineer
Guillaume Davenne, technician
Zakaria Mana, engineer
Nathalie Michielsen, research engineer
Céline Monsanglant-Louvet, research engineer

Publications

LEMAC's publications (till 2003) recorded in this website

LPMA's publications (since 2003) recorded in this website

The co-authored book “History and Reviews of Aerosol Science” (2005) includes a chapter on the research carried out by LPMA and its partners between 1980 and 2001.

The table of contents

Contact

Francois Gensdarmes, head of laboratory

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91191 Gif-sur-Yvette Cedex
France

By phone: +33 (0)1 69 08 55 06

François-Xavier Ouf, engineer
Samuel Peillon, engineer
Sébastien Pontreau, technician
Stéphane Roussel, technician
Audrey Roynette, technician
Philippe Sillon, technician

Facilities and techniques

Facilities

Icare, test bench for performing tests using calibrated radioactive aerosols, with caesium or plutonium markers;

Baccara, test bench for research on the metrology of radon.

Lec, ISO Class 8 dust-monitoring cleanroom.

Bise, air duct for research on the suspension of contaminants by air flow.

Cepia, test chamber for research on the performance of personal and environment aerosol samplers.

Measuring equipment

For aerosol characterisation :

Condensation Nuclei Counter (CNC), Nephelometer and Optical Counters (COP),

Scanning Mobility Particle Sizer (SMPS), Engine Exhaust Particle Size Spectrometer (EEPS), Differential Mobility Spectrometer (DMS),

Diffusion battery, Aerodynamic Particle Sizer (APS), Aerosizer, Electrical Low Pressure and Cascade Impactors (ELPI, Andersen), Coulter Counter,

Tapered Element Oscillating Microbalance (TEOM).

For aerosol generation:

Atomizers, Piezoelectric ceramics, Vibrating Orifices, Rotating brush, Fluidized bed, Vortex Shaker, Voltage pulse generator, Evaporation/condensation generator.

For radioactivity:

MINI 20 proportional counter, □, □ and □ spectrometry, Measurement of the activity concentration of radon

Partnership and research networks

LRGP (CNRS and Nancy University joint laboratory),
CORIA (Rouen University, CNRS and INSA joint laboratory),
LISA (Paris-Est Créteil University, Paris Diderot University and CNRS joint laboratory),
CERTES (Paris-Est Créteil University),
LPGP (Paris-Sud University and CNRS joint laboratory),
INRS,
CEA,
LNE,
AREVA,
EDF,
ONERA

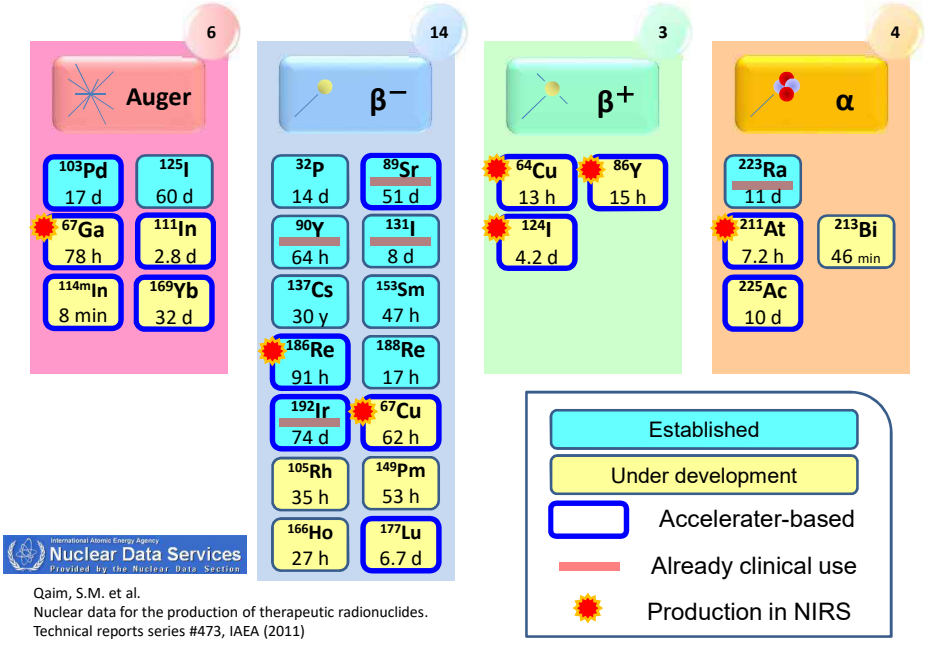
Alpha-emitter Astatine-211; Production and Utilization

Laboratory works and implementation

National Institute of Radiological Sciences (NIRS)
National Institutes for Quantum and Radiological Science and Technology (QST)

Dr. Kotaro NAGATSU

Candidate radionuclides for RNT

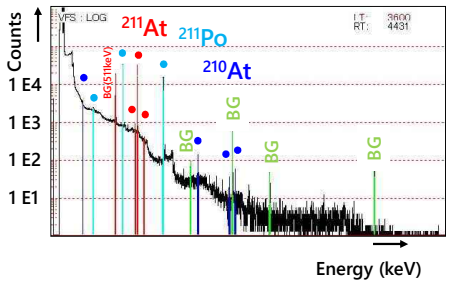


Nuclear Data Services

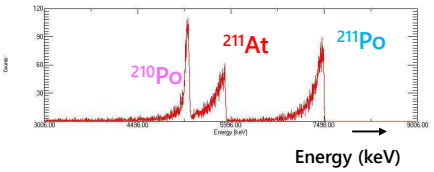
Qaim, S.M. et al.
Nuclear data for the production of therapeutic radionuclides.
Technical reports series #473, IAEA (2011)

Production; Results

Gamma



Alpha



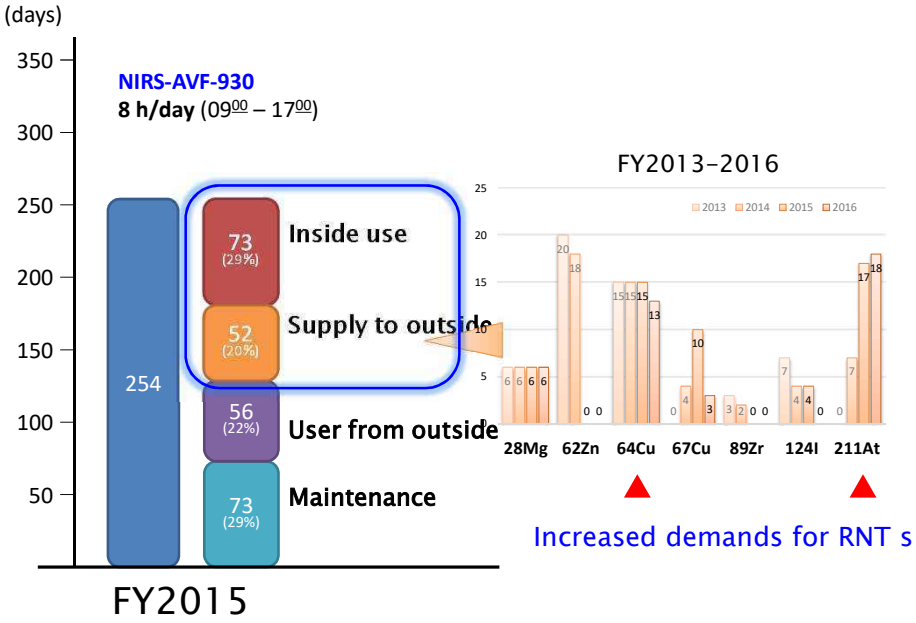
120 h from EOB (~17-$T_{1/2}$ of ^{211}At)

Items	Results
Bombardement	α 28.5 MeV 13 eμA × 2 h
Yield at target	600 μCi/μAh*
Yield	9.5±0.2 mCi**
Purity	>99% (+^{211}Po) (@5 h from EOB)
^{210}At/ ^{211}At	0.0022% @ EOB
^{210}Po mix rate	0.32 nCi/μAh (0.67 ppm of ^{211}At**)
Purification time	~1.5 h

* decay corrected
** decay uncorrected

Recent Condition
12 eμA x 3 h
= 12–14 mCi/ $CHCl_3$ 0.5 cc
@1h EOB

RI Production by Accelerator AVF-930

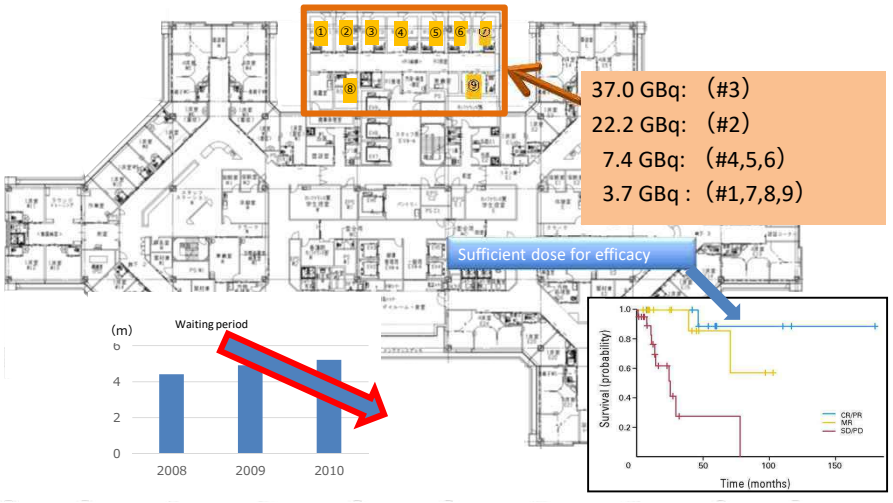


Increased demands for RNT s

Fukushima Medical University
Fukushima Global Medical Science Center
Targeted Alpha Therapy

FMU website

Radionuclide Therapy Ward in Medical Center



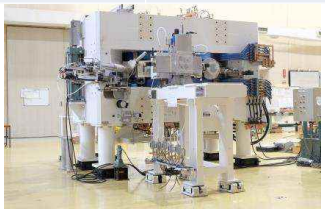
Two cyclotrons for production of radionuclides

HM-20S

Proton	Energy	20 MeV
	Current	150 μA
Deuteron	Energy	10 MeV
	Current	50 μA
Max. Targets		8 (4/Port)
Power		55 kW

MP−30

Proton	Energy	15 - 30 MeV
	Current	100 μA
Deuteron	Energy	16 MeV
	Current	50 μA
Alpha	Energy	32 MeV
	Current	30 μA
Max. Targets		Depend on Requirement
Power		150 kW



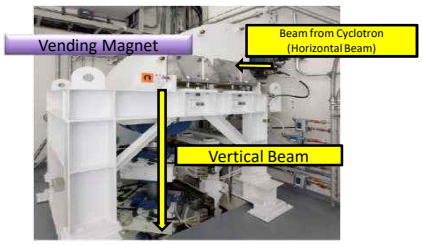
Cyclotron (MP-30, Sumitomo Heavy Industries, Ltd.)

