PET Assessment of Tumor Hypoxia

Farrokh Dehdashti, M.D.
Mallinckrodt Institute of Radiology
Washington University
St. Louis, Missouri

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Hypoxia

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- **Tumor microenvironment plays a critical role in tumor progression and treatment resistance**
- **An important microenvironment parameter relevant to treatment outcome is tumor hypoxia**
- **Up to 50–60% of locally advanced solid tumors may exhibit areas of hypoxic and/or anoxic tissue heterogeneously distributed within the tumor mass**
- **Tumor hypoxia results from an imbalance between the supply and use of oxygen in tumor tissues**

Imaging Hypoxia – Basic Science to Clinical Application

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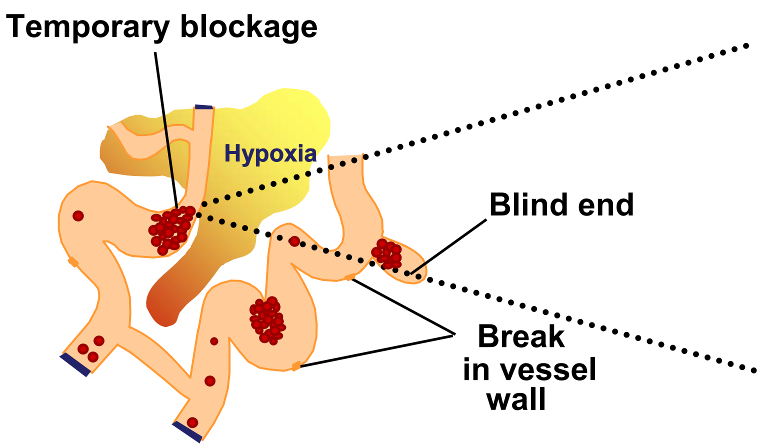
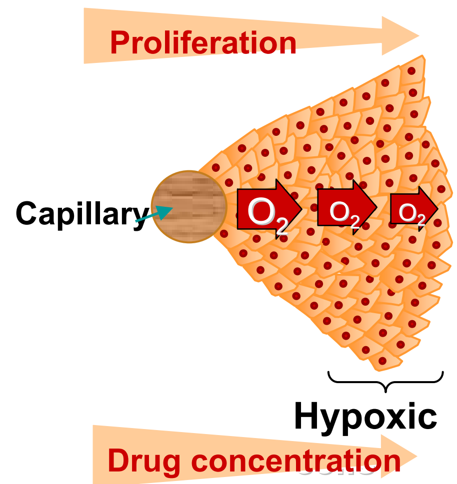
Hypoxia – reduction of tissue oxygen supply below physiological levels





<u>Hypoxic Tumor</u>: Induces gene expression
- Resistant to traditional radiotherapy
- Resistant to chemotherapy
- Increases metastatic potential
- Increases tumor aggressiveness

Brown JM. Mol Imaging Today 2000; 6:157-162

Pathophysiology of Hypoxia

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- **Most common types of hypoxia**
  - **Acute hypoxia (perfusion-limited) – caused by inadequate blood flow due to abnormal and disorganized vasculature in the tumor**
  - **Chronic hypoxia (diffusion limited) – caused by increase in diffusion distances with tumor expansion**
  - **Anemic hypoxia – caused by reduced oxygen transport capacity of blood subsequent to tumor-associated or therapy-associated anemia**

Clinical Studies: Hypoxia in Cancers

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- Cervical Cancer
  - Oxygen status of the tumor is the single most important prognostic factor, independent of
    - Various patient demographics (such as age and menopausal status)
    - Pretreatment tumor characteristics (such as clinical tumor stage and size, histological type, and differentiation)
  - Determination of hypoxia carries information independent of current clinicopathological parameters and can provide important prognostic information

Clinical Studies: Hypoxia in Cancers

- Head & Neck Cancer
  - Oxygen status of the tumor determines the likelihood of locoregional failure and affects the prognosis independent of other known prognostic variables
  - Pretreatment tumor oxygenation status is predictive of radiation response
- Soft tissue tumors
  - Hypoxia was an indicator for a poorer disease specific, disease-free and overall survival, higher recurrence rate

Treatment of Hypoxia

- Several hypoxia-targeted therapies, including high-oxygen-content gas breathing, radiosensitizers and hypoxic cytotoxins, have been developed to overcome hypoxia-mediated radio/chemoresistance
  - Increased tumor control rate is often accompanied by more severe side effects
- Hypoxic-measuring tools are needed:
  - To identify hypoxic tumors and predict patient outcome
  - To select treatments on an individual basis
  - To monitor therapy and evaluate early response to treatment

