Supporting Information

Radiohalogenated Prostate-Specific Membrane Antigen (PSMA)-Based Ureas as Imaging Agents for Prostate Cancer

Ying Chen,[1] Catherine A. Foss,[1] Youngjoo Byun,[1] Sridhar Nimmagadda,[1] Mrudula Pullambhatla,[1] James J. Fox,[1] Mark Castanares,[2] Shawn E. Lupold,[3] Alan P. Kozikowski,[4] John W. Babich,[5] Ronnie C. Mease,[1] Martin G. Pomper[1,2]

[1]Russell H. Morgan Department of Radiology and Radiological Sciences, [2]Department of Pharmacology & Molecular Sciences, [3]Department of Urology, Johns Hopkins Medical Institutions, Baltimore, MD 21231, [4]Department of Medicinal Chemistry and Pharmacognosy, University of Illinois at Chicago, IL 60612 and [5]Molecular Insight Pharmaceuticals, Inc., Cambridge, MA 02142

Corresponding Author: Martin G. Pomper, M.D., Ph.D.
Johns Hopkins Medical Institutions
1550 Orleans Street, 492 CRB II
Baltimore, MD 21231

410-955-2789 (T)
443-956-5055 (F)
mpomper@jhmi.edu

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Methods of Molecular Modeling. Molecular modeling studies were performed using Discovery Studio Programs (Version 2.0) from Accelrys Inc. (San Diego, CA) and figures were generated using DS2.0 or PyMOL programs.

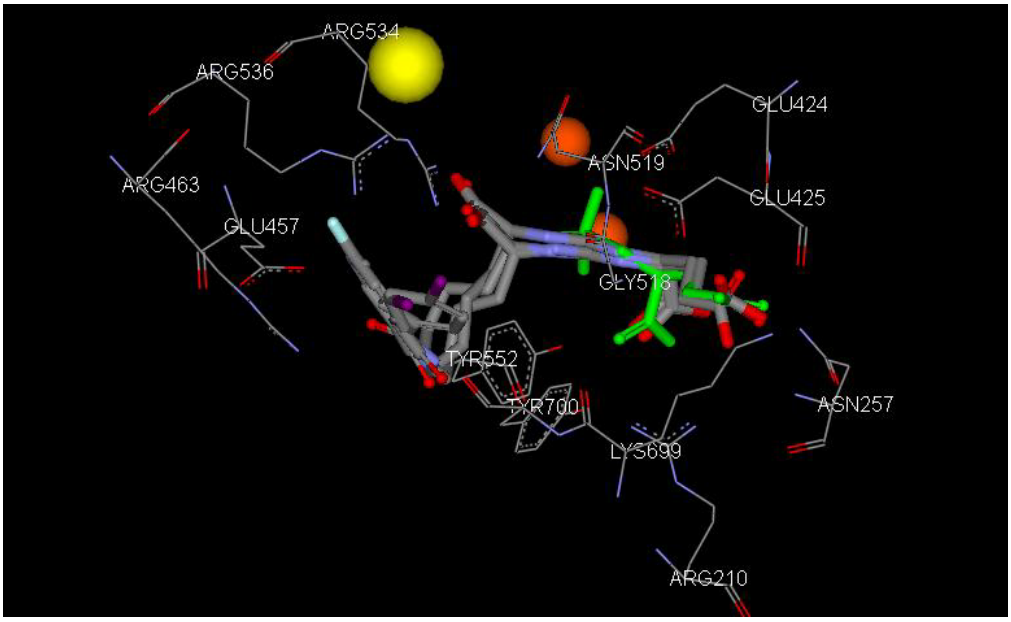
Protein Preparation. The 3-D coordinates of GCPII for docking studies were prepared as GCPII crystal structures in complex with 2-PMPA (PDB ID: 2PVW) or compound **3** (PDB ID:3D7H) through a clean-up process implemented in Discovery Studio 2.0 (DS 2.0), which can correct for structural disorder, fix bond order and connectivity of amino acid residues. The CHARMm forcefield that was applied to the protein and the binding site for docking studies was obtained through an automated method by using the option of finding sites from receptor cavities. Two zinc ions and one chloride ion in the active site were typed as Zn^{2+} and Cl^{-}, with formal charges of +2 and -1, respectively.

