Immuno-PET imaging with ^{89}Zr-rituximab in CD20+ B-cell lymphoma

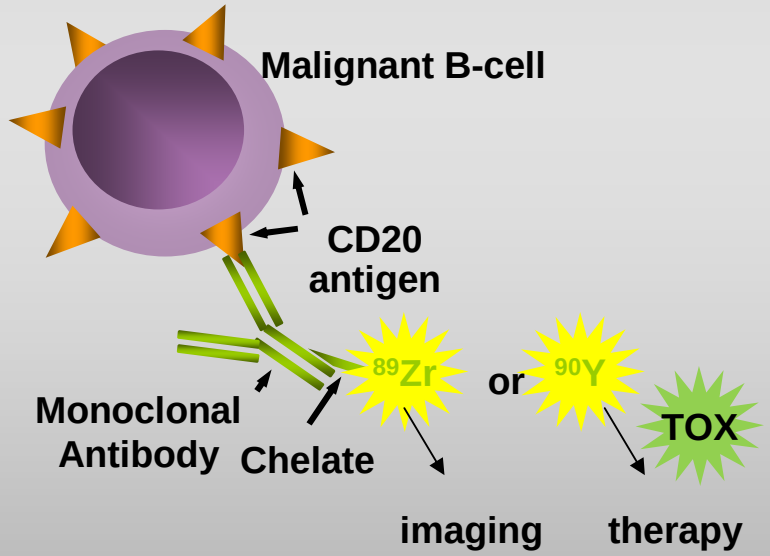
Kristoff Muylle MD

Jules Bordet Cancer Institute
Department of Nuclear medicine
Brussels
Belgium

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Immunohistochemical staining in vivo

- **^{89}Zr-rituximab-PET/CT combines the high sensitivity of PET/CT with the specificity of the chimeric monoclonal antibody (mAb) rituximab for the CD20-antigen expressed on the surface of CD20+ B-cell non-Hodgkin's lymphoma (NHL).**
- **Zirconium-89 (^{89}Zr)**
  - **a positron emitter with a half-life of 78.4 hours, which is compatible with the time needed for intact mAb to achieve optimal tumour-to-background ratios.**
  - **produced in a cyclotron by a (p,n) reaction on natural yttrium-89 (^{89}Y)**
  - **residualizes in the target cell after internalization (cfr. ^{68}Ga, ^{64}Cu, ^{86}Y)**



Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Background

- **Stable labeling of mABs/rituximab with ^{89}Zr using a bifunctional chelate (derivative of desferrioxamine B)**
  - **multi/6-step synthesis (Verel I. et al, J Nucl Med. 2003;44:1271–1281.)**
    - **relatively complicated and timeconsuming**
    - **challenging with respect to Good Manufacturing Practice (GMP) compliancy**
  - **2-step synthesis (Perk L. et al, Eur J Nucl Med Mol Imaging (2010) 37:250–259.)**
    - **allows efficient, easy and rapid preparation of optimally performing 89Zr-labeled mAbs**
    - **facilitates further exploration of 89Zr-immuno-PET as an imaging tool.**

- **Similar in vitro stability and biodistribution in NHL-bearing nude mice suggest that ^{89}Zr-labeled mAb can be safely used for monitoring ^{90}Y-(DOTA)-labeled mAb biodistribution in a clinical setting. (Perk L. Eur J Nucl Med Mol Imaging (2006) 33:1337–1345.)**

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Single Centre Pilot Study (YZIRIT): objectives

- **Primary objectives:**
  - **Evaluation of the safety of ^{90}Y-rituximab treatment in patients with B-cell lymphoma who are in PR or progressive disease, when using the Zevalin therapeutic regimen.**
  - **Evaluation of the safety of ^{89}Zr-rituximab PET/CT-imaging**
- **Other study objectives are:**
  - **Evaluation of the efficacy of ^{90}Y-rituximab treatment by assessment of metabolic response status (by FDG-PET/CT-imaging) and progression-free survival.**
  - **Evaluation of the efficacy/accuracy of ^{89}Zr-rituximab PET/CT-imaging**
  - **Diagnostic comparison of ^{89}Zr-rituximab-PET/CT with FDG-PET/CT**
  - **Evaluation of the influence of infusion/predose of unlabelled (cold) rituximab on the distribution of the radioimmuno-conjugate.**

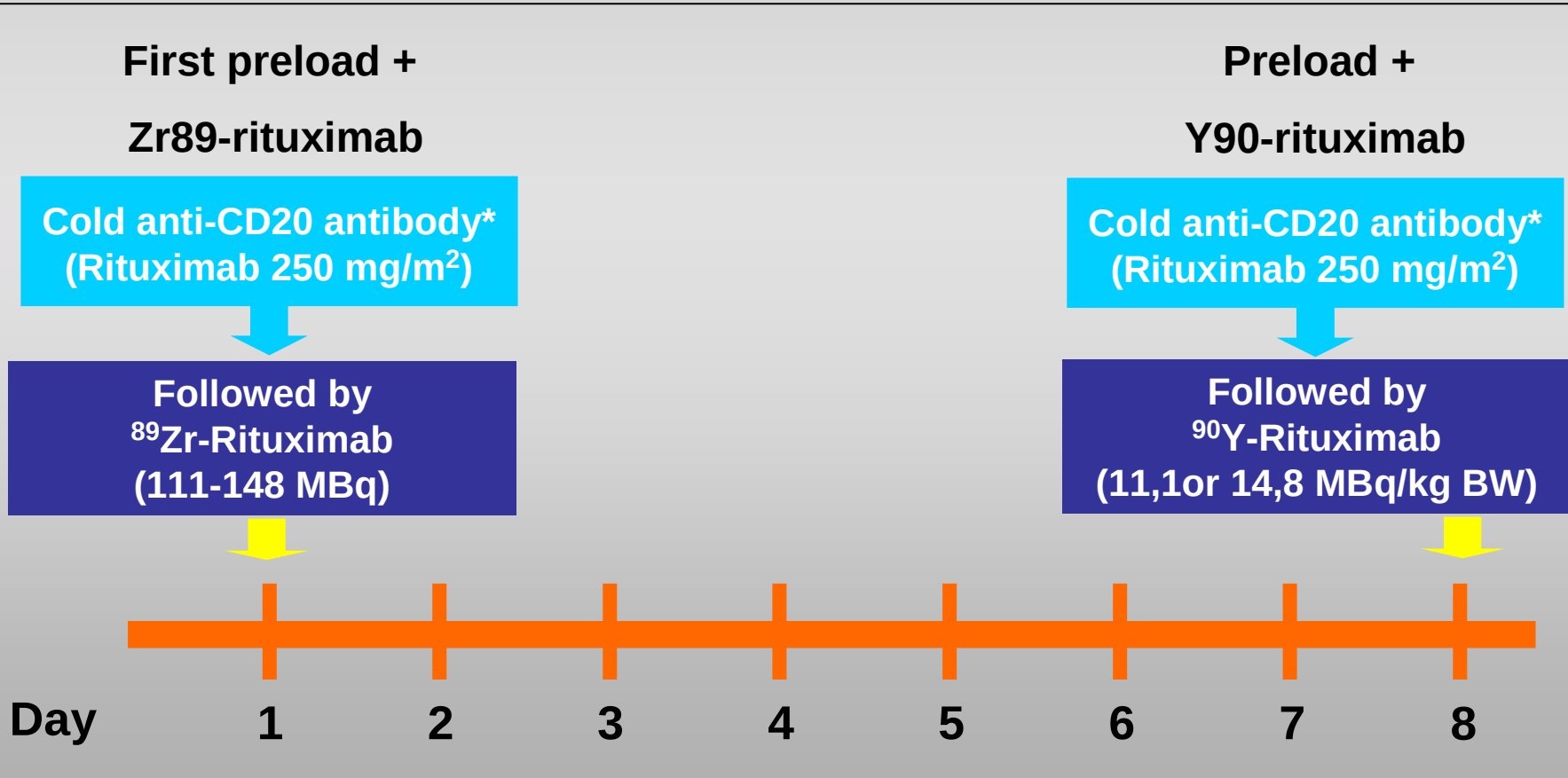
Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

