Prospects for Enhancing Efficacy of Radioimmunotherapy

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Abstract Radioimmunotherapy has been in use for more than 20 years and has progressed significantly since its efficacy has first been demonstrated in hematology. Yet it still has limitations that prevent its large-scale clinical use. This chapter reviews recent developments to overcome these limitations including new antibody specificities, pretargeting methods, fractionated injections, and the use of alpha emitters. Immuno-PET is also likely to assist in selecting patients for radioimmunotherapy, optimizing injected activities, and noninvasively monitoring therapy efficacy.

Keywords Radioimmunotherapy · Fractionation · Pretargeting · Alpha-RIT · Theranostic

Abbreviations

ADC	Antibody drug conjugate
AML	Acute myeloid leukemia
ARC	Antibody radionuclide conjugate

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BsMAb	Bispecific monoclonal antibody
CEA	Carcinoembryonic antigen
EGFR	Epidermal growth factor receptor
HSG	Histamine-succinyl-glutamine
LET	Linear energy transfer
MRD	Minimal residual disease
MTD	Maximum tolerated dose
NHL	Non-Hodgkin B-cell lymphoma
PSMA	Prostate-specific membrane antigen
RIT	Radioimmunotherapy
SPECT	Single-photon emission computed tomography
PET	Positron emission tomography

1 Introduction

Clinical development of radioimmunotherapy (RIT) started in the 1980s and progressed rapidly due to advancements in recombinant humanized or human antibodies and in the development of radiolabeling methods and/or superior chelating agents. The first clinical application was for non-Hodgkin B-cell lymphoma (NHL) because the radiosensitivity of this type of cancer allows good efficacy for a relatively moderate tumor dose delivery. Two radioimmunoconjugates targeting the CD20 antigen have been approved: ^{131}I-tositumomab (Bexxar; GlaxoSmithKline) which was subsequently discontinued and ^{90}Y-ibritumomab tiuxetan (Zevalin; Spectrum Pharmaceuticals) which continues to be used both in the US and in Europe. While most clinicians agree that this last radioimmunoconjugate has demonstrated clinical efficacy, it has not been successfully adopted by the hemato-oncologist community.

For more radioresistant solid tumors, the clinical efficacy of RIT remains limited, and up to now, no radioimmunoconjugate has been yet approved.

In parallel with the clinical development of radioimmunoconjugates, also termed antibody radionuclide conjugates (ARCs), some pharmaceutical companies have developed antibody drug conjugates (ADCs) for treatment of several types of cancer [1]. A recent review summarized the results of 11 studies including 598 patients treated with 6 ADCs and 9 studies including 377 patients treated with 5 ARCs [1]. While it was obviously not possible to statistically compare the results of both modalities, the objective was to roughly estimate their respective toxicity and clinical efficacy. Toxicity was generally less frequent with ADCs (less than 20%) than with ARCs but led to more uncomfortable side effects. Hematologic toxicity was higher with ARCs than with ADCs. Clinical efficacy was roughly comparable.

There is no doubt that RIT still has limitations preventing its large-scale clinical use. These limitations can be partially overcome by using fractionation of the injected activities and combination therapy with nonradioactive drugs that have non-overlapping toxicity and synergistic effects. Finally, the use of alpha-emitting

radionuclides could dramatically improve the clinical efficacy for microscopic tumors or clusters of malignant cells disseminated throughout the body.

2 Current Limitations of RIT

The current limitations of RIT are technical, logistical, and societal. Until now the majority of clinical studies have used a single injection for treatment of the most common large bulk tumors. Under these conditions tumor uptake was low or very low resulting from an inefficient weak dose. However, it has been clearly documented that dose delivery to tumors increases with decreasing tumor mass [2, 3]. For treatment of medullary thyroid cancer using a pretargeting technique, a tumor dose as high as 174 cGy/mCi (4.7 cGy/MBq) has been calculated for a small resected tumor of 1.8 g. By extrapolating this value to an injected activity of 100 mCi (3700 MBq) comparable to the activity of ^{131}I administered for treatment of metastases of differentiated thyroid carcinoma, a tumoricidal absorbed tumor dose of 174 Gy would have been obtained. Moreover, a serious problem for macroscopic tumors is the accessibility of circulating antibody to cells of the inner hypoxic areas [4]. Thus there is a consensus that the best situation for an efficient RIT would be a dissemination of small-size tumors or some clusters of malignant cells in the body.

Another serious limitation of RIT is the need for a reliable supply chain for the radionuclide. Big pharma companies do not have such a supply chain and are generally not familiar with coupling radionuclides to antibody molecules. That is probably why they prefer the use of chemotherapeutic drugs which they control very well for antibody drug conjugates. Changing from chemotherapeutic drugs to radionuclides would require them to secure radionuclide supply in the event of a very efficient RIT, for example, with an overall survival gain of 6 months to 1 year, which is longer than that generally observed for many chemotherapeutic drugs.

Finally, RIT may cause concern among patients due to the use of radioactivity and may require secondary myelodysplasia/acute/leukemia risk management by oncologists, even though such a risk is limited to heavily pretreated patients.

