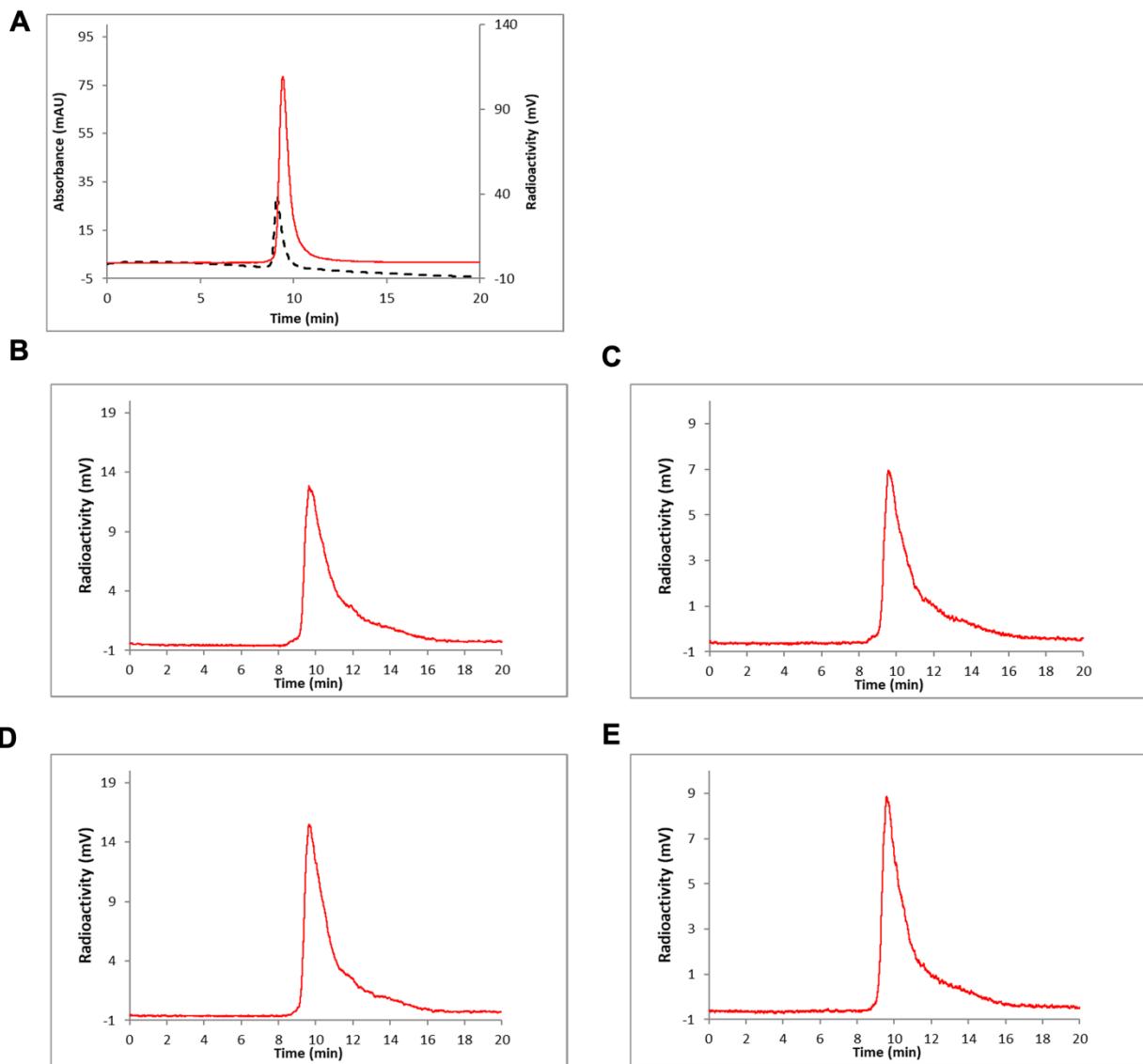


Supplementary Information File

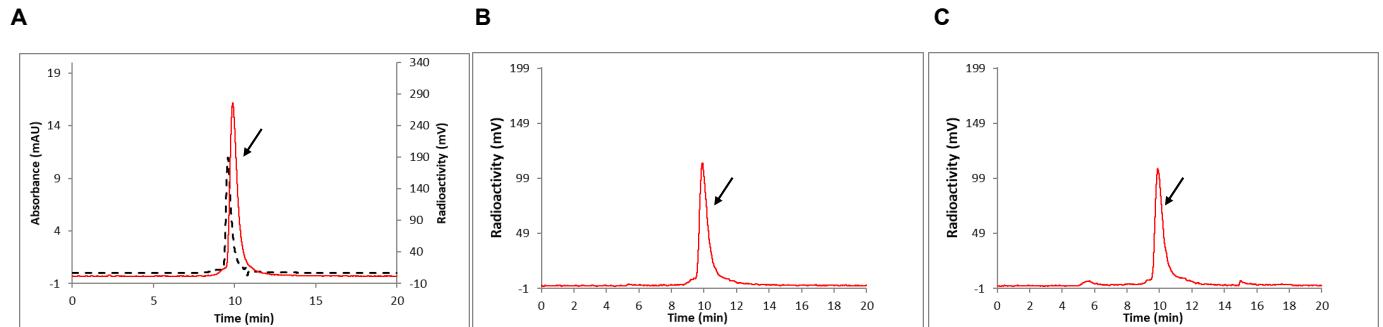
Precision Radiolabeled B-Cell Maturation Nanobody for Targeted PET Imaging and Radioligand Therapy of Disseminated Multiple Myeloma

Behnaz Ghaemi¹, Colleen P. Olkowski¹, Falguni Basuli², Jianfeng Shi², Ryan Young³, Dickran Kazandjian⁴, Ola Landgren⁴, Elizabeth Hill³, Peter L. Choyke¹ and Orit Jacobson^{1*}

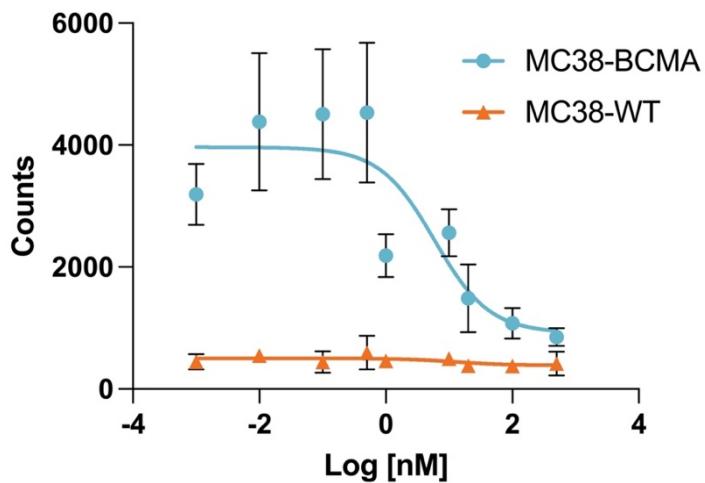
¹*Molecular Imaging Branch, Center for Cancer Research, National Cancer Institute, NIH, Bethesda, Maryland, 20892;* ²*Chemistry