Acceleration energy

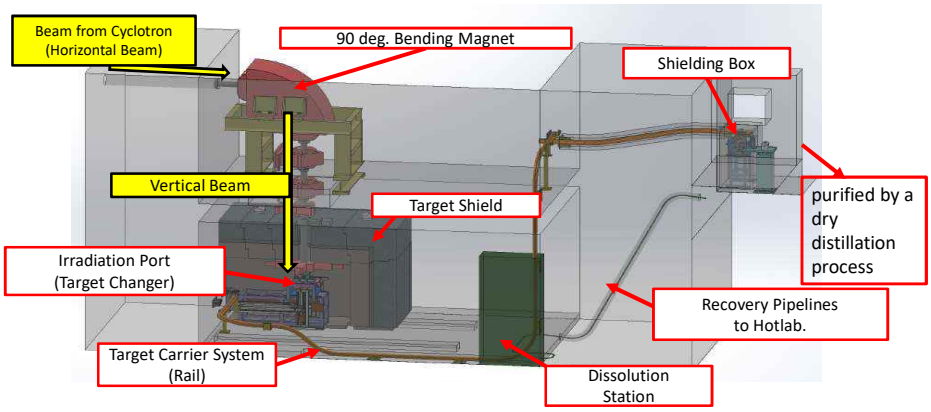
Proton	15 - 30 MeV
Deuteron	8 - 15 MeV
Alpha	32 MeV

For Nuclear Reaction of

(p,x) (d,x) (α,x)



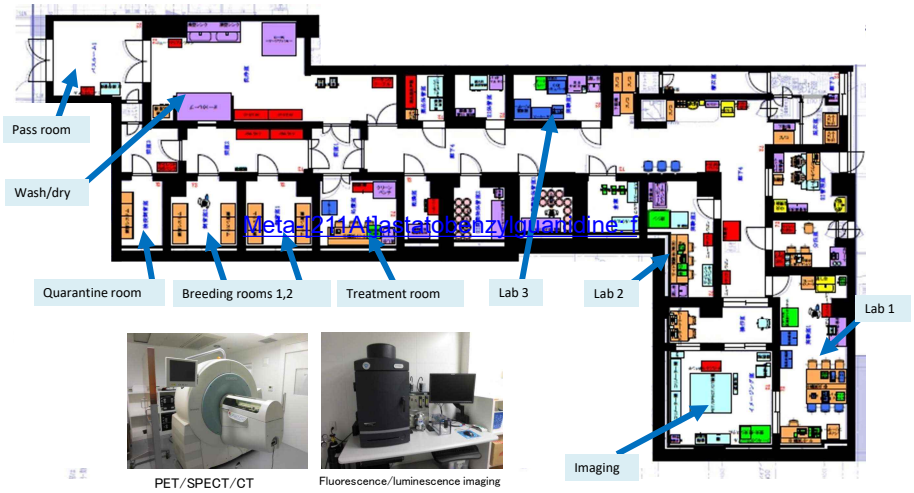
MP-30 Cyclotron



Special Features:

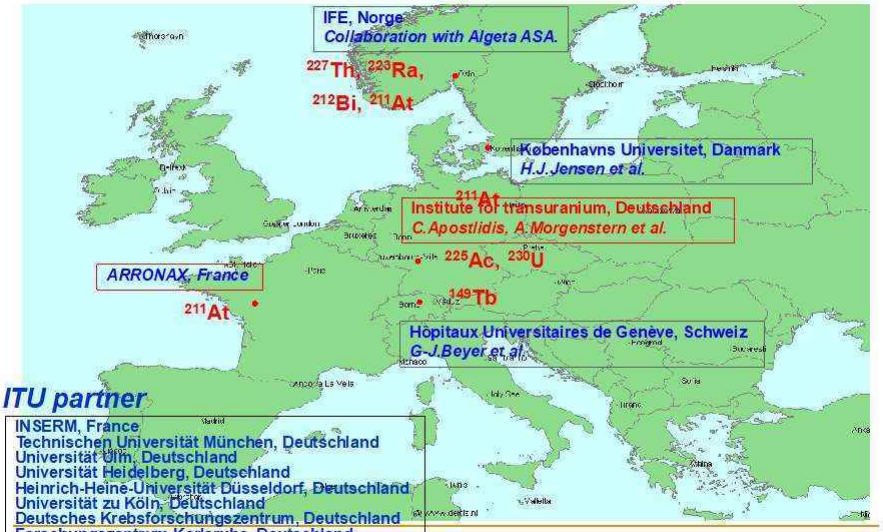
Vertical Irradiation System & Automatic Target Transport System

Preclinical facility for study with radionuclide



参考資料7

Production group of α-emitter in Europe



ITU partner

INSERM, France
Technischen Universität München, Deutschland
Universität Ulm, Deutschland
Universität Heidelberg, Deutschland
Heinrich-Heine-Universität Düsseldorf, Deutschland
Universität zu Köln, Deutschland
Deutsches Krebsforschungszentrum, Deutschland
Forschungszentrum Karlsruhe, Deutschland
Universiteit Gent, Belgie
Virga Jesseziekenhuis, Belgie
Universitätsspital Basel, Schweiz

Ruusia, Poland

Production facilities of α-emitter in Japan

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[3] 日本アイソトープ協会 （2017） アイソトープ等流通統計 2017（抜粋）

[4] IAEA Technical Report Series No.15-A Basic Toxicity Classification of Radionuclides (1963) （抜粋）

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展望 TENBO

α線内用療法の現状と展望

細野　眞
Hosono Makoto
（近畿大学高度先端総合医療センター）

1 はじめに

放射性同位元素（RI）内用療法（内照射療法，核医学治療）とは非密封放射性核種による内部放射線治療であり，放射性核種を含んだ薬剤を病巣（がん，あるいは良性疾患）に選択的に取り込ませて放射線を照射するものである。英語ではUnsealed radionuclide therapy，Radionuclide therapy，Targeted radionuclide therapyなどと呼ばれることが多い。

従来は，RI内用療法にβ線を放出する核種が主として用いられており（同時にγ線を放出する核種もある），甲状腺機能亢進症・分化型甲状腺癌に対する^{131}I（ヨウ素）は1940年代から臨床に利用されてきた歴史を持つ[1]。また，褐色細胞腫・神経芽腫など神経内分泌腫瘍に対する^{131}I-MIBG（カテコラミン類似体）も国内外で約30年来用いられてきた[2]。これらは組織の特異的な取込み機序を応用した古典的かつ優れた分子標的療法と言える。また，転移性骨腫瘍の疼痛緩和療法として向骨性放射性薬剤が1990年前後から世界的に広く使われ，主なものは，^{89}Sr（ストロンチウム，販売名メタストロン），^{153}Sm（サマリウム）-EDTMP（販売名Quadramet）である[3]。さらに，B細胞性非ホジキンリンパ腫治療薬としてRI標識抗CD20モノクローナル抗体が1990年代に入って登場し，米国で2002年に^{90}Y（イットリウム）-ゼヴァリンが，2003年に^{131}I-Bexxar（ベキサール）が相次いで認可された[4]。2010年前後からは，ソマトスタチン受容体を発現する神経内分泌腫瘍に対するRI標識ソマトスタチンアナログ治療がPeptide receptor radionuclide therapy（PRRT）として海外で盛んに行われるようになり，^{177}Lu（ルテチウム）-DOTA-[Tyr3]-octreotate（^{177}Lu-DOTATATE）などが代表的な薬剤である[5]。

現在（2013年6月時点），日本国内で保険収載されている内用療法薬剤は，甲状腺機能亢進症・分化型甲状腺癌に対する^{131}I，固形癌骨転移疼痛緩和の^{89}Sr（2007年保険収載），B細胞性非ホジキンリンパ腫に対する^{90}Y-ゼヴァリン（2008年保険収載）である。

最近，α放出核種であるRadium-223（^{223}Ra，塩化ラジウム-223，Xofigo®，以前はAlpharadinと呼ばれていた）が，転移性骨腫瘍に対する放射性薬剤として登場し，症状を緩和し，骨関連事象（病的骨折など）の出現を遅らせ，生命予後を延長し，さらに副作用の少ない優れた抗腫瘍薬として，欧米での第Ⅲ相臨床試験の結果が2011～12年にかけて報告され，2013年5月15

表 1　臨床利用可能な α 核種の例

核種		半減期	娘核種		崩壊系列	元素分類
^{223}Ra	ラジウム	11.4 日	^{219}Rn		アクチニウム系列	アルカリ土類金属
^{211}At	アスタチン	7.21 時間	^{211}Po	^{207}Bi	アクチニウム系列	ハロゲン
^{212}Bi	ビスマス	60.6 分	^{212}Po	^{208}Tl	トリウム系列	窒素族
^{225}Ac	アクチニウム	10.0 日	^{221}Fr		ネプツニウム系列	アクチノイド
^{213}Bi	ビスマス	45.6 分	^{213}Po	^{209}Tl	ネプツニウム系列	窒素族
^{149}Tb	テルビウム	4.15 時間	^{149}Gd	^{145}Eu	—	ランタノイド

日に米国食品医薬品局（FDA）の承認を得たことから，^{223}Ra を含めた α 放出核種による内用療法に注目と期待が集まるようになった。本稿では，α 放出核種による内用療法について概説する。

2　なぜ α 放出核種による内用療法か

β 放出核種による内用療法が前述のように ^{131}I による甲状腺癌治療，^{90}Y 及び ^{131}I 標識抗体による非ホジキンリンパ腫などにおいて有効であることは議論の余地がないが，様々ながんの治療において内用療法の果たす役割はまだ限定的であると言わざるを得ない。RI 標識抗体による放射免疫療法にしても数多くのがんに対してこの四半世紀試みられたが，いまだに固形癌に対して有効性を確立したものはなく，実用化に至ったのは非ホジキンリンパ腫に対するものだけである。そこで内用療法の有効性を高める研究が精力的に行われ，がんに親和性の高い新規化合物の開発とともに，核種自体についても，様々な物理学的化学的性質を持つ核種が内用療法への応用を試みられ，その中で α 放出核種も取り上げられた[6]。また，世界各地の原子炉や加速器で生成する核種を医療に応用しようという動きも自然なことであった。**表 1** に臨床利用可能な α 核種の例を示す。

α 線の大きな特徴は，高い線エネルギー付与（linear energy transfer：LET）と短い飛程にある。例えば，転移性骨腫瘍治療で用いられている代表的な核種である ^{89}Sr や ^{153}Sm の β 放出核種と α 放出核種である ^{223}Ra を比較すると（**表 2**），^{223}Ra の放射線のエネルギーは ^{89}Sr に比べて数十倍高く，α 線の LET は β 線の LET のほぼ 400 倍（80 keV/μm vs 0.2 keV/μm）である。このように α 線の LET が高いため，DNA 二重鎖切断を起こして損傷の修復がしにくいので，生物学的効果比（RBE）も高く，RBE は用いる指標によって異なる値を示し得るが，α 線の細胞不活化作用に関する RBE は 3.8 であるとの報告がある。なお，ここで留意すべきは，α 線の放射線加重係数が 2007 年 ICRP 勧告等で 20 とされているが，それは放射線防護の見地から設定されている値であり，RBE とはかなり大きく違う点である。α 線の LET が高いことから，酸素増感比（OER）が小さいため低酸素細胞にも有効で，細胞周期依存性が小さいため放射線感受性の低い S 後期細胞にも有効と考えられる。