3 Prospects to Improve RIT

3.1 Fractionation of Injections

The rationale for using fractionated instead of single-dose RIT was reported in 2002 by DeNardo et al. [5]. The main advantage of injected activity fractionation is to reduce hematologic toxicity as a consequence of faster and more efficient bone marrow repair than tumor cell repair. Several preclinical studies over many years have tended to validate this concept [6]. The number of clinical studies is more limited. Two in particular, using well-known radioimmunoconjugates in a substantial number

Table 1 Clinical studies with single or fractionated injected activities

	^{90}Y-Ibritumomab tiuxetan		^{177}Lu-hJ591 (ATL101)	
	Single1	Fractionated2	Single3	Fractionated4
Number of patients	59	74	47	44
Injected activity	15 MBq/m^2 up to max 1200 MBq	11.1 MBq/m^2 × 2 up to max 888 x2	65–70 mCi/m^2	40–45 mCi/m^2 × 2
Interval time between two injections	NA	8–12 w	NA	2 w
Thrombocytopenia Gr 3/4	48%	56.4%	65.7%	Global hematol tox (plts+neutro):73.5%
Neutropenia Gr 3/4	32%	36.4%	65.6%	
ORR	87%	94.4%	NA	NA
CR/CRu	56%	58.3%	NA	NA
PFS	*26 m*	*40.2 m*	NK	NK
OS	Median OS not reached	Median OS not reached	*21.8 m*	*42.9 m*

NA Not applicable, *ORR* Overall response rate, *NK* Not known, CR/CRU Complete/unconfirmed complete response, *PFs* Progression-free survival, *Os* Overall survival

of patients, have provided important information for future applications (Table 1). The first study used the approved ^{90}Y-ibritumomab tiuxetan (Zevalin®) radioimmunoconjugate in 74 patients as an initial therapy for follicular lymphoma [7]. The patients were sequentially injected twice with an activity of 11.1 MBq/m^2 (not exceeding twice 888 MBq) 2–12 weeks apart. Another study used the same radioimmunoconjugate in 59 patients, again as an initial treatment for follicular lymphoma, with a single activity of 15 MBq/m^2 (not exceeding 1200 MBq) [8]. The fractionated radioimmunotherapy therefore used a cumulative activity 48% higher than in the single-dose radioimmunotherapy. The hematologic toxicity was roughly comparable between the single and fractionated studies with grade 3/4 thrombocytopenia and neutropenia of 48 and 56% and 32 and 36%, respectively. The clinical efficacy was clearly improved with fractionation, with an overall response rate of 94 vs 87% with single-dose therapy and more impressively a progression-free survival of 40 vs 26 months. These studies using ^{90}Y-ibritumomab tiuxetan illustrate a clear advantage of activity fractionation compared to single-dose activity and allow the overall injected activity to be significantly increased while maintaining the same level of toxicity.

A second study used the ^{177}Lu-J591 (ATL101) DOTA radioimmunoconjugate in 44 patients with metastatic prostate cancer [9]. The patients were sequentially injected twice with an activity of 1480–1665 MBq/m^2 2 weeks apart. Another study used the same radioimmunoconjugate in 47 patients in the same indication of metastatic prostate cancer with a single activity of 2405–2590 MBq/m^2 [10]. The fractionated radioimmunotherapy used a cumulative activity 26% higher than in the single-dose radioimmunotherapy.

The hematologic toxicity was difficult to compare between the two approaches because with fractionation only global toxicity was evaluated with 73.5% grade 3/4. However, compared to 66% of grade 3/4 thrombocytopenia and neutropenia with single-dose activity, a clear higher toxicity with fractionation does not appear significant.

However, the clinical efficacy was clearly improved with fractionation showing an overall survival of 43 vs 22 months with the single-dose activity. The predominant bone metastases in prostate cancer did not allow evaluation of overall response rate. It is obviously not possible to statistically compare the results of both single and fractionated studies in these two clinical indications using these two methodological approaches. Only a rough estimate of efficacy and toxicity can be drawn. It appears that fractionation is clearly preferable to single activity, allowing the injected activity to be substantially increased and consequently improving clinical efficacy without impairing hematologic toxicity. However, fractionation needs to be optimized for each radioimmunoconjugate. Two parameters should be taken into consideration, namely, the level of fractionated activity and the time interval between two sequential injections. It is well known that following irradiation, bone marrow repair is faster and more efficient than tumor repair. Consequently, it is logical to wait for 6–8 weeks, i.e., the time required for hematologic recovery, before reinjection. Determining the level of injected activity is more difficult, and the choice is somewhat empirical. In preclinical studies it is easy to test a range of injected activities; however extrapolating these results to the clinical situation is questionable. In clinical studies, testing a selected activity requires months to years to accrue sufficient patient numbers to estimate the toxicity and clinical efficacy. This is why the choice of the level of activity is relatively empirical.

3.2 Combinations with Other Therapeutic Agents

The rationale for combining RIT with other systemic therapies, especially chemotherapy, is to take advantage of potentially radiation-enhancing drugs and the non-overlapping drug-limiting toxicity of each agent. It is well established that for a large tumor burden, the tumor dose delivered by RIT does not exceed 15–40 Gy, which is not sufficient for an efficient tumor-killing effect. The situation is different for small or microscopic tumors for which much higher tumor doses can be delivered. One way to increase RIT efficacy is to combine it with systemic drugs with a different and if possible synergistic tumor-killing effect. Many preclinical animal studies using human cancer xenograft models in nude mice have clearly shown a significant benefit of such a combination in terms of tumor shrinkage and survival time [11]. However, the extrapolation of these results to clinical studies in predicting efficacy should be made with caution. Hence the only way to assess the real benefit of combining RIT and chemotherapy is to refer to clinical studies performed with specific radioimmunoconjugates, chemotherapeutic drugs, and clinical situations. Only a limited number of RIT +/− combined therapy studies have been performed.