and Synthesis Center, National Heart, Lung, and Blood Institute, NIH, Rockville, Maryland, 20850;* ³*Lymphoid Malignancies Branch, Center for Cancer Research, National Cancer Institute, NIH, Bethesda, Maryland, 20892;* ⁴*Sylvester Myeloma Institute, Sylvester Comprehensive Cancer Center, Department of Medicine, Myeloma Division University of Miami, Florida, 33146*



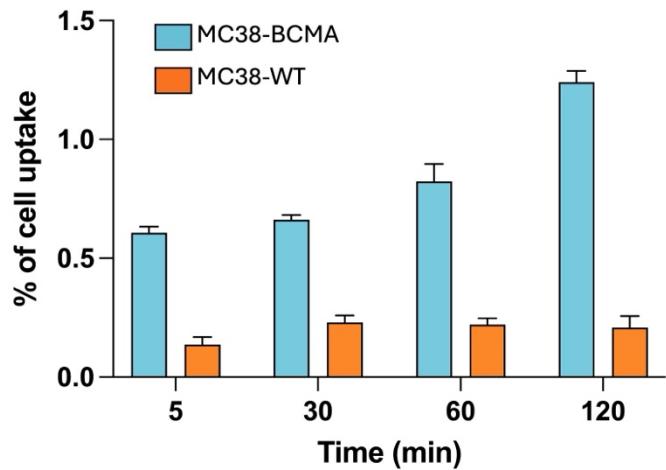
Supplemental Figure S1. HPLC chromatogram of $[^{18}\text{F}]$ F-FPy-BCMA-Nb in (A) PBS, mouse serum (B) 0 hour, (C) 3 hour; Whole human serum (D) 0 hour and (E) 3 hours.