Ligand Preparation. All of the 2-D chemical structures were drawn with ChemDraw v10.0 (CambridgeSoft Corp. Cambridge, MA). The corresponding 3D structures were generated using the Corina program (http://www.mol-net.de) and the coordinates were transferred into DS 2.0 for docking studies.

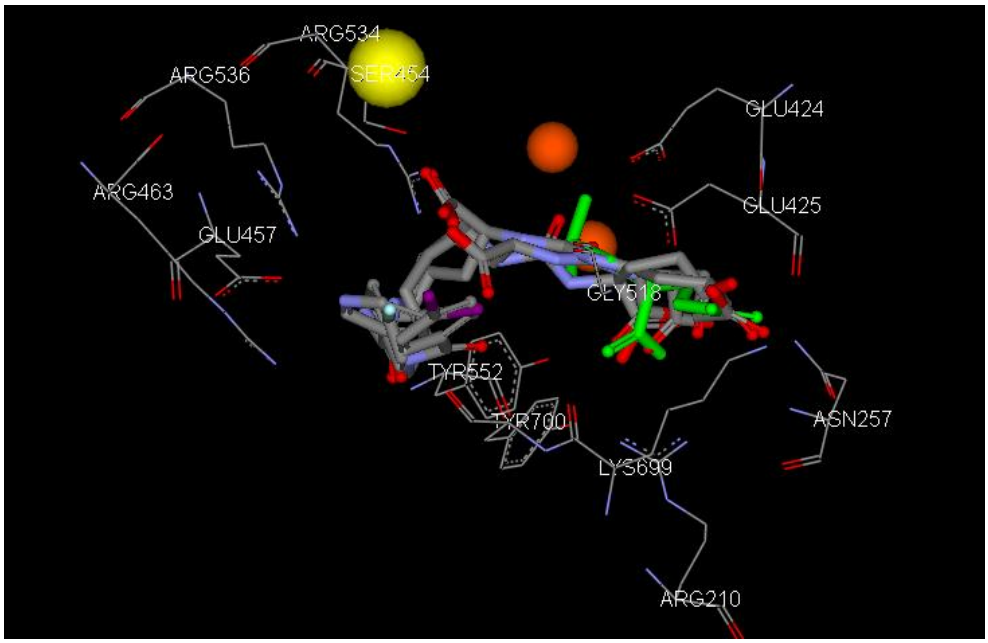
Docking Studies with CDOCKER. Docking studies of compounds **3**, **6** and **8** were performed with two conformers of 2PVW using the CDOCKER module implemented in DS 2.0 by modifying the default settings (Top hits: 20, random conformations: 20, random conformation dynamics steps: 1000, random conformations dynamics target temperature: 1000, orientation to refine: 20, maximum bad orientations: 800, orientation vdW energy threshold: 300, simulation heating steps: 2000, heating target temperature: 700, cooling steps: 5000, cooling target temperature: 300, Grid extension: 8, ligand partial charge: CHARMm). The best pose of each ligand with high CDOCKER energy was used for generating the overlay structures (Supporting Figures 1

and 2) with crystal ligand **3** from GCPII complex (PDB ID: 3D7H). Seven water molecules within 3Å from crystal ligand **3** were included for docking studies with 3D7H.

