[^{18}F]FACBC and [^{18}F]FLT PET imaging for gliomas

MSKCC NON-THERAPEUTIC/DIAGNOSTIC PROTOCOL

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Please Note: A Consenting Professional must have completed the mandatory Human Subjects Education and Certification Program.

Participating Institution(s)	PI's Name	Site's Role
Columbia University	Andrew Lassman, MD	Data Analysis

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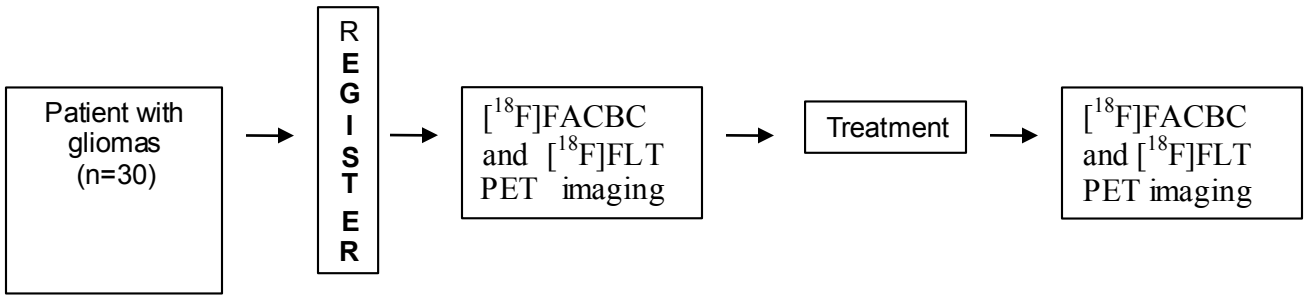
1.0 PROTOCOL SUMMARY AND/OR SCHEMA

We will perform [^{18}F]FACBC PET and [^{18}F]FLT PET of the brain on 30 patients receiving small molecule inhibitors for treatment of recurrent malignant gliomas. In this setting small molecule inhibitors includes antibodies and other agents used to target specific receptors or signal transduction cascades. We are currently using several regimens involving agents that act on critical signal transduction pathways, on and off protocol, for the treatment of recurrent gliomas. Evaluation of treatment response with conventional neuroimaging is a challenge because imaging changes seen with MRI or CT are usually seen late, 4 to 8 weeks after the initiation of treatment. Patients will agree to two [^{18}F]FACBC PET studies and two [^{18}F]FLT PET studies, one of each at baseline and one of each after treatment. In patients planned for surgery after treatment, the second set of imaging studies will be done prior to surgery.

The [^{18}F]FACBC PET and [^{18}F]FLT PET studies will be performed under the Radioactive Drug Research Committee (RDRC) guidelines as defined and established by the Food and Drug Administration (FDA). The RDRC mechanism is appropriate because this is an exploratory protocol with no intent for treatment decisions to be made based on the imaging findings. This protocol fits under the RDRC umbrella because there is a lack of pharmacologic effect of both FLT and FACBC in humans, both radiotracers have been used in humans previously, and both will be administered at below a potentially harmful radiation dose (Appendix A). We will obtain organ/tissue radiation dosimetry and assess subjects for signs of radiopharmaceutical toxicity following i.v. injection of [^{18}F]FACBC and [^{18}F]FLT.

RDRC guidelines can be found at: http://www.fda.gov/CDER/regulatory/RDRC/

SCHEMA



2.0 OBJECTIVES AND SCIENTIFIC AIMS

2.1 Determine and compare the biodistribution, clearance, and dosimetry of [^{18}F]FACBC and [^{18}F]FLT tissue/organs within the field of view of the dynamic PET imaging studies prior-to and during anti-AKT and/or anti-VEGF directed therapies alone or in combination with radiation for glioma.

2.2 Compare [^{18}F]FACBC PET and [^{18}F]FLT PET results with MRI imaging in patients with recurrent gliomas . All MRI's will be performed as part of the patient's standard clinical care.

2.3 Explore if [^{18}F]FACBC PET and [^{18}F]FLT PET imaging can be related to molecular markers (AKT, VEGFR, and related signaling/biologic changes by immunohistochemistry and/or analysis of flash frozen tissue).assessed on resected tumor samples, if the tissue samples are available from the initial and any subsequent resections.

3.0 BACKGROUND AND RATIONALE

High grade gliomas are the most common primary brain tumors in adults. Despite novel chemotherapeutic and biologic agents, prognosis has changed little in the past 30 years. Patients with glioblastoma multiforme (GBM) have an average survival of 1 year while those with anaplastic astrocytomas are expected to survive only 2 to 5 years.[1] The addition of concurrent and adjuvant temozolomide to radiation treatment is one of the rare advances but has only lengthened median survival for patients with GBM from 12 to 14 months. [2] The hope of researchers today is that a better understanding of the molecular biology of gliomas will provide clinicians with more effective therapies.