Phase I clinical trials assessing three radioimmunoconjugates, combined or not, with three chemotherapeutic drugs have been performed (Table 2). In patients treated with a maximum tolerated dose (MTD) of 614 MBq/m^2 of an anti-carcinoembryonic antigen antibody labeled with yttrium-90 (T84–66), combined or not with 5-fluorouracile in, respectively, 21 and 22 patients with metastatic CEA-producing

Table 2 Clinical studies with combined or RIT alone

	^{90}Yttrium-anti-CEA chimeric T84.66		^{90}Yttrium-clivatuzumab		^{177}Lu-J591	
	RIT alone	RIT+ 5-FU	RIT alone	Fract RIT + gemcitabine	RIT alone	Fract RIT docetaxel
Number of patients	22	21	9	17	12	15
Injected MTD	614–814 MBq/m^2	614 MBq/m^2	740 MBq/m^2	444 × 3 MBq/m^2	2220–2775 MBq/m^2	
Thrombocytopenia Gr 3/4	10%	24%	55%	74%	75%	
Neutropenia Gr 3/4	20%	19%	44%		67%	
Overall response rate	0	0	22%	16%	11% ≥50% PSA decline	73.3% ≥50% PSA decline
Stabilization and mixed response	32%	57%	ND	42%	46% PSA Stabilization	
Overall survival	ND	ND	4.3 m	8 m	ND	ND

malignancies, thrombocytopenia was slightly higher for combination therapy (24 vs 10%), while neutropenia was the same (19% and 20%) [12, 13]. There was no objective response in either situation and a slightly higher mixed or stable response with combination treatment (57% vs 32%).

A second radioimmunoconjugate, clivatuzumab, is an anti-PAM4 reactive mucin antibody labeled with yttrium-90. Treatment of pancreatic carcinoma patients with clivatuzumab alone or combined with gemcitabine (17 and 9 patients, respectively) at the MTD in a fractionated mode (444 MBq x3) showed that the hematologic toxicity was roughly comparable, even though the interpretation of this parameter is difficult to assess because the combination study of thrombocytopenia and neutropenia results was merged [14, 15]. There was no real difference in the response rate but a tendency toward a longer overall survival with the fractionated and combined study (8 vs 4.4 months).

Finally, an anti-PSMA antibody, labeled with lutetium-177 (J591) at the MTD, was compared against combined therapy with docetaxel (15 and 12 patients, respectively) for the treatment of prostate cancer [16, 17]. While these results should be treated with caution due to the small number of patients, combination therapy resulted in a trend toward improved clinical efficacy without altered toxicity. Promotion to phase II trials will require a substantial increase in patient numbers and data and most likely a number of years.

3.3 Pretargeting Using Bispecific Antibodies

Pretargeting may be achieved by a primary injection of an unlabeled bispecific monoclonal antibody (BsMAb), followed by a second injection of a radiolabeled bivalent hapten-peptide [18, 19]. Using this strategy, the radiolabeled bivalent peptide binds more avidly to the BsMAb attached to the antigen expressed at the tumor cell surface, whereas nontargeted hapten-peptide in the circulation clears rapidly through the kidneys. After the promising phase I/II studies, encouraging clinical results have been obtained using an anti-CEA chimeric hMN-14x734 BsMAb and ^{131}I-di-DTPA peptide in a prospective multicentric phase II study performed in 45 patients with progressive metastatic medullary thyroid carcinoma (MTC) [20]. This study demonstrated a disease control rate of 76.2% (durable stabilization plus objective response) according to RECIST, with 1 case of durable complete response of at least 40 months (2.4%) and 31 durable stable disease cases of ≥6 months (73.8%). After RIT, 21 of 37 assessed patients (56.7%) showed a ≥100% increase in serum biomarker concentration doubling time or prolonged decrease in serum biomarker concentration. As expected for these patients with a high frequency of diffuse bone marrow involvement, high-grade 3 and 4 hematologic toxicity was observed in 54.7% of patients and myelodysplastic syndrome reported in two cases, including one treated heavily previously.

New-generation recombinant humanized trivalent BsMAb and bivalent histamine-succinyl-glutamine (HSG) peptides have been produced. These can be

labeled with a variety of radionuclides, including yttrium-90 and lutetium-177 for therapeutic purposes [21–23]. This new-generation pretargeting system using anti-CEA × anti-HSG BsMAb TF2 and ^{177}Lu-IMP288 has been performed and optimized in two clinical trials in patients with metastatic colorectal carcinoma and lung carcinoma [24, 25]. Different schedules were studied to define the optimal molar doses of TF2 and IMP-288 and the optimal delay between the two infusions.

