Demonstration of highly specific toxicity of the $\alpha$-emitting radioimmunoconjugate $^{211}$At-rituximab against non-Hodgkin’s lymphoma cells

E Aurlien$^1$, RH Larsen$^2$, G Kvalheim$^3$ and ØS Bruland$^1$

$^1$The Norwegian Radium Hospital, Department of Oncology, Montebello, N-0310 Oslo; $^2$University of Oslo, Department of Chemistry, Section D, Box 1033 Blindern, N-0315 Oslo; $^3$The Norwegian Radium Hospital, Clinical Stem Cell Laboratories, Montebello, N-0310 Oslo, Norway

Summary The ability of an $\alpha$-emitter conjugated to a chimaeric anti-CD20 monoclonal antibody to kill selectively human B-lymphoma cells in vitro is reported. Two B-lymphoma cell lines RAEL and K422, and normal haematopoietic progenitor cells from human bone marrow aspirates were incubated with $^{211}$At-rituximab (Rituxan® or MabThera™) and plated in clonogenic assays for survival analyses. Following 1 h incubation with $^{211}$At-rituximab, in concentrations which gave an initial activity of 50 kBq ml$^{-1}$, a high tumour cell to normal bone marrow cell toxicity ratio was obtained; 4.1 to 1.0 log cell kill. Biodistribution studies of $^{211}$At-rituximab in Balb/c mice showed similar stability as that of the iodinated analogue. The data indicate that testing of $^{211}$At-rituximab in human patients is warranted. © 2000 Cancer Research Campaign

Keywords: clonogenic survival; lymphoma cells; bone marrow; $\alpha$-emitter; radioimmunoconjugate; rituximab

Most patients with relapsed non-Hodgkin’s lymphoma (NHL) are incurable by current conventional treatment. High-dose chemotherapy with stem cell support is effective only to patients with chemosensitive disease (Armitage, 1993). Hence, new treatment modalities are needed. Various immunotherapeutic approaches, including the use of native monoclonal antibodies (MoAbs), immunotoxins and radioimmunoconjugates, have been investigated (for review see Multani and Grossbard, 1998). MoAbs against the CD20 antigen given to patients with relapsed B-cell NHL yielded response rates of about 30–50% (McLaughlin et al, 1998). When conjugated to the $\beta$-emitting isotopes $^{131}$I (Press et al, 1995; Kaminski et al, 1996) or $^{90}$Y (Knox et al, 1996), responses in up to 86% were seen (Multani and Grossbard, 1998; Liu et al, 1998). Radioimmunotherapy is dose-limited mainly by haematological toxicity, unless stem cell support is given.

The $\alpha$-emitter $^{211}$At has several favourable physical properties compared to the currently used $\beta$-emitters. Thus the half-life ($t_{1/2}$) of $^{211}$At is only 7.2 h, whereas $^{131}$I and $^{90}$Y have half-lives of 8.0 days and 64 h, respectively. Alpha particles from $^{211}$At have an average energy of 6.8 MeV and their range in soft tissue is only 55–80 $\mu$m, i.e. a few cell diameters. Because of the low range of the $\alpha$-radiation in tissues, need for patient shielding is greatly reduced, allowing $\alpha$-radioimmunotherapy to be given on an outpatient basis (Larsen et al, 1999). Moreover, the high LET (linear energy transfer) of $^{211}$At, about 97 keV $\mu$m$^{-1}$, is close to the optimum value for high RBE (relative biological effectiveness) (Brown, 1986). Alpha emitters are also generally associated with low oxygen enhancement ratio and virtual absence of dose-rate effects (Barendsen et al, 1965; Hall, 1994). The short range of the $\alpha$-emitters implies that the radiation dose is delivered to the cell or within its close vicinity. This fact, together with the targeting ability of the MoAb, makes $\alpha$-particle-emitting immunoconjugates attractive for radioimmunotherapy against dispersed cells and micrometastases.

The present study was performed to explore the ability of $^{211}$At-rituximab to selectively inactivate NHL tumour cells with acceptable bone marrow damage.