Measurement of Tumor Hypoxia

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- **Invasive methods**
  - Oxygen electrodes
  - *In vitro* analysis of tumor tissue
    - CA-IX expression and hypoxia- induced factor HIF-1 alpha
    - DNA Comet assay
    - Immunohistochemistry with antibodies- pimonidazole immunostaining
    - Hemoglobin-O_2 cryospectrophotometry

- **Noninvasive methods**
  - Imaging by MRI
    - » Oxygen related MR signals
    - » P-31 metabolites
    - » Oxygen mimetic molecules e.g., F-19 nitroimidazole
  - Imaging by SPECT
    - » ^{131}IAZA - requires lengthy waits between injection and imaging
    - » ^{99m}Tc-HL91 - retention in nonviable myocardium similar to normal tissue
  - Imaging by PET

Measurement of Tumor Hypoxia

- Oxygen electrodes
  - Direct measurement of hypoxia
  - Strong correlation between pre-therapy oxygen measures and outcome in several tumors (cervical cancers, head and neck cancers)
  - Invasive, technically demanding to use
  - Require accessible tumors
  - Subject to sampling errors

Measurement of Tumor Hypoxia

- Imaging by positron emission tomography (PET)
  - Noninvasive
  - Accessible and non-accessible tumors
  - Entire tumor sampled
  - Excellent resolution
  - Repeated measurements possible

Measurement of Tumor Hypoxia with FMISO

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- **[^{18}F] fluoromisonidazole (FMISO), analogs of the radiosensitizer misonidazole**
  - **Has been well characterized as a probe for hypoxic cells *in vitro* and *in vivo***
  - **Undergoes bioreduction in hypoxic cells and traps within the cell**
  - **Its uptake is inversely proportional to O2 level in animal tumor models**
  - **Imaging hypoxic cells in human tumors, such as non-small cell lung cancer (NSCLC), head & neck (H & N) cancer, etc.**

Measurement of Tumor Hypoxia with FMISO

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- **Soft tissue sarcoma: no significant correlation between FMISO uptake and oxygen electrode measurement** **(Bentzen et al. Radiother Oncol 2003; 67:339)**
- **H & N: strong correlation with FMISO uptake (T/M) and PO2 ≤ 5 mm Hg ($p < 0.001$)** **(Zimmy et al. Eur J Nucl Med Mol Imaging 2006)**
- **H & N: pretherapy FMISO uptake shows a strong trend to be an independent prognostic factor** **(Rajendran et al. Clin Cancer Res. 2006 Sep 15;12(18):5435-41)**

Measurement of Tumor Hypoxia with FMISO

- **H & N cancer: FMISO-PET and hypoxia-targeted therapy with tirapazamine**
  - **Patients (n = 53) with stage III or IV squamous cell ca of H & N randomized to chemotherapy with or without tirapazamine**
  - **45/53 participated in PET imaging arm**
  - **In patients treated with chemo alone: 1/10 (10%) without hypoxia and 8/13 (61%) with hypoxia recurred locally**
  - **In patients treated with chemo & tirapazamine: 0/3 without hypoxia and 1/19 (5%) patients with hypoxic tumors recurred locally**

Rischin et al. J Clin Oncol 2006; 24:2098

Measurement of Tumor Hypoxia with FMISO

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- **H & N cancer: FMISO-PET and hypoxia-targeted therapy with tirapazamine**
  - **Repeat FMISO-PET 4-5 weeks after therapy in 29/32 with baseline tumor hypoxia**
    - **4/13 patients treated with chemo alone were FMISO+, all 4 recurred**
    - **2/16 patients treated with chemo & tirapazamine were FMISO+, none recurred**