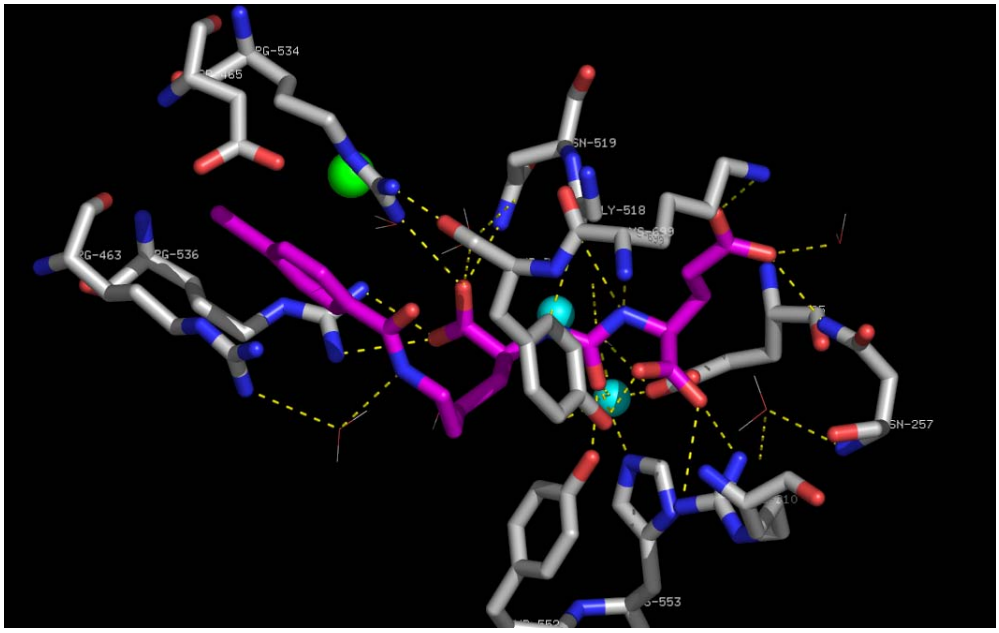
High-Performance Liquid Chromatography (HPLC). HPLC analyses of compounds **3**, **6** and **8** were performed with an Econosphere C18 column (10μ, 250 mm × 10 mm) eluted with CH_3CN/H_2O/TFA or $MeOH/H_2O$/TFA systems. Compounds were detected at UV 254 nm.



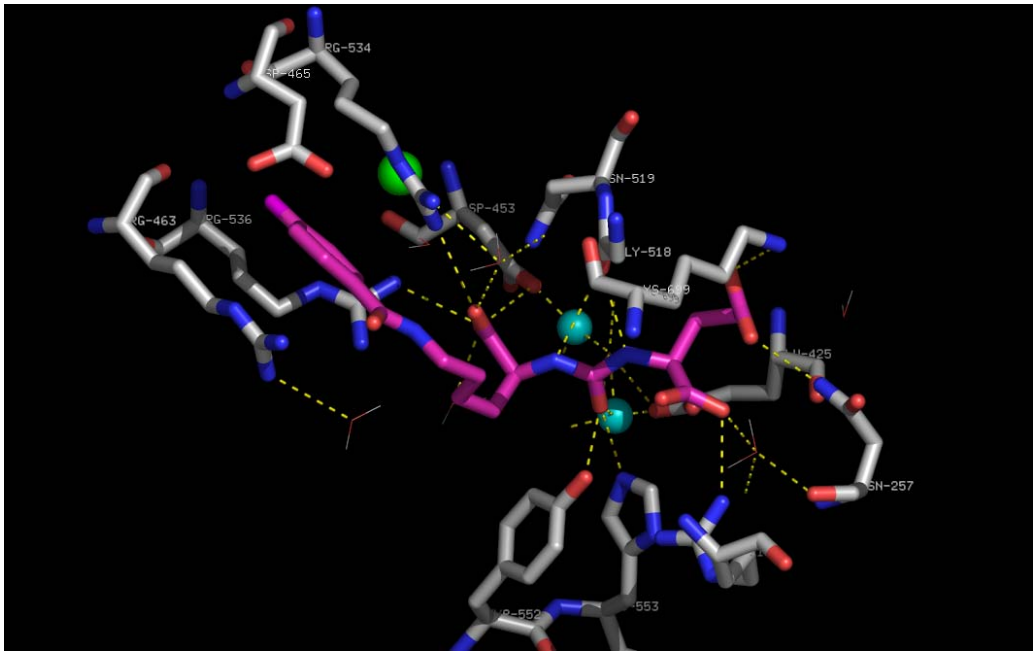
Supporting Figure 1. Overlay of **3**, **6** and **8** with 2-PMPA in the binding conformer mode of PSMA (PDB ID:2PVW).



