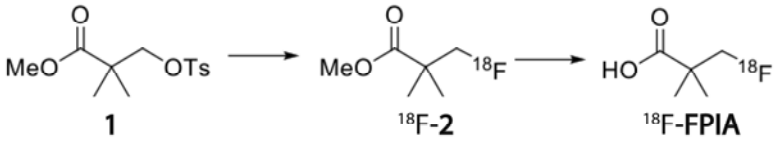
SUPPLEMENTAL METHODS

Radiosynthesis

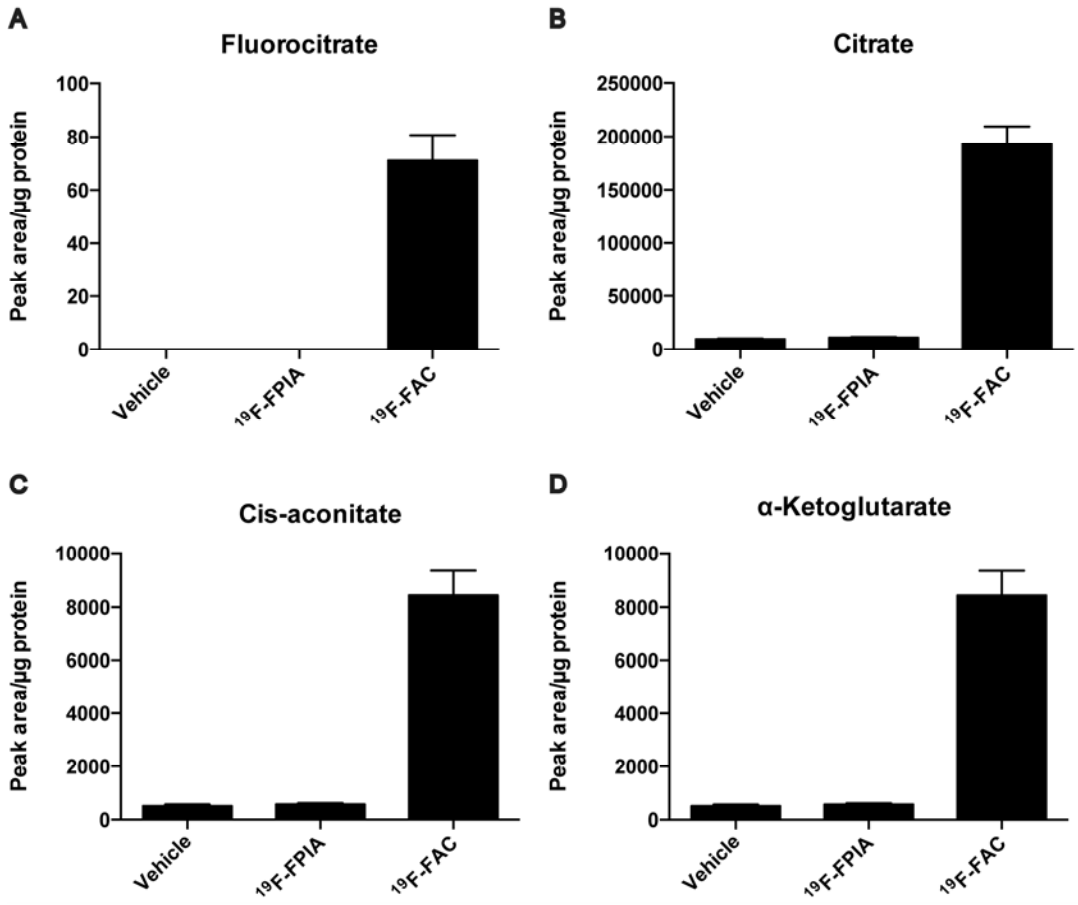
The radiosynthesis of ^{18}F-FPIA was automatically performed on a Siemens Explora RN + LC platform. Aqueous ^{18}F-fluoride was trapped into a QMA cartridge preconditioned with water (1 mL) and eluted into a 5-mL Wheaton vial with $KHCO_3$ (100 μL of a 12 mg/mL stock solution in water) and K222 (400 μL of a 18 mg/mL stock solution in water). The fluoride was dried at 105°C, and an azeotrope of MeCN (0.5 mL × 2) was used to aid drying. Precursor **1** (8 mg) in DMF (450 μL) was added, and the reaction mixture was heated at 120°C for 10 min and then cooled to 30°C. The reaction mixture was quenched with water (4 mL), and labeled intermediate ^{18}F-**2** was isolated by semipreparative HPLC (Phenomenex Gemini 5μ C18 110A [100 × 10 mm, 5 micron] column, isocratic 20% EtOH/water method, flow rate of 5 mL/min, retention time [rt] of 10 min). NaOH (1 M, 200 μL) was added and the mixture heated at 60°C for 5 min and then cooled to 45°C. Ethanol was removed at 45°C under a vacuum for 30 min, and the mixture was neutralized with HCl (1 M, ~200 μL).