Rischin et al. J Clin Oncol 2006; 24:2098

Measurement of Tumor Hypoxia with FMISO

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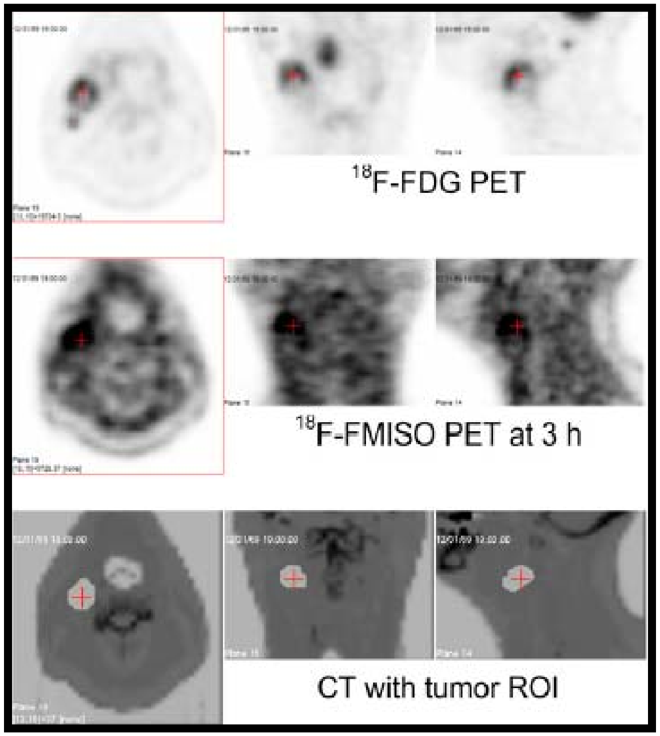
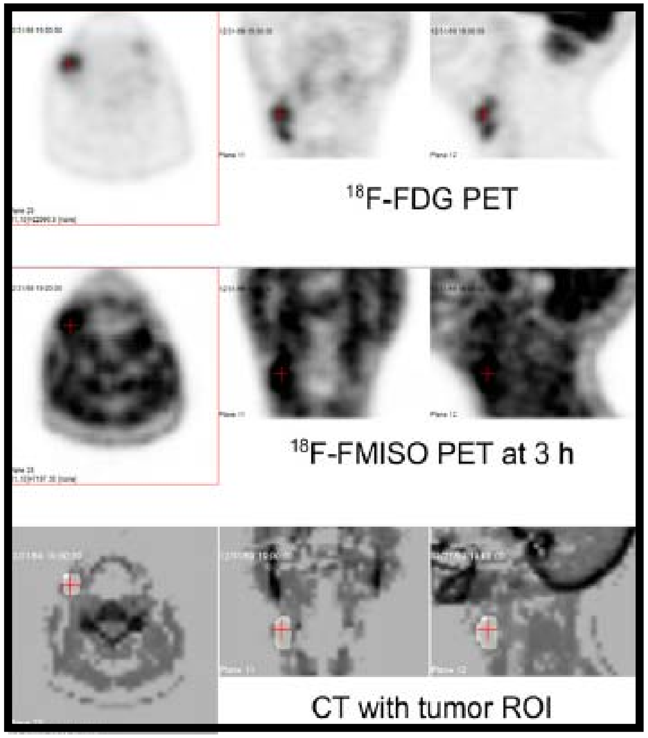
H & N cancer: positive correlation of between the average T/B and trapping rate constant (correlation coefficient 0.86)

T/B=1.74



Hypoxic: Right Tonsillar Ca

T/B=1.19



Normoxic: Right Base of Tongue Ca

Wang et al. J Nucl Med 2010; 51:37-45

Measurement of Tumor Hypoxia with FMISO

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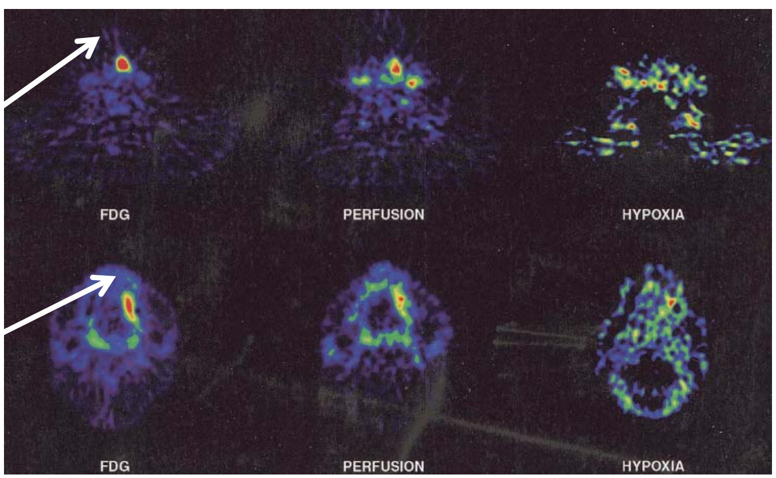
- **Inadequate imaging characteristics**
  - Insufficient tissue extraction (low lipophilicity-low membrane permeability)
  - Slow blood clearance
  - Low hypoxic/normoxic tissue activity ratio (1.4 at 2 hrs)