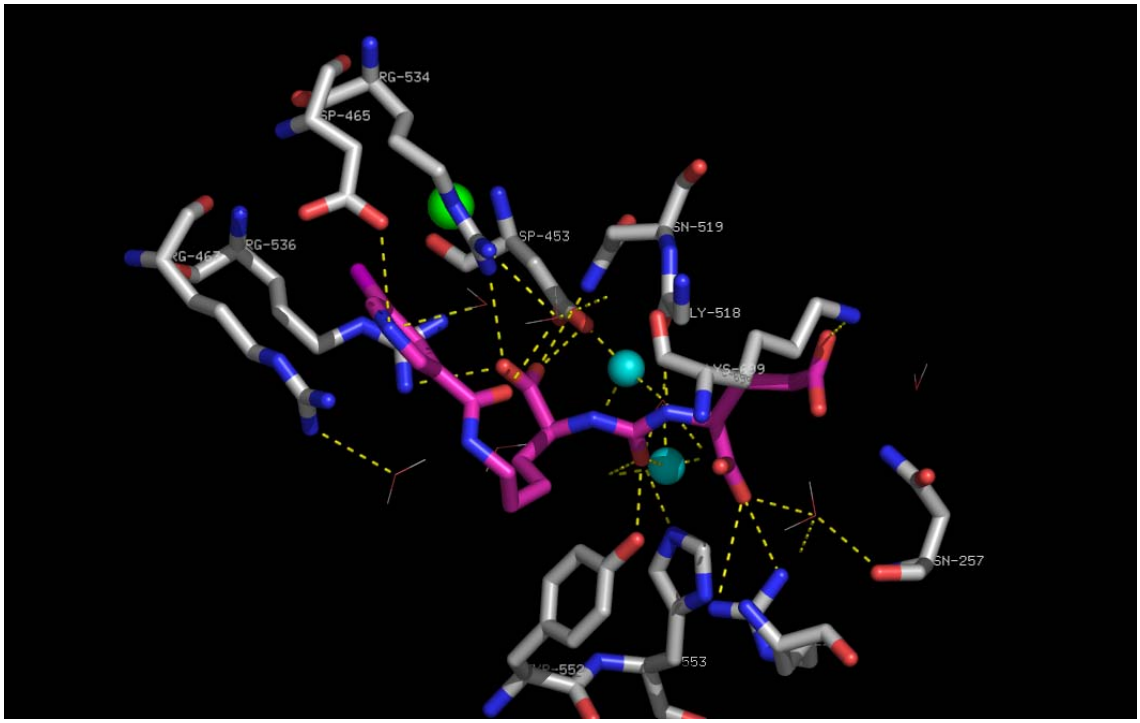
Supporting Figure 2. Overlay of **3**, **6** and **8** with 2-PMPA in the stacking conformer mode of PSMA (PDB ID:2PVW).



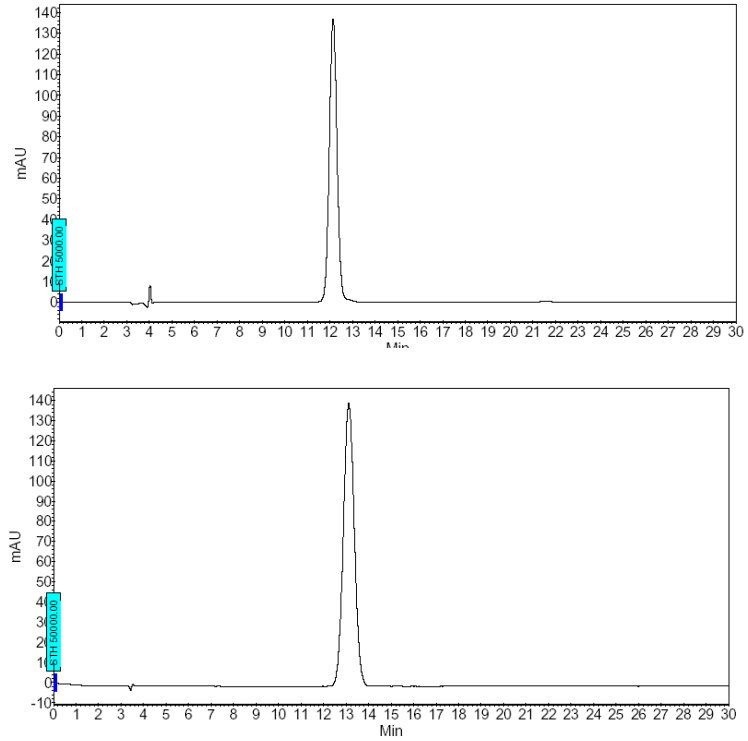
Supporting Figure 3. Crystal ligand **3** with PSMA. Zinc (cyan sphere), chloride (green sphere), dotted yellow lines (polar interaction range), and water molecules (lines) are shown.