Small molecule inhibitors that target specific signal transduction and proliferation pathways have become an important component of glioma clinical trials. It is challenging to assess the impact of small molecule inhibitors in gliomas at the molecular level for a number of reasons. Many of these agents, by nature of their action are cytostatic. Interpreting response with standard anatomic imaging is therefore unsatisfactory; small molecule inhibitors could potentially act on gliomas without necessarily shrinking the size of the tumor. Current measures of efficacy are based on patient survival and not on whether the involved pathways have in fact been inhibited. However, the inherent variability in the molecular genotype of gliomas renders response rates based on clinical outcomes difficult to interpret. The only definitive way to determine if signal transduction pathways have been affected would be to obtain and analyze tissue. This is technically feasible and has been done, but is particularly difficult in the brain since tissue is infrequently obtained during treatment with a chemotherapeutic or biologic agent.[3] Imaging that can reliably assess the metabolic and functional status of brain tumors non-invasively

would be an important biomarker for the effects of small molecule inhibitors on specific signaling pathways in human brain tumors.

Tyrosine kinase inhibitors (TKI) have been the subject of intense study in treatment of malignant gliomas, particularly in light of their success in the treatment of patients with CML, GIST and a subgroup of patients with NSCLC. [4] The majority of high grade gliomas overexpress EGFR, but EGFR inhibitors are effective in only 15 to 20% of patients. In addition to receptor tyrosine kinases, downstream molecules involved in signal transduction are important potential targets in cancer treatment. Numerous agents are under development and already in clinical trials. The most important of these interfere with either the PI3K or RAS cascades. The assessment of response in these trials largely depends on conventional anatomic based MR imaging. Recently, the use of VEGFR or VEGF targeted agents, such as bevacizumab (Avastin) and VEGF-Trap, has become common in patients with recurrent [5, 6] and newly diagnosed[7] malignant gliomas. However, it remains challenging to interpret MR scans in patients treated with such agents because contrast enhancement reduction may not reflect true reduction of tumor burden.[8, 9]

The availability of a non-invasive biomarker for molecular effects in vivo would represent an important step forward in the assessment of treatment response with this class of drugs. [^{18}F]FACBC PET and [^{18}F]FLT PET imaging may serve this function because of their potential dependence on AKT signaling. The AKT effector, mTOR (mammalian target of rapamycin), is an important regulator of cell growth and proliferation. Through an unknown mechanism, mTOR senses the availability of intracellular amino acids and controls phosphorylation and activation of downstream targets.[10] A protein critical to this process in cancer cells is the L-type amino acid transporter, LAT1. In human gliomas, LAT1 expression correlates with poor survival and is also a potential target of anti-cancer agents.[11] Overexpression of LAT1 contributes to tumor growth by facilitating entry of neutral amino acids that are necessary building blocks for unchecked proliferation.[11] The increased requirement for intracellular amino acids is sensed by mTOR and leads to activation of downstream pathways. Conversely, mTOR regulates LAT1 gene expression and the trafficking of LAT1 transporter protein to the cell membrane. [12] Increased LAT1 expression is a function of AKT pathway dysregulation and in turn contributes substrate for enhanced cell proliferation.

LAT1, in its' dual roles as a downstream effector of mTOR and as a neutral amino acid transporter, provides a unique opportunity to assay AKT pathway dysregulation by non-invasive imaging with radiolabeled amino acids. [^{18}F]FACBC (1-amino-3-[^{18}F]fluorocyclobutyl-1-carboxylic acid) is a non-metabolized amino acid analogue that is not incorporated into protein. It is an ideal neutral amino acid for studying LAT1-mediated transport across the blood-brain (tumor) barrier and across tumor cell membranes.

The thymidine analog [^{18}F]3'-deoxy-3'-fluorothymidine (FLT) is trapped intracellularly during DNA synthesis and used in brain PET imaging by The University of Washington, Wayne State University, UCLA, MSKCC, and others to image proliferation. The uptake of FLT correlates with thymidine uptake, thymidine kinase activity, and the percentage of cells in S-phase. Preclinical studies have identified the potential of FLT as a PET imaging agent for cellular proliferation. Cellular proliferation

has a number of potential advantages over imaging with FDG-PET, particularly the low background activity of normal brain tissue in contrast to the high FDG uptake (glucose utilization) by normal brain tissue. It is more specific to malignant processes since high energy metabolism, measured by FDG PET, is also associated with a variety of other processes such as inflammation and tissue healing. Cellular proliferation occurs early in response to treatment and is likely to provide earlier and more definitive evidence of response than changes in glucose metabolism, which can be compounded by a variety of issues, including cellular repair. In the case of cytostatic agents, which stop cell division but not necessarily lead to cell death, tumor cellular proliferation drops, but tumor energy metabolism may not change. All these considerations provide an impetus for PET cellular proliferation imaging, especially as a tool for measuring early response.[13]