Second Generation ^{18}F-labeled nitroimidazoles Hypoxic Agents

- [^{18}F]fluoroerythronitroimidazole (^{18}F-FETNIM)
  - More hydrophilic (*limits diffusion in tumors*), metabolically stable, traps in hypoxic tumor tissue, more rapidly clears from well-oxygenated tissues, and correlated with OS in lung ca
  - *High blood concentration* (lower tissue/plasma ratios than FMISO, T/B=0.93 for H & N ca, 1.5

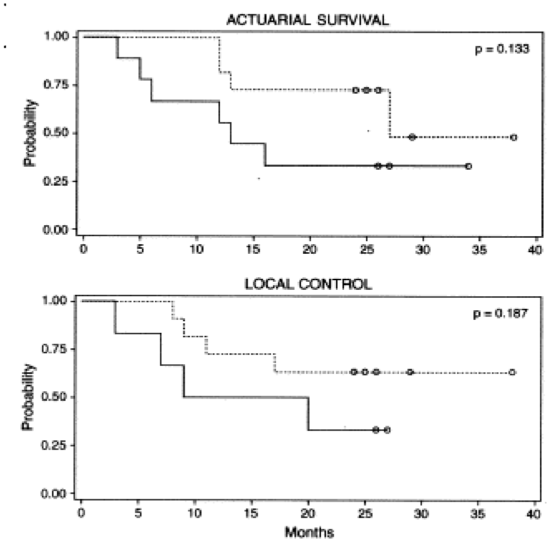
Left false vocal cord

Left tongue



Normoxic

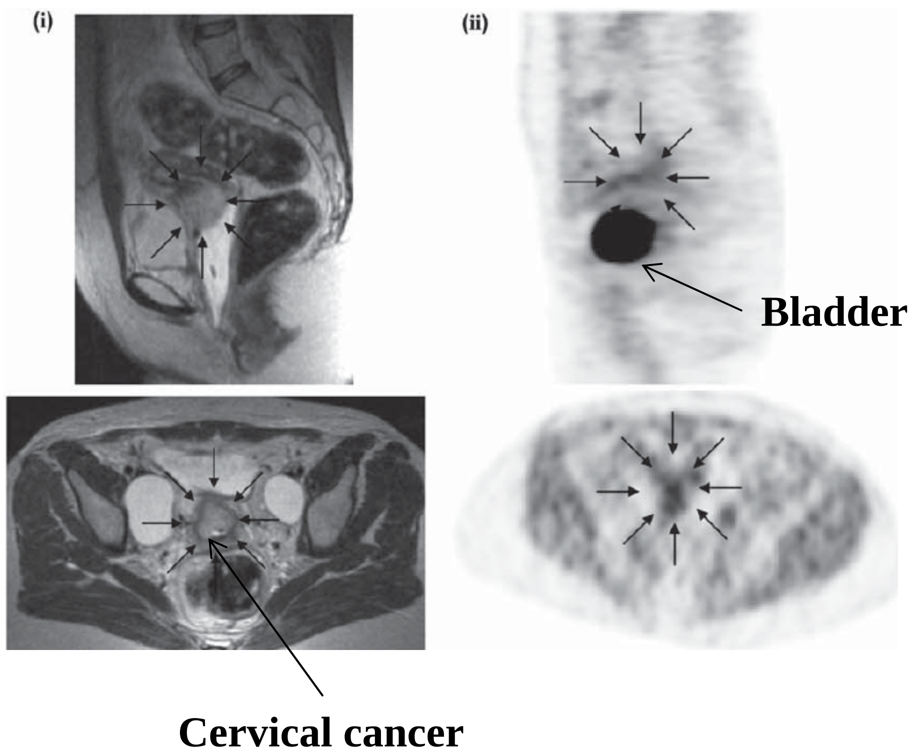
Hypoxic



Imam SK. Cancer Biother Radiopharm 2010; 25:365-74; Li et al. Clin Lung Cancer 2010; 11:335-40; Yang et al. Radiology 1995; l94:795-80; Lehtiö et al. Int J Radiation Oncology Biol Phys 2004; 59:971-82