YZIRIT: inclusion criteria

- **Histologically confirmed (according to the REAL/WHO classification) CD20 positive lymphomas**
- **FDG + lesions on baseline FDG-PET/CT**
- **Patients with a PR or PD**
- **Failed at least one regimen of standard treatment/chemotherapy**
- **Age 18 years or older**
- **World Health Organization (WHO) performance status of 0 to 2**
- **Absolute Neutrophil Count (ANC) of 1.5 x 10^9/L or higher**
- **Haemoglobin (Hb) of 9 g/dl or higher**
- **Platelet count of 100 x 10^9/L or higher**
- **Life expectancy of at least 6 months**
- **Written informed consent obtained according to local guidelines**
- **Peripheral blood stem cell harvested before RIT**

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

YZIRIT: (Zevalin) Therapeutic Regimen



Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

YZIRIT: Study Design

- A baseline ^{18}FDG-PET/CT was performed 1 to 4 weeks before the ^{89}Zr-rituximab immuno-PET/CT.
- Injection of 250 mg/m2 rituximab followed by injection of 111-148 MBq ^{89}Zr-rituximab; immuno-PET/CT 1 hour, (1day), 3 days and 6 days p.i.
- Farmacokinetics 5, 15, 30 min & 1, 2, 4, 16, 24, 72 et 144 h p.i.
- 1 week later: Injection of 250 mg/m2 rituximab followed by injection of 11,1-14,8 MBq/kg ^{90}Y-rituximab
- Farmacokinetics 5, 15, 30 min & 1, 2, 4, 16, 24, 72 et 144 h p.i.
- Evaluation of hematotoxicity by weekly blood samples during 12 weeks
- Response assesment by FDG-PET/CT 3 months after RIT
- Follow-up by FDG-PET/CT

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Accuracy: comparison with FDG-PET/CT

- Six patients with relapsed CD20+ B-cell non-Hodgkin's lymphoma (1 mantle cell and 5 follicular lymphomas) were included in this study.
- Similarly to the Zevalin® treatment schedule, each patient received a first infusion of unlabelled (cold) rituximab at 250 mg/m² followed by the injection of 3-4 mCi ^{89}Zr-rituximab and one week later, the same infusion of cold rituximab followed by radioimmunotherapy with ^{90}Y-rituximab (0.3-0.4 mCi/kg).
- ^{89}Zr-rituximab-PET/CT was performed at 4 time points: 1 hour, 24 hours, 3 days and 6 days after intravenous administration of ^{89}Zr-rituximab.
- A baseline ^{18}FDG-PET/CT was performed 1 to 4 weeks before the ^{89}Zr-rituximab immuno-PET/CT.
- Standard uptake values (SUV) were assessed for all PET-positive lesions and compared for both tracers.

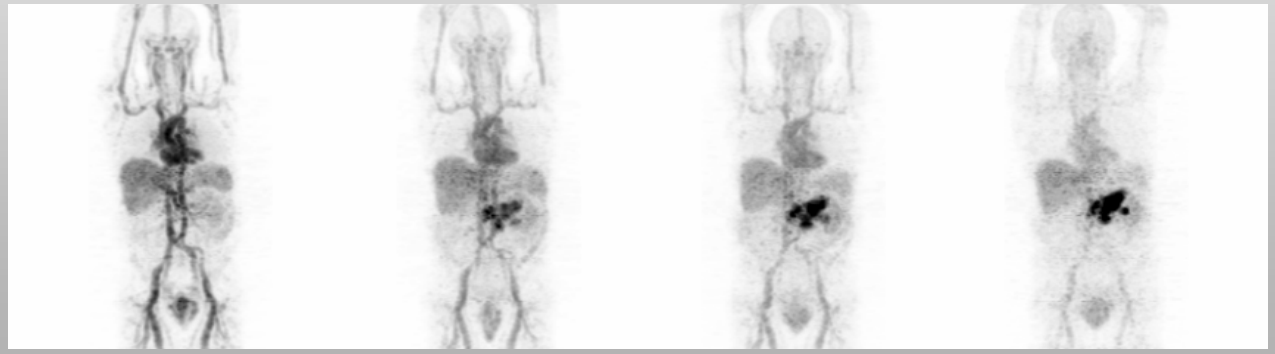
Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Accuracy: comparison with FDG-PET/CT

- ^{18}FDG-PET/CT revealed 24 hypermetabolic lesions (SUVmax: 8±4, range: 2.1 - 15.9) in the 6 evaluated patients.
- All FDG-positive lesions showed significant uptake on ^{89}Zr-rituximab-PET/CT, with highest SUV on the late images (6 days post injection of ^{89}Zr-rituximab; SUVmax: 8.9±5.3, range: 2,6 - 26,1).

^{89}Zr-rituximab Immuno-PET/CT

Patient with an intra-abdominal relapse of a follicular lymphoma.



1 hour p.i. | 1 day p.i. | 2 days p.i. | 6 days p.i.

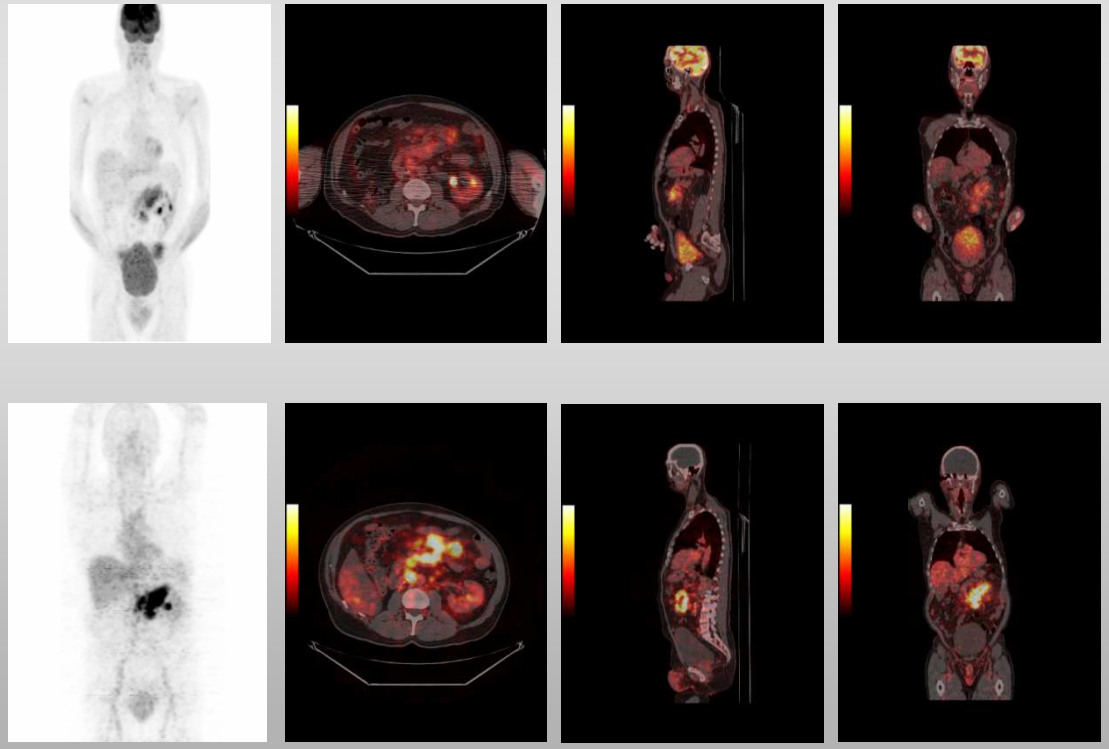
Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Accuracy: comparison with FDG-PET/CT

Comparison between FDG-PET/CT and Zr89-rituximab Immuno-PET/CT 6 days p.i.