MATERIALS AND METHODS

Haematopoietic cells and lymphoma cell lines

Fresh mononuclear bone marrow (BM) cells from healthy volunteers were used. BM may contain various amounts of B-lymphocytes (CD20$^+$), from < 1% up to 15–20%. Peripheral blood progenitor cells (PBPC) were harvested in the recovery phase following chemotherapy and G-CSF (in patients with other diagnoses than NHL), and enriched CD34$^+$ haematopoietic cells (from PBPC) were used. Two B-cell NHL cell lines in exponential growth, RAEL (high-grade) and K422 (low-grade) (Dyer et al, 1990), expressing the CD20 antigen, were studied.

Monoclonal antibody

A chimaeric anti-CD20 MoAb with murine variable regions and human IgG1-kappa constant regions, IDEC-C2B8 or rituximab, was used. This MoAb is marketed by IDEC Pharmaceutical Corporation, San Diego CA and by Genentech Inc, San Francisco CA in the USA under the name of Rituxan® and, in Europe by Hoffmann-La Roche Ltd, Basel, Switzerland under the name MabThera™. The CD20 antigen is expressed at high density on malignant B-cells as well as normal B-lymphocytes, but not on early B-cells and stem cells. Within the time-frame of $^{211}$At decay, the CD20 antigen is virtually stable on the cell surfaces (Reff et al, 1994).
Radionuclides and labelling

Astatine-211 was produced in a cyclotron at the Department of Physics, University of Oslo (Larsen et al, 1994 a) using the $^{210}$Bi($\alpha$, 2n) $^{211}$At reaction. Commercially available $^{125}$I (New England Nuclear, Billerica MA, USA) was used for the iodine labelling. Rituximab was radiolabelled with $^{211}$At and $^{125}$I and purified as previously described for other proteins (Larsen et al, 1994 b). The resulting radioimmunoconjugates, $^{211}$At-rituximab and $^{125}$I-rituximab, were sterile-filtered prior to the biological experiments, using a Millex-GV 0.22 μm filter (Millipore, Bedford MA, USA).

In the present experiments the specific activity varied from 10–75 MBq mg$^{-1}$. The specific binding fraction of $^{211}$At-labelled rituximab was determined in a one-point assay. Cell-bound radioimmunoconjugate was measured in triplicate after incubation at room temperature for 1–2 h, using approximately 10$^7$ cells in 0.3 ml medium. An average of 36.4% of the labelled antibody (range 17.3–54.1%) bound to antigen-positive RAEL cells, whereas non-specific binding of $\leq$1.8% was measured on the antigen-negative osteosarcoma cell line OHS. A binding fraction of 36% in the one-point assay corresponds to an immunoreactive fraction at infinite antigen excess (Lindmo et al, 1984) of about 50–60%, as verified experimentally (data not shown).

Cell killing activity of $^{211}$At-rituximab

Different amounts of $^{211}$At-rituximab were added to a fixed concentration of 10$^7$ cells ml$^{-1}$. Initial radioactivity concentrations (kBq ml$^{-1}$) in the test tubes were counted in a LKB Wallac 1277 Gammamaster gammacounter (Wallac Oy, Turku, Finland). Two experimental series with different incubation time were performed: A 1 h incubation series to study the efficacy of cell-bound radioimmunoconjugate; and a 4 h incubation series to simulate the prolonged circulation in vivo of IgG-type radioimmunoconjugates. Four hours are similar to the effective half-life of the radiopharmaceutical in the body, as indicated in mice experiments. After the incubation, under soft stirring for 1 or 4 h, the cells were washed in culture medium and centrifuged three times. Then survival was measured in clonogenic assays, and compared to that observed in the absence of radioactivity.

The number of clonogenic tumour cells remaining after exposure to $^{211}$At-rituximab was assessed by a modified version (Kvalheim et al, 1987) of the Courtenay and Mills soft agar clonogenic assay (Courtenay and Mills, 1978). For evaluation of toxicity on haematopoietic cells the CFU-c (colony forming unit cell) assay with METHOCULT H4433 medium (Stem Cell Technologies Inc, Vancouver, Canada) was used. The toxicity of the labelled MoAbs was also tested on more primitive (stem cell- near) haematopoietic cells, using the LTC-IC (long-term culture initiating cell) assay (Eaves et al, 1991). Briefly, the cells were seeded on a pre-irradiated (20 Gy) human bone marrow stroma mono-layer in the medium MYELOCULT H5100 (Stem Cell Technologies Inc), supplemented with 50 μM hydrocortisone, and cultured for 5 weeks before secondary culture in CFU-c assay.