Second Generation ^{18}F-labeled nitroimidazoles Hypoxic Agents

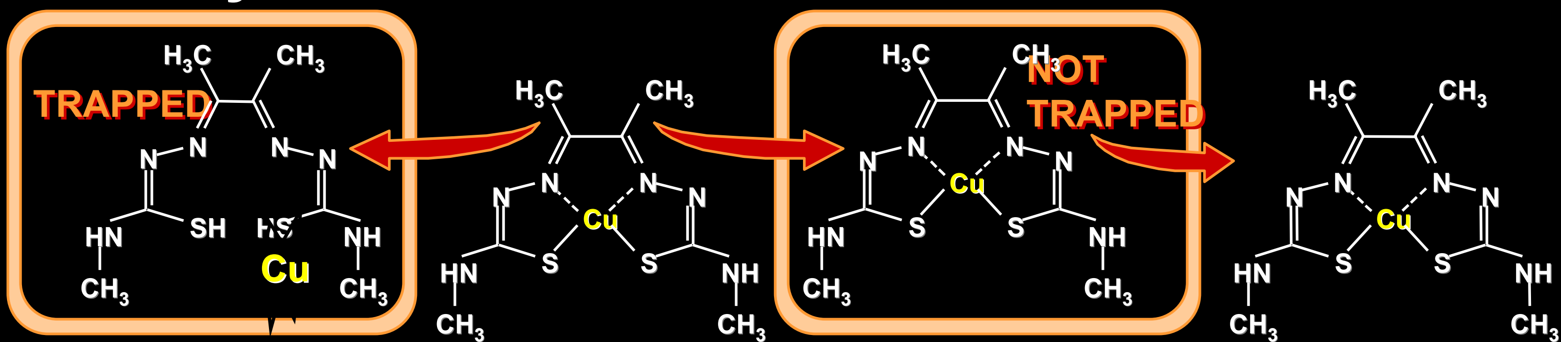
- ^{18}F-fluoroazomycin arabinoside (^{18}F-FAZA)
  - Less lipophilic than FMISO, peak activity within 60 min
  - Superior biokinetics, more rapidly clears from well-oxygenated tissues
  - Data suggesting binding to hypoxic tumor tissue is reversible



Imam SK. Cancer Biother Radiopharm 2010; 25:365-74;
Schuetz et al. Acta Oncologica 2010; 49: 941-7

PET Imaging Agents – Cu(ATSM)

Theory:

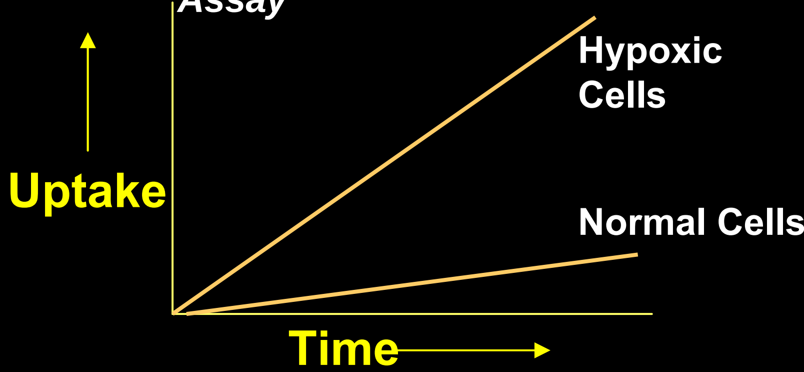


Hypoxic cell ($-O_2$)

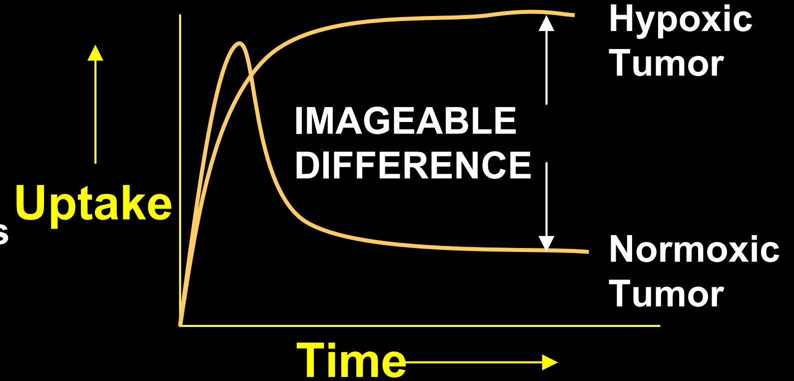
Normal cell ($+O_2$)