Shields et al [14] published the first human studies using FLT-PET in a patient with non–small cell lung cancer, demonstrating the high image quality and low background afforded by FLT. Good images are obtained from injection of 5 mCi or less of FLT, and imaging can be started as soon as 45 to 60 minutes after injection. Recently, the radiation dosimetry has been published for FLT [15], showing that patient imaging is feasible with clinically acceptable radiation exposure for the subject. This is a critical issue for FLT because it is likely to be used in clinical applications that require repeated studies to measure response to therapy. These studies paved the way for a series of pilot studies examining FLT-PET imaging for a variety of tumors. The initial study by Shields et al. [14] reported "good" static images with a 5 mCi dose of FLT. We elected to use a 10 mCi dose for the proposed dynamic imaging studies based on current dosimetry estimates; the 10 mCi dose is still below guideline radiation exposures, as discussed in the protocol.

Most early series focused on testing the feasibility of FLT-PET imaging and comparing FLT uptake with in vitro measures of tumor proliferation, typically the Ki-67 (MIB-1) index. Some studies also compared FLT-PET with FDG-PET, given the established clinical role of FDG for staging in the tumor types studied. Studies have shown good correlation between FLT uptake and the Ki-67 index for a variety of tumors.[16] [17] [18] In cases where FDG-PET was also performed, the correlation for FLT uptake versus Ki-67 index was much better than the correlation for FDG uptake versus Ki-67.

Although preclinical models demonstrate the potential utility of FLT-PET for measuring therapeutic response, limited data are available for this use in humans. Preliminary studies used FLT to monitor neoadjuvant breast cancer treatment and showed that FLT could measure changes early in the course of treatment. The appropriate clinical application of FLT is for characterizing known tumors and assessing their response to treatment through serial imaging studies. Both mouse and human trials on gliomas have shown that inhibition of DNA synthesis assays can be used to monitor the therapeutic effect of inhibiting AKT signaling in gliomas. FLT PET is therefore a potentially important marker of glioma proliferation. [19] [20] [21] One study also suggests that FLT PET can predict outcome for patients with gliomas treated with bevacizumab.[22]

Imaging brain tumors with radiolabeled amino acids has been shown to be advantageous over the past two decades, particularly in Europe, where [^{11}C]methionine has been extensively used. It is now widely recognized that [^{11}C]methionine provides better differentiation of brain lesions (particularly tumors) than [^{18}F]FDG. [23] The rationale for imaging brain tumors with [^{18}F]FACBC, rather than with FDG

or methionine,, includes the following: 1) the interpretation of FDG images of solid tumors is often confounded by the multiplicity of factors that increase glycolysis (factors such as tissue hypoxia and the infiltration of inflammatory cells in addition to tumor proliferation rate) 2) [^{18}F]FDG images of brain tumors are often difficult to interpret because of the relatively "high" metabolic activity of surrounding brain tissue which results in relatively "low" tumor-to-brain activity ratios (contrast) 3) [^{18}F]FDG images, in contrast to [^{11}C]methionine and [^{18}F]FACBC images, are not useful for identifying tumor infiltration of adjacent brain tissue (beyond the margin of contrast enhancement) 4) significantly greater contrast between tumor and normal brain tissue is seen with [^{18}F]FACBC compared to methionine 5) the interpretation of methionine images (and images of other naturally occurring amino acids) are confounded by the presence of radiolabeled metabolites that contribute to image background; [^{18}F]FACBC is not metabolized in mammals (including humans) and is excreted unchanged in the urine 6) most importantly for routine clinical applications, tumor imaging with [^{18}F]FACBC has substantial logistical and cost-effective benefits in a busy nuclear medicine department in comparison to imaging with [^{11}C]methionine. [24]