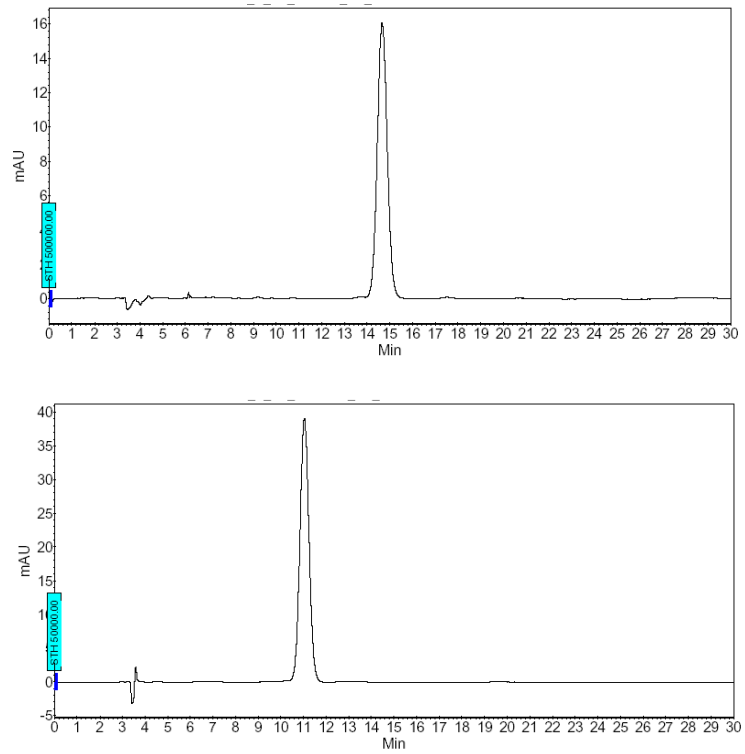
Supporting Figure 4. The best pose of **6** with 3D7H. Zinc (cyan sphere), chloride (green sphere), dotted yellow lines (polar interaction range), and water molecules (lines) are shown.



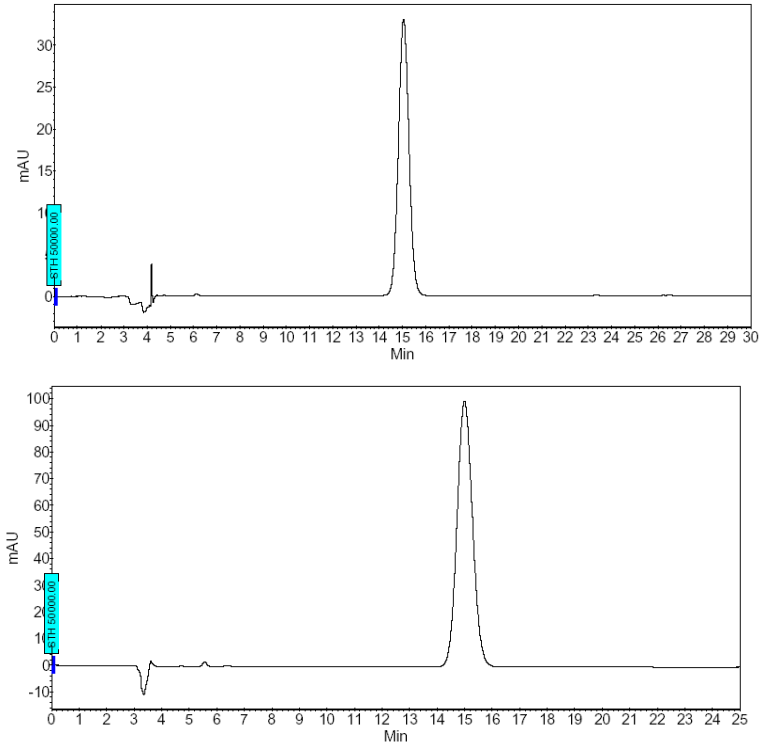
Supporting Figure 5. The best pose of **8** with 3D7H. Zinc (cyan sphere), chloride (green sphere), dotted yellow lines (polar interaction range), and water molecules (lines) are shown.