Therapeutic gain

The relationship between the initial concentration of radioactivity (kBq ml$^{-1}$) and clonogenic survival was, in the range tested, best fitted by quadratic regression. The concentration of $^{211}$At-rituximab giving a survival of 10% or 37% ($A_{10}$ or $A_{37}$) was calculated from the survival curves. A therapeutic gain factor (TGF) for tumour cells relative to haematopoietic cells exposed to $^{211}$At-rituximab, was calculated using the following equation:

$$TGF_{37} = \frac{A_{37}, BM}{A_{37}, tumour}$$

Biodistribution

Biodistribution was studied in Balb/c mice. Astatine-211 and $^{125}$I were examined in a ‘paired label’ fashion where 30 kBq of $^{125}$I-labelled and 120 kBq of $^{211}$At-labelled rituximab in 100 μl were co-injected into the tail vein of each animal. Approximately 4 μg rituximab were injected per mouse. At various time-points three mice were sacrificed, the organs were dissected out and weighed, the radioactivity contents were measured and the percentage of injected dose per g of tissue was determined.

Statistical analyses and curve estimations

SPSS 8.0, EXCEL 97 for Windows, and SigmaPlot (Scientific Graphic Software, version 2, Jandel Corporation) were used in the statistical analyses, plots and graphs. The input values for the survival curves were calculated by the mean log$_{10}$ anti-log of surviving fractions for separate doses in the individual experiments (presented in the Figures). Survival curves were fitted to the data using quadratic regression of log$_{10}$ survival. The 95% confidence intervals (CI) of the survival curves were calculated using log$_{10}$ of the 95% CI from each point on the survival curves.

RESULTS

Cell-bound radioactivity

Cell-bound radioactivity (mBq cell$^{-1}$), measured after 1 h incubation, is presented in Figure 1A for normal BM and CD34$^+$ cells and in Figure 1B for the RAEL cells. It is seen that, as expected, much more radioactivity was bound to the tumour cells than to the normal BM cells.
Survival of lymphoma cells and haematopoietic cells following exposure to $^{211}$At-rituximab

Clonogenic survival vs initial activity concentration of $^{211}$At-rituximab is shown in Figure 2. The data in Figure 2A represent experiments with BM cells from five different donors tested in CFU-c, four of them also examined in LTC-IC assays. In addition, one experiment with enriched CD34$^+$ cells is included in the figure. In Figure 2B four individual experiments with RAEL cells are shown. Comparison of Figure 2A and 2B (1 h measurements) demonstrates a highly significant, favourable survival of the BM cells compared to the malignant RAEL cells. From the survival curves the following mean $\Lambda_{10}$ values were estimated: 50.0 kBq ml$^{-1}$ for BM$_{\text{CFU-c}}$, 39.1 for BM$_{\text{LTC-IC}}$, 203.5 for CD34$^+$, and 9.0 for RAEL. An initial activity concentration of 50 kBq ml$^{-1}$ yielded corresponding log cell kill values of 1.00, 1.24, 0.20 and 4.13. In Table 1 the values are given with 95% CI.

Table 1: $\Lambda_{10}$ values$^a$ and log cell kills$^a$ following exposure to $^{211}$At-rituximab

| Cells | BM$_{\text{CFU-c}}$ | BM$_{\text{LTC-IC}}$ | CD34$^+$ | PBPC$_{\text{CFU-c}}$ | RAEL | K422 |
|-------|---------------------|---------------------|----------|---------------------|------|------|
| Incubation time – 1 h | | | | | | |
| $\Lambda_{10}$ (kBq ml$^{-1}$) | 50.0 | 39.1 | 203.5 | – | 9.0 | – |
| Log cell kill | 1.00 | 1.24 | 0.20 | – | 4.13 | – |
| at 50 kBq ml$^{-1}$ | (0.91–1.10) | (1.09–1.61) | (0.12–0.28) | | (3.95–4.50) | |
| Incubation time – 4 h | | | | | | |
| $\Lambda_{10}$ (kBq ml$^{-1}$) | 22.8 | – | – | – | 5.6 | 4.3 |
| Log cell kill | 1.01 | – | – | – | 0.61 | (5.5–6.0) |
| at 25 kBq ml$^{-1}$ | (0.86–1.14) | | | | (3.18–3.96) | (4.1–4.6) |