FDG-PET

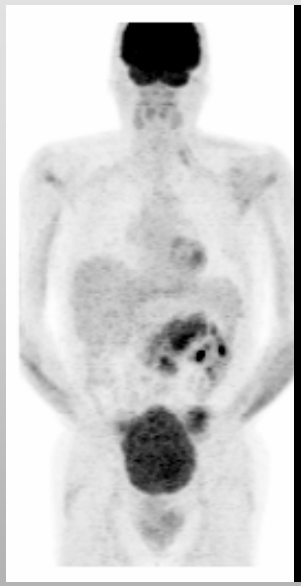
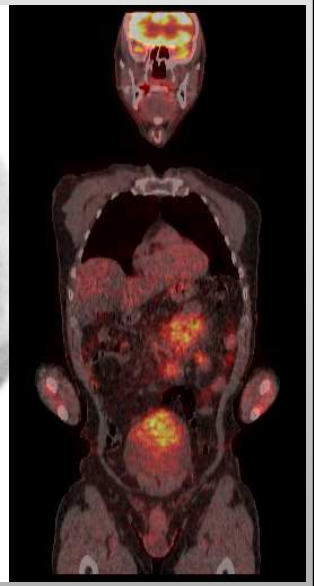
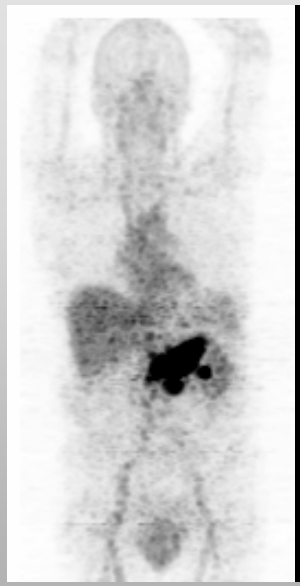
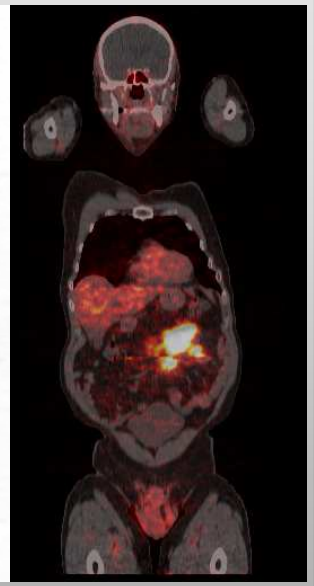
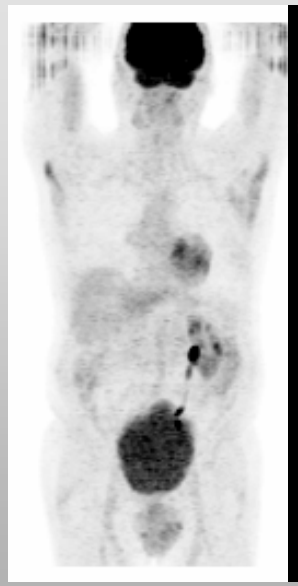
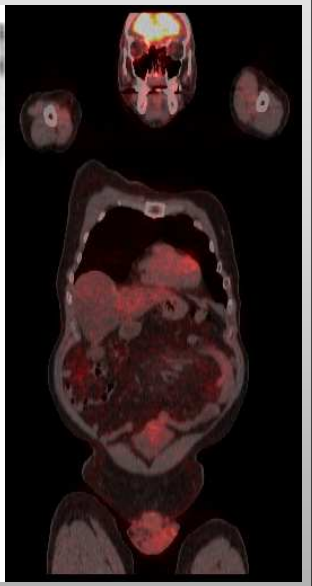
IMMUNO-PET



- Moreover, in 2 out of 6 patients, ^{89}Zr-rituximab-PET/CT revealed 8 supplementary CD20+ lesions which were strictly negative on ^{18}FDG-PET/CT and corresponded to particularly small (≤ 1cm) lymph nodes and mesenteric nodules on CT.

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

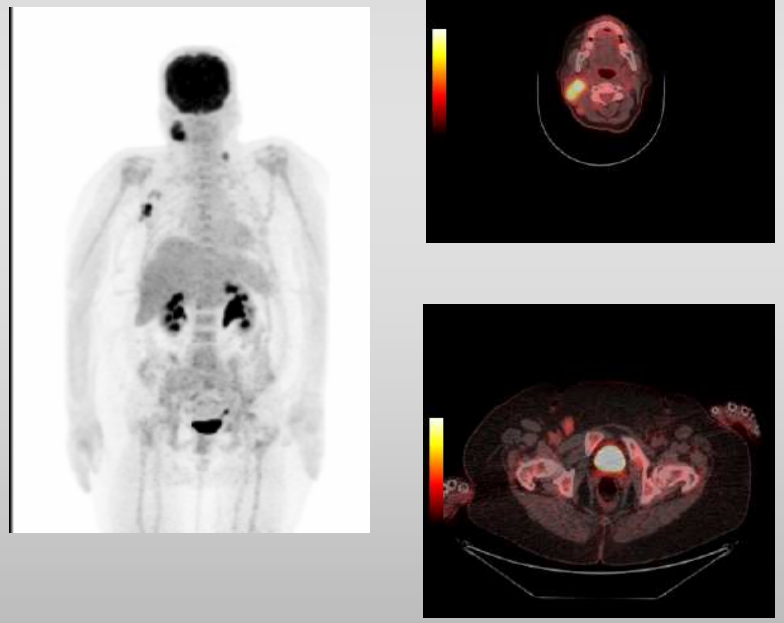
Accuracy: comparison with FDG-PET/CT

FDG-PET/CT before treatment with ^{90}Y-rituximab	Immuno-PET/CT 6 days p.i. of Zr89-rituximab	FDG-PET/CT 6 months after treatment with ^{90}Y-rituximab (0,4 mCi/kg) showing a complete remission
 	 	 
MIP Coronal slice	MIP Coronal slice	MIP Coronal slice

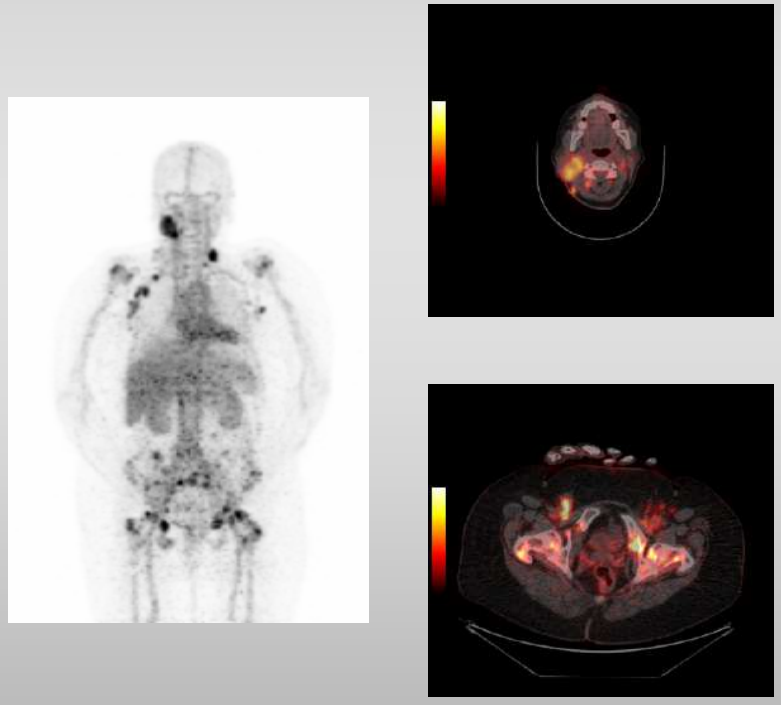
Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Accuracy: comparison with FDG-PET/CT

18FDG-PET/CT



immuno-PET/CT with 89Zr-rituximab 6 days p.i.