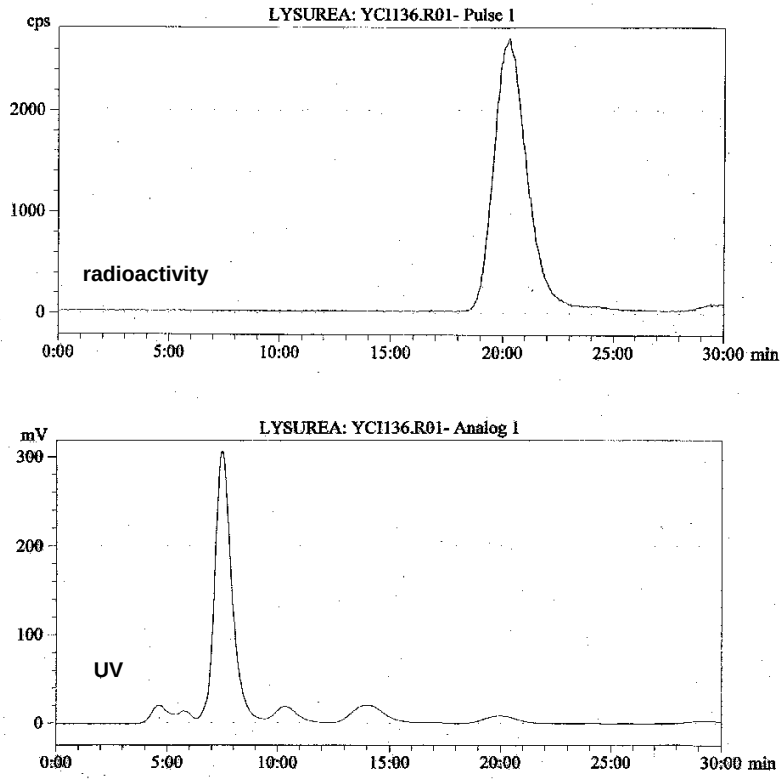
Supporting Figure 6. HPLC Analysis of compound **3**. Top: CH_3CN/H_2O/TFA = 25/75/0.1, 4 mL/min; bottom: MeOH/H_2O/TFA = 40/60/0.1, 4 mL/min.