表 2　向骨性放射性同位元素の物理学的性質[13]

核種	半減期（日）	放出放射線当たりの平均エネルギー（MeV）	組織中の平均飛程（mm）
^{89}Sr	50.5	0.58	2.4
^{153}Sm	1.9	0.22	0.55
^{223}Ra	11.4	27.4*	＜0.10

* 子孫核種の放出エネルギーを含む

また α 線の飛程は非常に短く，例えば ^{223}Ra で 100 μm 以下と，細胞数個分の長さである（**図 1**）。このため α 放出核種が腫瘍にうまく局在すれば，周囲の正常組織の不要な被ばくが少なくなる。その一方で，α 放出核種を腫瘍部位

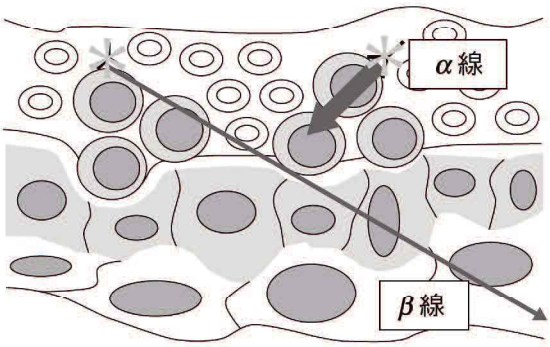

図1　組織中のα線とβ線
α線の飛程は数十μmと標的細胞に留まるが，β線は正常組織にも到達する

に選択的に運ぶドラッグデリバリーの手法が不可欠である。なぜならば，β放出核種の場合ならば，組織内の飛程が数mmあるので腫瘍組織に結合したβ放出核種が，血流や結合部位が少ない腫瘍組織をも照射できる（クロスファイヤー効果）が，α放出核種は飛程の短さゆえに，それを期待できないからである。後述の^{223}Ra（塩化ラジウム-223）はカルシウムと同様に核種そのものが骨に結合して標的部位に照射するが，多くの場合は核種と標的をマッチさせるドラッグデリバリーの手法が不可欠であり，その1つが腫瘍に特異的なモノクローナル抗体に結合し，それを体内に投与し，抗体の力を借りて腫瘍組織まで運搬する放射免疫治療である。1996年にBismuth（ビスマス）-213標識抗CD33抗体が骨髄性白血病に使用されたのが，α放出核種標識抗体がヒトに投与された初めての例とされる[7]。

このような内用療法におけるα放出核種の応用は，外部放射線治療の分野での粒子線の応用にシンクロするとも考えられる。

3　塩化ラジウム-223による骨転移治療の現状

最近，α放出核種である^{223}Ra（塩化ラジウム-223，Xofigo®）が，転移性骨腫瘍に対する抗腫瘍薬としてノルウェーのAlgeta社（www.algeta.com）によって開発された。^{89}Srが骨転移疼痛緩和の対症療法薬であるのに対して，^{223}Raは抗腫瘍薬として生命予後の改善を示す。現在Algeta社とバイエル社とのパートナーシップによって国際市場への導入が進められている。

元素としてラジウムはカルシウム，ストロンチウムと同様のアルカリ土類金属であり，骨に親和性がある。^{223}Ra（半減期11.4日，α：5.716 MeV）は骨代謝の亢進した骨転移部位に集積して，病巣を照射する。そのα線の飛程が組織中で100 μm以下と短いため骨髄の被ばくが少なく，腫瘍に選択的に高い線量を与えることができる。^{223}Raから娘核種の^{219}Rn，^{215}Po，^{211}Pb，^{211}Bi，^{207}Tlと壊変を経て安定核種^{207}Pbに至るまでα線とともにβ線，γ線も放出する（**図2**）。^{223}RaはActinium-227（アクチニウム，半減期21.77年）から取り出すことができ，半減期11.4日と医薬品として製造して世界中に運搬するのにも適し，医療現場でも扱いやすい。

^{223}Ra開発の経緯に関しては，2005年に乳癌と前立腺癌の骨転移症例を対象にした第Ⅰ相臨床試験の結果が報告され[8]，2007年に前立腺癌の骨転移症例を対象にした第Ⅱ相臨床試験の結果が報告された[9]。さらに，2008年から実施された第Ⅲ相臨床試験ALSYMPCA（ALpharadin in SYMptomatic Prostate Cancer）で去勢抵抗性（ホルモン抵抗性）前立腺癌多発骨転移症例における大規模な臨床試験が欧米で実施され，全生存期間を延長し，骨関連事象（病的骨折など）発現までの期間を延長することが2011～12年にかけて公表された[10,11]。報告によれば，全生存期間中央値は^{223}Ra群の14.9か月と全く治療効果のないプラセボ群の11.3か月，骨関連事象発現期間中央値は^{223}Ra群の15.6か月，プラセボ群の9.8か月と^{223}Raによって有意な延長を示した。一方副作用に関しては，α線は生物学的効果が大きいので副作用が強いのではないかとの予想もあるかもしれないが，実際には軽微であり，grade 3～4の好中球減少は1.8%，grade 3～4の血小板減少は4.1%と骨髄抑

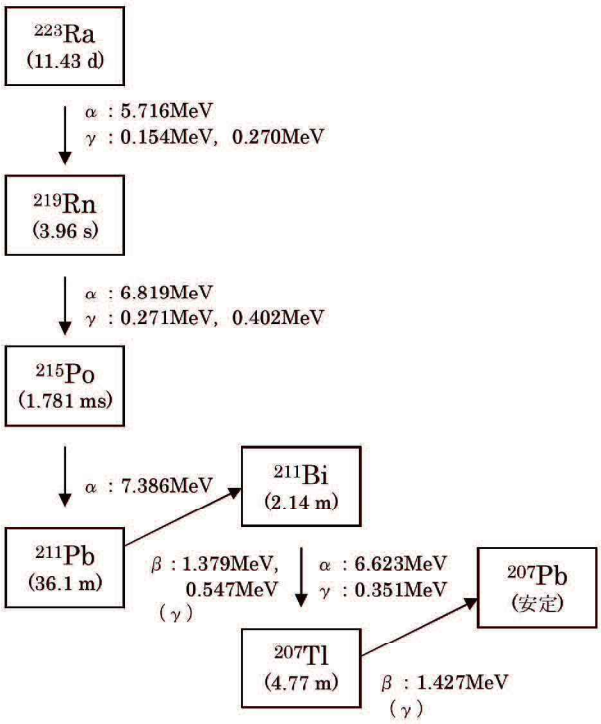

図 2　^{223}Ra 壊変系列

制が軽度にとどまり，ほかには悪心・嘔吐・下痢が主なものであった。この報告を受けて，欧米で去勢抵抗性前立腺癌多発骨転移に対する医薬品としての申請がなされ，米国では 2013 年 5 月 15 日に承認された。今後の展開として，乳癌やその他の固形癌への適応も検討され，また ^{223}Ra と化学療法を併用してより高い治療効果を得ることも考えられている。

α 放出核種である ^{223}Ra の医療の国内導入の可否に関しては，2010 年以前には懐疑的な見方があった。それは，免除レベル取り入れによる法令改正（放射線障害防止法 2005 年 6 月 1 日施行）以前には α 放出核種というだけで，定義数量が 4 群の中で最も厳しい 3.7 kBq（第 1 群）であったり，^{226}Ra（半減期 1,600 年）/^{222}Rn（ラドン，半減期 3.824 日）から半減期が長いのではないかとの連想があったりしたことが原因かもしれない。法令改正前の定義数量（第 1 群）3.7 kBq は，法令改正後は，下限数量 100 kBq となり取り扱いやすくなった。いずれにしても，^{223}Ra はもともと医療法施行規則に基づく告示の別表にも掲載され，医療法の枠内で用いることのできる核種である。

日本核医学会は，ワーキンググループ「α 線を用いたがんの最小侵襲治療法のあり方について」（代表 井上登美夫）において 2008 年頃から α 放出核種の内用療法への応用を検討し[12]，また平成 22 年度厚生労働科学研究費補助金研究「医療放射線の安全確保と有効利用に関する研究」（研究代表者 細野　眞）は，^{223}Ra の有効性と放射線防護の要件を検討して，^{223}Ra を国内医療へ導入するための環境整備を進めた[13]。また ^{223}Ra 内用療法実施において，管理は通常は光子を測定して行うので，^{223}Ra から放出される光子に対するサーベイメータ等の計測器の特性を検討することが重要である[14]。

4　α 線内用療法の展望

α 線内用療法を推進するに当たって幾つかの重要な要素があるが，放射線生物学，核種製造と放射性薬剤合成，線量評価の各分野の研究が代表的なものであろう。

放射線生物学の分野では，α 線の生物学的効果について取り組むべきテーマは多く，^{223}Ra を例にとると，なぜ ^{223}Ra が転移性骨腫瘍に対して効くのかについては解明されていないことも多い。α 線の飛程が組織中で 100 μm 以下であるので，骨の沈着部位から腫瘍細胞に十分に照射されるのかどうかという疑問がある。ある程度はクロスファイヤー効果が寄与しているのであろう。また，α 線照射によって組織中に産生されるサイトカインが直接照射を受けない腫瘍細胞にも作用を与えるという Bystander effect の関与も考えられている。

核種製造と放射性薬剤合成の分野では，加速器や原子炉から効率的に α 放出核種を製造する技術が重要であるのはもちろんであるが，α 放出核種固有の性質，つまり安定核種になるまで複数の壊変を伴い，元素の物理学的化学的性質が遷移するという点に対応して，元素の性質

が変化しても安定にリガンドに結合する手法の開発が不可欠である。通常のキレートでは核種の化合物への安定な結合が得られにくいので，リポソームを使ってα放出核種を化合物に結合させる手法などが試みられつつある[15]。

線量評価として，放射線生物学的な組織内での微細な評価（microdosimetry）が基礎となるが，イメージングに基づいて生体内での核種の分布・動態を確認し[16]，適切な量と種類の放射性薬剤を処方することも，これからの内用療法に不可欠な要素である。今後，個別化医療を実現するためにイメージングを用いた患者さんごとの線量評価が求められるようになるであろう。

このようなα線を含めて内用療法の諸課題について世界中の専門家たちが一堂に会して熱い議論を行っているのが，International Symposium on TArgeted Radiotherapy and Dosimetry（ISTARD）であり，2012年10月のヨーロッパ核医学会（EANM，European Association of Nuclear Medicine，開催地ミラノ）の際に4th ISTARDが同時開催された。2004年，2006年にはそれぞれヘルシンキ，アテネでEANMと合わせて，2009年にはトロントでSNM，Society of Nuclear Medicineと合わせて開催された。最先端の内用療法を知る上で欠かせない学会である。ISTARDには様々な分野の専門家が参加しているが，とりわけ線量評価を担っている医学物理の専門家が存在感を発揮しつつある。

5 まとめ

α線内用療法は，科学の進歩がもたらした新しいがん治療として注目され，その開発や実施には，医療従事者はもちろん，放射線生物学，核種製造と放射性薬剤合成，線量評価などの高度な分野の専門家が密接に連携することが必要であり，産業界や行政を含めた多数の関係者を巻き込む巨大プロジェクトとして，社会に大きなインパクトを与えようとしている。

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REVIEW

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Targeted alpha therapy using short-lived alpha-particles and the promise of nanobodies as targeting vehicle

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ABSTRACT

Introduction: The combination of a targeted biomolecule that specifically defines the target and a radionuclide that delivers a cytotoxic payload offers a specific way to destroy cancer cells. Targeted radionuclide therapy (TRNT) aims to deliver cytotoxic radiation to cancer cells and causes minimal toxicity to surrounding healthy tissues. Recent advances using α-particle radiation emphasizes their potential to generate radiation in a highly localized and toxic manner because of their high level of ionization and short range in tissue.

