Supplementary Materials for the Manuscript Entitled

"Monitoring Breast Tumor Lung Metastasis by U-SPECT-II/CT with an Integrin $\alpha_v\beta_3$ -Targeted Radiotracer ^{99m}Tc-3P-RGD₂"

By

Yang Zhou¹, Guoqiang Shao^{1,2}, and Shuang Liu¹*

¹School of Health Sciences, Purdue University, IN 47907, USA

²Department of Nuclear Medicine, Nanjing First Hospital, Nanjing Medical University, Nanjing, China

*To whom correspondence should be addressed. School of Health Sciences, Purdue University, 550 Stadium Mall Drive, West Lafayette, IN 47907. Phone: 765-494-0236; Fax 765-496-1377; Email: <u>liu100@ purdue.edu</u>

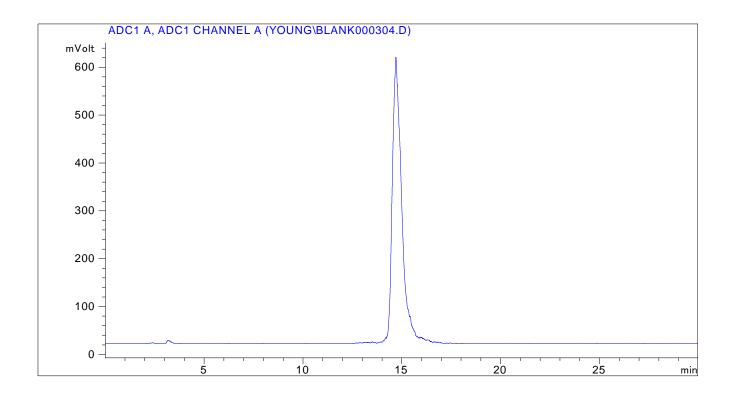


Figure SI1. Typical radio-HPLC chromatogram of ^{99m}Tc-3P-RGD₂ to illustrate its radiochemical purity.

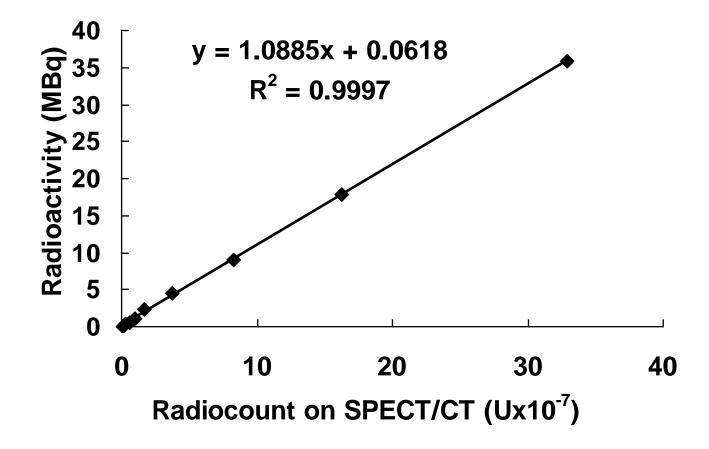


Figure SI2. Relationship between the radioactivity counts from the SPECT/CT fusion images and those from γ -counter. Obviously, there was a linear relationship with R² being 0.9997. The conversion formula was y = 1.0885x + 0.0618, where y represents the radioactivity (MBq) from γ -counter, and x is the radioactivity (U×10⁻⁷) from the SPECT/CT.

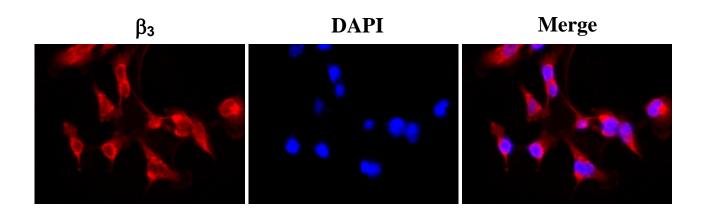


Figure SI3. Integrin β 3 immunostaining of MDA-MB-231 human breast cancer cells. MDA-MB-231 cells were seeded into 8-well chamber slides. Cells were allowed to attach and spread for >24 h. The tumor cells were fixed in -20 °C methanol for 5 min and rinsed with PBS. Cells were blocked with 10% goat serum for 30 min at room temperature, and then incubated with hamster anti-integrin β_3 antibody (1:100, BD Biosciences, San Jose, CA) for 1 h at room temperature followed by incubation with Cy3-conjugated goat anti-hamster secondary antibody (1:100, Jackson ImmunoResearch Inc., West Grove, PA). After washing with PBS, the slides were mounted with Dapi-Fluoromount-G (SouthernBiotech, Birmingham, AL). Primary antibody replaced by PBS only was served as the negative control. Fluorescence was visualized with an Olympic BX51 fluorescence microscope (Olympus America Inc., Center Valley, PA).