Detection of irradiation-induced, membrane heat shock protein 70 (Hsp70) in mouse tumors using Hsp70 Fab fragment.

Abstract: The major stress-inducible heat shock protein 70 (Hsp70) is frequently overexpressed in highly aggressive tumors, and elevated intracellular Hsp70 levels mediate protection against apoptosis. Following therapeutic intervention, such as ionizing irradiation, translocation of cytosolic Hsp70 to the plasma membrane is selectively increased in tumor cells and therefore, membrane Hsp70 might serve as a therapy-inducible, tumor-specific target structure. Based on the IgG1 mouse monoclonal antibody (mAb) cmHsp70.1, we produced the Hsp70-specific recombinant Fab fragment (Hsp70 Fab), as an imaging tool for the detection of membrane Hsp70 positive tumor cells in vitro and in vivo. The binding characteristics of Hsp70 Fab towards mouse colon (CT26) and pancreatic (1048) carcinoma cells at 4 °C were comparable to that of cmHsp70.1 mAb, as determined by flow cytometry. Following a temperature shift to 37 °C, Hsp70 Fab rapidly translocates into subcellular vesicles of mouse tumor cells. Furthermore, in tumor-bearing mice Cy5.5-conjugated Hsp70 Fab, but not unrelated IN-1 control Fab fragment (IN-1 ctrl Fab), gradually accumulates in CT26 tumors between 12 and 55 h after i.v. injection. In summary, the Hsp70 Fab provides an innovative, low immunogenic tool for imaging of membrane Hsp70 positive tumors, in