Areas covered: We review the importance of targeted alpha therapy (TAT) and focus on nanobodies as potential beneficial vehicles. In recent years, nanobodies have been evaluated intensively as unique antigen-specific vehicles for molecular imaging and TRNT.

Expert opinion: We expect that the efficient targeting capacity and fast clearance of nanobodies offer a high potential for TAT. More particularly, we argue that the nanobodies' pharmacokinetic properties match perfectly with the interesting decay properties of the short-lived α-particle emitting radionuclides Astatine-211 and Bismuth-213 and offer an interesting treatment option particularly for micrometastatic cancer and residual disease.

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1. Introduction

1.1. Targeted radionuclide therapy

The evolution of modern medicine during the second half of the twentieth century has improved the clinical outcome of patients with numerous forms of cancer. Today, the treatment of cancer generally consists of surgery, systemic chemotherapy, radiation therapy (including external beam radiation), immunotherapy, antihormone therapy, targeted radionuclide therapy (TRNT). The choice depends upon the location and grade of the tumor and the stage of the disease, as well as the general state of the patient. Presently, tumor reduction by chemotherapy is increasingly being used in combination with surgery in multiple cancer types. Chemotherapy interacts with vital processes of the cell cycle or cell metabolism, thereby stopping or reversing cancer growth. Chemotherapy does not distinguish cancer cells from certain healthy cells, making it a less specific treatment option. External beam radiation is not suited for disseminated disease and immunotherapy often has to deal with specific resistance issues.[1]

The main objective of TRNT is the ability to selectively deliver cytotoxic radiation to cancer cells that causes minimal toxicity to surrounding healthy tissues, using optimized vehicles that deliver a nuclear payload into the tumor cells. TRNT is a growing and favorable treatment option for cancer. Currently, two principal categories can be distinguished. First, there are agents that accumulate naturally in tumor tissue. Examples are Iodine-131 (^{131}I) for the treatment of differentiated thyroid cancer [2] and Strontium-89 (^{89}Sr) and Radium-223 (^{223}Ra) for the treatment of bone metastases.[3,4] ^{131}I and ^{89}Sr are both β^--particle-emitting radionuclides, while ^{223}Ra is an α-particle-emitting radionuclide. The second category includes agents that target tumor-associated antigens that are aberrantly present in malignant tissue. Examples are Yttrium-90 (^{90}Y)- and Lutetium-177 (^{177}Lu)-octreotide as radiolabeled peptides to treat somatostatin-overexpressing neuroblastoma.[5–7] In addition, monoclonal antibodies (mAbs) are also used as vehicles to target tumor-associated antigens and hereby providing a specific internal radiotherapy.[8] The only regulatory-approved radiolabeled mAb is ^{90}Y-ibritumomab to treat non-Hodgkin lymphoma.[9,10]

Thus, a radiopharmaceutical usually consists of two parts: a targeting biomolecule that specifically determines the localization of the radiopharmaceutical and a radionuclide that delivers the mechanism of action through its decay. Today, radiopharmaceuticals are used as either diagnostics for non-invasive imaging through the detection of γ-rays using positron emission tomography (PET) or single-photon emission computerized tomography (SPECT), and/or as therapeutics to deliver radiation to the targeted tumor cells. When

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Article highlights

- Due to the short range in tissue and high linear energy transfer of α-particles, targeted alpha therapy (TAT) is ideal for micrometastatic or residual disease
- Nanobodies are the smallest antibody-derived antigen-binding fragments and have superior characteristics compared to classical mAbs and their derived fragments for *in vivo* cell targeting
- Nanobodies are being evaluated intensively as both diagnostic tracers for nuclear imaging and vehicles for TRNT
- The combination of the short-lived α-particle emitters ^{211}At and ^{213}Bi and nanobodies offer new possibilities for their application in TAT, which will most likely be demonstrated by ongoing and planned research work.

The box summarizes key points contained in the article.

radiopharmaceuticals are employed both for diagnosis and therapy, they are referred to as 'theranostic agents.' This combined diagnostic–therapeutic procedure uses a diagnostic test to determine whether a patient may benefit from a specific therapeutic drug, allowing personal, structural, and functional characterization of a tumor during therapy. Moreover, the therapy response could be measured throughout the therapy.

In general, there are three types of radiation that can be used for TRNT: β$^-$-particles, Auger electrons, and α-particles. Each radionuclide is characterized by its own decay properties, tissue range, half-life, and chemistry, proposing the opportunity to adapt the features of the radionuclide to a particular type of cancer and in the long run to the needs of an individual patient.[11] Until now, TRNT has been mainly explored using β$^-$-particle-emitting radionuclides. β$^-$-Particles have a low linear energy transfer (LET) (0.2 keV/μm), producing repairable DNA damage including single- or double-stranded DNA breaks, base chemical modifications, and protein crosslinks. In case of low-LET radiation, like for example β$^-$-particles, the damage caused by direct ionization of the target might only be sublethal, if dosed insufficiently high. Indirect effects caused by reactive oxygen species (ROS) also contribute to the eventual damage. β$^-$-Particles have a relatively long range in tissue (1–10 mm), causing cytotoxic damage in surrounding nontargeted cells, referred to as 'crossfire effect.' This might be useful for the treatment of heterogeneous, bulky tumors, but it has the disadvantage of damaging surrounding normal tissue. Most progress with β$^-$-particle radiation has been made in hematological malignancies, while the progress in epithelial-derived tumors has been slow. One of the shortcomings of low-LET β$^-$-particle-emitters is that much more of the radioactivity need to reach the tumor tissue to effectively kill it, compared to high-LET α-particles. A single α-particle is sufficient to destroy the cell nucleus, as cell death due to the α-radiation is largely independent of oxygenation or active cell proliferation. β$^-$-Particles on the other hand need much more hits at the level of the cell nucleus as they produce sparse ionization and individual DNA lesions, mostly repairable. This disadvantage is one of the reasons for the average success of agents labeled with β$^-$-particle-emitting radionuclides in clinical trials. Theoretically, Auger-electron emitters present multiple advantageous characteristics, making it an attractive candidate for TRNT. Auger emitters have a short effect range (subcellular, order of nanometers), a LET of 4–26 keV/μm, and are able to produce a high level of cytotoxicity due to Auger electron cascades. These cascades, by which electrons, carrying a characteristic kinetic energy, are ejected from atoms in response to a downward transition by another electron in the atom. In contrast to α-radiation, Auger radiation is of low toxicity when decaying outside the cell nucleus, such as in the cytoplasm or outside of cells, and will therefore cause little damage to nontargeted cells. Some studies have shown that Auger electrons can be effective when targeted only to the cell membrane.[12] However, it is generally considered that the radioisotope needs to be delivered close to the cell nucleus in order to be effective, which makes internalization into the cell crucial.[13]

1.2. General considerations of targeted alpha therapy

The selection of the appropriate radionuclide depends on its decay properties, namely the physical half-life and emission characteristics. For the management of bulky and heterogeneous tumors, treatment with β$^-$-particle-emitting radionuclides might be the preferred approach. However, for the eradication of small-volume tumors and small clusters of cancer cells, agents that emit high-energy α-particles would be more beneficial due to their highly specific toxic load to the targeted tumor cells and their short range in tissue. Thus, the main strength of targeted alpha therapy (TAT) is the potential to deliver radiation in a highly localized and toxic manner, because of their high level of ionization produced and short range in tissue.[14] An α-particle consists of a ^{4}He nucleus; therefore, it is much heavier than other subatomic particles emitted from decaying radionuclides and nuclear reactions. The main characteristics of currently available α-particle-emitting radionuclides are summarized in Table 1.[12] With a charge of +2, α-particles are effective ionization agents with a high LET (50–230 keV/μm) at a short range of 50–100 μm in tissue. They induce clusters of DNA damage such as double-stranded DNA breaks and base chemical modifications that evoke a large number of cellular responses and pathways that include apoptosis, autophagy, necrosis, and cell-cycle arrest. This type of damage is difficult to repair by the cell. Moreover, the damage is independent from the generation of indirect ROS, leaving their effectiveness potentially unabated by tumor hypoxia.[12] These characteristics make α-emitters effective in eradicating small clusters or isolated cancerous cells with little exposure to surrounding healthy tissue. Thus, TAT is of high interest for the treatment of micrometastatic and minimal residual disease after surgery. Moreover, the concept of TAT has moved from bench to bedside, with increasing clinical experience in, for example, ovarian cancer, metastatic prostate cancer, gliomas, and acute myeloid leukemia (Table 2). A median survival of 8.9 months could be achieved after intravenous administration of the α-immunoconjugate, Bismuth-213 (^{213}Bi)-cyclic diethylenetriaminepentaacetic acid anhydride (cDTPA)-9.2.27, in patients with metastatic melanoma in a phase I trial.[15] Using TAT to treat metastatic melanoma, α-particles reach the endothelial cell nuclei, causing cell death and leading to capillary closure and interruption of nutritional support to the tumor. If enough capillaries are closed down, the tumor might regress and could even disappear. Thus, this

Table 1. Main characteristics of the currently available α-particle-emitting radionuclides.

