Supplemental Information

Noninvasive Imaging and Quantification of Radiotherapy-Induced PD-L1 Upregulation with ⁸⁹Zr-Df-atezolizumab

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Figure S1. Setup for irradiation of mice in lead shield.



Figure S2. Many lymph nodes were clearly visible on ⁸⁹Zr-Df-atezolizumab PET images.



Figure S3. Staining of spleen tissue verified high PD-L1 and immune cell presence. Magnification: 60x. Scale bar = $20 \mu m$.



Figure S4. Immunofluorescent staining of peripheral H460 tumor slices before and after fractionated radiotherapy and the spleen at 20x magnification. Scale bar = $100 \mu m$.



Figure S5. Comparison of bone uptakes of ⁸⁹Zr-Df-atezolizumab. (A) Statistically higher accumulation of the tracer was noted following fractionated radiotherapy in all bones/joints as determined through PET ROI analysis. *p<0.05; **p<0.01. (B) Statistically higher accumulation was noted in the hip and coccyx relative to the arm, particularly following radiotherapy. *p<0.05; **p<0.01, relative to accumulation in arm. (C) Similar trends were noted in ex vivo gamma-counting biodistribution studies of tracer accumulation in the femur, with higher values measured after radiotherapy. *p<0.05; **p<0.01, relative to non-irradiated femur accumulation in same tumor model. (D) Approximate location of ROIs in PET images.