Three cohorts of three patients were included in the first part of a phase I/II clinical trial designed to optimize and assess anti-CEA × anti-HSG BsMAb TF2 in CEA-expressing lung cancer patients. Patients underwent a pre-therapeutic imaging session S1 (44 or 88 nmol/m2 of TF2 followed by 4.4 nmol/m^2 and 185 MBq of ^{111}In-IMP288) and, 1–2 weeks later, a therapy session S2 (240 or 480 nmol/m^2 of TF2 followed by 24 nmol/m^2, 1.1 GBq/m^2, ^{177}Lu-IMP288). The pretargeting delay was 24 or 48 h. According to the pharmacokinetic and imaging analysis, the best dosing parameters corresponded to the shorter pretargeting delay (24 h) and to the highest TF2 molar doses. While toxicity was quite limited in the eight patients evaluated, treatment efficacy was minimal in this optimization part of the study, with only two cases of disease stabilization for only short periods of time [25]. Thus, to improve treatment efficacy, the injected activity should be increased for the second part of the study, which is planned with an activity escalation. Overall, it was not expected that a single therapy cycle would be sufficient to deliver antitumor therapeutic doses and the use of shorter half-life and higher intrinsic toxicity radionuclides, such as yttrium-90, could be preferable to that of lutetium-177. Taking into account these data, a prospective phase I study is ongoing, to assess fractionated injection of ^{90}Y-IMP288 in metastatic colorectal carcinoma patients.

3.4 Alpha-Emitting Radionuclides

Due to their high linear energy transfer (LET), α-particles deliver a high fraction of their energy inside the targeted cells leading to highly efficient killing, making them particularly suited for targeting of isolated tumor cells and minimal residual disease (MRD). Moreover, α-particle cytotoxicity is considered to be independent of the dose rate and oxygenation [26]. Among the large number of identified α-emitting radionuclides, only few of them exhibit physical characteristics adapted for RIT. ^{213}Bi is available through a ^{225}Ac/^{213}Bi generator, but its short half-life (T1/2) of 45.6 min makes it difficult to use. While ^{225}Ac (T1/2 = 10 days) appears clinically more suitable, its decay produces a series of alpha-emitting daughter nucleons that are released from the chelating agent which then increase irradiation of normal tissues. With an intermediate half-life of 7.2 h and 100% of decays leading to the emission of an α-particle, ^{211}At,which is available from cyclotron production, may be a better candidate, although its availability and chemistry remain to be improved [27].

The first clinical report of alpha-RIT was performed using an anti-CD33 monoclonal antibody labeled with ^{213}Bi. The CD33 antigen is a 67 kDa glycoprotein expressed on most myeloid leukemias and clonogenic leukemia progenitors but not

on normal stem cells. Anti-CD33 RIT has been developed using the murine M195 and the HuM195 (lintuzumab) humanized antibodies by the Scheinberg group at the Memorial Sloan-Kettering Institute. A phase I dose-escalation study assessing ^{213}Bi-lintuzumab was conducted in 18 patients with relapsed and refractory acute myeloid leukemia (AML) or chronic myelomonocytic leukemia treated with 10.36–37.0 MBq/kg ^{213}Bi-RIT [28]. No significant non-hematologic toxicity was observed. Dose-limiting toxicity, defined as grade 4 leukopenia for more than 35 days from the beginning of therapy, was observed in one patient treated at the 37 MBq/kg dose level following relapse after allogeneic transplantation. ^{213}Bi-HuM195 was retained in areas of leukemic involvement (bone marrow, liver, and spleen). The estimated total absorbed dose to the marrow, and therefore to CD33$^+$ target cells, ranged from 6.6 to 73 Sv, whereas the total dose to the liver, spleen, and blood ranged from 2.4 to 23.5 Sv, 2.9 to 36.8 Sv, and 1.1 to 11 Sv, respectively. Absorbed dose ratios between the bone marrow, liver, spleen, and the whole body were approximately 1000 times higher for ^{213}Bi-HuM195 than those for the β-emitting immunoconjugates. An antileukemic effect was observed: 15/18 patients had leukemic blasts in the blood before treatment, and 14 of them showed reductions in circulating blasts after α-RIT. Even at the lowest activity level, patients showed elimination of more than 99% of peripheral blasts. Up to three logs of circulating leukemia cells were killed, and four patients (27%) had complete eradication of peripheral leukemia cells. Fourteen of the 18 patients (78%) experienced reductions in the percentage of bone marrow leukemia cells 7–10 days after α-RIT. Among the four patients with complete elimination of peripheral blood blasts, three also experienced reductions in bone marrow blasts (Table 3).

The major obstacles to the widespread clinical use of ^{213}Bi-lintuzumab are the short half-life of ^{213}Bi and the requirement of an on-site ^{225}Ac/^{213}Bi generator. On the other hand, the much longer-lived ^{225}Ac ($T_{1/2}$ = 10 days) can serve as an in vivo generator (atomic nanogenerator) of four α-particles. A phase I trial evaluating ^{225}Ac-lintuzumab was conducted on 18 patients with relapsed or refractory AML [29]. Patients were treated with a single infusion of 0.5–4 μCi/kg (18.5–150 kBq/kg) of ^{225}Ac-lintuzumab. The MTD was determined to be 3 μCi/kg (110 kBq/kg). Serious non-hematologic toxicity was observed in three patients (transient grade 3 liver function abnormalities), but there was no evidence of radiation-induced nephrotoxicity. Peripheral blasts were eliminated in 10 of 16 evaluable patients (63%) but only at doses of 1 μCi/kg (37 kBq/kg) or more. Bone marrow blast reductions were observed in 10 of 15 evaluable patients (67%) 4 weeks after treatment.

