

SUPPLEMENTAL METHODS

Pharmacokinetics and Dosimetry

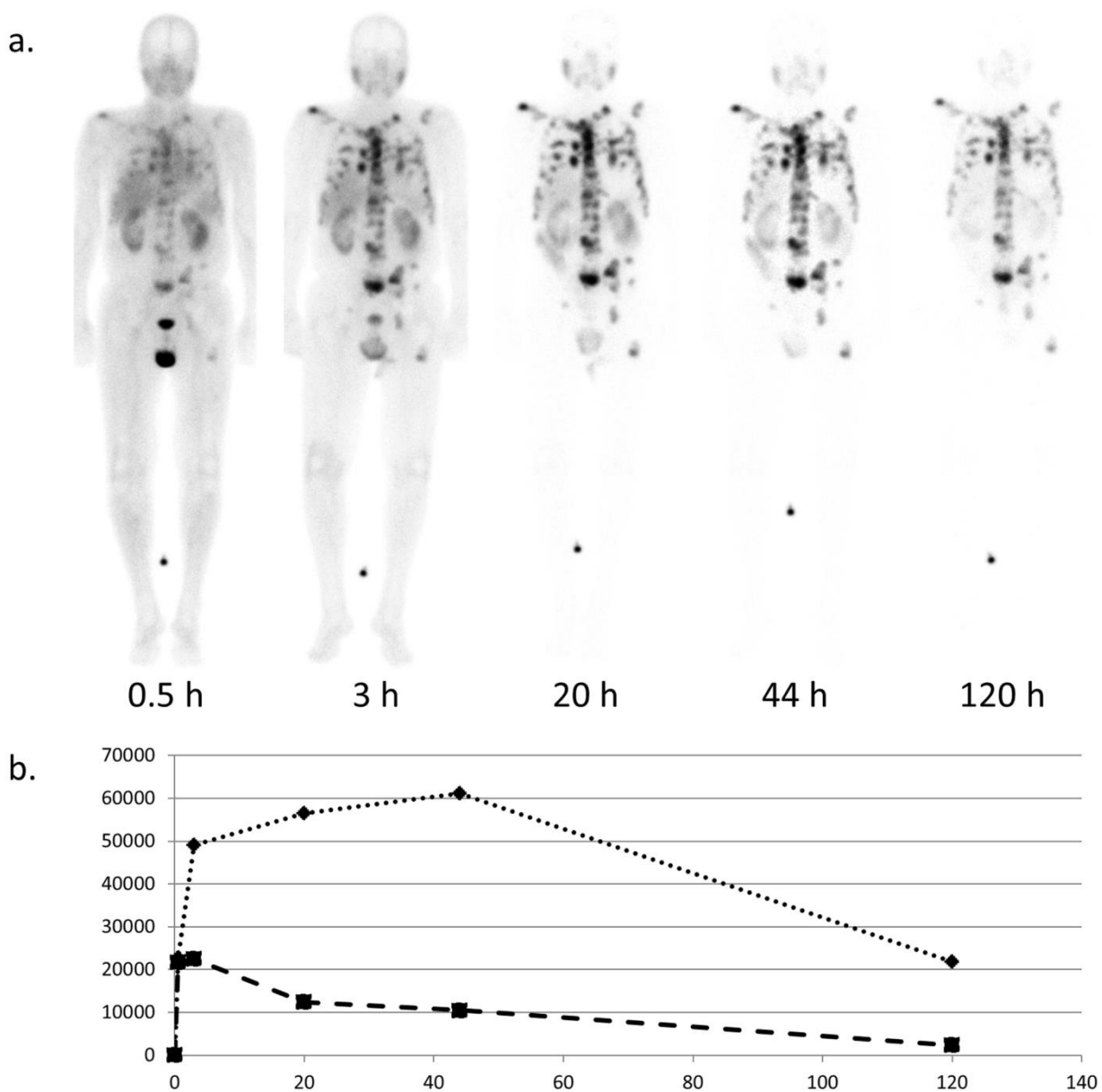
Blood samples were obtained at 30min, 3h, 20h, 44h, 5-8d after the therapeutic injection and the activity of 1ml blood was measured in a well counter (Bertold, Germany). Then the blood was centrifuged and the activity of plasma was measured. The blood/plasma ratio was calculated and compared with the patient's individual hematocrit. Urine was collected 0-4h, 4-24h, 24-44h p.i., measured for volume and activity concentration and then the cumulative renal clearance was calculated. Decay correction between sampling and gamma counting was performed, respectively. Blood and urine samples from 34 treatment cycles of 23 patients could be obtained. However, due to the increased rate of incontinence after prostate surgery and sometimes poor vein conditions only 24 complete test series for blood and 17 reliable test series for urine could be evaluated. The fecal clearance was not measured directly but estimated by imaging.