$^a$Mean (95%CI); $^b$Initial activity concentration
Table 2 Therapeutic gain factor at 37% cell survival (TGF$_{37}$) following exposure to $^{211}$At-rituximab

| Incubation time | RAEL to 211At-rituximab | K422 to 211At-rituximab |
|-----------------|--------------------------|--------------------------|
| 1 h             | BM$_{CFU-c}$ 5.2 ± 0.7   | BM$_{CFU-c}$ 4.2 ± 1.2   |
|                 | CD34$^+$ 32.8 ± 2.0      | CD34$^+$ 26.8 ± 3.9      |
|                 | PBPC$_{CFU-c}$ -         | PBPC$_{CFU-c}$ -         |
| 4 h             | BM$_{CFU-c}$ 2.3 ± 0.5   | BM$_{CFU-c}$ -           |
|                 | CD34$^+$ 5.0 ± 2.8       | PBPC$_{CFU-c}$ 3.0 ± 0.7 |
|                 | PBPC$_{CFU-c}$ -         | PBPC$_{CFU-c}$ 6.5 ± 3.7 |

$^a$Mean ± SD

Table 3 Biodistribution of $^{211}$At- and $^{125}$I-rituximab in mice

| Tissue   | 211At-rituximab | 125I-rituximab |
|----------|-----------------|----------------|
|          | 1 h             | 3.5 h          |
|          | 211At-rituximab | 211At-rituximab |
| Blood    | 36.2 ± 2.9      | 37.9 ± 2.1     |
| Lung     | 15.5 ± 3.6      | 15.7 ± 3.8     |
| Liver    | 11.5 ± 1.5      | 13.0 ± 1.7     |
| Heart    | 10.5 ± 0.8      | 10.1 ± 0.8     |
| Spleen   | 8.1 ± 2.7       | 8.7 ± 1.9      |
| Kidney   | 7.9 ± 0.8       | 9.1 ± 0.8      |
| Muscle   | 1.0 ± 0.4       | 1.2 ± 0.3      |
| Bone     | 2.8 ± 0.7       | 2.9 ± 0.3      |

|          | 211At-rituximab | 125I-rituximab |
|----------|-----------------|----------------|
|          | 10.75 h         | 10.75 h        |
| Blood    | 27.1 ± 1.2      | 28.5 ± 0.6     |
| Lung     | 11.6 ± 1.3      | 11.1 ± 1.2     |
| Liver    | 6.7 ± 0.7       | 8.2 ± 1.2      |
| Heart    | 8.9 ± 1.3       | 8.4 ± 0.5      |
| Spleen   | 7.3 ± 0.6       | 7.2 ± 0.2      |
| Kidney   | 6.5 ± 1.3       | 7.7 ± 0.9      |
| Muscle   | 1.6 ± 0.1       | 1.8 ± 0.1      |
| Bone     | 3.0 ± 0.2       | 3.2 ± 0.2      |

|          | 23.5 h          | 23.5 h         |
|----------|-----------------|----------------|
| Blood    | 17.4 ± 1.4      | 20.1 ± 2.0     |
| Lung     | 8.5 ± 1.9       | 8.7 ± 1.8     |
| Liver    | 4.4 ± 1.2       | 4.2 ± 3.4     |
| Heart    | 6.8 ± 0.9       | 6.9 ± 0.9     |
| Spleen   | 4.8 ± 0.8       | 5.0 ± 0.6     |
| Kidney   | 4.1 ± 0.1       | 5.2 ± 0.6     |
| Muscle   | 1.5 ± 0.3       | 1.8 ± 0.3     |
| Bone     | 2.0 ± 0.4       | 2.9 ± 0.2     |

$^a$Values expressed as % of i.v. injected dose of $^{211}$At- and $^{125}$I-rituximab per gram tissue, mean ± SD

Therapeutic gain

Both 1 h and 4 h incubation with $^{211}$At-rituximab gave favourable therapeutic windows between tumour cells and haematopoietic cells. The average TGF$_{37}$ ± SD, is presented in Table 2. In the 1 h series TGF$_{37}$ between RAEL and BM$_{CFU-c}$ was 5.2 in average. TGF$_{37}$ for tumour cells vs BM$_{CFU-c}$ in the 4 h series were 2.3 for RAEL and 3.0 for K422.