Alpha-RIT using a^{212}Pb/^{212}Bi generator has also been assessed in a phase I trial using an anti-HER2 radiolabeled mAb intraperitoneally injected in patients with HER2-positive peritoneal carcinomatosis for which no standard therapy is available [30]. ^{212}Pb-TCMC-trastuzumab was delivered intraperitoneally within less than 4 h after administration of trastuzumab (4 mg/kg intravenously). The five activity levels assessed in this study (7.4, 9.6, 12.6, 16.3, and 21.1 MBq/m^2) showed minimal toxicity. The lack of substantial toxicity was consistent with the dosimetry results (mean equivalent dose to the marrow, 0.18 mSv/MBq). Further studies are required to assess ^{212}Pb-TCMC-trastuzumab efficacy.

Table 3 Clinical studies with alpha particle emitting radionuclides

	^{213}Bi		^{225}Ac	^{211}At		^{212}Pb
	Anti-CD33	Anti-chondroitin sulfate proteoglycan	Anti-CD33	Anti-tenascin (IC)	Anti-NaPi2B (IP)	Trastuzumab (IP)
Type of cancer	Myeloid leukemia	Metastatic melanoma	Myeloid leukemia	Brain tumor	Ovarian cancer	Ovarian cancer
Number of patients	18	38	18	18	9	16
Thrombocytopenia gr 3/4	NA	0	Gr 4 in one patient	0	0	0
Non-hematologic toxicity	0	0	Gr3 liver in three patients	22% (seizures)	0	0
Response rate	Bone marrow blasts reduction in 78%	Partial response: 10% Stable: 40%	Bone marrow blasts reduction in 67%	NK	NK	No objective response
MTD	Not reached	Not reached	110 kBq/kg	NK	NK	Not reached

IC Intracavitary, *IP* Intraperitoneal, *MTD* Maximum tolerated dose, *NK* Not known

Astatine-211, an α-emitting radionuclide with a physical half-life of 7.2 h, also appears relevant for RIT. Preclinical studies recently showed that anti-CD45 ^{211}At-RIT and bone marrow transplantation prolonged survival in a disseminated acute myeloid leukemia murine model [31]. Biodistribution studies showed excellent localization of the ^{211}At-anti-murine CD45 mAb 30F11 to the marrow and spleen within 24 h. In syngeneic hematopoietic stem cell transplantation studies, ^{211}At-RIT improved the median survival of leukemic mice in a dose-dependent fashion with minimal toxicity. ^{211}At-RIT feasibility was reported in two clinical trials. The first study assessed anti-tenascin ^{211}At-RIT followed by chemotherapy in patients with glioblastoma [32]. The radioimmunoconjugate was injected into the resection cavity with a maximum activity of 347 MBq (9.4 mCi). Six patients out of 18 experienced reversible grade 2 neurotoxicity but no grade 3–4 toxicities were observed. Maximum tolerated activity was not reached, and observed median survival favorably compared with that of historical control groups. In the second study, ^{211}At-MX35 F(ab')$_2$ was assessed in women in complete response after a second-line chemotherapy for recurrent ovarian carcinoma in a phase I study [33]. MX35 F(ab')$_2$ was labeled with ^{211}At via the *N*-succinimidyl 3-(trimethylstannyl)-benzoate reagent. Nine patients underwent laparoscopy 2–5 days before ^{211}At-RIT. Before RIT infusion, the abdominal cavity was inspected to exclude the presence of macroscopic tumor growth or major adhesions. Patients were infused with ^{211}At-MX35 (22.4–101 MBq/L) in the dialysis solution via the peritoneal catheter. The estimated absorbed dose was

15.6 ± 1.0 mGy/MBq/L to the peritoneum, 0.14 ± 0.04 mGy/MBq/L to the red bone marrow, and 24.7 ± 11.1 mGy/MBq/L to the unblocked thyroid. This dose decreased when the thyroid was blocked (1.4 ± 1.6 mGy/MBq/L). No adverse effects were reported.

These first clinical results of alpha-RIT appear very promising, and larger phase II clinical trials have been performed in patients with minimal residual disease to fully demonstrate efficacy. However, large clinical trials will require access to higher production levels of alpha-emitting radionuclides.

3.5 Theranostic Approaches: Imaging of Radiolabeled Antibodies to Improve RIT Procedures

For more than two decades, mAbs have been labeled with γ-emitting radionuclides, such as ^{131}I, ^{177}Lu, or ^{111}In, and subsequently used in planar or single-photon emission computed tomography (SPECT) imaging procedures to try and improve RIT using dosimetry procedures. Indeed, optimization studies performed using new-generation pretargeting systems in both colorectal carcinoma and lung carcinoma patients [24, 25] assessed the potential of ^{111}In-IMP288 imaging to predict ^{177}Lu-IMP288 dosimetry. In an optimization PRIT study using anti-CEA × anti-HSG BsMAb TF2 in 20 patients with colorectal carcinoma, Schoffelen et al. reported that individual high-activity doses in PRIT could be safely administered by predicting the radiation dose to the red marrow and kidneys, based on dosimetric imaging obtained with a test dose of TF2 and ^{111}In-IMP288 [24]. These results were confirmed by the phase I/II clinical trial using the same pretargeting system in CEA-expressing lung cancer patients showing that a pre-therapeutic imaging session using ^{111}In-IMP288 accurately predicted pharmacokinetics as well as absorbed doses of the therapeutic session using ^{177}Lu-IMP288, potentially allowing for patient selection and dose optimization [25].