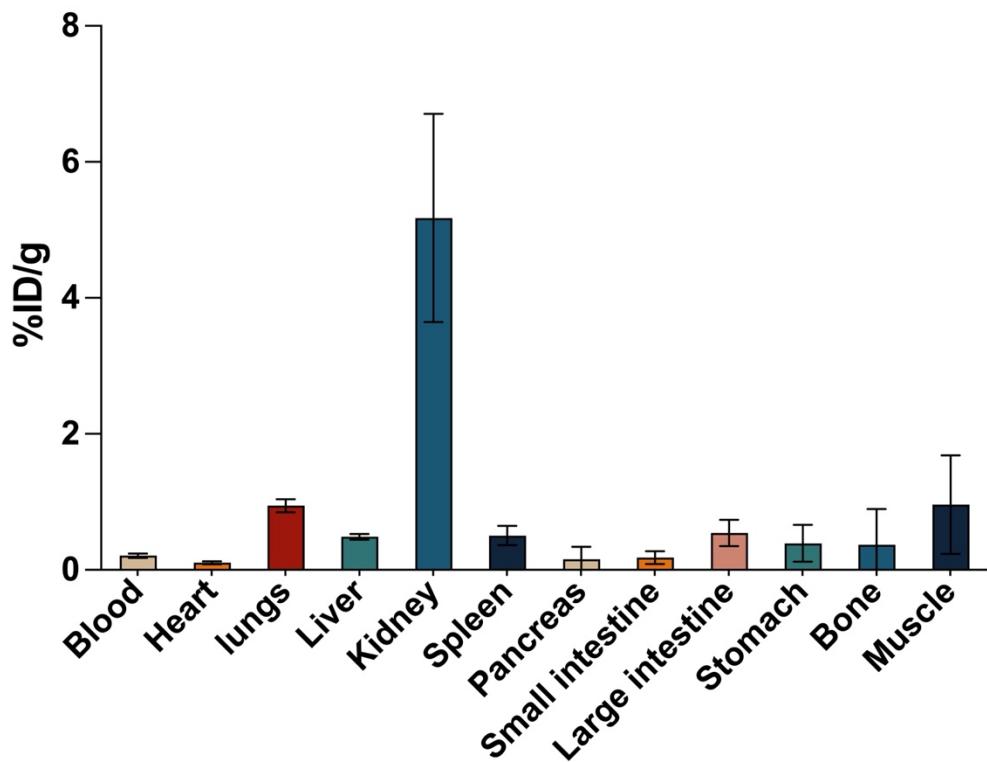
Supplemental Figure S2. (A) HPLC chromatogram of $[^{131}\text{I}]$ -BCMA-Nb in PBS and mouse serum (B) 0 hour and (C) 3 hours.



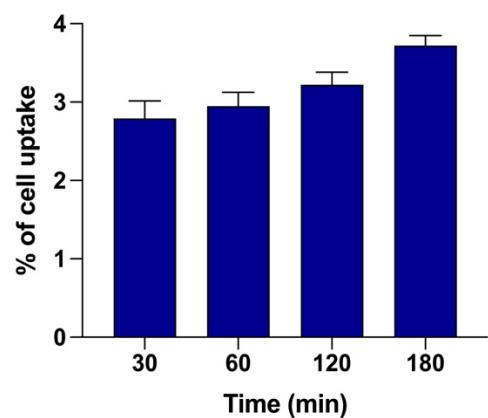
Supplemental Figure S3. Competitive cell-binding assays, $[^{18}\text{F}]$ -FPY-BCMA-Nb as competitor for 1 hour incubation time in MC38-WT and MC38-BCMA cells.



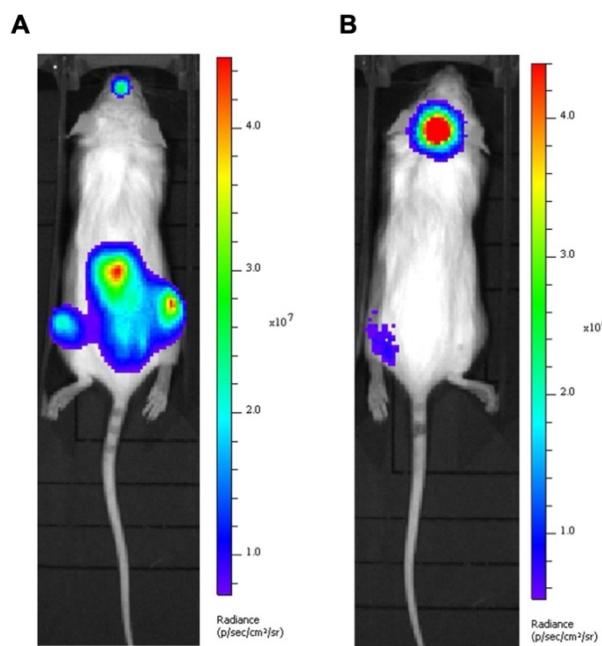
Supplemental Figure S4. Cell uptake of $[^{18}\text{F}]$ -FPY-BCMA-Nb up to 2 hours post incubation with MC38-WT and MC38-BCMA cells.