To estimate radiation dosimetry we selected 4 patients in acceptable general clinical condition and without incontinence, which agreed to and tolerated the serially performed scintigraphy scans. The tumor load of these patients was scored visually (low-intermediate-high). Each patient was evaluated twice with serially performed imaging respectively, once during the first and 8-10 weeks later during the second treatment cycle. Planar anterior and posterior whole-body-scans were obtained with a dual head gamma camera (GE Hawkeye Millennium) with medium energy parallel whole collimator and a scan speed of 15 cm/min at the time points 0.5h, 3h, 20h, 44h and one delayed image 5-8 days p.i. using only the higher 208keV \pm 10% [187-228 keV] photo peak window, respectively. A known calibrated activity of Lu-177 was placed between the legs during the whole-body-scan (Supplement-Figure-1). 3D SPECT/CT images of one bed position at kidney level were acquired at 20h and were used to visually rule out major overlay between kidneys and intestine but were not used for internal radiation dosimetry. Geometric mean (GM) images were generated using a MATLAB script (MathWorks, Massachusetts, USA) and evaluated with ROI (region of interest) technic in PMOD (PMOD Technologies Ltd, Zürich, Switzerland). A rectangular ROI covering the whole body area was drawn on the first planar GM image. As the first time point was imaged before voiding, the total counts of the first whole body ROI, after correction of decay, was assumed to correlate to the injected activity with a calibration factor. Parotid, submandibular, and lacrimal glands were segmented using an ellipsoidal ROI while liver, kidneys, spleen and bladder were segmented using a freehand ROI on the geometric mean image of the second time point and propagated to all other time points. The location of each ROI was manually adjusted to cope with incomplete