While providing reliable information, this modality suffers from several drawbacks including poor sensitivity, poor spatial resolution, and complex scatter correction due to the collimator. Accurate quantitative information could be better achieved using positron emission tomography (PET) for mAb imaging. The improved spatial resolution of PET makes the delineation of tumors and organs better than with SPECT. Additionally, exact attenuation correction, precise scatter correction, and, last but not the least, high sensitivity combined with the possibility of performing true whole body imaging in a reasonable time constitute additional key factors for the superiority of PET over SPECT or planar imaging. As for therapeutic emitters, marrying mAbs and PET emitters requires an appropriate match between the biologic half-life of the protein and the physical half-life of the isotope [34]. ^{89}Zr and ^{124}I with their long half-life of 78 and 100 h are well suited to the labeling of larger molecules such as intact immunoglobulins. ^{64}Cu with an intermediate half-life of

12.7 h can also be used for labeling of a large number of molecules of different sizes. Within the scope of a "theranostic" approach, pairs of beta+/beta-emitting radionuclides (^{124}I/^{131}I, ^{86}Y/^{90}Y, ^{64}Cu/^{67}Cu, ^{44}Sc/^{47}Sc) are very promising because the same distribution is expected both for dosimetry imaging and therapy with the same elements. Animal studies showed that immuno-PET could be useful for visualizing CD138-expressing tumors with ^{124}I-B-B4 in the context of treatment of metastatic triple-negative breast cancer that cannot benefit from hormone therapy or anti-Her2/neu immunotherapy [35]. PET distribution of the ^{124}I-B-B4 radiolabeled mAb correlated well with the biodistribution data analyzed on sacrificed animals. Moreover, it has been recently demonstrated that ^{64}Cu-cetuximab immuno-PET represented EGFR expression levels in an esophageal squamous cell carcinoma model, ^{177}Lu-cetuximab RIT effectively inhibited tumor growth, and that ^{64}Cu-/^{177}Lu-PCTA-cetuximab may be useful as a diagnostic tool in patient selection and a potent RIT agent for EGFR-positive tumors [36]. Similarly, Rizvi et al. conducted a prospective clinical study to evaluate the biodistribution and radiation dosimetry of ^{90}Y-ibritumomab tiuxetan (Zevalin®) using ^{89}Zr-ibritumomab tiuxetan [37]. Patients with relapsed or refractory aggressive B-cell (CD20-positive) NHL underwent a PET scan at 1, 72, and 144 h after injection of 70 MBq ^{89}Zr-ibritumomab tiuxetan and again 2 weeks later after coinjection of 15 MBq/kg or 30 MBq/kg of ^{90}Y-ibritumomab tiuxetan. Biodistribution of ^{89}Zr-ibritumomab tiuxetan was not influenced by simultaneous therapy with ^{90}Y-ibritumomab tiuxetan, and the correlation between predicted pre-therapy and absorbed therapy organ doses as based on ^{89}Zr-ibritumomab tiuxetan images was high. These results are similar to previous data presented by Perk et al. [38] and confirm the potential value of pre-therapy ^{89}Zr-immuno-PET to enable individualized treatment by optimizing RIT dose schedules and limit unnecessary toxicity for patients.

4 Conclusion

While radiolabeled mAbs have demonstrated encouraging results in the treatment of hemopathies and several solid tumors, randomized clinical trials in stratified patients need to be performed to confirm efficacy. Treatment of solid tumors by RIT should be developed in combination with several other drugs and in repeated courses of treatment, just as chemotherapy is used. Combinations of all possible new developments, including new antibody specificities, pretargeting methods, fractionated injections, and the use of alpha emitters, are needed to improve RIT efficacy in radioresistant solid tumors. Immuno-PET is likely to assist in selecting patients for RIT, optimizing injected activities, and noninvasively monitoring therapy efficacy.