The radiopharmaceutical synthesis, imaging procedures and data analysis for [^{18}F]FACBC PET have been established at MSKCC for brain tumor patient studies under IRB 03-028 (funded NIH grant - R21 CA093501; PI R. Blasberg). These pilot studies in 17 patients demonstrated that [^{18}F]FACBC has better brain tumor imaging characteristics than [^{11}C]-methionine and [^{18}F]FDG. They also determined the biodistribution and clearance of [^{18}F]FACBC in different tissues/organs using whole body PET, and calculated whole body and critical organ radioactivity exposure . Dosimetry information regarding [^{18}F]FACBC PET when used in conjunction with [^{18}F]FLT PET are listed in Appendix A. In addition, we have performed serial scans on patients with recurrent GBM enrolled in trial 04-010 involving concurrent treatment with the EGFR inhibitor gefitinib and the mTOR inhibitor RAD001. Interpretation of brain MRI scans for patients on 04-010 has been difficult because enhancing disease on MRI is often not amenable to measurement by traditional criteria. In addition, in one patient who was classified as a “responder” because of unequivocal improvement in the brain MRI, the [^{18}F]FACBC PET demonstrated clear progression of disease, which was subsequently confirmed. This suggests that the initial MRI results were a false positive for response and that [^{18}F]FACBC PET may be useful in identifying progression earlier than MRI, and may also suggest that [^{18}F]FACBC PET may serve as a biomarker for treatment effect, in this case, lack of effect. Correlation with tissue would allow confirmation of this hypothesis.

We hypothesize that LAT1 upregulation can be imaged and measured using [^{18}F]FACBC PET. We propose to exploit the observation that the LAT1 amino acid transporter is upregulated by activation of mTOR (downstream of AKT), and that LAT1 upregulation will lead to an increase in entry of neutral amino acids in GBMs. LAT1 strongly correlates with high proliferative potential and poor survival in gliomas. [11] Its upregulation results in an influx of amino acids that can contribute to growth. Comparing FACBC PET and FLT findings with tissue analysis will enable us to determine if imaging results are concordant with molecular findings; i.e. if increased LAT 1 expression correlates with higher amino acid signal with PET imaging.

A number of agents targeting tyrosine kinases or specific components of downstream signaling cascades are currently under study in IRB approved clinical protocols for the treatment of brain tumors

at MSKCC. In addition, several of these studies incorporate surgery after initial treatment with an experimental agent in order to assess inhibition of specific pathways in treated tumor tissue. In both the surgical and non-surgical protocols, we would aim to study the role of amino acid PET to assess treatment response. The surgical trials would provide us with pathological tissue that could then be analyzed and compared with [^{18}F]FACBC PET and [^{18}F]FLT PET results. In this manner, we propose to generate a bridge between tissue analysis and brain PET imaging in the future.

4.0 OVERVIEW OF STUDY DESIGN/INTERVENTION

4.1 Design

4.1.1 We will perform [^{18}F]FACBC PET and [^{18}F]FLT PET imaging on 30 patients with gliomas scheduled for treatment with pathway inhibitor agents such as receptor tyrosine kinase inhibitors, antibodies (e.g., bevacizumab), VEGF-Trap, etc. Patients with measurable disease on MRI will undergo PET imaging at baseline (prior-to) and after approximately 1 month of treatment. The primary objective of this study is to determine the biodistribution, clearance, and dosimetry of [^{18}F]FACBC and [^{18}F]FLT in tissue/organs within the field of view of the dynamic PET imaging studies prior-to and during therapy. A comparison of biodistribution parameters (see below) obtained prior-to and during therapy will be performed.

4.1.1.1 Patients eligible for cytoreductive surgery for recurrent malignant glioma often enroll onto a surgical sub-study of a clinical trial involving a novel therapeutic agent. Such patients typically receive therapy for about 1 week, then undergo resection (with the tissue used to interrogate molecular effects of the investigational drug), then resume treatment with the investigational drug after recovering from surgery. We will recruit eligible patients with recurrent malignant glioma who are in such a surgical substudy for [^{18}F]FACBC PET and [^{18}F]FLT PET imaging studies. These patients will have [^{18}F]FACBC PET and [^{18}F]FLT PET imaging at baseline and then again during treatment prior to surgical resection, and then approximately 1 month after initiation of treatment. Note that this analysis is ENTIRELY EXPLORATORY. Only when tissue is available and consent has been obtained, will tissue be analyzed for AKT, VEGFR, and related signaling/biologic changes by immunohistochemistry and/or analysis of flash frozen tissue. This component is a pilot sub-study, consistent with RDRC guidelines, that seeks to investigate whether anti-AKT and/or anti-VEGF directed therapies (alone or in combination with radiation) alter [18F]FACBC and [18F]FLT biodistribution and dosimetry of tissues within the field of view (head). As such, tissue will be analyzed only when avaialble and no additional tissue is collected for the purposes of this protocol. Given the inability to predict at this time the number or quality of tissue specimens,

it is difficult to pre-specify specific analyses. Intraoperative MRI-guided stereotaxy will be used to sample tumor tissue from high- and low-uptake regions when available. Tissue will be analyzed for LAT1 expression and for AKT activity and correlated with imaging. Subjects also registered to MSKCC protocol #09-060 (Michelle Bradbury, PI) will have the pre-operative FLT-PET performed on 09-060 rather than this protocol to coordinate care, maximize institutional resources, and minimize visits for the patient,

4.2 Intervention: PET scan imaging

4.2.1 All subjects will be pre-registered at MSKCC and will have signed the consent form prior to participation in this study.