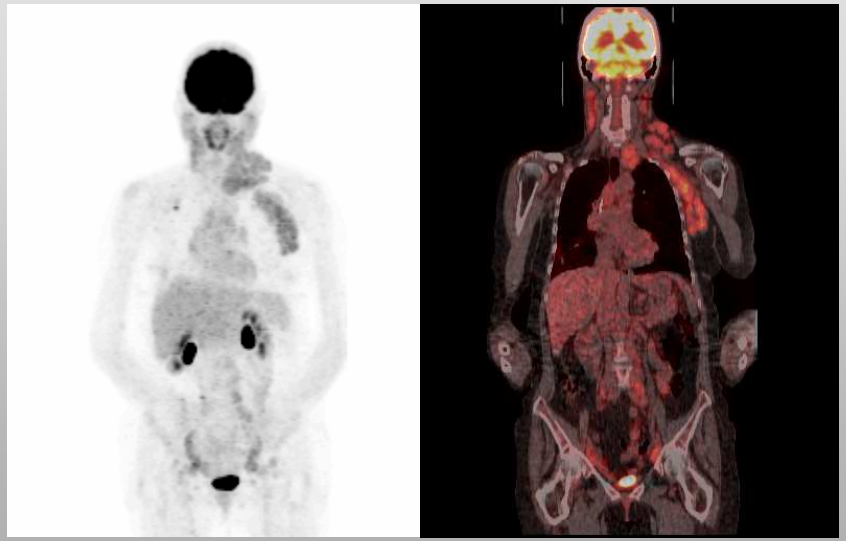
The preliminary results of this pilot study suggest that ^{89}Zr-rituximab-PET/CT is more accurate than ^{18}FDG-PET/CT for the detection of viable lymphoma in patients with predominantly indolent NHL.

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Perspectives: lymphoma with low avidity for FDG

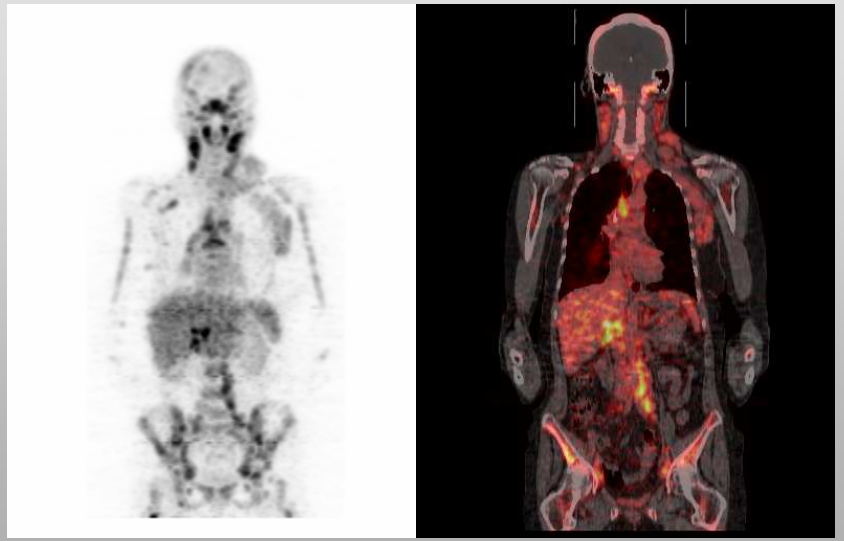
• possible interest of ^{89}Zr-rituximab-PET/CT in lymphoma with low avidity for FDG (e.g. patient with small lymphocytic lymphoma) ; cfr. bone marrow infiltration and several LN M+ not seen on FDG-PET

18FDG-PET/CT



immuno-PET/CT with 89Zr-rituximab

6 days p.i.



Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

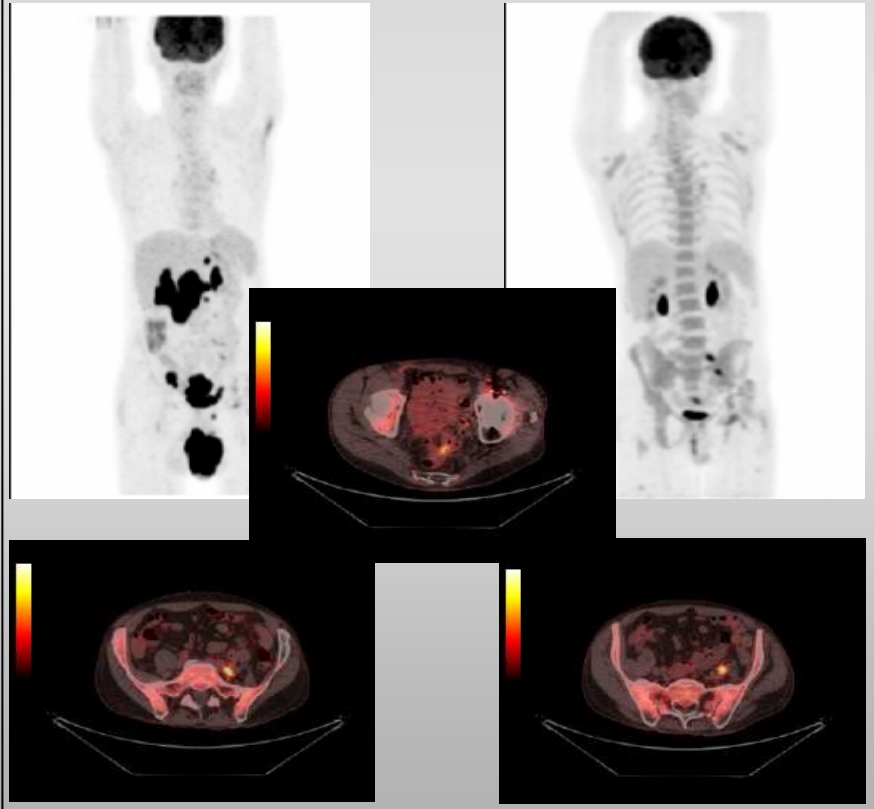
Perspectives: clinical decision making

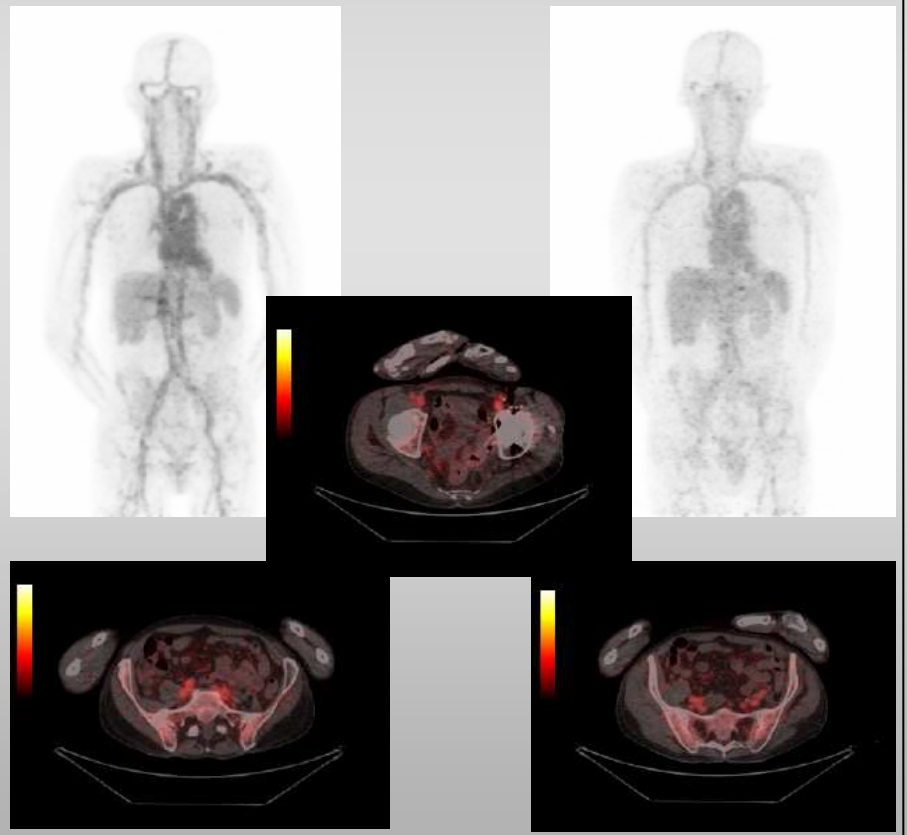
18FDG-PET/CT

Baseline **After treatment**

immuno-PET/CT with 89Zr-rituximab

3 days p.i. **6 days p.i.**

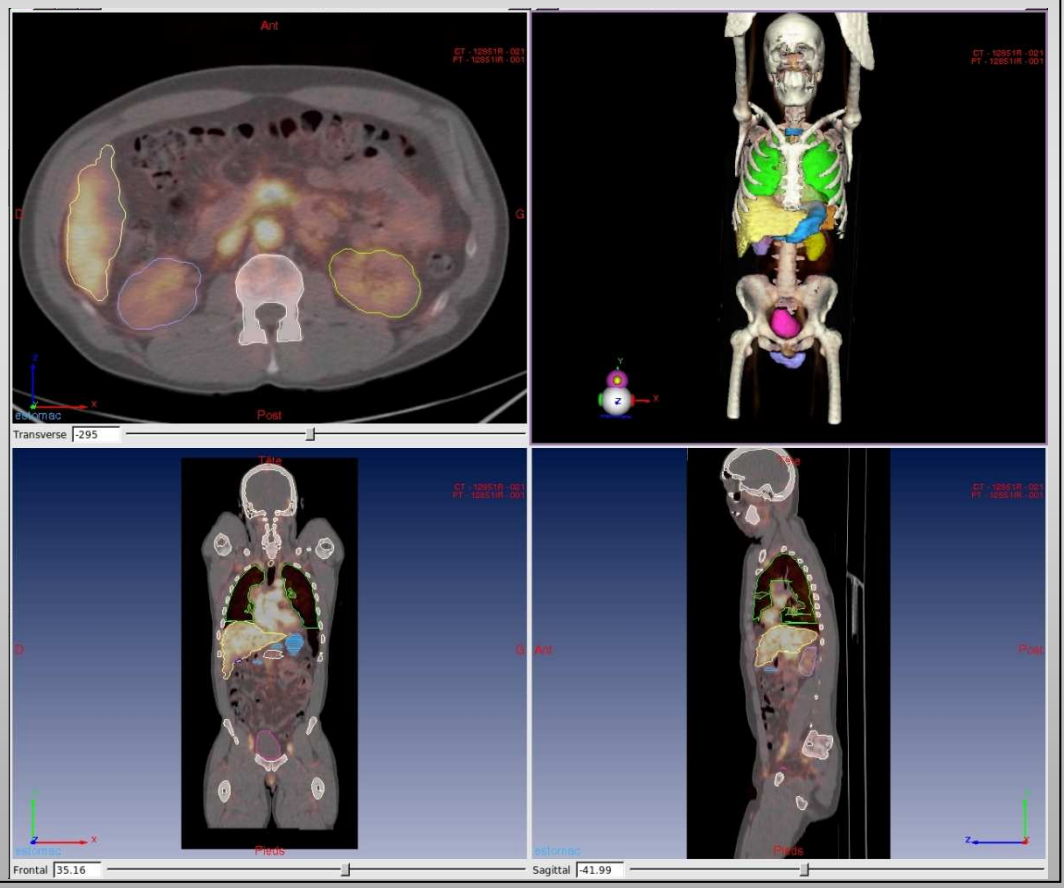




Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Perspectives: 3D dosimetry

- 3D delineation of organs and lesions for dosimetry as a prelude to radioimmunotherapy with 90Y-rituximab opens the door for:
- dose-response correlation
- prediction of treatment outcome
- better selection of patients for receptor-targeted therapy
- patient tailored image-guided therapy.



Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

- Evaluation of the influence of high preload of cold rituximab (250mg/m^2) before the administration of RIT.
- This preload is assumed to clear circulating B-lymphocytes from the blood
- Does this common practice really enhances tumour targetting?

•***Aim***

To compare the distribution of ^{89}Zr-rituximab in 5 patients with histologically confirmed B-cell lymphoma (CD20+) in PR or PD after at least 1 line of therapy, with a positive FDG-PET. Treatment with rituximab has to be stopped at least 6 months before inclusion in this protocol.

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

- ***Methodology***

1) **Diagnostic/dosimetric phase I**: Baseline ^{89}Zr-rituximab PET/CT-imaging: injection of the ^{89}Zr labelled rituximab (3 mCi) without a preload of unlabelled rituximab.
2) **Diagnostic/dosimetric phase II (3 weeks later):** administration of a preload (250mg/m2of unlabelled (cold) rituximab followed by the injection of the ^{89}Zr labelled rituximab (3 mCi).
3) **Therapeutic phase (1 week later)**: the same infusion of 250mg/m2 followed by the slow IV-injection of ^{90}Y labelled rituximab (0.3 mCi/kg if platelet count: 100000 ≤ 150000/mm^3 and 0.4 mCi/kg if platelet count: > 150000/mm^3).