Basic Science:

Single Cell Suspension Assay

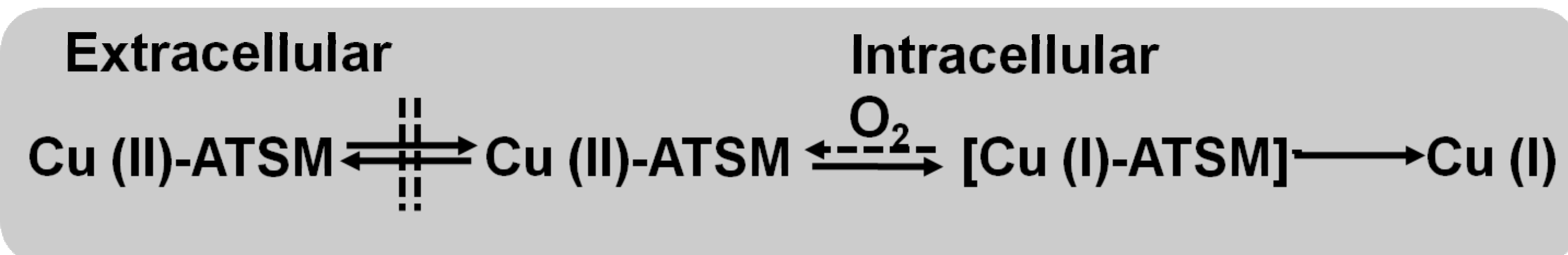


PET Imaging of Tumor-Bearing Rodent



Measurement of Hypoxia with Cu-ATSM

- **Copper labeled dithiosemicarbazone complex (Cu-ATSM)** **(Fujibayashi et al., 1989)**
  - **Highly lipophilic - high membrane permeability, reduced and retained only in hypoxic cells (mitochondria in nontumor and microsomal in tumors)**
  - **Highly extracted, but rapidly washes out of normoxic tissue** **(John et al., 1990, Taniuchi et al., 1995, Fujibayashi et al., 1997)**
  - **Good hypoxic/normoxic tissue activity ratio (hypoxic/normoxic of 4.0 at 15 min)**



Uptake of ^{64}Cu(ATSM) in EMT6 Tumor Cell Line *In Vitro* at Varying Degrees of O_2

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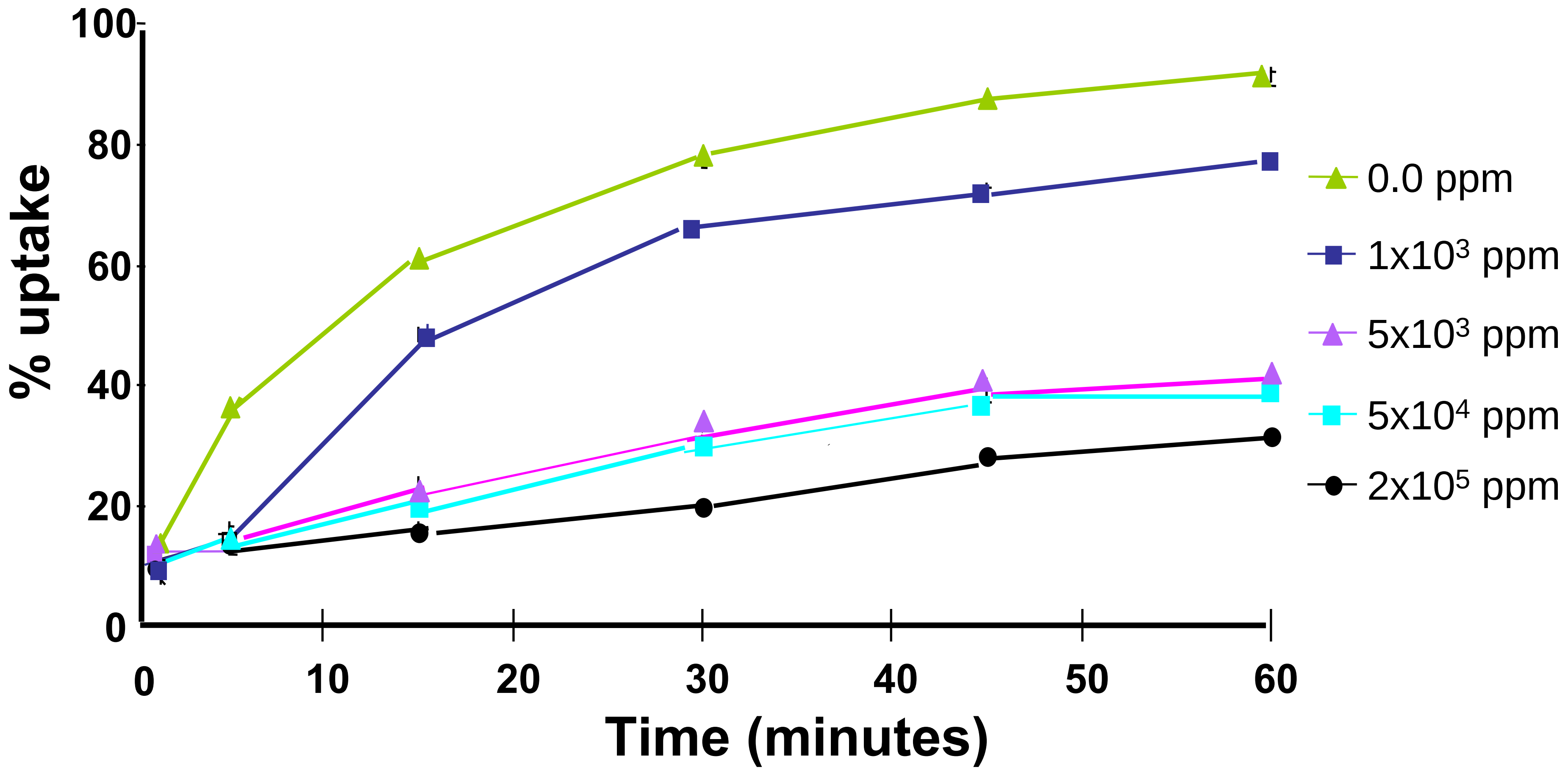