Isotope	Daughter isotopes*	Physical half-life	Maximum energy (keV)	Occurrence (%)	Associated emissions
^{211}At	–	7.2 h	5.867	α (41.8%)	α, γ, LEE
	^{211}Po	516 ms	7.450	α (100%)	
^{225}Ac	–	10 days	5.830	α (100%)	α, γ, Auger, β$^-$
	^{221}Fr	4.9 min	6.341	α (100%)	
	^{217}At	32.3 ms	7.069	α (99.98%)/β$^-$ (0.01%)	
	^{213}Bi	45.6 min	6.051	α (2.2%)/ β$^-$(97.8%)	
	^{213}Po	4.2 μs	8.377	α (100%)	
^{213}Bi	–	45.6 min	6.051	α (2.2%)/β$^-$ (97.8%)	α, γ, Auger, β$^-$
	^{213}Po	4.2 μs	8.377	α (100%)	
^{212}Bi	–	61 min	5.870	α (36%)/β$^-$ (64%)	α, γ, Auger, β$^-$
	^{212}Po	298 ns	8.785	α (100%)	
^{227}Th	–	18.72 days	6.038	α (100%)	α, γ, Auger, β$^-$
	^{223}Ra	11.4 days	5.871	α (100%)	
	^{219}Rn	4 s	6.819	α (100%)	
	^{215}Po	1.8 ms	7.386	α (100%)	
	^{211}Bi	2.14 min	6.623	α (99.7%)/β$^-$ (0.3%)	
^{212}Pb	–	10.64 h		β$^-$ (100%)	β$^-$
	^{212}Bi	61 min	5.870	α (36%)/β$^-$ (64%)	α, γ, Auger, β$^-$
	^{212}Po	0.3 μs	8.785	α (100%)	
^{223}Ra	–	11.4 days	5.871	α (100%)	α, γ, Auger, β$^-$
	^{219}Rn	4 s	6.819	α (100%)	
	^{215}Po	1.8 ms	7.386	α (100%)	
	^{211}Bi	2.14 min	6.623	α (99.7%)/β$^-$ (0.3%)	

*Generated α-particle emitter after decay of the conjugated parent.
LEE: Low-energy electron emission; NS: yield not significant.

subtype of TAT targets specifically the vasculature and has been referred to as 'tumor anti-vascular α-therapy (TAVAT).'[16] TAT has been compared to β$^-$-particle-emitting radionuclides in several clinical trials, highlighting their promising therapeutic potential. For example, investigators compared ^{131}I-labeled bisphosphonates with their Astatine-211 (^{211}At)-labeled counterparts for pain relief in patients with bone metastasis.[17] In addition, Henriksen et al. explored the bone-seeking properties of ^{223}Ra and compared it with those of the β$^-$-particle-emitting radionuclide ^{89}Sr.[3] The conclusion of both studies was that α-particle radiation showed a lower toxic effect to the healthy bone marrow compared to β$^-$-particle emitters, which is attributed to the reduced crossfire effect. This and other studies indicated that the strength and short distance of high-LET α-particles make them more suitable than low-LET β$^-$-particles in particular circumstances. Despite its positive features, the translation of TAT into the clinic has been slow, mainly due to the limited radionuclide availability and the short physical half-life and daughter α-particles of some of the available α-emitters. Furthermore, several other issues concerning α-particle emitters should be addressed as well, which are discussed in the following paragraphs.

1.2.1. Radiolysis

Radiolysis is the dissociation of molecules by nuclear radiation. The magnitude of energy deposits by volume of α-particle emitters is two times greater than that of β$^-$-emitters such as ^{90}Y or ^{131}I. Because of this, the potential impact of radiolysis effects when using α-particles is noticeably higher. Hence, the radiolabeling of certain vectors with an α-particle emitter using high levels of radioactivity while maintaining appropriate biological properties may be challenging.[51] Studies by Zalutsky et al. indeed emphasize the potential importance of radiolysis-mediated effects on the chemistry of α-particle-emitting radiopharmaceuticals and the need to evaluate their labeling chemistry and stability at high doses required for clinical use.[63,64]

1.2.2. The radiation-induced biological bystander effect

The radiation-induced biological bystander effect (RIBBE) is a process whereby nontargeted healthy cells are damaged, not as a result of directly being hit by radiation, but via the radiation-induced death or stress of neighboring cells. As α-particle-emitting radionuclides have a range in tissue that is equivalent to only a few cell diameters, the physical crossfire effect will be limited. To date, the majority of studies of RIBBE have been performed *in vitro* using single-cell or multicellular systems *ex vivo* or in artificial three-dimensional human tissue systems. Boyd et al. demonstrated that cell death in adjacent cells after treatment with α-particle-emitting radionuclides might be enhanced via RIBBE.[65] Furthermore, evidence on the *in vivo* effectiveness of RIBBE has been limited, but new findings indicate that they may affect tumor development in susceptible mouse models. For example, Mancuso et al. demonstrated that DNA double strand breaks and apoptotic cell death could be induced by bystander responses in mouse cerebellum after X-ray exposure of the remainder of the body.[66] Mice were whole-body exposed or irradiated with individual cylindrical lead shields providing protection of heads. Whole-body-irradiated animals developed cerebellar tumors. A high percentage of mice (62%) died of aggressive disease by 23 weeks, with median survival of 14 weeks. Significantly, they also observed a remarkably increased medulloblastoma rate (39%) in lead shielded-irradiated mice, indicating that bystander effects are factual *in vivo* events with carcinogenic potential. However, the underlying mechanisms are incompletely characterized and it remains unclear how processes involving oxidative metabolism and stress-inducible proteins lead to (oxidative) DNA damage in bystander cells.[67]

1.2.3. Distribution of recoil daughters in the body

Another important aspect that should be taken into account is the unstable bond of daughter isotopes upon α-decay due to the different chemical properties of the daughters. This could result in an immediate loss of the daughter atom from the chelating chemistry.[68] In addition, the recoil energy of the recoiling daughters is more than 1000 times higher than the binding energy of any chemical compound, which will lead to the rupture of the chemical bonds of the daughter atom with the targeting vehicle, as well as to the ionization of the surrounding medium. The released daughter isotopes that are often themselves α-emitters might cause substantial harm since they will no longer be bound to the targeting vehicle. Therefore, it is of utmost importance to study the fate of both mother and daughter isotopes. For instance, the biodistribution of the bone-targeting radiopharmaceutical

Table 2. Vehicles used in targeted α-particle therapy in preclinical and clinical settings.

Radionuclide	TAT agent	Indication	Antigen	Reference (preclinical data)	Reference (clinical phase)
^{225}Ac	Anti-CD33 IgG (HuM195)	Leukemia	CD33	[18]	I [19,20]
^{225}Ac	Anti-HER2 IgG (trastuzumab)	Ovarian cancer	HER2	[21]	–
^{227}Th	Anti-HER2 IgG (trastuzumab)	Breast and ovarian cancer	HER2	[22,23]	
^{227}Th	Anti-CD20 IgG (rituximab)	Non-Hodgkin lymphoma	CD20	[24,25]	
^{213}Bi	Anti-CD33 IgG (HuM195)	Leukemia	CD33	[26,27]	I and I/II [28,29]
^{213}Bi	Anti-CD20 IgG (rituximab)	Non-Hodgkin lymphoma	CD20	[30,31]	I [32]
^{213}Bi	Plasminogen activator inhibitor type 2	Breast cancer, pancreatic cancer	Urokinase plasminogen activator receptor	[33–35]	
^{213}Bi	Anti-MUC1 IgG (C595 IgG)	Ovarian cancer, pancreatic cancer	MUC1	[36,37]	
^{213}Bi	Substance P	Glioblastoma	Neurokinin type-1 receptor		0/I [38,39]
^{213}Bi	Anti-NG2 IgG (9.2.27 IgG)	Melanoma	NG2 proteoglycan	[40,41]	I [15,42,43]
^{213}Bi	Anti-CD138 IgG	Multiple myeloma	CD138	[44]	
^{213}Bi	Anti-PSMA IgG (J591 IgG)	Prostate cancer	PSMA	[45]	
^{213}Bi	C6.5K-A scFv, C6.5K-A diabody	Breast and ovarian carcinomas	HER2	[46]	
^{212}Pb/^{212}Bi	Anti-HER2 IgG (TCMC-trastuzumab)	Ovarian cancer	HER2	[47,48]	[48–50]
^{211}At	Chimeric 81C6 IgG	Glioblastoma	Tenascin-C	[51,52]	II [53]
^{211}At	MX35 F(ab')$_2$	Ovarian cancer	NaPi2b	[54]	I [55]
^{211}At	Anti-FRA IgG (Mov18)	Ovarian cancer	Folate receptor alpha	[56]	
^{211}At	Anti-EGFRvIII IgG	Glioblastoma	EGFRvIII	[57]	
^{211}At	Anti-HER2 C6.5 diabody	Breast cancer	HER2	[58]	
^{211}At	$Z_{HER2:342}$ and $(Z_{HER2:4})_2$ affibody molecules	Breast and ovarian carcinomas	HER2	[59]	
^{223}Ra	^{223}Ra-chloride	Skeletal breast and prostate cancer metastases	Hydroxyapatite	[60]	I–III [61,62]

NG2: Neural/glial antigen 2; PSMA: prostate-specific membrane antigen; EGFRvIII: epidermal growth factor receptor variant III.

^{223}Ra, which naturally targets the hydroxyapatite matrix in the bone, has been studied extensively *in vivo*.[3,69] Although the daughter isotopes are not intrinsically bone-seeking, the rapid cascade of α-particle-emitting daughters will deliver high doses to bone metastases. However, their short half-life appears to prevent them from causing major damage to healthy tissue. An *in vivo* study demonstrated that less than 2% of the daughters migrate away from the bone surface within 6 h after administration of ^{223}Ra, and after 3 days, this number has dropped down to less than 1%.[3] Another example is the decay of actinium-225 (^{225}Ac) with the formation of potentially disadvantageous radiotoxic daughter products such as ^{213}Bi. It is critically important to reduce the redistribution of the daughter isotopes to nontarget tissues and to diminish systemic radiotoxic events. Therefore, the ^{225}Ac 'nanogenerator' approach was designed in which the delivery system is engineered to be internalized into the targeted tumor cell.[70] McDevitt and colleagues demonstrated the ability to safely and efficiently use ^{225}Ac as a potent tumor-selective generator in both established solid carcinomas and disseminated cancers.[71] Although these results were very promising, additional development of this modality is warranted to optimize the stability of the nanogenerator to maximize the retention of the tumor while avoiding uptake in healthy organs.

1.2.4. Dosimetry

Radiation dosimetry is the measurement of the absorbed dose delivered by the ionizing radiation and provides a basis for understanding the effects and efficacy of different radiation-based treatments. One of the major impediments of TRNT is the heterogeneous distribution of the radiopharmaceutical in normal and tumor tissues. In the case of α-particle radiation, their short path length and high LET need to be taken into account, posing an enormous challenge on the methods needed for relevant dosimetry.[72] For high-LET irradiation, the effect of a single incident in the nucleus of the cell is so abundant that the variations in absorbed dose (specific energy) to the nucleus can be very large and therefore might be a misleading index of the biologic effect. The clinical quantification of the absorbed doses with the γ-camera is only able to give an estimate about the uptake of the radiopharmaceutical in whole organs and in macroscopic tumors, while quantification of absorbed doses in smaller compartments in organs or microscopic tumors is barely feasible. Thus, small-scale dosimetry or microdosimetry, which takes into account the stochastic nature of energy deposited in small targets, would generate improved dosimetric calculations for α-particle radiation. Due to the limited clinical experience with α-particles to date, unknown maximum tolerable doses in humans are the major issue in TAT. In mice, absorbed doses of α-particle radiation can be calculated in tissues at a macroscopic level (organs and substructures) using Monte Carlo techniques based on fundamental physical principles.[73,74] In addition to that, Bäck and colleagues developed the α-camera, which is a quantitative imaging technique developed to detect α-particles in tissues *ex vivo* at suborgan level, to get a better view on the biodistribution of internal α-radiation on a cellular level.[75] The high-resolution (35 μm or less) α-camera was able to measure the activity distribution on a cellular level by virtue of the short path length of α-particles, making it a promising tool in the evaluation of future TAT.