Biodistribution in mice

Table 3 presents the tissue distribution of $^{211}$At-rituximab and $^{125}$I-rituximab in eight organs of the mice, within a time-frame of 1–23.5 h. The biodistribution of $^{211}$At-rituximab was similar compared to that of $^{125}$I-rituximab, and revealed the general characteristics of IgG radiolabelled MoAbs, i.e. high initial activity levels in blood and blood-rich tissues.

DISCUSSION

The reported experiments were designed to assess targeted $\alpha$-emitter exposure to dispersed B-lymphoma cells and haematopoietic progenitor cells.

The normal bone marrow cells generally showed a low sensitivity following exposure to $^{211}$At-rituximab. BM contains various amounts of B-lymphocytes, ranging from < 1% up to 20%, which may explain the considerable variation in cell-bound activity observed in different experiments. The difference in toxicity on the BM cells, reflected in the survival curve (Figures 2A and 2C), could possibly be explained by crossfire irradiation of the antigen-negative clonogenic cells from B-cells in the pellets before seeding. This is further supported by the very low toxicity observed with CD34$^+$-enriched cells, which had low binding of $^{211}$At-rituximab, due to the virtual absence of B-cells in this cell population. Furthermore, PBPC products, which contain about 0.5–1% B-cells, also showed a trend towards lower cytotoxicity from the radioimmunoconjugate compared to BM, although the difference was not statistically significant. Neither was the survival significantly different between BM cells tested concurrently in the CFU-c and the LTC-IC assay.

Astatine-211-labelled rituximab was highly cytotoxic to both high- and low-grade NHL cells when measured after 4 h incubation, and no significant difference in survival between the two cell lines was found (Figure 2D).

In our experiments, A$_{19}$ following 1 h incubation was 3.8 and 19.5 kBq ml$^{-1}$ for RAEL and BM$_{CFU-c}$, respectively, whereas the calculated value for CD34$^+$ cells was as high as 100.9 kBq ml$^{-1}$. Hence, A$_{19}$ for CD34$^+$ cells was almost identical to the values previously reported for human cell lines exposed to non-specific $^{211}$At-BSA (bovine serum albumin) (Larsen et al, 1994a).

The biodistribution profile indicated that the quality of the immunoconjugate had not been significantly reduced by labelling with $^{211}$At and also that the stability of the conjugate in vivo was adequate. As with $^{131}$I-labeled compounds a blocking agent should be used for therapeutic purposes to minimize radiation dose to the thyroid for $^{211}$At (Larsen et al, 1998).

The current study confirms previous reports indicating that $\alpha$-emitting immunoconjugates may have selective anti-tumour effect. One study by Harrison and Royle (1987) used $^{211}$At conjugated to a T-cell antibody (OX7) to treat a T-cell lymphoma in mice and increased the median survival time of mice and probably cured more than half of the animals (Harrison and Royle, 1987). Later Behr and colleagues (1999) compared the therapeutic efficacy and toxicity of the $\alpha$-emitter $^{211}$Bi with that of the $\beta$-emitter $^{90}$Y, linked to a monovalent Fab$^\prime$ fragment, in a human colon cancer xenograft model in nude mice (Behr et al, 1999).

They concluded that radioimmunotherapy with $\alpha$-emitters may be therapeutically more effective than conventional $\beta$-emitters. Recently pharmacokinetics and dosimetry of a $^{211}$Bi-labelled anti-CD33-antibody in patients with leukaemia was reported (Sgouros et al, 1999).

Clinical studies with radioimmunoconjugates ($^{131}$I and $^{90}$Y) in lymphomas have mainly involved murine MoAbs. Using a chimeric MoAb like rituximab in radioimmunotherapy could...
strongly reduce HAMA (human anti-mouse antibody) responses in patients. However, the prolonged biological half-life of the humanized MoAbs could cause problems with increased radiation dose to normal tissues, and hence a decreased therapeutic index, when isotopes with longer half-life (days) are utilized (Bruland, 1995; Multani and Grossbard, 1999). The use of a relatively short-lived isotope like 211At will make those differences in biological half-life less relevant.