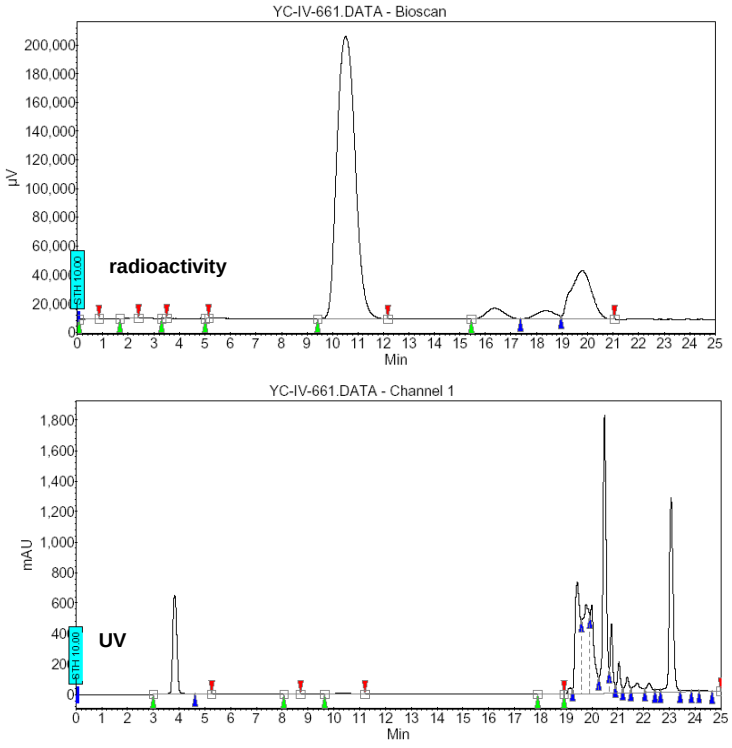
Supporting Figure 7. HPLC Analysis of compound **6**. Top: CH_3CN/H_2O/TFA = 15/85/0.1, 4 mL/min; bottom: MeOH/H_2O/TFA = 30/70/0.1, 4 mL/min.



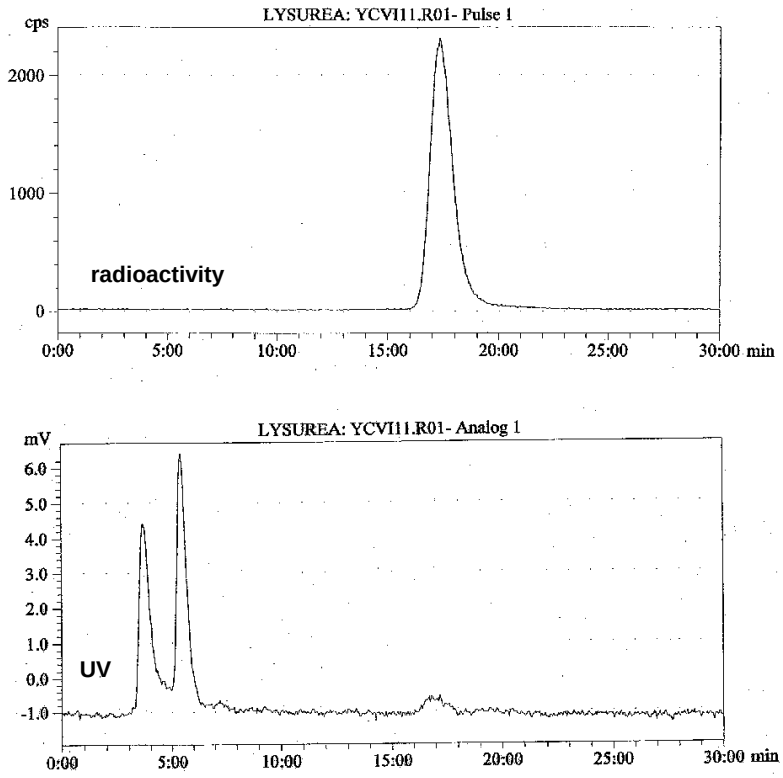
Supporting Figure 8. HPLC Analysis of compound **8**. Top: CH_3CN/H_2O/TFA = 15/85/0.1, 4 mL/min; bottom: MeOH/H_2O/TFA = 30/70/0.1, 4 mL/min.



Supporting Figure 9. HPLC separation of [^{125}I]**3**. Top: radioactive detector,bottom: UV 254 nm detector ([^{125}I]**3** retention time = 20 min)



Supporting Figure 10. HPLC separation of [^{18}F]**6**. Top: radioactive detector; bottom: UV 254 nm detector ([^{18}F]**6** retention time = 10.5 min)



Supporting Figure 11. HPLC separation of [^{125}I]**8**. Top: radioactive detector; bottom: UV 254 nm detector ([^{125}I]**8** retention time = 17 min)