4.2.2 The morning of the PET imaging studies, all patients will be asked to avoid high protein foods (meat, fish, poultry, cheese, eggs, beans and nuts) and to record the types and servings of the foods they eat. Some patients may also be asked to avoid high protein foods the day prior to imaging. These same patients will also be asked to refrain from eating lunch on the day of the study to reduce muscle uptake of the [^{18}F]FACBC. All patients will also be urged to drink several glasses of water to attain good hydration and urine output. Patients will be placed comfortably on the couch and positioned in the PET scanner. [18F]FACBC and [^{18}F]FLT will be prepared under RDRC guidelines by the MSK Radiochemistry Core and assessed for quality control following "good manufacturing practice" criteria. The radiopharmaceutical will immediately be brought to the Nuclear Medicine Radiopharmacy for dispensation in the PET suite. Patients will receive 370 MBq (10 mCi) [^{18}F]FACBC or [^{18}F]FLT by i.v. infusion over approximately 5 minutes.

4.2.3 It is our intent for the [^{18}F]FACBC PET to be performed first and the [^{18}F]FLT PET performed either the next day or within approximately one week. However, the order and timing may be reversed or modified depending on patient, radiopharmaceutical, and scanner availability.

4.2.4 Patients with gliomas will undergo one [^{18}F]FACBC and/or one [^{18}F]FLT PET scan as initial baseline scan(s) prior to initiating new antineoplastic therapy.

4.2.5 For patients with gliomas, subsequent [^{18}F]FACBC and [^{18}F]FLT PET scans will be performed approximately 1 following the initiation of antineoplastic therapy.

4.2.6 A venous catheter will be placed in a superficial hand or arm vein for administration of the radiopharmaceuticals. A second venous catheter will be placed in the opposite hand or arm for venous blood sampling. If a central venous catheter is present, it will be used for blood sampling or radiopharmaceutical administration, and only a single venous catheter will be placed. One blood sample will be obtained prior to injection of

[^{18}F]FACBC to measure plasma amino acid concentrations at the time of the study. Sequential blood samples will be obtained following [^{18}F]FACBC or [^{18}F]FLT infusion and assayed for whole blood and plasma radioactivity. All catheters will be removed at the end of the day.

4.2.7 The [^{18}F]FACBC or [^{18}F]FLT study will involve a 60-minute emission dynamic scan of the head to obtain kinetic information on tumor uptake for modeling. After the initial dynamic scan, the patient will void completely. The volume of urine will be recorded, and a urine sample will be assayed for urine radioactivity by research staff.

4.2.8 Two subsequent head scans will be obtained at 90 and 120 minutes for both the [^{18}F]FACBC and [^{18}F]FLT studies.

4.2.9 Venous blood samples will be drawn in green top tubes 30, 60, 90, and 120 minutes post-radiopharmaceutical injection from a vein in the arm opposite that used to inject the radiopharmaceutical by research staff. [^{18}F] radioactivity in the [^{18}F]FACBC and [^{18}F]FLT blood/plasma/urine samples will be measured by research staff.

4.2.10 The volume of urine voided between scans will be recorded by research staff. Frequent urination will be encouraged to reduce bladder exposure and provide a brief "relaxation" period between sequential sets of scans, and will facilitate individual patient acceptance and compliance during the study.

5.0 CRITERIA FOR SUBJECT ELIGIBILITY

The criteria for patient selection will include patients registered at Memorial Hospital with a diagnosis of a glioma who are undergoing treatment with a signal transduction pathway inhibitor. Patients will have no known medical contraindications to PET scans. . There will be no exclusions based on age, sex or ethnic background; a pregnancy test will be required of women of child-bearing age. Pregnant women will be excluded from this study.

5.1 Subject Inclusion Criteria

5.1.1 Registered patient at MSKCC.

5.1.2 Child-bearing age females must be non-pregnant (documented by a negative pregnancy test within the last 2 weeks)„ non-lactating, and must be using adequate contraception or be surgically sterile.

5.1.3 Patients with gliomas

5.1.3.1 Patients planning to start anti-AKT and/or anti-VEGF directed therapies.

5.1.3.2 Patients with measurable disease on MRI or CT neuroimaging.

5.2 Subject Exclusion Criteria

5.2.1 Patient is incontinent of urine or stool (which would make them unable to tolerate lying still for 60 minutes).

5.2.2 Patient cannot tolerate lying still for a 60 minute session in the PET tomograph.

6.0 RECRUITMENT PLAN

6.1 Recruitment of patients will be through the participating investigators and referrals from staff physicians at Memorial Hospital. A detailed description of the study procedures will be provided to each referring physician and a simpler version to each patient by the principal investigator or one of the senior investigators of this research proposal.