As pointed out, 211At also has several advantages from a radiation-protection point of view. More than 99% of its radiation energy is from α-particles. The short half-life (7.2 h) could reduce the hospitalization time, and the short tissue-range of the α-particles (maximum 80 μm) would simplify the protection of the staff and environment (Larsen et al., 1999).

A BM progenitor cell survival of 5% should be sufficient for recovery without stem-cell support. This survival level of BMCFU-s was observed at an initial activity concentration of 70.3 kBq ml⁻¹ following 1 h incubation. The corresponding log cell kill of RAE1 was as high as 4.8. Consequently there may be a significant therapeutic window if non-myeloablative doses of 211At-rituximab are explored clinically.

In conclusion, our study shows that rituximab labelled with 211At exerts selective cytotoxicity to NHL tumour cells in vitro. The effect of the radioimmunoconjugate will now be investigated in patients with relapsed B-cell lymphomas.

ACKNOWLEDGEMENTS

The work was financially supported by The Norwegian Cancer Society, grant no. 97014 and 96070. Rituximab (MabThera™) was a gift from Roche Norway A/S. Thanks are due to Dr LS Rusten, Clinical Stem Cell Laboratories, the Norwegian Radium Hospital, Oslo, Norway, for instruction in the LTC-IC method, and to Mr E Olsen, Department of Physics, University of Oslo for performing cyclotron irradiations to produce 211At. We are indebted to Professor A Pihl for constructive criticism of the manuscript.

