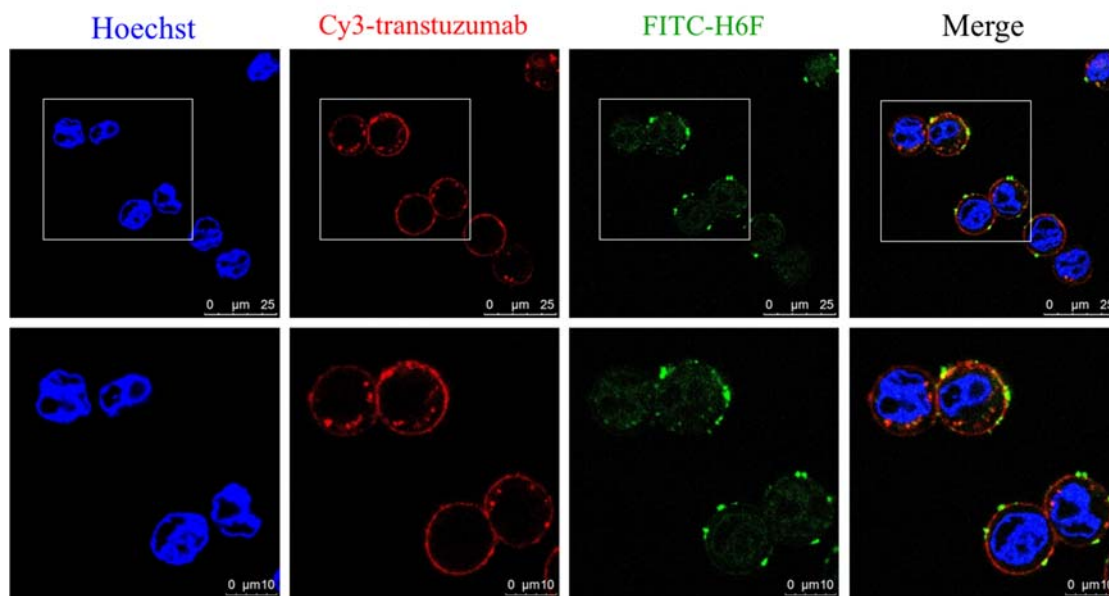
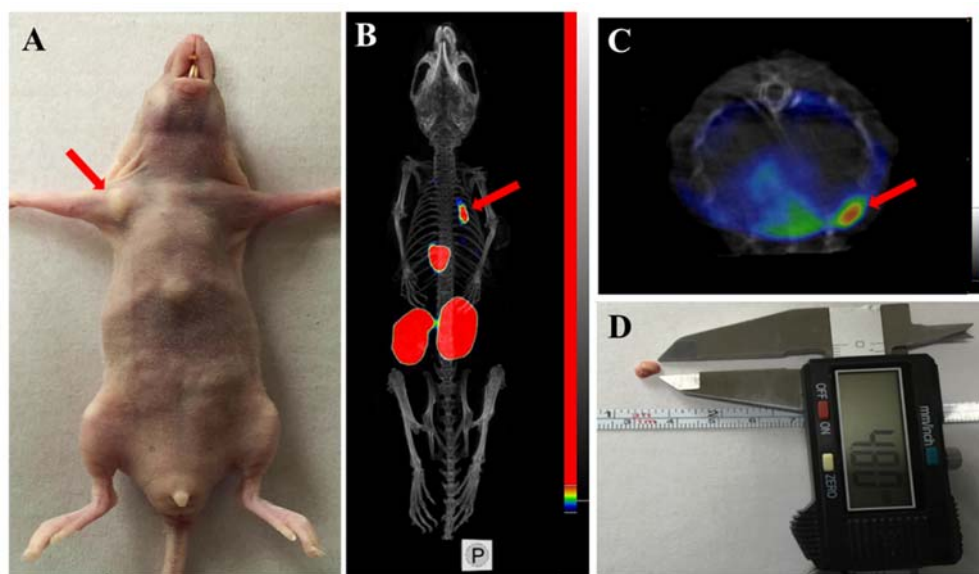