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References

1. Chatal JF, Kraber-Bodere F, Bodet-Milin C, Rousseau C. Therapeutic immunoconjugates. Which cytotoxic payload: chemotherapeutic (ADC) or radionuclide (ARC) ? Curr Cancer Therapy Rev. 2016;12:54.
2. Siegel JA, Pawlyk DA, Lee RE, Sasso NL, Horowitz JA, Sharkey RM, et al. Tumor, red marrow, and organ dosimetry for 131I-labeled anti-carcinoembryonic antigen monoclonal antibody. Cancer Res. 1990;50:1039s–42s.
3. Bardiès M, Bardet S, Faivre-Chauvet A, Peltier P, Douillard JY, Mahé M, et al. Bispecific antibody and iodine-131-labeled bivalent hapten dosimetry in patients with medullary thyroid or small-cell lung cancer. J Nucl Med. 1996;37:1853–9.
4. Zanzonico P. Radioimmunotherapy of micrometastases: a continuing evolution. J Nucl Med. 1992;33:2180–3.
5. DeNardo GL, Schlom J, Buchsbaum DJ, Meredith RF, O'Donoghue JA, Sgouros G, et al. Rationales, evidence, and design considerations for fractionated radioimmunotherapy. Cancer. 2002;94:1332–48.
6. Schlom J, Molinolo A, Simpson JF, Siler K, Roselli M, Hinkle G, et al. Advantage of dose fractionation in monoclonal antibody-targeted radioimmunotherapy. J Natl Cancer Inst. 1990;82:763–71.
7. Illidge TM, Mayes S, Pettengell R, Bates AT, Bayne M, Radford JA, et al. Fractionated ^{90}Y-ibritumomab tiuxetan radioimmunotherapy as an initial therapy of follicular lymphoma: an international phase II study in patients requiring treatment according to GELF/BNLI criteria. J Clin Oncol. 2014;32:212–8.
8. Scholz CW, Pinto A, Linkesch W, Lindén O, Viardot A, Keller U, et al. (90)Yttrium-ibritumomab-tiuxetan as first-line treatment for follicular lymphoma: 30 months of follow-up data from an international multicenter phase II clinical trial. J Clin Oncol. 2013;31:308–13.
9. Batra JS, Karir BS, Vallabhajosula S, Christos PJ, Hodes G, Date PR, et al. Fractionated dose radiolabeled antiprostate specific membrane antigen (PSMA) radioimmunotherapy (177Lu-J591) with or without docetaxel for metastatic castration-resistant prostate cancer (mCRPC). ASCO Meeting Abstr. 2015;33:194.
10. Tagawa ST, Milowsky MI, Morris M, Vallabhajosula S, Christos P, Akhtar NH, et al. Phase II study of Lutetium-177-labeled anti-prostate-specific membrane antigen monoclonal antibody J591 for metastatic castration-resistant prostate cancer. Clin Cancer Res. 2013;19:5182–91.
11. Tschmelitsch J, Barendswaard E, Williams C, Yao TJ, Cohen AM, Old LJ, et al. Enhanced antitumor activity of combination radioimmunotherapy (131I-labeled monoclonal antibody A33) with chemotherapy (fluorouracil). Cancer Res. 1997;57:2181–6.
12. Wong JYC, Chu DZ, Yamauchi DM, Williams LE, Liu A, Wilczynski S, et al. A phase I radioimmunotherapy trial evaluating 90yttrium-labeled anti-carcinoembryonic antigen (CEA) chimeric T84.66 in patients with metastatic CEA-producing malignancies. Clin Cancer Res. 2000;6:3855–63.
13. Wong JYC, Shibata S, Williams LE, Kwok CS, Liu A, Chu DZ, et al. A phase I trial of 90Y-anti-carcinoembryonic antigen chimeric T84.66 radioimmunotherapy with 5-fluorouracil in patients with metastatic colorectal cancer. Clin Cancer Res. 2003;9:5842–52.
14. Gulec SA, Cohen SJ, Pennington KL, Zuckier LS, Hauke RJ, Horne H, et al. Treatment of advanced pancreatic carcinoma with 90Y-Clivatuzumab Tetraxetan: a phase I single-dose escalation trial. Clin Cancer Res. 2011;17:4091–100.
15. Ocean AJ, Pennington KL, Guarino MJ, Sheikh A, Bekaii-Saab T, Serafini AN, et al. Fractionated radioimmunotherapy with (90) Y-clivatuzumab tetraxetan and low-dose gemcitabine is active in advanced pancreatic cancer: a phase 1 trial. Cancer. 2012;118:5497–506.
16. Bander NH, Milowsky MI, Nanus DM, Kostakoglu L, Vallabhajosula S, Goldsmith SJ. Phase I trial of 177lutetium-labeled J591, a monoclonal antibody to prostate-specific membrane antigen, in patients with androgen-independent prostate cancer. J Clin Oncol. 2005;23:4591–601.