2. The current developments

2.1. A milestone for TAT: radium-223

Radium (Ra) and polonium (Po) were first described by Marie and Pierre Curie in 1898 while investigating the radioactive

properties of a complex ore, which had radioactive emissions in excess. ^{223}Ra and ^{89}Sr are bone-targeting radiopharmaceuticals with hydroxyapatite ($Ca_5[PO_4]_3OH$) as target, which is an essential component of the inorganic bone matrix. Ra, barium (Ba), Sr, and calcium (Ca) are all chemicals in the alkaline earth metal family on the periodic table and each will localize in the areas of osteoblastic metastases. ^{223}Ra is currently the most commonly used radioisotope for medical therapeutics, showing an increased survival in patients with metastatic castration-resistant prostate cancer [61] and has a half-life of 11.4 days (Table 1). ^{223}Ra is the first α-emitter approved by the US Food and Drug Administration.[76] In addition, ^{223}Ra is the first α-particle-based therapy that results in pain relief and extends survival in patients with progressive castration-resistant prostate cancer and bone metastasis in the absence of visceral metastasis. Thus, ^{223}Ra is naturally incorporated in areas of increased bone turnover in bone metastases.[77] More than 90% of patients with metastatic resistant prostate cancer have radiologic evidence of bone metastases. ^{223}Ra dichloride has been evaluated in two phase I trials and three double-blind phase II trials. The phase III ALSYMPCA (Alpharadin in the Treatment of Patients With Symptomatic Bone Metastases in Castration-Resistant Prostate Cancer) trial showed an improved overall survival of 3 months and pain relief in patients with osseous metastasis.[61] The success of ^{223}Ra as a therapeutic further stimulates TAT-based preclinical and clinical research. In a way, ^{223}Ra could be considered as a game changer in nuclear medicine, as it might facilitate the future use of additional high-LET particle emitters.

2.2. Other promising α-particle-emitting radionuclides

Besides ^{223}Ra, many other α-particle emitters have suitable characteristics for therapeutic applications (Table 2). ^{211}At, ^{213}Bi, lead-212 (^{212}Pb)/bismuth-212 (^{212}Bi), and ^{225}Ac are the most frequently used α-particle-emitting radionuclides in clinical molecular targeting applications to date.[78]

2.2.1. Actinium-225

^{225}Ac is a parent α-particle emitter in a decay cascade that produces three net α-particle isotopes, ^{221}Fr (half-life 4.8 min), ^{217}At (half-life 32.3 ms), and ^{213}Bi (half-life 45.6 min), making it a very effective and potent option for TAT (Table 1). ^{225}Ac has a half-life of 10 days and can be produced by natural decay of ^{233}U in Oak Ridge National Laboratory, USA [79] or by accelerator-based methods in Karlsruhe.[80] However, the latter production of ^{225}Ac also results in the production of ^{227}Ac which decays with a half-life of 21.772 years. The biggest disadvantage concerning ^{225}Ac is its cost, which might reach to $1200/mCi. In addition, the recoiled daughters of ^{225}Ac can do significant damage to healthy tissue when not retained at the tumor site. Encapsulation in a nano-carrier, fast uptake of the α-particle-emitting radionuclides in tumor cells, and local administration are some approaches to minimize toxic effects caused by α-particle-emitting daughters.[68] On the other hand, the relatively long half-life of ^{225}Ac allows a centralized production and shipment of the irradiated targets to further users so that any investigator is able to exploit the power of this α-particle. Furthermore, ^{225}Ac decays to ^{213}Bi, of which the latter also results in a 440 keV γ-ray emission that can be useful for imaging of the therapeutic biodistribution. It should be remarked that it is uncertain whether the measured radioactive decay represents intact radiopharmaceutical or released daughter radioisotopes. Moreover, ^{225}Ac can be conjugated to peptides or antibodies, using an optimized radiochemistry with standard widely available macrocyclic bifunctional chelators.[81,82] *In vivo* experiments showed that the ^{225}Ac complex with 1,4,7,10-tetraazacyclododecane-*N*,*N*′,*N*″,*N*‴-tetra-acetic acid (DOTA) was more stable than the ^{225}Ac complex with 4,7,10,13,16-hexaazacyclohexadecane- *N*,*N*′,*N*″,*N*‴,*N*⁗,*N*⁗′-hexaacetic acid.[70] The biodistribution aspects of ^{225}Ac-labeled mAbs and other carriers, together with their pharmacokinetic properties, radiobiology, and dosimetry, have been reviewed by Miederer et al.[70] A successful phase I trial has demonstrated that a humanized anti-CD33 mAb HuM195 conjugated to ^{225}Ac (Actimab-A) is safe to use at doses ≤0.1 MBq/kg [19] (Table 2).

2.2.2. Bismuth-213

^{213}Bi is most often produced through an ^{225}Ac-generator. The principal drawbacks of using ^{213}Bi are its very short physical half-life of 46 min and limitations regarding availability and cost as for ^{225}Ac. Pippin and colleagues were the first to label ^{213}Bi with mAbs.[83] Moreover, McDevitt and colleagues labeled ^{213}Bi via the bifunctional metal cDTPA complex with a humanized mAb (HuM195) directed against CD33, a glycoprotein expressed on the majority of myeloid leukemia cells.[26] In subsequent studies, the stability of this radiopharmaceutical has been improved to achieve a clinically applicable ^{213}Bi-CHX-A-DTPA-HuM195.[84] A phase I clinical study on 18 patients with acute myelogenous leukemia (AML) or chronic myelomonocytic leukemia showed no significant extramedullary toxicity, although myelosuppression was seen in all patients.[28] The phase I/II trials showed that sequential administration of cytarabine and ^{213}Bi-CHX-A-DTPA-HuM195 was reported to be tolerable and produced remissions in some patients with AML, although myelosuppression was again a common adverse effect.[29] The responses in this high-risk population persisted up to 12 months. In addition, patients with non-Hodgkin lymphoma, malignant melanoma, and glioblastoma have been enrolled in clinical trials with other ^{213}Bi-labeled compounds, showing its relevant potential for TAT (Table 2).

2.2.3. Astatine-211

^{211}At is an α-particle-emitting radionuclide with a physical half-life of 7.2 h and its decay does not result in the production of any relevant daughter isotopes. The first branch decays to ^{211}Po (half-life 526 ms), after which it decays through α-particle radiation to stable ^{207}Pb. In the second branch, ^{211}At α-decays to ^{207}Bi, which then results in stable ^{207}Pb after emission of X-rays. Theoretically, this offers significant advantages for TAT regarding minimal toxicity and quantitative α-particle emission. However, additional clinical research is needed in order to confirm this as a real advantage. The chemical features of ^{211}At are similar to those of iodine, its nearest halogen neighbor, but ^{211}At contrarily also tends to behave as a metalloid. Moreover, the exact behavior of ^{211}At is far from understood due to the limited

knowledge of the chemistry of elemental ^{211}At and the lack of any stable equivalent, which excludes the use of conventional analytical techniques for its characterization.[85] Reasonable yields (0.8–2.5 GBq) of ^{211}At are obtained via the bombardment of natural bismuth targets with α-particles through the ^{209}Bi(α, 2n) ^{211}At nuclear reaction in a cyclotron.[86] The 7.2-h half-life of ^{211}At is well suited for a multistep synthetic procedure. Consequently, a wide variety of tumor-associated antigens that are aberrantly expressed on the cancer cell surface have been targeted by ^{211}At-labeled radiopharmaceuticals.[87,88] To date, ^{211}At has been investigated bound to antibodies, thymidine analogs,[89] biotin analogs,[90] colloids,[91] melanin precursors,[92] substrate carriers,[93] and bisphosphonate complexes.[94] Only two clinical studies have been reported so far with ^{211}At-labeled molecules.[53,55] The first clinical study for the treatment of recurrent brain tumor provides a proof-of-concept for regional targeted radiotherapy with ^{211}At-labeled mAbs.[53] This clinical study demonstrated that the regional administration of ^{211}At-ch81C6 was feasible, safe, and resulted in a possible therapeutic benefit for patients with malignant brain tumors. In the second reported clinical study of ^{211}At using the MX35 F(ab')$_2$, the compound was delivered successfully through intraperitoneal administration without observed toxicity.[55] These two clinical trials showed no subjective toxicity related to the immunoconjugate and the overall outcomes were highly encouraging. However, there are no clinical data on the toxicity of ^{211}At-labeled immunoconjugates after intravenous administration. Further clinical evaluation of ^{211}At-labeled compounds in metastatic tumors or residual disease is warranted.

3. Vehicles for TAT

The attractive feature of TRNT is its adaptable nature. The radionuclide and the targeting vehicle should in principle be matched to each other in the context of the route of administration, disease stage, target accessibility, and site of action. The selection of both the optimal tumor-associated antigen and the targeting vehicle is a crucial step in the development of a new probe for TRNT. The ideal antigen should be overexpressed on cancer cells, while the expression levels on normal, healthy cells should be extremely low.[95] Examples of biomarkers that are targeted in TAT studies are epidermal growth factor receptor variant III, human epidermal growth factor receptor 2 (HER2), folate receptor alpha, tenascin-C, CD20, CD33, and prostate-specific membrane antigen (Table 2). The vehicle molecules should be optimized to provide a high degree of selectivity and specificity toward the target site or 'biomarker.' Below, a section of important vehicles are discussed.

mAbs are Y-shaped proteins that contain two identical *fragment antigen-binding* (Fab) fragments and a *fragment crystallizable* (Fc) region (Figure 1). They are produced by plasma cells (mature, activated B cells) and are recruited by the immune system to identify and destroy foreign objects. Moreover, they have the capacity to bind any potential antigen epitope with high affinity, including tumor-associated biomarkers. Today, a variety of preclinical and clinical investigations were conducted using mAbs labeled with α-particle-emitting radionuclides (Table 2). The melanoma trials (Table 2) using ^{213}Bi-cDTPA-9.2.27 show that solid tumors can be regressed by TAVAT. Moreover, these clinical results demonstrated that TAVAT for melanoma patients were locally efficacious and nontoxic up to 1.4 mCi. In the ^{213}Bi-HuM195 phase I study described above, the authors provided a proof-of-concept for the use of α-particle immunotherapy to treat myeloid leukemia. Although ^{213}Bi-HuM195 was well tolerated and 14 (78%) of 18 patients had reductions in the percentage of bone marrow blasts, myelosuppression was seen in all treated patients.[28] Similarly, myelosuppression and liver function abnormalities were observed in a phase I/II trial investigating antileukemic effects of ^{213}Bi-HuM195 after partial cytoreductive chemotherapy.[29] These toxicities could be explained by the suboptimal pharmacokinetic properties of mAbs as vehicles for TAT. The high molecular weight of mAbs (150 kDa) and the presence of an Fc-region result in a long serum half-life (several days or weeks) and in interactions with Fc-receptors in myeloid and hepatic sinusoidal cells, resulting in higher bone marrow toxicity and accumulation in the liver. Improvement in antibody engineering has led to the development of antibody fragments that are smaller and devoid of Fc, such as 25-kDa single-chain Fv (scFv), Fab (50 kDa), F(ab')$_2$ (110 kDa), diabodies (55 kDa), and minibodies (80 kDa) without compromising their affinity and specificity (Figure 1).[96]

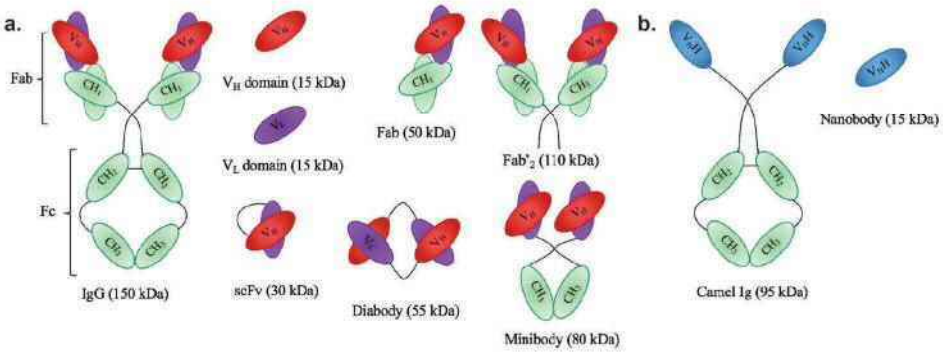

Figure 1. Schematic representation of antibodies and their derived antigen-binding fragments. a. Conventional mAb and the derived Fab, scFv, Fv domains V_L or V_H, Fab'$_2$, minibody and diabody. b. Camelid heavy-chain-only antibody and its V_HH (also known as nanobody).