6.2 The signed IRB consent form will be brought to the patient protocol accrual (PPA) system in the Data Management Resource Group of the Dept. of Biostatistics and Epidemiology at MSKCC for registration at MSKCC, and the imaging study requisition for the imaging protocol will be brought to the Nuclear Medicine Service, Department of Radiology.

6.3 All study participants will receive $50 per visit to cover travel/food expenses. In addition, participants who live more than 50 miles from the hospital are eligible to be reimbursed up to $350 per night for a hotel stay per study visit.

7.0 ASSESSMENT/EVALUATION PLAN

7.1 Reconstructed images will be obtained from the PET data; corrections will be made for randoms, scatter, dead-time, detector inhomogeneity and attenuation. Image data will be calibrated to nCi/cc and decay corrected to the time of injection. Comparisons will be made with the most recent MRI study obtained as part of the patient's standard radiographic assessment. The PET and MRI scans will be digitally co-registered using established methods at MSKCC to facilitate this comparison.

7.2 The co-registered MRI images will be used to define the tumor margins and the digitally transformed analysis will be quantitative. Volumes of interest (VOI) will be drawn on the MRI and transferred to the digitally registered [^{18}F]FACBC and [^{18}F]FLT PET scans; VOI's on remote normal white and gray-matter regions of the brain will also be drawn. The mean of the voxel values (nCi/cc) within each VOI will be determined for each of the dynamic frames of the [^{18}F]FACBC and [^{18}F]FLT studies. An established measurement of tumor and normal brain radioactivity, normalized to body weight and injected radioactivity dose, the Standard Uptake

Value (SUV) will be calculated [25]. Radioactivity-time profiles will be generated and fitted to established kinetic models for evaluating [^{18}F]FACBC and [^{18}F]FLT accumulation. For [^{18}F]FACBC and [^{18}F]FLT, values of the blood to brain influx constant (K_1) and the distribution volume (V_d) in tumor and remote normal brain tissue will be estimated. For [^{18}F]FLT, the rate-determining flux of FLT-trapping (K_i) in tumor and remote normal brain will also be estimated. In both studies, the rapidly equilibrating plasma compartment (Vp) will also be estimated. Established kinetic modeling and dynamic data fitting routines, currently existing at MSK, will be used to estimate the above parameters: 1) SAAM2 [University of Washington, Seattle, WA 98195-5061, USA; Email: mailto:saam2@u.washington.edu], and 2) Matlab [The MathWorks, Inc., 3 Apple Hill Drive, Natick, MA 01760-2098].

7.3 The registered image sets will be displayed simultaneously, facilitating the identification of the correspondence between the lesion(s) in each modality. Volumes of interest created on one modality will be applied to the other modalities, thus enabling direct quantitative comparisons.

7.4 We will be performing a pre-treatment – during-treatment comparison in the brain tumor patients. Paired-test comparisons of the above parameters will be performed. There is no treatment-response assessment being performed. Therefore, any issue of "threshold" criteria does not apply.

7.5 All tissue analyses are entirely exploratory and will only be performed on available tissue. Given the inability to predict at this time the number or quality of tissue specimens, it is difficult to pre-specify specific analyses. As above, there is no treatment-response assessment being performed. Therefore, any issue of "threshold" criteria does not apply.

7.6 Radiation dosimetry for critical tissues/organs within the PET imaging field of view will be calculated using standard MIRD-based equations.

8.0 TOXICITIES/SIDE EFFECTS

7.1 8.1 PET Scanning. Potential risks to the patient are small, and present more of an inconvenience (multiple scans on consecutive days). The PET has a large bore to accommodate the whole body and the environment is much less confining than the head cage of a MR tomograph. For the [^{18}F]FACBC and the [^{18}F]FLT scan, the scanning period of the head will be 60 min. followed by two 15 minute scans at 90 and 120 minutes.

8.2 Radiation Exposure. The radiation exposure from diagnostic [^{18}F]FACBC and [^{18}F]FLT PET studies is within acceptable limits as established by FDA Guidelines (Code of Federal Regulations, Section 361.1). Dosimetry estimates for both [^{18}F]FACBC obtained from data obtained at MSKCC under IRB protocol 03-028 and [^{18}F]FLT are listed in Appendix A. Dosimetry estimates for the combined FACBC plus FLT PET studies is also listed in Appendix A. The dosimetry table in Appendix A provides the centigray dose per mCi of injected FACBC and FLT in columns 2 and 3. The dose per PET/CT scan after a 10 mCi administration for each tracer in columns 4 and 5 and finally the total anticipated patient dose for 2 FACBC and 2 FLT PET/CT

scan procedures. Note that the total dose organ dose is below 5cGy (3cGy to total body, marrow, lens of the eye and testes) for any single scan and below a cumulative dose of 15 cGy (5cGy to total body, marrow, lens of eye and testes) for the sum of all procedures. The organ receiving the largest dose is the liver with a dose of 6.86 cGy for all scans proposed within this experimental protocol.