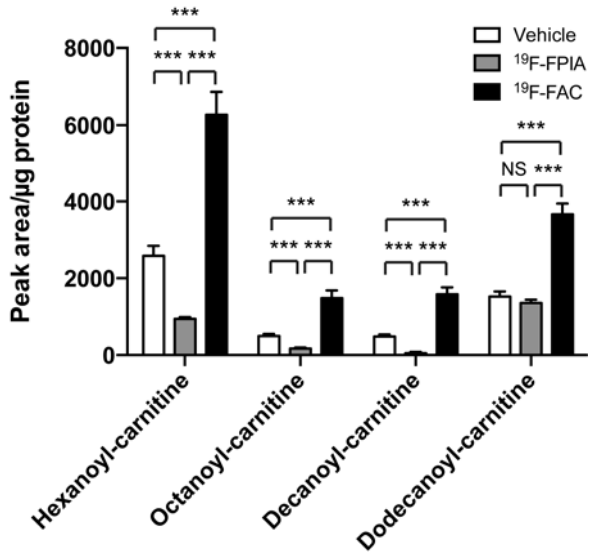
Dynamic PET imaging studies

Dynamic ^{18}F-FPIA imaging scans were obtained on a dedicated small-animal PET/SPECT/CT scanner (Inveon [Siemens Medical Solutions USA, Inc.]; matrix size, 256 × 256 × 159) following a bolus intravenous injection of ~3.7 MBq of the radiotracer into tumor-bearing mice. Dynamic scans were acquired in list mode format over 60 min.

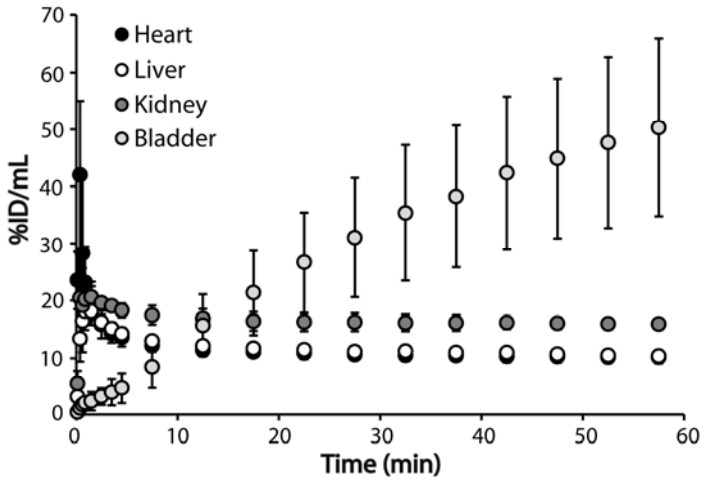
The acquired data were then sorted into 0.5-mm sinogram bins and 19 time frames for image reconstruction (4 × 15 s, 4 × 60 s, and 11 × 300 s), which was done by iterative reconstruction (2-dimensional ordered-subsets expectation maximization). The Inveon Research Workplace software was used for visualization of radiotracer uptake in the tumor; 30- to 60-min cumulative images of the dynamic data were used to define 3-dimensional volumes of interest. The count densities were averaged for all volumes of interest at each time point to obtain a time versus radioactivity curve (TAC). Tumor TACs were normalized to injected dose, measured by a VDC-304 dose calibrator (Veenstra Instruments), and expressed as percentage injected dose per milliliter of tissue. The area under the TAC, calculated as the integral of %ID/mL from 0 to 60 min, and the normalized uptake of radiotracer at 60 min ($\%ID/mL_{60}$) were also used for comparisons.