Smaller engineered mAb derivatives are more rapidly delivered to the tumor and mediate more effective tumor penetration. Because of their smaller size and lack of Fc, they are more rapidly cleared from the circulation, which is indirectly proportional to the level of kidney retention. Therefore, their administration results in fast tumor uptake with high tumor-to-background ratios. One study reported the successful conjugation of ^{213}Bi to anti-HER2 C6.5 scFv and diabody molecules. However, a lack of tumor-specific therapeutic effect was shown, probably resulting from instability of the scFv and diabody molecules *in vivo*.[46] Here, it was concluded that the physical half-life of 45.6 min of ^{213}Bi was too short to allow the systemically administered diabody to specifically localize in an established solid tumor. In a subsequent study, ^{211}At was coupled to the stable *N*-succinimidyl-*N*-(4-[^{211}At] astatophenethyl) succinamate and subsequently conjugated to the C6.5 diabody (Table 2).[58] Here, the somewhat longer physical half-life of ^{211}At matches more closely to the rapid tumor targeting and rather fast systemic clearance of the C6.5 diabody. In the ^{211}At-MX35 F(ab')$_2$ phase I trial, therapeutic doses were reached for the treatment of ovarian cancer.[55] However, 50% of the initial activity concentration of this radionuclide remained in the peritoneal fluids 24 h after injection, indicating a higher toxicity risk related to this immunoconjugate.

Besides antibodies and antibody derivatives, ligands (e.g. folate), synthetic protein scaffolds (e.g. affibodies), and substrate analogs (e.g. peptides) can also be used as targeting agents in order to specifically deliver the toxic radionuclide. [97–99] Affibody molecules are small single domain proteins with a molecular weight of 6.5 kDa that are derived from one of the immunoglobulin binding domains of staphylococcal protein A.[100] Previous research demonstrated that affibody molecules can bind to their targets within minutes after administration. The binding kinetics of affibodies are similar to that of nanobodies, but faster than the larger sized mAb and its derived fragments. With regard to TAT, affibody molecules directed against the membrane protein HER2 ($Z_{HER2:342}$ and the bivalent version $[Z_{HER2:4}]_2$) were radiolabeled with ^{211}At using the precursor *N*-succimidyl-*para*(trimethylstannyl) benzoate. Based on preliminary results, the authors concluded that the labeling chemistry needs to be improved before this strategy can be translated to clinical studies.[59]

So far, significant improvements have been made in the development and application of optimized vehicles for TAT. While these preliminary results are promising, there is still considerable room for improvement, mainly in the development of new coupling chemistries and elucidation and optimization of the *in vivo* biodistribution.

4. Nanobodies: potential vehicles to specifically deliver toxic α-radiation

Recently, there has been a growing interest in the use of nanobodies as vehicles for TRNT. Nanobodies are the smallest, antigen-binding fragments from unique heavy-chain-only antibodies naturally occurring in *Camelidae* (Figure 1). [101] Several applications of nanobodies as *in vivo* diagnostic tracers have been and are currently being developed. [102] Nanobodies have many favorable characteristics as targeted tracers, including high stability in harsh conditions, such as elevated temperatures and extreme pHs offering the potential to use a broader range of radiochemistry methods. Other favorable characteristics include high affinity and specificity for their cognate antigen and facile production (Figure 2(a,b)). As such, nanobodies have been developed as efficient radiotracers directed against a variety of membrane-bound biomarkers [103] in various animal models of cancer,[104–107] inflammation,[108] and cardiovascular diseases [102] using SPECT/PET. Because of their exceptional targeting specificity that is unaffected by labeling with various radionuclides, nanobodies have become valuable vehicles for both nuclear imaging and TRNT.[105–107] Furthermore, nanobodies possess various advantages over mAbs. First, the molecular weight of nanobodies (15 kDa) is one-tenth of that of conventional Abs (150 kDa), making it possible to recognize and bind hidden isotopes. Second, nanobodies have a low immunogenicity because of their rapid blood clearance and high sequence identity to human variable domains of the heavy chain. Furthermore, previous studies by our group demonstrated that nanobodies efficiently penetrate tumor tissues and bind tumor antigens rapidly and specifically *in vivo*.

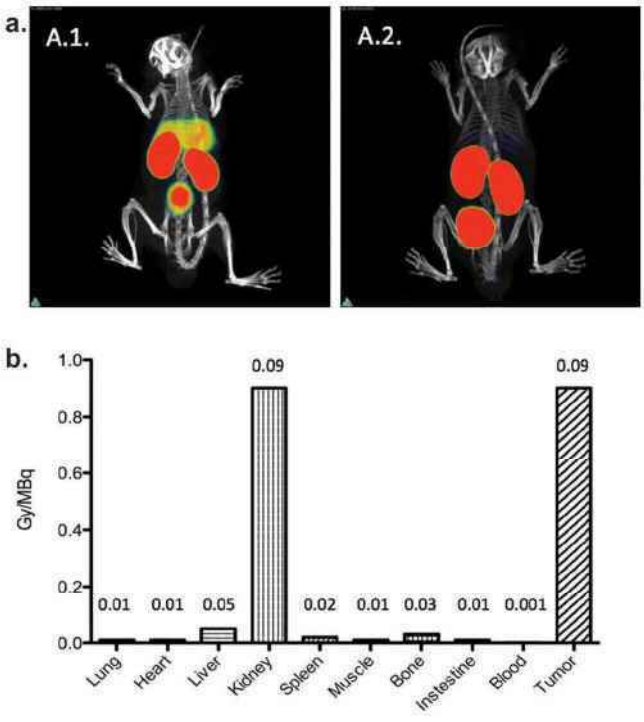

Figure 2. Nanobodies possess numerous advantageous characteristics, including their high antigen specificity (a) and high tumor targeting potential (b). **a.** ^{99m}Tc-labeled-nanobody targeting the complement receptor of the Ig superfamily, CRIg, expressed on Kupffer cells in the liver. 3D-rendered SPECT/micro-CT images of naive wild-type (A.1.) and CRIg$^{-/-}$ mice (A.2.) 1 h after intravenous injection of ^{99m}Tc-labeled-nanobody. Representative images for 3 mice per group are shown. Figures adapted with permission from.[115] **b.** Dosimetry calculation of untagged ^{177}Lu-DTPA-anti-HER2 nanobody coinfused with 150 mg/kg Gelofusin, in HER2^{pos} tumor xenografted mice. Radiolabeling of nanobodies is characterized by significant retention of radioactivity at the kidneys, due to the charged-based aspecific tubular reuptake after glomerular filtration. Figure adapted with permission from.[109].

Meanwhile, there is very little nonspecific binding to other tissues, which, along with the rapid blood clearance, results in high tumor-to-background ratios as early as 1 h after injection.[109] Therefore, the nanobody technology could provide an adequate solution to the off-target toxicity problem caused by long blood circulation, as is observed during mAb-based TRNT. A first-in-human PET study with a GMP-grade HER2-targeting nanobody-based tracer for breast cancer has recently been completed at our university hospital [110] and new clinical trials with nanobodies targeting HER2 and tumor-associated macrophages are planned for 2016. The first clinical study confirmed the fast clearance of nanobodies in patients, with only 10% of the injected activity remaining in the blood at 1 h p.i. (Figure 3(a)). In addition, high tumor-to-background ratios could be observed in 17 out of 19 primary tumors, with mean standard uptake values ranging between 0.7 and 11.8 (Figure 3(b)). Furthermore, the utility of nanobodies as vehicles for TRNT has been investigated in preclinical models using the β^--particle-emitting radionuclide ^{177}Lu. The most relevant *in vivo* study demonstrated that ^{177}Lu-labeled anti-HER2 nanobody efficiently targeted HER2^{pos} s.c. xenografts in a 5-day follow-up study, while radioactivity levels in normal organs were low (Figure 2(b)).[109] Weekly i.v. administrations of ^{177}Lu-labeled anti-HER2 nanobody in mice with small HER2^{pos} tumors completely prevented tumor growth, while tumors grew exponentially in untreated mice or in mice receiving a control, nontargeting nanobody. In addition, TRNT using a ^{177}Lu-labeled anti-5T2 multiple myeloma nanobody led to an inhibition of disease progression in treated mice compared to control animals. [111] These proof-of-concept TRNT studies show that nanobodies display a more beneficial toxicity profile than mAbs and can deliver a specific lethal radiation dose to a developing tumor. The low molecular weight of nanobodies, below the kidney cut-off for glomerular filtration, and the subsequent charged-based nonspecific tubular reuptake result in significant accumulation and retention of radioactivity in the kidneys. To avoid potential kidney-related toxicities, strategies were tested to reduce renal retention. Both the removal of nonessential positively charged amino acids in the nanobody sequence and co-infusion with positively charged amino acids or the plasma expander Gelofusin were able to lower kidney retention significantly. [107,109] Another approach to reduce the kidney retention is to use optimized radiolabeling procedures. For instance, Zalutsky and colleagues labeled an anti-HER2 nanobody with iodine-131 (^{131}I), using the prosthetic group *N*-succinimidyl-4-guanidinomethyl-3-iodobenzoate (SGMIB). SGMIB is a prosthetic group used for antibody and small-protein radioiodination and possesses improved properties as a group that stabilizes ^{131}I and maximizes the retention in tumor cells.[112,113] Remarkably, ^{131}I-SGMIB-anti-HER2-nanobody was not retained in the kidneys, while tumor targeting was maintained. In addition, Zalutsky and coworkers recently labeled an anti-HER2 nanobody with ^{211}At, using this similar residualizing agent, referred to as *N*-succinimidyl-3-[^{211}At]astato-4-guanidinomethylbenzoate (SAGMB).[114] Paired-label biodistribution studies directly compared the *in vivo* behavior of ^{211}At-SAGMB-nanobody to that of its ^{131}I analog SGMIB-nanobody in athymic mice, showing excellent preservation of HER2 binding after ^{211}At labeling in combination with high internalization and optimal tumor uptake. Further investigation of this ^{211}At-SAGMB-nanobody compound is warranted.