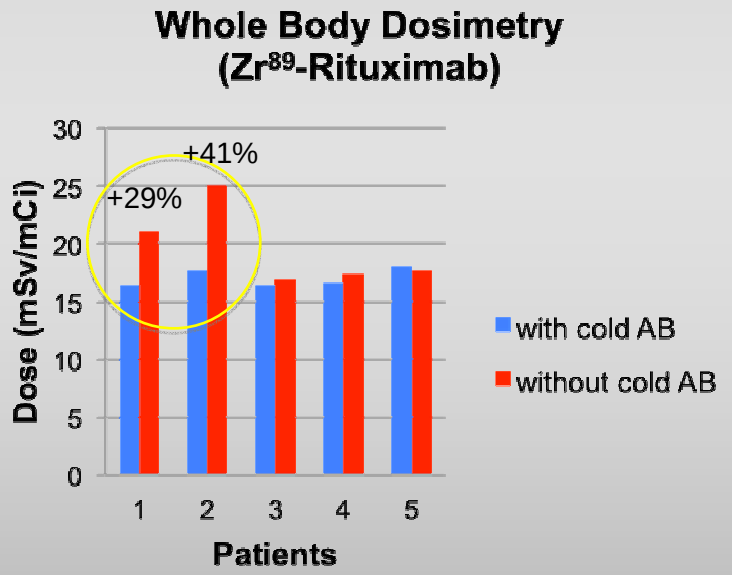
- ***Imaging / Biodistribution***

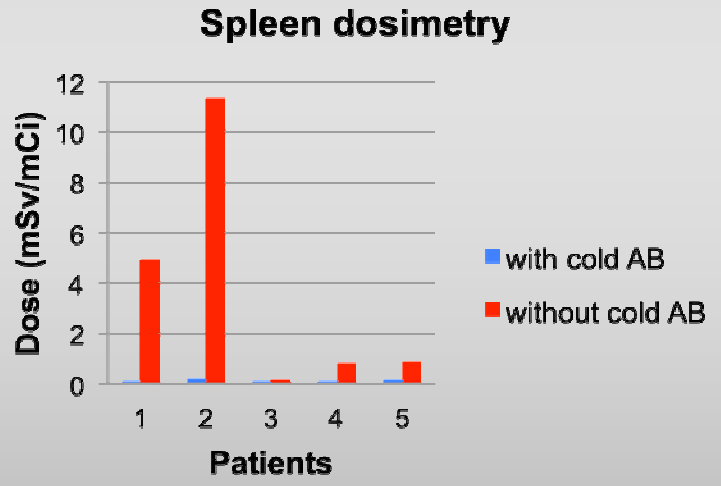
Whole body PET/CT-scans (low dose CT) are done at 3 time points starting within 1 hour and at 72 and 144 hours after both i.v. injections of ^{89}Zr-rituximab

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

Results



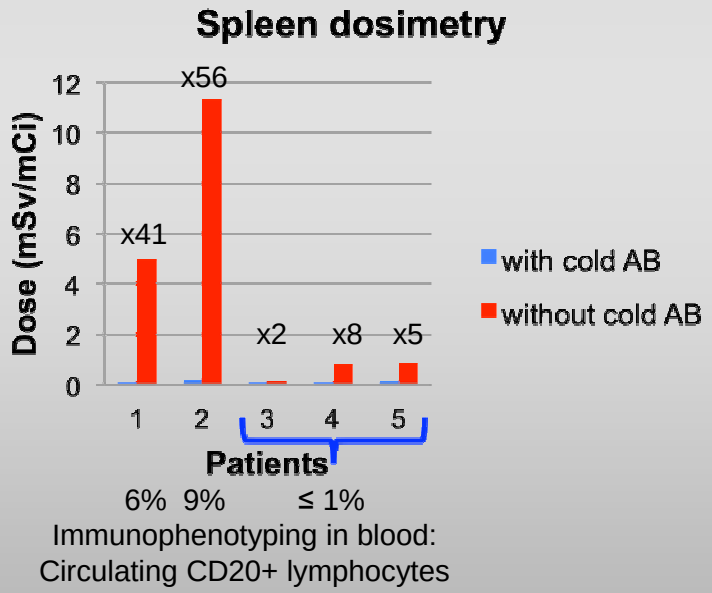


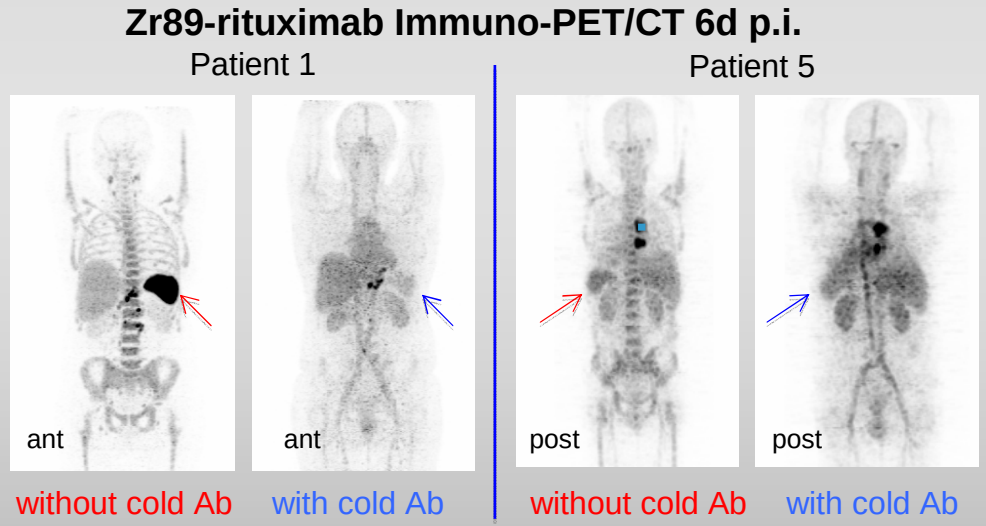
- \+ preload: similar whole body radiation doses for all patients
- \- preload: ➡ increase of whole body radiation dose in 2 out of 5 patients
  similar whole body doses in 3 patients

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

Results



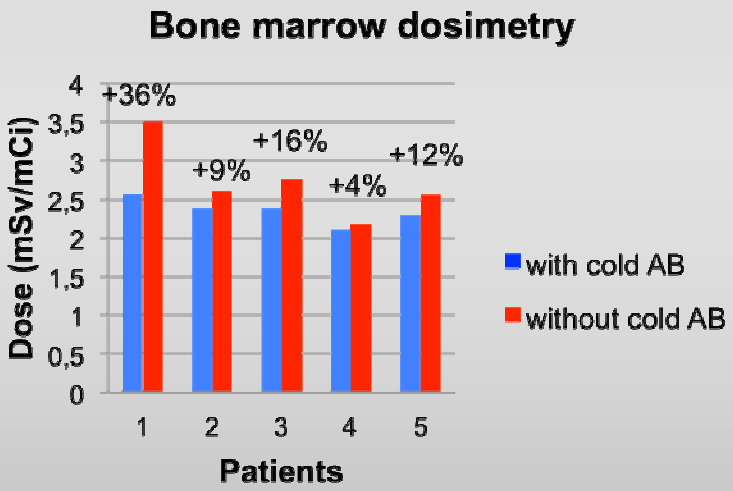


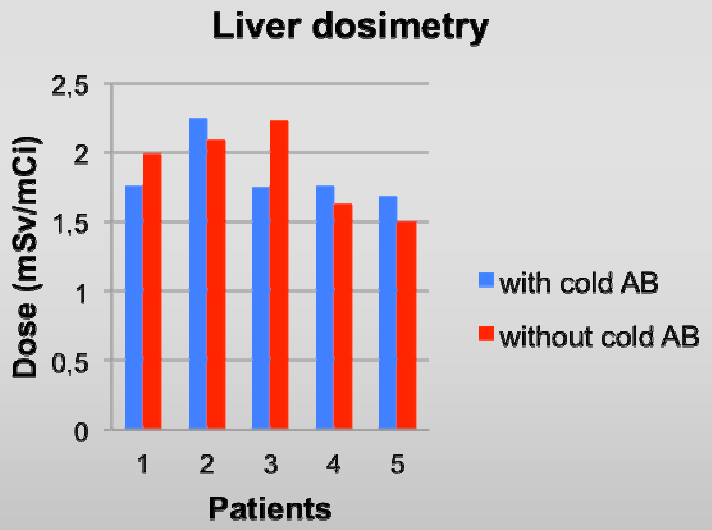
- Influence of a preload of rituximab on the distribution of the radioconjugate, especially the uptake in the spleen, highly depends on the amount of circulating CD20+ lymphocytes.
- Preload: minor influence on the radiation dose to the spleen in patients with B-cell depletion.