8.3 The dosimetry estimates in Appendix A have led us to select a conservative 370 MBq (10 mCi) dose of [^{18}F]FACBC for this study. This will maintain the radioactivity exposure to all normal tissue (including whole body, blood forming organs and gonads) well below the annual radiation dose limit of 5 cSv established by the FDA (Code of Federal Regulations, Section 361.1). In the event that our measurements and dosimetry estimates for [^{18}F]FACBC, [^{18}F]FLT and the combined study exceed the annual radiation dose limit of 5 cSv established by the FDA, we will reduce the administered dose to insure that the annual radiation dose limit of 5 cSv is not exceeded. Similarly, with an administered activity of 10 mCi, none of the normal-organ doses for an [^{18}F]FACBC, [^{18}F]FLT and the combined study exceed the RDRC limit of 5 rad for a single study, and none of the normal-organ doses exceed the RDRC limit of 15 rad per year.

8.4 Patients will be observed closely for any signs of radiopharmaceutical toxicity such as change in vital signs or well being.

9.0 PRIMARY OUTCOMES

Basic measurements of tumor and normal brain radioactivity (SUV) will be obtained. In addition, the dynamic PET data will be fit to one or more kinetic models to estimate the plasma volume (V_p), blood-to-tissue influx constant (K_1) and tissue volume of distribution (V_d) for FACBC and FLT, and the flux of FLT-trapping (K_i) as a measure of thymidine kinase activity.

10.0 CRITERIA FOR REMOVAL FROM STUDY

10.1 If the patient is no longer able to participate in the protocol and imaging schedule.

10.2 If the patient's primary physician and the PI consider that further participation in the protocol would not be in the best interest of the patient.

10.3 If at any time the patient is found to be ineligible for the protocol as designated in the section on Criteria for Patient/Subject Eligibility (e.g., a change in diagnosis), the patient will be removed from the study.

11.0 BIOSTATISTICS

11.1 We will compare the changes in 4 imaging parameters for [18F]FACBC (SUV, K1, Vd, and Vp) and 5 parameters (SUV, Ki, K1, Vd and Vp) for [18F]FLT. Since we will be comparing pre-treatment vs during treatment values in the same patient, paired t-tests will be performed. This is a small pilot study involving 30 patients with brain tumors. Any clinical, tissue, or other correlations are entirely exploratory in nature and are not the purpose of this study.

12.0 RESEARCH PARTICIPANT REGISTRATION AND RANDOMIZATION PROCEDURES

12.1 Research Particpant Registration

Confirm eligibility as defined in the section entitled Criteria for Patient/Subject Eligibility.

Obtain informed consent, by following procedures defined in section entitled Informed Consent Procedures.

During the registration process registering individuals will be required to complete a protocol specific Eligibility Checklist.

All participants must be registered through the Protocol Participant Registration (PPR) Office at Memorial Sloan-Kettering Cancer Center. PPR is available Monday through Friday from 8:30am – 5:30pm at 646-735-8000. The PPR fax numbers are (646) 735-0008 and (646) 735-0003. Registrations can be phoned in or faxed. The completed signature page of the written consent/verbal script and a completed Eligibility Checklist must be faxed to PPR.

12.2 Randomization

There will be no randomization in this study.

13.0 DATA MANAGEMENT ISSUES

A Research Study Assistant (RSA) will be assigned to the study. The responsibilities of the RSA include project compliance, data collection, abstraction and entry, data reporting, regulatory monitoring, problem resolution and prioritization, and coordinate the activities of the protocol study team.

The data collected for this study will be entered into a secure database (CRDB). Source documentation will be available to support the computerized patient record.

13.1 Quality Assurance

Weekly registration reports will be generated to monitor patient accruals and completeness of registration data. Routine data quality reports will be generated to assess missing data and inconsistencies. Accrual rates and extent and accuracy of evaluations and follow-up will be monitored periodically throughout the study period and potential problems will be brought to the attention of the study team for discussion and action

Random-sample data quality and protocol compliance audits will be conducted by the study team, at a minimum of two times per year, more frequently if indicated.