5. Conclusion

TAT is an emerging and promising treatment modality that has the ability to specifically kill isolated cancer cells or cell

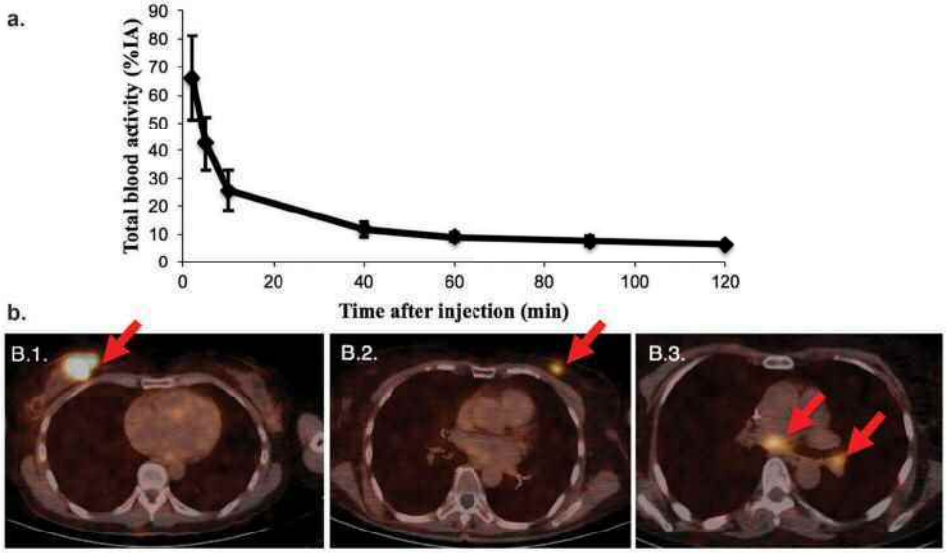

Figure 3. Diagnostic tumor imaging using ^{68}Ga-HER2-nanobody in patients with HER2^{pos}-breast cancer. **a.** Time-activity curve of total blood activity, expressed in % of injected activity (%IA) (n=20). **b.** Fusion PET/CT images of the uptake of ^{68}Ga-HER2-nanobody in breast carcinoma lesions. (B.1.) Patient with the highest tracer uptake (SUVmean 11.8) in a primary breast carcinoma. (B.2.) Patient with moderate tracer uptake in the left breast, which is easily discernable from background (SUVmean 4.9). (B.3.) Patient with invaded lymph nodes in the mediastinum and left hilar region. Lesions are indicated by red arrows. Figures are adapted with permission from.[110].

clusters and might only cause little damage to healthy non-target tumor cells. The combination of preclinical and clinical data affirms the potential of TAT. However, more research is needed to identify the ideal combination of targeting vehicle and α-particle radionuclide, its specific way of linking both, and all this optimized toward specific target expression, disease stage, target accessibility, and site of action.

6. Expert opinion: nanobodies coupled to α-particle-emitting radionuclides in cancer therapy

There is an unmet need to treat minimal residual disease and micrometastatic spread of tumor cells, as the current cancer treatment options like chemotherapy, surgery, and external beam radiotherapy are less effective once the tumor has metastasized. Targeted α-particle therapy or TAT allows, due to the high LET of the associated radioactivity, precise delivery of a highly toxic radiation to target cells with reduced harm to normal untargeted cells in the vicinity. This strategy might be ideal for the treatment of small malignant cell populations that are located in the proximity of essential normal tissue structures and could be used in addition to other existing treatment modalities. Increased production and evaluation of α-particle emitters has improved their availability, enhancing the development for new TATs. Currently, TAT has mainly been explored using mAbs. However, the high molecular weight of mAbs and the presence of the Fc-region result in a long serum half-life and interactions with cells containing Fc-receptors. Consequently, the systemic administration of radiolabeled mAbs results in a prolonged presence of radioactivity in blood and highly perfused organs, and unwanted radiation exposure of nontargeted cells. Unsurprisingly, myelotoxicity has been shown to be a limiting factor in several preclinical and clinical studies. Moreover, the dose delivered to carcinomas is often inadequate, owing to the limited penetration of mAb-based vehicles. Hence, we claim that mAbs are not the ideal vehicles to couple with an α-particle emitter. Ab engineering is an interesting approach to overcome some of the limitations of mAbs. Nanobodies in particular have emerged as excellent Ab fragments, as they exhibit high affinity and specificity, fast diffusion and clearance kinetics *in vivo*, high tumor-to-normal-tissue ratios, and a high stability. Moreover, nanobodies have already proven their value in both diagnostic and therapeutic applications. We believe that nanobodies, with their improved properties compared to full-size mAbs and larger Ab-fragments, could be ideal vehicles for TAT.

A key element in the design of radiopharmaceuticals is attuning the properties of the therapeutic radionuclide with those of the tumor-targeting vehicle. The main goal here is to optimize the vehicle in such a way that it fits the characteristics of the α-particle-emitting radionuclide, resulting in optimal tumor targeting and minimal exposure of normal organs. Due to their half-life in the range of minutes to hours, ^{213}Bi and ^{211}At could be ideal radioactive partners for fast and specific targeting nanobodies. However, both radioisotopes have both their advantages and disadvantages. Currently, the most important limitation of ^{211}At is the limited availability of accelerators that are able to generate the 28 MeV α-particle beam required to produce useful levels of ^{211}At.[86] Therefore, production and supply of sufficient amounts of ^{211}At is still challenging, although over the past few years some progress has been made in the recruitment of new cyclotrons for commercial ^{211}At production. Today, about 30 cyclotrons in the world have the beam characteristics (28 MeV) capable for the production of ^{211}At. Furthermore, Lindegren and colleagues developed a fully automated procedure that enables automatic, reproducible, rapid, high-yield production of clinically relevant amounts of ^{211}At and ^{211}At-labeled radiopharmaceuticals.[116] To date, only two clinical trials have been reported using ^{211}At-labeled molecules (Table 2). In the first clinical trial, the median survival for patients with glioblastoma multiforme, anaplastic astrocytoma, and oligodendroglioma was 54, 52, and 116 weeks after ^{211}At-labeled chimeric antitenascin 81C6 therapy.[53] In the second phase I study, ovarian cancer patients were injected with ^{211}At-MX35 F(ab')$_2$. Intraperitoneal administration of this immunoconjugate showed that it was possible to achieve therapeutic absorbed doses (15.6 ± 1.0 mGy/[MBq/L]) in the peritoneal peritoneum, where the microscopic tumor clusters are situated, without significant toxicity.[55]

Targeting vehicles can be astatinated via a variety of prosthetic groups.[85] However, many prosthetic groups fail to deliver relevant amounts of astatinated end product, as well as proper *in vivo* stability. In addition, automatable chemistries with high radiochemical yields are yet to be developed. Therefore, many ^{211}At-labeled compounds labeled have been abandoned in the past. To this, a more in-depth understanding of the chemistry of ^{211}At is required to provide future, useful astatinated radiopharmaceuticals. The production of ^{213}Bi is more straightforward, through the actinium-225/bismuth-213 generator system. However, the use of ^{213}Bi has been limited by the availability of ^{225}Ac. In numerous clinical studies, ^{213}Bi ($t_{1/2}$ = 46 min) has shown to be effective to treat patients with malignant melanoma, metastatic breast cancer, prostate cancer, pancreatic cancer, and other metastatic diseases. The labeling of targeting vehicles with ^{213}Bi is generally performed using straightforward chelating agents such as DTPA and DOTA. In addition, ^{213}Bi decays via a branched pathway by α and β emissions to stable ^{209}Bi, leading to low toxicity due to the minimal recoil energy the daughter experience upon α-decay. However, the short half-life of ^{213}Bi might eventually limit its clinical applicability, as relevant therapeutic doses of ^{213}Bi need to be available on a regular basis. Based on these characteristics and on the corresponding features of nanobodies, we claim that nanobodies are ideal for radiolabeling with short-lived radionuclides such as ^{211}At and ^{213}Bi.

α-Particle recoiling daughter isotopes pose serious problems during TAT as they can do significant harm to healthy tissue when they are not retained at the tumor site. Different approaches to limit the distribution of recoiling daughter isotopes have been found such as encapsulation in a nano-carrier and fast internalization of the α-particle inside the tumor cells. In general, monovalent nanobodies only show limited degree of internalization inside tumor cells after binding. However, it has been shown that internalization can be stimulated by the development of multivalent nanobody constructs, which

would augment the amount of α-particles trapped inside the tumor cell.[117]

In order to become valuable, some aspects concerning nanobody-based TAT need to be considered.[1] In general, nanobody targeting is characterized by only moderate absolute uptake in tumor tissue (compared to longer circulating mAbs) and fast blood clearance. To this, it will be important to assess the maximum dose that can be delivered to target tissues. The fast clearance and very specific way of targeting of nanobodies allows repeated injections. In the past, we have shown that therapeutic doses can be reached through fractionated administration using ^{177}Lu-labeled nanobodies.[109,111] We therefore believe that therapeutic TAT doses will be achieved by means of repeated administration. The high LET of α-particles will have their beneficial effect on tumor tissues, but can in parallel cause toxicity in tissues with elevated uptake or retention. It is known that nanobodies can interact with the negatively charged lumen of kidney tubuli during filtration from blood. It is therefore of upmost importance to assess the effect of nanobody-TAT at the level of the kidneys. Based on previous published work, there are several countermeasures that can be taken to reduce renal retention of radiolabeled nanobodies. Kidney retention can be reduced significantly by removal of the nanobodies' amino-acid tag or through co-infusion with the plasma expander Gelofusin and positively charged amino acids.[107,109] In addition, it has been shown that the linker between radioisotope and targeting vehicle can have a dramatic influence on the degree of kidney retention. Recently, anti-HER2 nanobodies were radiolabeled with ^{131}I using the prosthetic group SGMIB. It was shown that while the retention in tumor cells was maintained, a complete absence of kidney retention was observed.[112] Interestingly, this exact prosthetic group can be used for astatination of nanobodies. The short path length of α-particles causes a heterogenous distribution in both tumor and tissues, which can lead to very localized toxicity (suborgan or subtissue level). Novel methods that allow micro- and small-scale dosimetry will be essential to realistically estimate dosimetry of TAT-based radiopharmaceuticals. Emerging strategies include the recent development of the α-camera that allows *ex vivo* imaging of α-particle deposits at a cellular level.

In conclusion, the superior characteristics of α-particle emitters ^{213}Bi and ^{211}At as toxic payload and nanobodies as targeting vehicles offer exciting possibilities in TAT. We therefore expect that the pairing of short-lived α-particle emitters and fast and specific targeting nanobodies will show their potential in the future.

Declaration of interests

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