Supplementary materials



Figure S1. Non-reducing SDS gel showing good association of the AC133 antibody and IR700 (A). Size exclusion chromatography showing that the unconjugated and the IR700-conjugated AC133 mAb eluted mainly as single, monomeric peaks; 10 μ g of the unconjugated and the conjugated mAb were run on an EnrichTM SEC 650 column (B).



Figure S2. Microscopic analyses demonstrating the resistance of AC133-negative WT U251 glioma cells to AC133-IR700-mediated PIT. All cultures were irradiated with NIR light. Scale bar, $50 \mu m$.



Figure S3. Therapeutic effect of a single round of AC133-IR700-mediated PIT on well-established s.c. CD133-OE U251 tumors. (A) Stability of the AC133-IR700 conjugate after incubation in mouse serum for 24, 48, or 72 h at 37 °C. Thereafter, the conjugate was diluted to assay concentrations and PIT experiments were conducted *in vitro* using CD133-OE U251 cells. As control, non-pre-incubated AC133-IR700 was mixed with mouse serum at similar concentrations immediately before PIT. Fitted curve showing the means of experiments performed in triplicates. (B) *In vivo* PIT experiments: When the tumors had reached a size of 50 mm³ (approx. 17 days after implantation), 100 µg of AC133-IR700 was injected i.v. and FMT as well as BLI were performed 24 h later. Thereafter, the tumor in the left flank, but not the one in the right flank, was irradiated with NIR light at a dose of 100 J/cm². Representative

images are shown. (C) Tumor growth delay after AC133-IR700-mediated PIT (n = 4 mice per group; **p < 0.01; two-way repeated measures ANOVA). (D) Representative photograph of a mouse 4.5 weeks after injection of AC133-IR700 followed 24 h later by NIR irradiation of the tumor in the right flank but not of that in the left flank. (E) Representative photographs of tumors harvested 4.5 weeks after PIT.