17. Tagawa ST, Whang YE, Kaur G, Vallabhajosula S, Christos PJ, Nikolopoulou A, et al. Phase I trial of docetaxel/prednisone plus fractionated dose radiolabeled anti-prostate-specific membrane antigen (PSMA) monoclonal antibody 177lu-J591 in patients with metastatic, castration-resistant prostate cancer (mCRPC). ASCO Meeting Abstr. 2014;32:5064.
18. Barbet J, Peltier P, Bardet S, Vuillez JP, Bachelot I, Denet S, et al. Radioimmunodetection of medullary thyroid carcinoma using indium-111 bivalent hapten and anti-CEA x anti-DTPA-indium bispecific antibody. J Nucl Med. 1998;39:1172–8.
19. Kraeber-Bodéré F, Rousseau C, Bodet-Milin C, Mathieu C, Guérard F, Frampas E, et al. Tumor immunotargeting using innovative radionuclides. Int J Mol Sci. 2015;16:3932–54.
20. Salaun P-Y, Campion L, Bournaud C, Faivre-Chauvet A, Vuillez J-P, Taieb D, et al. Phase II trial of anticarcinoembryonic antigen pretargeted radioimmunotherapy in progressive metastatic medullary thyroid carcinoma: biomarker response and survival improvement. J Nucl Med. 2012;53:1185–92.
21. Rossi EA, Goldenberg DM, Cardillo TM, McBride WJ, Sharkey RM, Chang C-H. Stably tethered multifunctional structures of defined composition made by the dock and lock method for use in cancer targeting. Proc Natl Acad Sci U S A. 2006;103:6841–6.
22. Sharkey RM, McBride WJ, Karacay H, Chang K, Griffiths GL, Hansen HJ, et al. A universal pretargeting system for cancer detection and therapy using bispecific antibody. Cancer Res. 2003;63:354–63.
23. Schoffelen R, van der Graaf WTA, Franssen G, Sharkey RM, Goldenberg DM, McBride WJ, et al. Pretargeted 177Lu radioimmunotherapy of carcinoembryonic antigen-expressing human colonic tumors in mice. J Nucl Med. 2010;51:1780–7.
24. Schoffelen R, Woliner-van der Weg W, Visser EP, Goldenberg DM, Sharkey RM, McBride WJ, et al. Predictive patient-specific dosimetry and individualized dosing of pretargeted radioimmunotherapy in patients with advanced colorectal cancer. Eur J Nucl Med Mol Imaging. 2014;41:1593–602.
25. Bodet-Milin C, Ferrer L, Rauscher A, Masson D, Rbah-Vidal L, Faivre-Chauvet A, et al. Pharmacokinetics and dosimetry studies for optimization of Pretargeted Radioimmunotherapy in CEA-expressing advanced lung Cancer patients. Front Med (Lausanne). 2015;2:84.
26. Chatal J-F, Davodeau F, Cherel M, Barbet J. Different ways to improve the clinical effectiveness of radioimmunotherapy in solid tumors. J Cancer Res Ther. 2009;5(Suppl 1):S36–40.
27. Guérard F, Gestin J-F, Brechbiel MW. Production of [(211)At]-astatinated radiopharmaceuticals and applications in targeted α-particle therapy. Cancer Biother Radiopharm. 2013;28:1–20.
28. Jurcic JG, Larson SM, Sgouros G, McDevitt MR, Finn RD, Divgi CR, et al. Targeted alpha particle immunotherapy for myeloid leukemia. Blood. 2002;100:1233–9.
29. Jurcic JG, Rosenblat TL. Targeted alpha-particle immunotherapy for acute myeloid leukemia. Am Soc Clin Oncol Educ Book. 2014:e126–31.
30. Meredith R, Torgue J, Shen S, Fisher DR, Banaga E, Bunch P, et al. Dose escalation and dosimetry of first-in-human α radioimmunotherapy with 212Pb-TCMC-trastuzumab. J Nucl Med. 2014;55:1636–42.
31. Orozco JJ, Bäck T, Kenoyer A, Balkin ER, Hamlin DK, Wilbur DS, et al. Anti-CD45 radioimmunotherapy using (211)At with bone marrow transplantation prolongs survival in a disseminated murine leukemia model. Blood. 2013;121:3759–67.
32. Zalutsky MR, Reardon DA, Akabani G, Coleman RE, Friedman AH, Friedman HS, et al. Clinical experience with alpha-particle emitting 211At: treatment of recurrent brain tumor patients with 211At-labeled chimeric antitenascin monoclonal antibody 81C6. J Nucl Med. 2008;49:30–8.
33. Andersson H, Cederkrantz E, Bäck T, Divgi C, Elgqvist J, Himmelman J, et al. Intraperitoneal alpha-particle radioimmunotherapy of ovarian cancer patients: pharmacokinetics and dosimetry of (211)At-MX35 F(ab')2 – a phase I study. J Nucl Med. 2009;50:1153–60.
34. Kraeber-Bodéré F, Bodet-Milin C, Rousseau C, Eugène T, Pallardy A, Frampas E, et al. Radioimmunoconjugates for the treatment of cancer. Semin Oncol. 2014;41:613–22.

35. Rousseau C, Ruellan AL, Bernardeau K, Kraeber-Bodéré F, Gouard S, Loussouarn D, et al. Syndecan-1 antigen, a promising new target for triple-negative breast cancer immuno-PET and radioimmunotherapy. A preclinical study on MDA-MB-468 xenograft tumors. EJNMMI Res. 2011;1:20.
36. Song IH, Lee TS, Park YS, Lee JS, Lee BC, Moon BS, et al. Immuno-PET imaging and radioimmunotherapy of 64Cu-/177Lu-labeled anti-EGFR antibody in esophageal squamous cell carcinoma model. J Nucl Med. 2016;57:1105.
37. Rizvi SNF, Visser OJ, Vosjan MJWD, van Lingen A, Hoekstra OS, Zijlstra JM, et al. Biodistribution, radiation dosimetry and scouting of 90Y-ibritumomab tiuxetan therapy in patients with relapsed B-cell non-Hodgkin's lymphoma using 89Zr-ibritumomab tiuxetan and PET. Eur J Nucl Med Mol Imaging. 2012;39:512–20.
38. Perk LR, Visser OJ, Stigter-van Walsum M, Vosjan MJWD, Visser GWM, Zijlstra JM, et al. Preparation and evaluation of (89)Zr-Zevalin for monitoring of (90)Y-Zevalin biodistribution with positron emission tomography. Eur J Nucl Med Mol Imaging. 2006;33:1337–45.