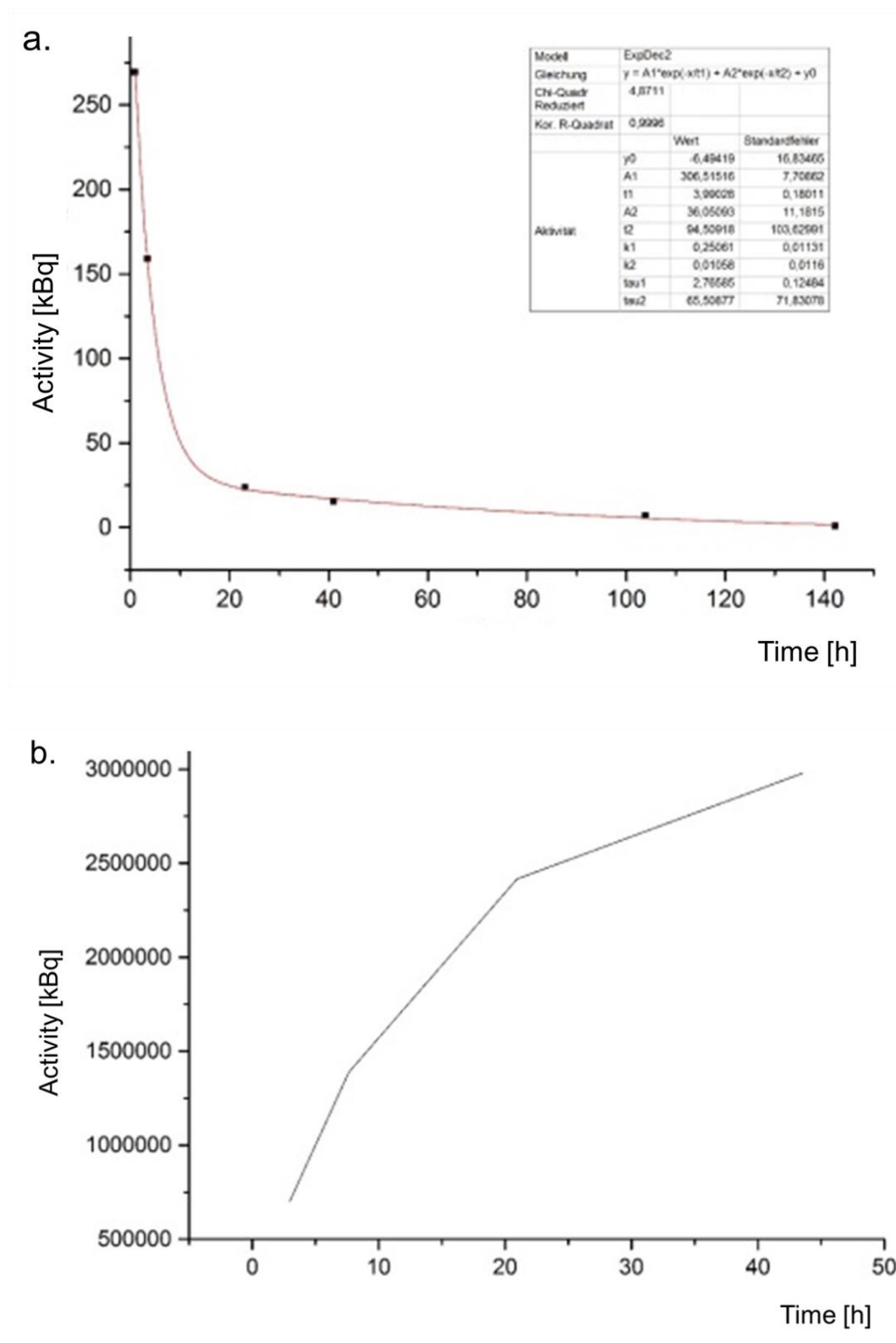
co-registration / motion correction for each time point. A circular ROI was drawn around the reference standard for each time point. The calibration factor was used to calculate the fraction of the injected dose (%ID) for each segmented ROI, which were used to generate time-activity-curves (TACs). The area-under-the-curve of that TAC was merged from three segmental phases: From 0 to first time point a linearly increasing activity was assumed. From first to last time point the best approximation of either a mono- or bi-exponential fit (using MATLAB code developed based on the curve fitting toolbox in MATLAB R2014b) was derived for all organs except salivary glands. The salivary glands and tumor lesions were integrated numerically using trapezoidal approximations (Supplement-Figure-1). From the last time point to infinity the area-under-the-curve was integrated using the fitted mono- or bi-exponential function. The cumulated activity was divided by the administered activity to achieve a normalized “residence time”. The TAC obtained from blood samples were used to calculate the perfusion/diffusion dependent residence time for red marrow using a previously described method that is valid for radiopharmaceuticals without specific binding to the red marrow (1). The residence times of kidney, liver, spleen, bladder, red marrow and WB-remainder were used as source organs in OLINDA/EXM (2) to calculate radiation doses for the adult male model using organ masses of the original phantom. The spherical model of OLINDA was used to calculate the doses for the salivary glands for individual gland volumes derived from PET/CT images acquired before the first treatment cycle. The reported tumor absorbed dose presents the average of 6 metastases per patient; in baseline PET/CT their volume was approx. 2ml (1-3 ml), no actual tumor volume was available during cycle-2. Background correction was not performed, making a slight overestimate of tumor absorbed doses possible.

The WB-remainder as a source organ in OLINDA is derived from the whole body ROI minus dedicated organ ROIs and thus includes also tumor uptake. Nevertheless, the geometric position of tumor lesions is neglected. To estimate the systematical error that might be introduced to the red marrow dose due to the lack of tumor as an dedicated source organ in OLINDA, we calculated the residency time of a huge abdominal LN bulk and did not attribute it to the “remainder body” but considered the activity to be localized in the pancreas. The red marrow dose was re-calculated using the new source organs.

1. Sgouros G. Bone marrow dosimetry for radioimmunotherapy: theoretical considerations. J Nucl Med. 1993 Apr;34(4):689-94.
2. Stabin MG, Sparks RB, Crowe E. OLINDA/EXM: the second-generation personal computer software for internal dose assessment in nuclear medicine. J Nucl Med. 2005 Jun;46(6):1023-7.



Supplemental Figure-1: (a) Depth-independent geometric-mean images of planar whole-body scans performed 0.5 – 120 h after injection of 6 GBq ^{177}Lu -PSMA-617 (gray scale normalized for decay by a standard activity placed between the legs). (b) Time-activity-curves derived from same sized regions-of-interest drawn to a sternal bone metastasis (dotted line) and left kidney (dashed line). The area under the curve correlates to the absorbed dose, respectively



Supplemental Figure-2: (a) Time-Activity-Curve derived from blood sampling during equilibrium between extra-cellular fluids and serum. Clearance was bi-phasic with half-lives of 4h and 90h. (b) Cumulative urine clearance trends to reach a plateau approximately 50h p.i.