Supplementary figure 1: ESI-Q-ToF-MS analysis of (A) untagged 2Rs15d, (B) untagged CHX-A\(^{-}\)-DTPA-2Rs15d and (C) untagged 1B4M-DTPA-2Rs15d. The reaction of CHX-A\(^{-}\)-DTPA to untagged 2Rs15d revealed a mixture of 1, 2 and 3 DTPA conjugated to untagged 2Rs15d. Using 1B4M-DTPA, a mixture of 2 and 3 DTPA to 2Rs15d was observed. The dominant conjugation ratio (chelator:nanobody) for both 1B4M-DTPA and CHX-A\(^{-}\)-DTPA to untagged 2Rs15d is 2:1.
Supplementary Figure 2: (Radio-)chromatographic analyses of Trastuzumab conjugates. (A) unconjugated Trastuzumab, (B) 1B4M-DTPA-Trastuzumab, (C) $^{177}$Lu-DTPA-Trastuzumab; (A,B) SEC on Superdex 75 10/30, (B) radio-SEC on Superdex 75 5/150GL; The R-times of the major peaks are shown in each graph.
Supplementary figure 3: Accumulation of radioactivity in kidneys in healthy Wistar rats (n=3 per condition) in function of time, after injecting $^{111}$In-labeled anti-HER2 nanobodies and gamma camera dynamic scintigraphy. (A) nanobody 2Rb17c, (B) nanobody 1R136d.