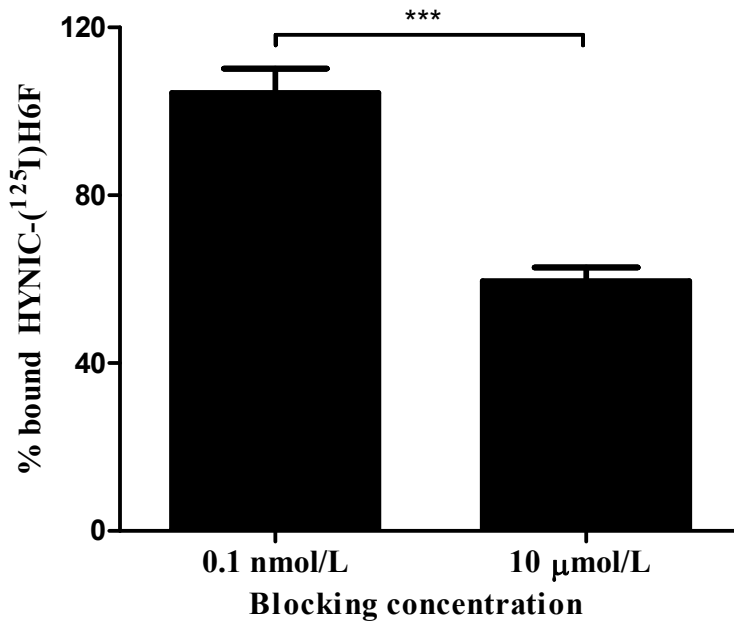
**Supplemental Figure 1.** (A) MS analysis of HYNIC-H6F. MALDI-TOF-MS:  $[M+H]^+$  m/z 1423.4 (calc. 1423.4); MW: 1422.4; yield: 68%. (B) HPLC analysis of  $^{99m}\text{Tc}$ -HYNIC-H6F after purification with Sep-Pak C18 cartridges (Waters). HPLC analysis (C18 column):  $t_R = 14.80$  min, purity > 95%. MALDI-TOF: matrix-assisted laser desorption/ionization time-of-flight. MS: mass spectrometry.



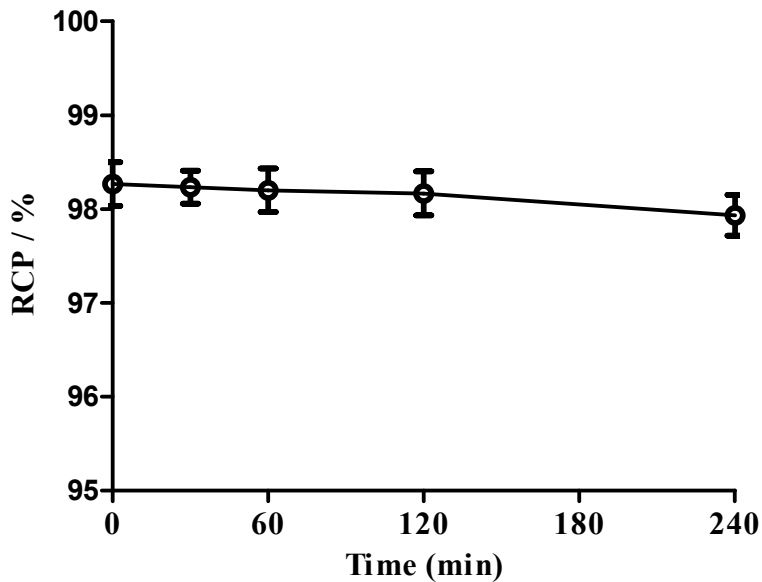
**Supplemental Figure 2.** Double fluorescent staining of MDA-MB-453 cells (HER2-positive) with Cy3-trastuzumab and FITC-H6F.



**Supplemental Figure 3.** (A, B, and C) SPECT/CT imaging of  $^{99m}\text{Tc}$ -HYNIC-H6F at 30 min p.i. in the MDA-MB-453 *in situ* tumor model. Red arrows mark the location of the MDA-MB-453 tumor on the mouse mammary fat pad. (D) The tumor size was measured by a Vernier caliper.



**Supplemental Figure 4.** Inhibition of HYNIC-(<sup>125</sup>I)H6F binding to the HER2 protein by HYNIC-H6F.



**Supplemental Figure 5.** Solution stability data for <sup>99m</sup>Tc-HYNIC-H6F in 25 mM phosphate buffer (pH = 7.4) containing cysteine (1.0 mg/mL).