Lewis et al., J Nucl Med 1999; 40:177-83

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

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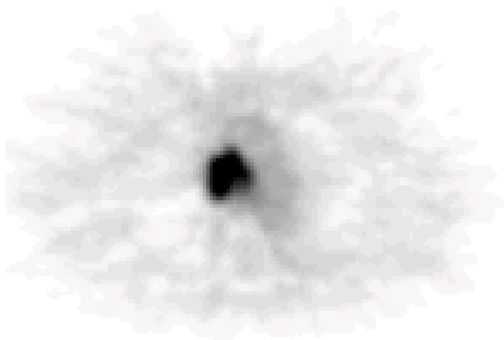
- ^{60}Cu-ATSM (T/M, not SUV) predictive of response to therapy by RECIST (n = 14)
  - FDG uptake was not significantly different in responders and nonresponders (P = 0.7) and did not correlate with ^{60}Cu-ATSM uptake (r = 0.04; P = 0.9)
- T/M differed significantly in responders (1.5 ± 0.4) and nonresponders (3.4 ± 0.8) (p = 0.002)
- Peak slope index was significantly lower in responders compared with nonresponders (1.7 ± 1.2%/min vs. 3.6 ± 0.95%/min) (p = 0.02)
  - All responders (n=8) had T/M < 3.0 & all nonresponders (n=6) had T/M ≥ 3.0