13.2 Data and Safety Monitoring

The Data and Safety Monitoring (DSM) Plans at Memorial Sloan-Kettering Cancer Center were approved by the National Cancer Institute in September 2001. The plans address the new policies set forth by the NCI in the document entitled "Policy of the National Cancer Institute for Data and Safety Monitoring of Clinical Trials" which can be found at: http://cancertrials.nci.nih.gov/researchers/dsm/index.html. The DSM Plans at MSKCC were established and are monitored by the Office of Clinical Research. The MSKCC Data and Safety Monitoring Plans can be found on the MSKCC Intranet at: *http://mskweb2.mskcc.org/irb/index.htm*

There are several different mechanisms by which clinical trials are monitored for data, safety and quality. There are institutional processes in place for quality assurance (e.g., protocol monitoring, compliance and data verification audits, therapeutic response, and staff education on clinical research QA) and departmental procedures for quality control, plus there are two institutional committees that are responsible for monitoring the activities of our clinical trials programs. The committees: *Data and Safety Monitoring Committee (DSMC)* for Phase I and II clinical trials, and the *Data and Safety Monitoring Board (DSMB)* for Phase III clinical trials, report to the Center's Research Council and Institutional Review Board.

During the protocol development and review process, each protocol will be assessed for its level of risk and degree of monitoring required. Every type of protocol (e.g., NIH sponsored, in-house sponsored, industrial sponsored, NCI cooperative group, etc.) will be addressed and the monitoring procedures will be established at the time of protocol activation.

14.0 PROTECTION OF HUMAN SUBJECTS

Risks: Potential risks to the patient include pain and discomfort related to phlebotomy and related to time spent undergoing [^{18}F]FACBC PET and [^{18}F]FLT PET scanning. Specific toxicities and side

effects related to [^{18}F]FACBC PET and [^{18}F]FLT PET imaging, including radiation exposure are described in section 8.

Benefits: We do not expect patients to derive any clinical benefit from this clinical trial. We hope in the future that knowledge from this trial will help in better evaluating treatment response in future patients with brain tumors.

Costs: Patients will not be charged for the radiotracer, PET study or any associated blood tests that will be done.

14.1 Privacy

MSKCC's Privacy Office may allow the use and disclosure of protected health information pursuant to a completed and signed Research Authorization form. The use and disclosure of protected health information will be limited to the individuals described in the Research Authorization form. A Research Authorization form must be completed by the Principal Investigator and approved by the IRB and Privacy Board.

14.2 Serious Adverse Event (SAE) Reporting

Any SAE must be reported to the IRB/PB as soon as possible but no later than 5 calendar days. The IRB/PB requires a Clinical Research Database (CRDB) SAE report be submitted electronically to the SAE Office at sae@mskcc.orgcontaining the following information:

Fields populated from the CRDB:

- Subject's name (generate the report with only initials if it will be sent outside of MSKCC)
- Medical record number
- Disease/histology (if applicable)
- Protocol number and title

Data needing to be entered:

- The date the adverse event occurred
- The adverse event
- Relationship of the adverse event to the treatment (drug, device, or intervention)
- If the AE was expected
- The severity of the AE
- The intervention
- Detailed text that includes the following information:
  - A explanation of how the AE was handled
  - A description of the subject's condition
  - Indication if the subject remains on the study
  - If an amendment will need to be made to the protocol and/or consent form

The PI's signature and the date it was signed are required on the completed report.

14.2.1
This protocol will be conducted through the RDRC.

In accordance with RDRC requirements, adverse events, annual report of patients accrued, and other RDRC requirements will also be reported to the appropriate regulatory bodies.

15.0 INFORMED CONSENT PROCEDURES

Before protocol-specified procedures are carried out, consenting professionals will explain full details of the protocol and study procedures as well as the risks involved to participants prior to their inclusion in the study. Participants will also be informed that they are free to withdraw from the study at any time. All participants must sign an IRB/PB-approved consent form indicating their consent to participate. This consent form meets the requirements of the Code of Federal Regulations and the Institutional Review Board/Privacy Board of this Center. The consent form will include the following:

1. The nature and objectives, potential risks and benefits of the intended study.
2. The length of study and the likely follow-up required.
3. Alternatives to the proposed study. (This will include available standard and investigational therapies. In addition, patients will be offered an option of supportive care for therapeutic studies.)
4. The name of the investigator(s) responsible for the protocol.
5. The right of the participant to accept or refuse study interventions/interactions and to withdraw from participation at any time.

Before any protocol-specific procedures can be carried out, the consenting professional will fully explain the aspects of patient privacy concerning research specific information. In addition to signing the IRB Informed Consent, all patients must agree to the Research Authorization component of the informed consent form.

Each participant and consenting professional will sign the consent form. The participant must receive a copy of the signed informed consent form.

16.0 REFERENCE(S)

See below

17.0 APPENDICES

Appendix A. [18F]FACBC and [18F]FLT Dosimetry

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