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

Results





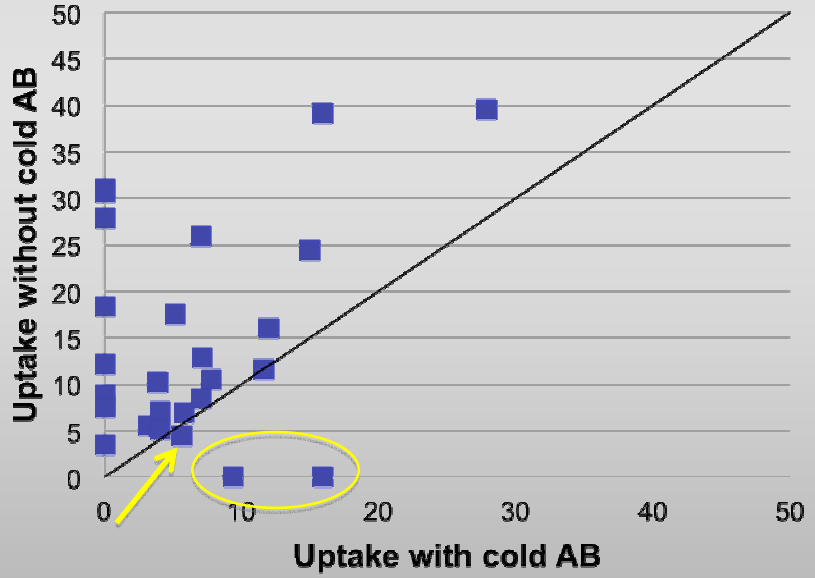
- Without preload: moderate increase of the bone marrow dose by 4-36%
- Preload: No significant influence on the radiation dose of the liver

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

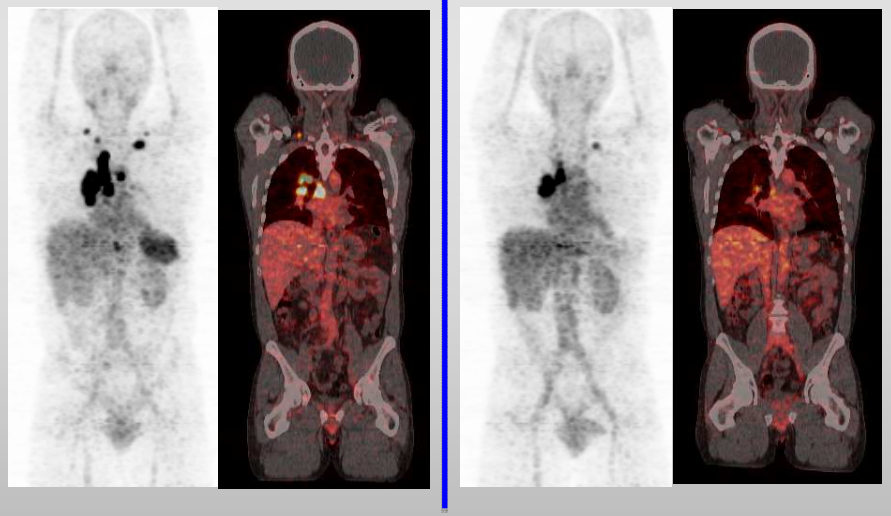
- ***Results***

Lesion uptake with AB vs without AB



Zr89-rituximab Immuno-PET/CT 6d p.i.

Without cold Ab preload **With cold Ab preload**

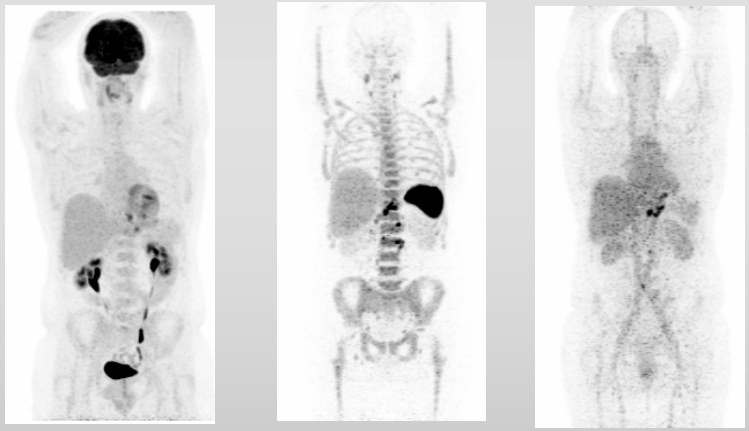


- Lesion uptake / tumor targetting is consistently higher without a preload, at least in patients with B-cell depletion…
- 3 lesions show less or no uptake without preload, all 3 in patients without B-cell depletion.

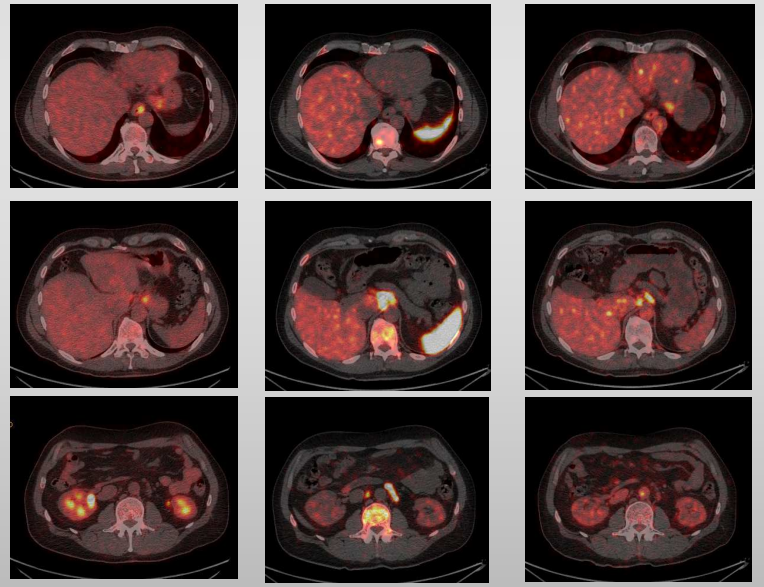
Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

FDG | ImmunoPET - preload | ImmunoPET + preload



FDG | ImmunoPET - Preload | ImmunoPET + preload



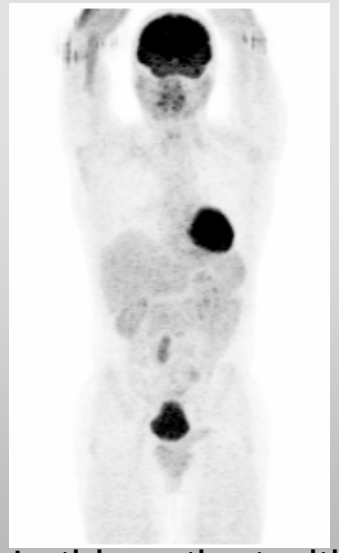
In this patient without B-cell depletion:

- Preload impairs uptake in involved lymph nodes < (partial) saturation with cold mAbs.
- Preload enhances uptake in the 2 visceral lesions < reducing the uptake in the spleen > higher residence time of the radioconjugate in blood > binding in less accessible regions.