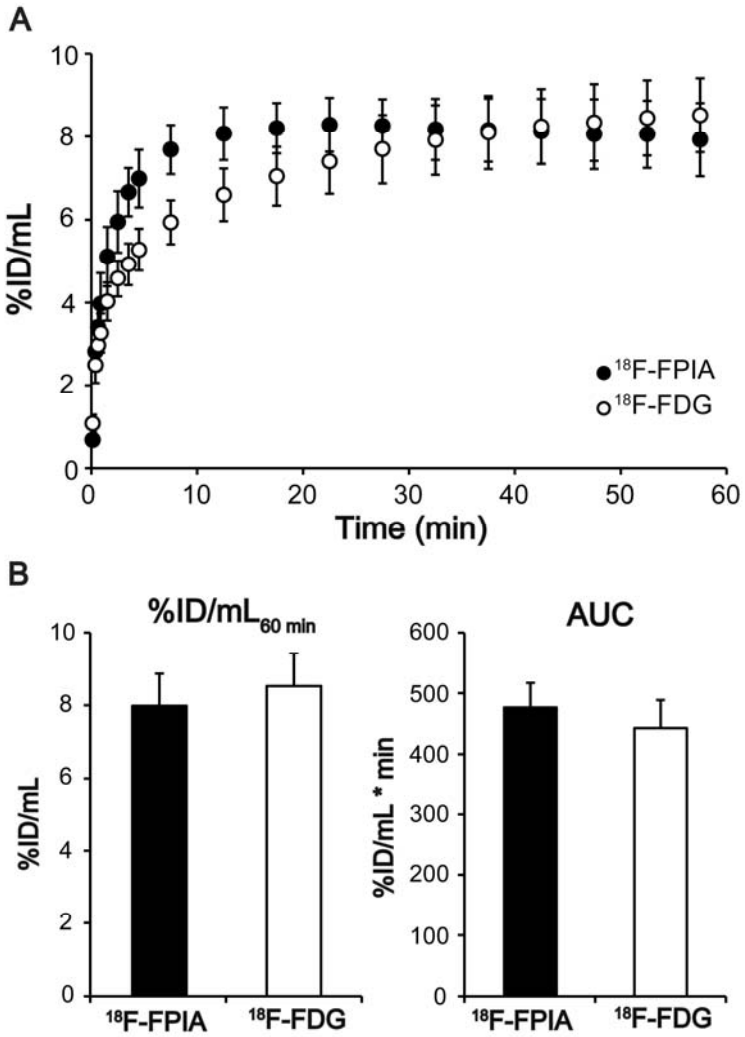
SUPPLEMENTAL FIGURE 1. Effect of exogenous ^{19}F-FPIA and ^{19}F-FAC on intracellular metabolite concentrations of fluorocitrate (A), citrate (B), cis-aconitate (C), and α-ketoglutarate pools (D) as analyzed by liquid chromatography–mass spectrometry. Data are mean ± SD (n = 4).



SUPPLEMENTAL FIGURE 2. Effect of exogenous ^{19}F-FPIA and ^{19}F-FAC on intracellular metabolite concentrations of medium-chain acyl-carnitine esters (6–10 carbon length). Data are mean ± SD (n = 4). ***P < 0.001. NS = not significant.



SUPPLEMENTAL FIGURE 3. ^{18}F-FPIA tissue pharmacokinetics. TACs obtained from 60-min dynamic PET imaging. Data are mean ± SD (n = 4 mice per group).



SUPPLMENTAL FIGURE 4. Dynamic ^{18}F-FPIA and ^{18}F-FDG PET image analysis in EMT6 tumors. (A) EMT6 tumor TAC obtained from 60-min dynamic PET imaging. Data are mean ± SD (*n* = 5 mice per group). (B) Semiquantitative imaging variables extracted from the TAC. Data are mean ± SD (*n* = 5 mice per group).