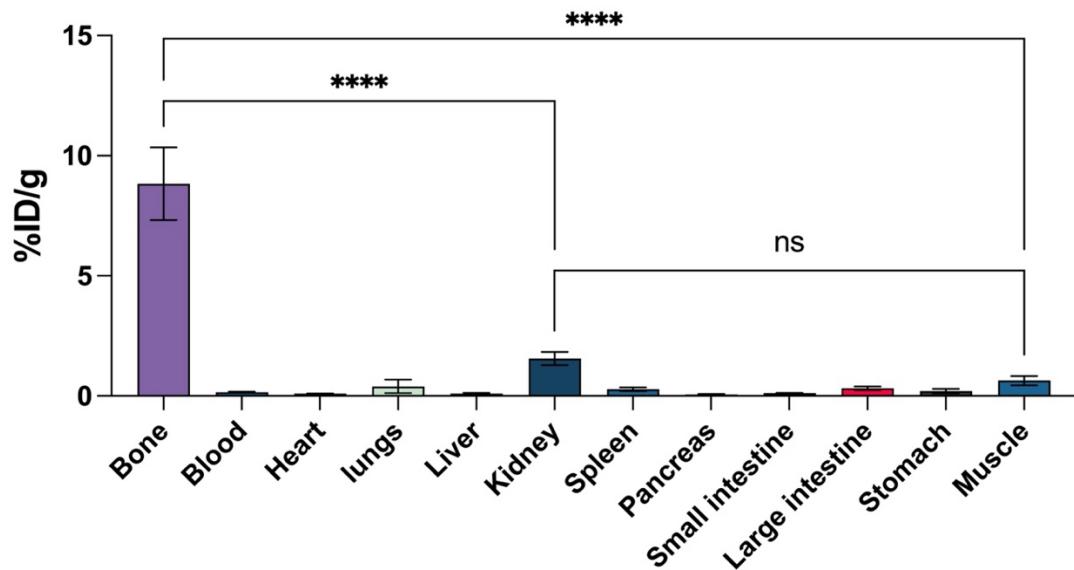
Supplemental Figure S5. Biodistribution analysis of $[^{18}\text{F}]$ -FPY-BCMA-Nb at 2 hours post-injection in athymic nude mice.



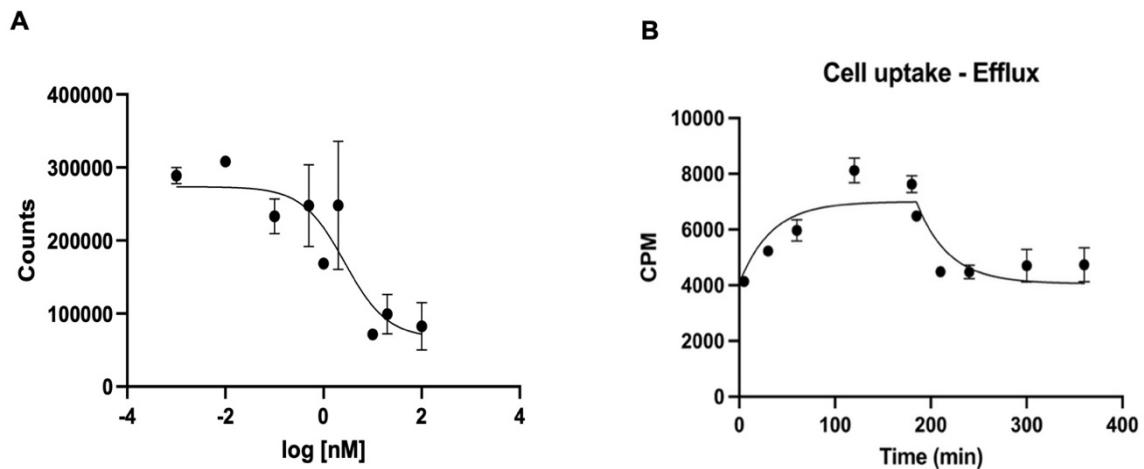
Supplemental Figure S6. Cell uptake of $[^{18}\text{F}]$ -FPY-BCMA-Nb up to 3 hours post incubation with H929 cells.