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

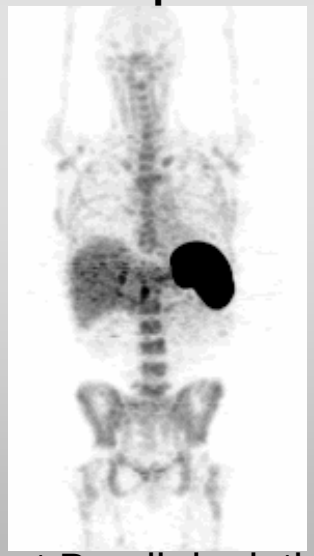
FDG-PET/CT



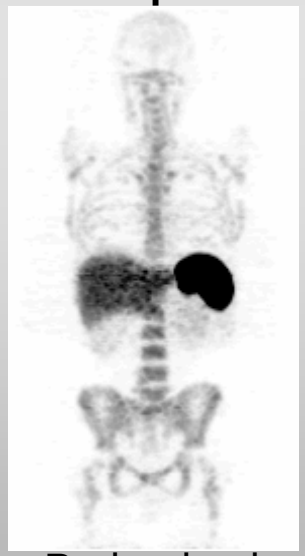
Zr89-rituximab Immuno-PET/CT 6d p.i.

Without cold Ab predose

1h p.i.

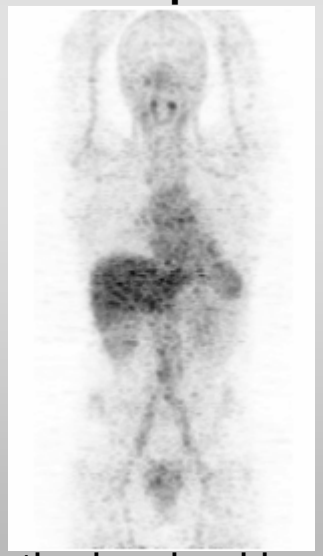


6d p.i.



With cold Ab predose

6d p.i.



In this patient without B-cell depletion: Preload enhances uptake in the involved lymp node
< clearing circulating B-lymphocytes from the blood > reducing the uptake in the spleen
> higher residence time of the radioconjugate in blood > higher uptake at the involved LN.

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Example: preload???

- Evaluation of the influence of high preload of cold rituximab (250mg/m^2) before the administration of RIT; does this common practice really enhances tumour targetting?

Influence of the preload:

1.In patients without B-cell depletion:

- reduces whole body radiation dose
- clears circulating B-lymphocytes from the blood
- reduces significantly the uptake in the spleen
- slower clearance of the radioconjugate from the circulation.
- enhances tumour targetting in some (especially visceral) lesions.

2.In patients with B-cell depletion (majority < previous R-chemo treatment(s):

- No influence on whole body radiation dose
- Lesion uptake / tumour targetting is consistently higher without a preload

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Perspectives: (pre)clinical evaluation of (new) drugs

^{89}Zr-labeled mAbs in (pre-)clinical trials:

Drug	Target	Tumor
• Cetuximab (Erbitux)	EGFR	Colorectal
• Bevacizumab (Avastin)	VEGF-A	Colorectal
• Trastuzumab (Herceptine)	Her2/neu	Breast
• ^{90}Y-Rituximab (RIT)	CD20	B-cell lymphomas
• Trastuzumab-DM1	Her2/neu	Breast

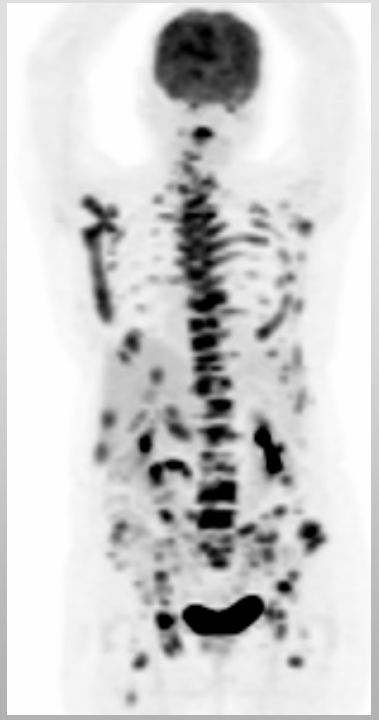
Possible interest in hemato-oncology:

- Immunotoxine: CD30, CD22, CD19,…
- Radioimmunotherapy: CD20, CD22, CD45,…

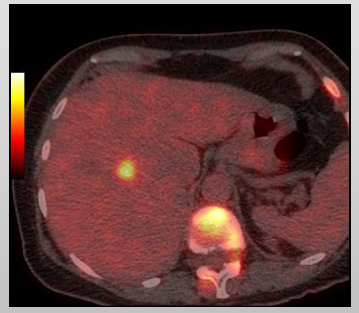
Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

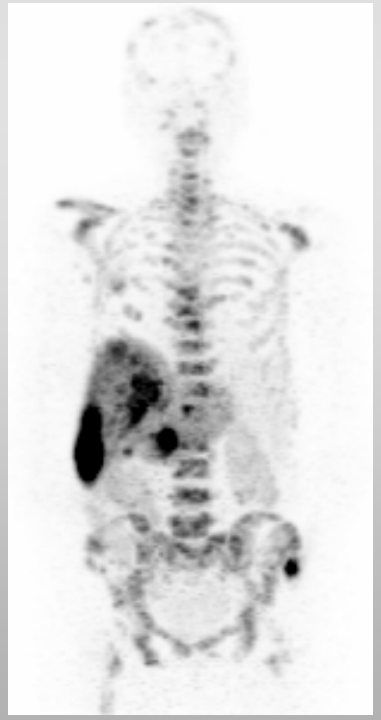
Perspectives: (pre)clinical evaluation of (new) drugs

^{89}Zr-labeled mAbs in (pre-)clinical trials:

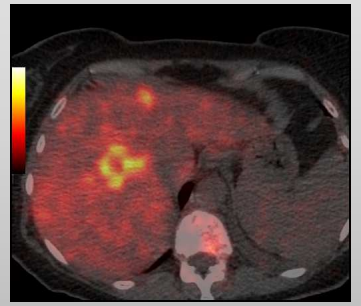


FDG
1 h p.i.





Zr89-trastuzumab
4 days p.i.



G. Gebhart & P. Flamen
Jules Bordet Institute, Brussels

Immuno-PET imaging with ^{89}Zr-rituximab in patients with CD20+ B-cell lymphoma

Conclusions

- ^{89}Zr-rituximab-PET/CT provides an excellent imaging tool for accurate quantification of CD20 antigen-expression, which is of particular interest for dosimetry as a prelude to radioimmunotherapy with ^{90}Y-Rituximab.
- The preliminary results of this pilot study suggest that ^{89}Zr-immuno-PET/CT is a promising imaging technique with perspectives in:
    - Accurate in vivo quantification of receptor-expression (dosimetry)
    - Clinical decision making (e.g. minimal residual FDG-uptake)
    - Evaluation and adaptation of current therapeutic regimens (e.g. predose)
    - Selection of patients for receptor-targeted therapy (e.g. immunotoxines)
    - Prediction of treatment outcome (solid tumours)
    - Individualised targeted therapy

Thanks

Grants

Les amis de Bordet 2007 & 2008

FNRS (Télévie) 2008-2010

National Cancer Plan 2009-2010

Department of Hemato-oncology
Dominique Bron
+
Co-workers

&

Department of Nuclear Medicine
Patrick Flamen
+
Co-workers

Collaboration

Jules Bordet Institute
Brussels
Belgium

VU University Medical Centre
Amsterdam
Netherlands

Guus van Dongen
+
Co-workers