REFERENCES

Armitage JO (1993) Treatment of non-Hodgkin’s lymphoma. N Engl J Med 328: 1023–1030
Barendsen GW, Koot CJ, Van Kersen GR, Bewley DK, Field SB and Parnell CJ (1965) The effect of oxygen on impairment of the proliferative capacity of human cells in culture by ionizing radiations of different LET. Int J Radiat Oncol Biol Phys 10: 317–327
Behr TM, Behe M, Stabin MG, Wehrmann E, Apostolidis C, Molinet R, Strutz F, Fayyazi A, Wieland E, Gratz S, Koch L, Goldenberg DM and Becker W (1999) High-linear energy transfer (LET) alpha versus low-LET beta emitters in radioimmunotherapy of solid tumors: therapeutic efficacy and dose-limiting toxicity of 213Bi-versus 90Y-labeled CO17-1A Fab’ fragments in a human colonic cancer model. Cancer Res 59: 2635–2643
Brown I (1986) Astatine-211: its possible applications in cancer therapy. Int J Radiat Appl Instrum A 37: 789–798
Bruland OS (1995) Cancer therapy with radio labelled antibodies. An overview. Acta Oncol 34: 1085–1094
Courtney VD and Mills J (1978) An in vitro colony assay for human tumours grown in immune-suppressed mice and treated in vivo with cytotoxic agents. Br J Cancer 37: 261–268
Dyer MJ, Fischer P, Nacheva E, Labadite W and Karpas A (1990) A new human B-cell non-Hodgkin’s lymphoma cell line (Karpas 422) exhibiting both t(14;18) and t(4;11) chromosomal translocations. Blood 75: 709–714
Eaves CJ, Cashman, JD and Eaves AC (1991) Methodology of long-term culture of human hemopoietic cells. J Tiss Cult Meth 13: 55–62
Hall EJ (1994) Linear energy transfer and relative biological effectiveness. In: Radiobiology for the Radiologist, 4th edn, pp 153–164. Philadelphia: JB Lippincott Company
Harrison A and Royle L (1987) Efficacy of astatine-211 labelled monoclonal antibody in treatment of murine T-cell lymphoma. National Cancer Institute Monogr: 157–158
Kaminski MS, Zasadny KR, Francis IR, Fenner MC, Ross CW, Milik AW, Estes J, Tuck M, Regan D, Fisher S, Glenn SD and Wahd RL (1996) Iodine-131-anti-B1 radioimmunotherapy for B-cell lymphoma. J Clin Oncol 14: 1974–1981
Knox SJ, Goris ML, Trlesker L, Neerin C, Davis T, Liles TM, Grillo LA, Chinn P, Varns C, Nimg SC, Fowler S, Deb N, Becker M, Marquez C and Levy R (1996) Yttrium-90-labeled anti-CD20 monoclonal antibody therapy of recurrent B-cell lymphoma. Clin Cancer Res 2: 457–470
Kvalheim E, Frostdt O, Pihl A, Nustad K, Pharo A, Uglandstad J and Funderud S (1987) Elimination of B-lymphoma cells from human bone marrow: model experiments using monodisperse magnetic particles coated with primary monoclonal antibodies. Cancer Res 47: 846–851
Larsen RH, Bruland OS, Hoff P, Alstad J, Lindmo T and Rofstad EK (1994a) Inactivation of human osteosarcoma cells in vitro by 211At-TP-3 monoclonal antibody: comparison with astatine-211 labeled bovine serum albumin, free astatine-211 and external-beam X rays. Radiat Res 139: 178–184
Larsen RH, Hoff P, Alstad J and Bruland OS (1994b) Preparation and quality control of 211At-labelled and 125I-labelled monoclonal antibodies. Biodistribution in mice carrying human osteosarcoma xenografts. Journal of Labelled Compounds and Radiopharmaceuticals XXXIV: 774–785
Larsen RH, Slade S and Zalutsky MR (1998) Blocking [211At]astatide accumulation in normal tissues: preliminary evaluation of seven potential compounds. Nucl Med Biol 25: 351–357
Larsen RH, Murud KM, Akabani G, Hoff P, Bruland OS and Zalutsky MR (1999) 211At- and 125I-labeled bisphosphonates with high in vivo stability and bone accumulation. J Nucl Med 40: 1197–1203
Lindmo T, Boven E, Cuitatt E, Fedorko J and Bunn PA Jr. (1984) Determination of the immunoreactive fraction of radio labeled monoclonal antibodies by linear extrapolation to binding at infinite antigen excess. J Immunol Methods 72: 77–89
Liu SY, Eary JF, Petersdorf SH, Martin PJ, Maloney DG, Appelbaum FR, Mathews DC, Bush SA, Durack LD, Fisher DR, Gooley TA, Bernstein ID and Press OW (1998) Follow-up of relapsed B-cell lymphoma patients treated with iodine-131-labeled anti-CD20 antibody and autologous stem-cell rescue. J Clin Oncol 16: 3270–3278
McLaughlin P, Grillo-Lopez AJ, Link BK, Levy R, Cruzman MS, Williams ME, Heyman MR, Bence-Bruker I, White CA, Cabañas F, Jain V, Ho AD, Lister J, Wey K, Shen D and Dallaire BK (1998) Rituximab chimeric anti-CD20 monoclonal antibody therapy for relapsed indolent lymphoma: half of patients respond to a four-dose treatment program. J Clin Oncol 16: 2825–2833
Multani PS and Grossbard ML (1998) Monoclonal antibody-based therapies for lymphoma. Adv Immunol 66: 23–29
Press OW, Eary JF, Appelbaum FR, Martin PJ, Nelp WB, Glenn S, Fisher DR, Porter B, Matthews DC and Gooley T (1995) Phase II trial of 131I-B1 (anti-CD20) antibody therapy with autologous stem cell transplantation for relapsed B cell lymphomas. Cancer 346: 336–340
Reff ME, Carner K, Chambers KS, Chinn PC, Leonard JE, Raab R, Newman RA, Hanna N and Anderson DR (1994) Depletion of B cells in vivo by a chimeric mouse human monoclonal antibody to CD20. Blood 83: 435–445
Sgouros G, Ballangrud AM, Jurcic JG, McDevitt MR, Humm JL, Mehta BM, Finn RD, Larson SM and Scheinberg DA (1999) Pharmacokinetics and dosimetry of an alpha-particle emitter labeled antibody: 213Bi-HuM195 (anti-CD33) in patients with leukemia. J Nucl Med 40: 1935–1946

© 2000 Cancer Research Campaign

British Journal of Cancer (2000) 83(10), 1375–1379

Anti-CD-20 α-particle-radioimmunotherapy 1379