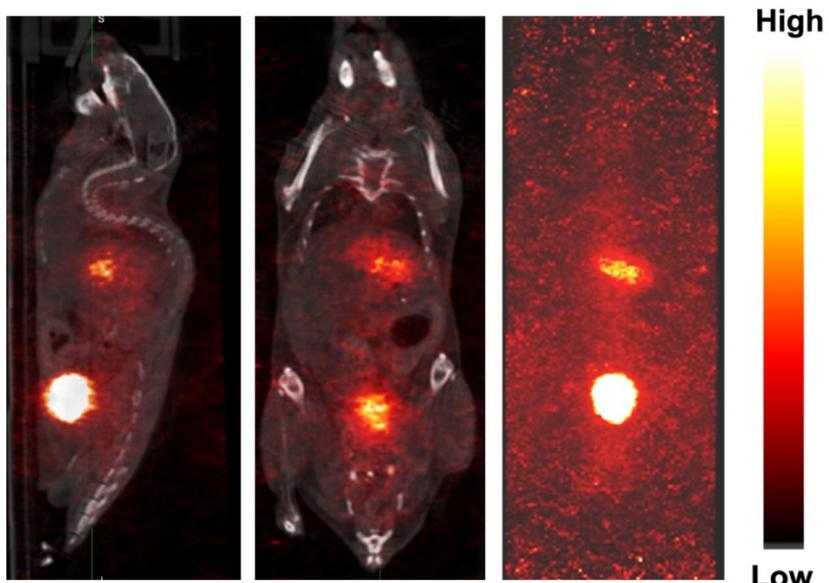
Supplemental Figure S7. Representative bioluminescence images of mouse with (A) H929 and (B) RPMI8226 tumors.



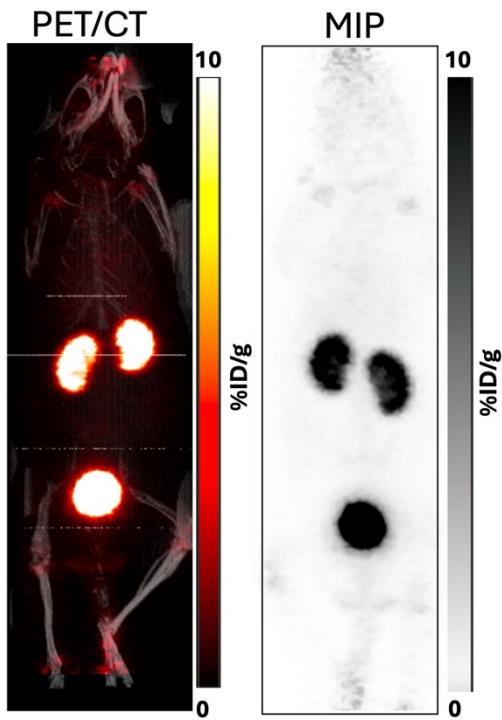
Supplemental Figure S8. Biodistribution analysis of $[^{18}\text{F}]$ F-FPY-BCMA-Nb at 3 hours post-injection in systemic MM-bearing athymic nude mice.



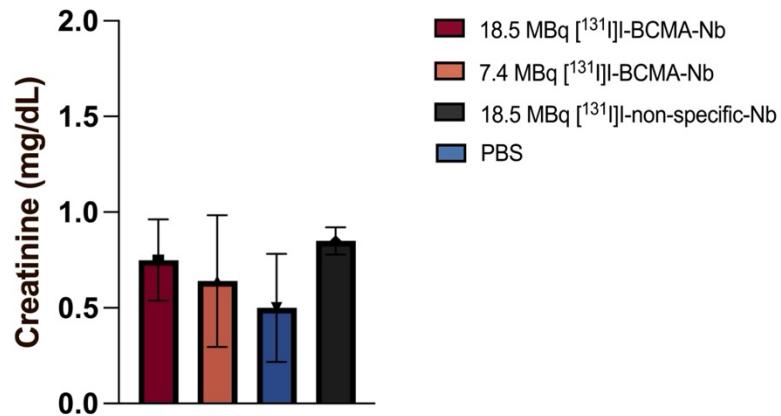
Supplemental Figure S9. (A) Competitive cell-binding assay and (B) Cell uptake and efflux studies of [^{131}I]I-BCMA-Nb in H929 cells.



