Supplementary methods and results for 2-deoxy-2-[^{18}F]fluoroglucose PET-CT studies

All animal studies and experimental procedures were approved by the local institutional animal care and use committees at Stony Brook University and conformed to both the Animal Welfare Act and Office of Laboratory Animal Welfare regulations. Female Sprague Dawley rats were used (Taconics, body weight (N=8): 274 ± 21g). The rats were fasted for ~ 2 hrs prior to PET-CT imaging.

Anesthesia and placement of cisterna magna catheters

For anesthesia, the animals were induced with 3% isofluorane in oxygen using an induction chamber and then received an intra-peritoneal injection of 40mg/kg phenobarbital (Nembutal® Sodium solution, OVATION, Pharmaceuticals, Inc., Deerfield, IL, USA). The animals were allowed to breathe spontaneously during the experiment. Non-invasive monitors (pulse-oximetry, respiratory rate and rectal temperature probe, SA Instruments, Inc., Stony Brook, NY) were placed to assure adequate oxygenation (O_2-saturation> 97%), ventilation (respiratory rate 50-65 breath per minute) and normal body temperature (36.5^{o}C-37.5^{o}C). The animals were placed in a stereotaxic frame. Under direct visualization using an operating microscope, a small polyethylene catheter was inserted into the cisterna magna (CM). The CSF catheter was secured, and the skin incision was closed with 3.0 silk.

PET-CT imaging

Following surgery, the rats were flipped supine and positioned on the scanner bed. An intra-peritoneal catheter was used for maintenance hydration (0.9% NaCl, 4cc/kg/hr) and supplemental anesthesia (Nembutal®, 5-10mg/kg I.P.) administered every ~1-2 hrs as guided by the respiratory rate. During imaging, body temperature was kept strictly within 36.5-37.5^{o}C using a computer assisted air heating system (SA Instruments, Inc., Stony Brook, NY). For rats receiving 18FDG into CSF, the CSF catheter was connected to PE 20 line filed with radioisotope and attached to a 1cc-syringe and micro-infusion pump (Baxter Model AS50 infusion Pump, Baxter Healthcare Corporation, Illinois, USA). For the CSF rats 20 µl (~0.20mCi) of 18FDG was infused at a rate of 1.6 µl/min (total infusion time: ~12.5min). For rats receiving 18FDG

intravenously (i.v.) a femoral venous catheter was placed. At the start of the PET scan, 0.5mCi 18FDG was administered as an i.v. bolus followed by a ~0.30 cc 0/9%NaCl flush.

PET data was acquired using the Inveon small-animal PET/CT scanner (Siemens Medical Solutions USA Inc). A CT image was acquired first for attenuation correction and anatomical fusion with the PET. PET acquisition commenced at injection and continued for 100 minutes for the CSF group and for 60 min for the i.v. group. Images were binned into 5 min frames and reconstructed using the 3D maximum likelihood-expectation maximization (ML-EM) iterative algorithm to reconstruct the PET images with CT based attenuation correction as well as all other quantitative corrections (scatter, randoms, detector efficiency, deadtime, decay, branching fraction) resulting in fully quantified image of radioactivity concentration, spatially registered to the CT image.

PET-CT data analysis

All 18FDG PET-CT data analysis was conducted using PMOD software (PMOD Version 3.908). The analysis for PET data obtained from the CSF group (N=5) was analyzed as followed : 1) regions of interest which included the brain and the nasal conchae were manually outlined on the CT images, 2) ROI of the CM + catheter, heart and lymph nodes were manually outlined on the summed 18FDG PET images, 3) The dynamic 18FDG PET data for each rat was normalized to 18F activity in the CM/CSF area and 4) the anatomical ROI were overlaid on the normalized dynamic 18FDG images and the time activity curves (TAC) extracted. The analysis of PET data from the IV group (N=4) was analyzed as follows: 1) regions of interest which included the brain and the nasal conchae were outlined on the CT images, 2) ROI including the heart and lymph nodes were outlined on the summed 18FDG PET images. 3) ROIs were overlaid on the dynamic 18FDG PET images and the TACs extracted. The brain and nasal conchae TACs were normalized to total uptake in whole brain and nasal conchae, respectively.