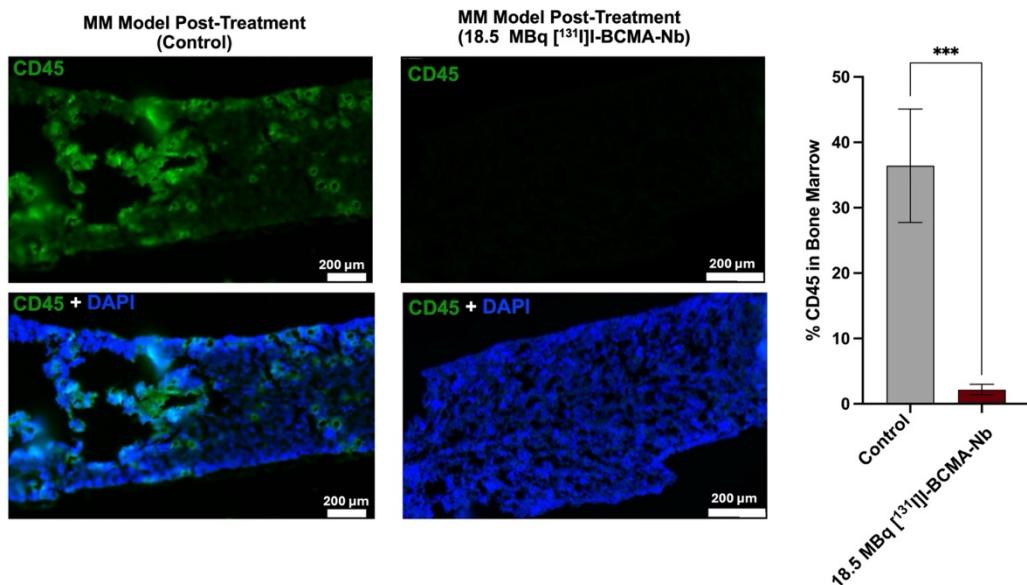
Supplemental Figure S10. Representative SPECT/CT images of a mouse with sagittal view (left), coronal view (middle) and SPECT MIP (right) of systemic H929 MM model, injected with 18.5 MBq [^{131}I]I-BCMA-Nb, highlighting radiotracer uptake in the spine, with no detectable thyroid uptake, at 3 hours post-injection.



Supplemental Figure S11. Representative PET/CT and MIP images in MM-bearing mice after treatment with 18.5 MBq [^{131}I]I-BCMA-Nb, acquired using [^{18}F]FPy-BCMA-Nb PET imaging to evaluate the presence of any residual or newly developed lesions following therapy. The scans were taken on Mediso nanoPET/CT system at 2 hours post-injection of [^{18}F]FPy-BCMA-Nb.



Supplemental Figure S12. Serum creatinine levels measured in all mice after completion of treatment across different groups. No significant differences were observed between groups, indicating the absence of treatment-related nephrotoxicity.



Supplemental Figure S13. Immunohistochemistry staining and corresponding quantitative analysis of CD45 in femoral bone marrow sections from H929 human multiple myeloma-bearing mice in the control group and after treatment with 18.5 MBq [¹³¹I]-BCMA-Nb. *** indicates $p < 0.001$.

Supplemental Table S1. Complete blood count (CBC) values measured before treatment across all groups. Data are presented as mean \pm SD.

	Pre-treatment				Unit
	PBS	7.4 MBq [^{131}I]-BCMA-Nb	18.5 MBq [^{131}I]-BCMA-Nb	18.5 MBq [^{131}I]-non-specific-Nb	
WBC Count	1.56 \pm 0.4	1.06 \pm 0.3	1.56 \pm 0.3	1.81 \pm 0.4	K/ μL
RBC Count	10 \pm 1.8	10 \pm 1.5	10.93 \pm 1.7	10.81 \pm 2.1	M/ μL
Hemoglobin	13.12 \pm 0.8	13.75 \pm 1.2	13.12 \pm 0.8	13.75 \pm 0.9	g/dL
Hematocrit	44.43 \pm 2.7	44.37 \pm 3.1	48.56 \pm 2.8	47.25 \pm 3.1	%
MCV	44.75 \pm 2.2	44.5 \pm 0.5	44.25 \pm 0.7	43.5 \pm 1.2	fL
Platelet	675	793	637.5	600	K/ μL