Dehdashti et al., Eur J Nucl Med Mol Imag 2003; 30:844-850

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

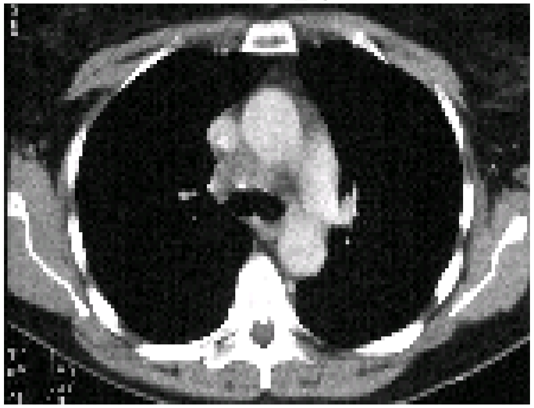
Responder

Pre-therapy FDG-PET

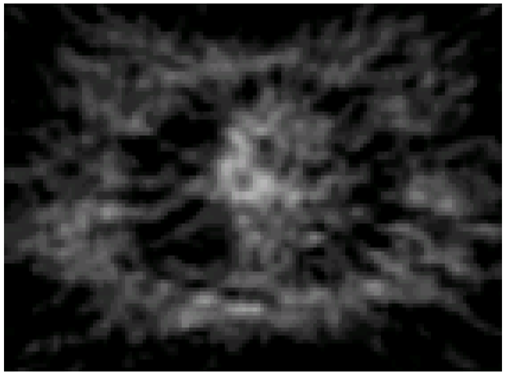


SUV = 4.9

Pre-therapy CT

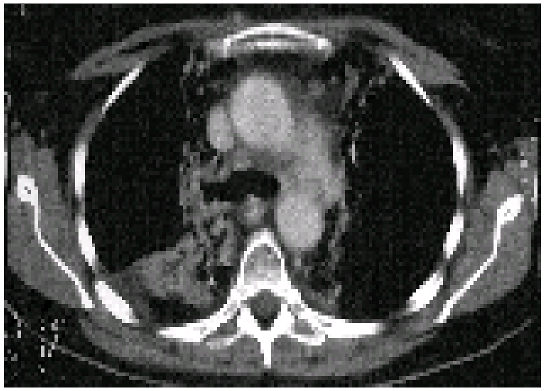


Pre-therapy Cu-ATSM-PET



T/M = 1.3

Post-therapy CT

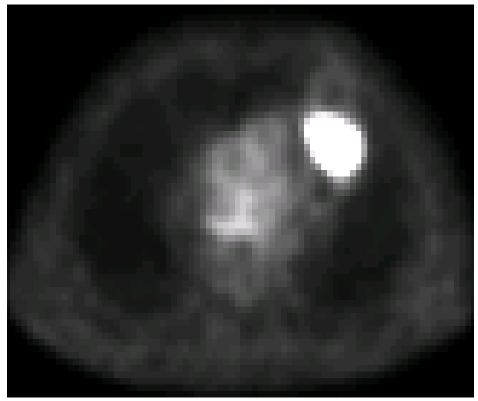


Dehdashti et al. ,Eur J Nucl Med Mol Imag 2003; 30:844-850

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

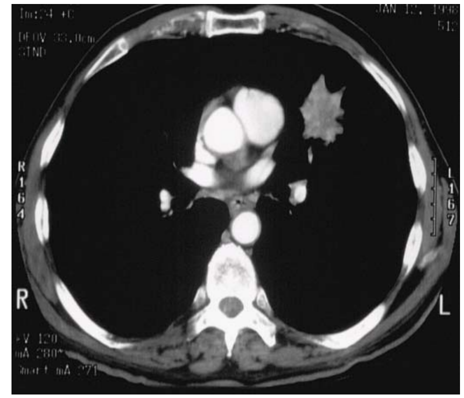
Non-Responder

Pre-therapy FDG-PET

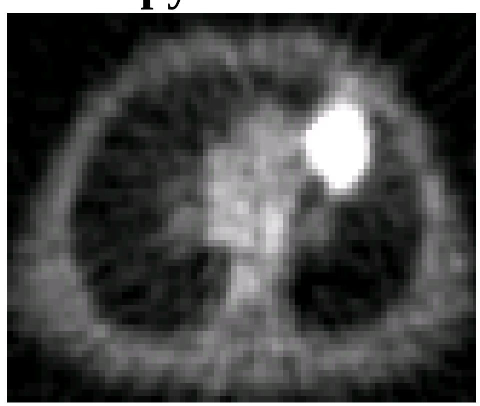


SUVmax = 17

Pre-therapy CT



Pre-therapy Cu- ATSM-PET



T/M = 3.6

Post-therapy CT

Dehdashti et al., Eur J Nucl Med Mol Imag 2003; 30:844-850

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

- **Rectal cancer: ^{60}Cu-ATSM prior to neoadjuvant chemoradiotherapy (n =17)**
  - **^{60}Cu-ATSM (T/M) was predictive of survival**
    - **Patients with T/M < 2.6 have better overall and progression-free survival than patients with T/M > 2.6 (both $p < 0.05$)**
  - **FDG uptake (n=11) did not correlate with ^{60}Cu-ATSM uptake (r=0.4; P=0.9)**
  - **No significant difference in mean tumor FDG uptake between patients with hypoxic tumors and those with normoxic tumors (P=0.3)**

Dietz et al. Dis Colon Rectum 2008; 51:1641-8

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

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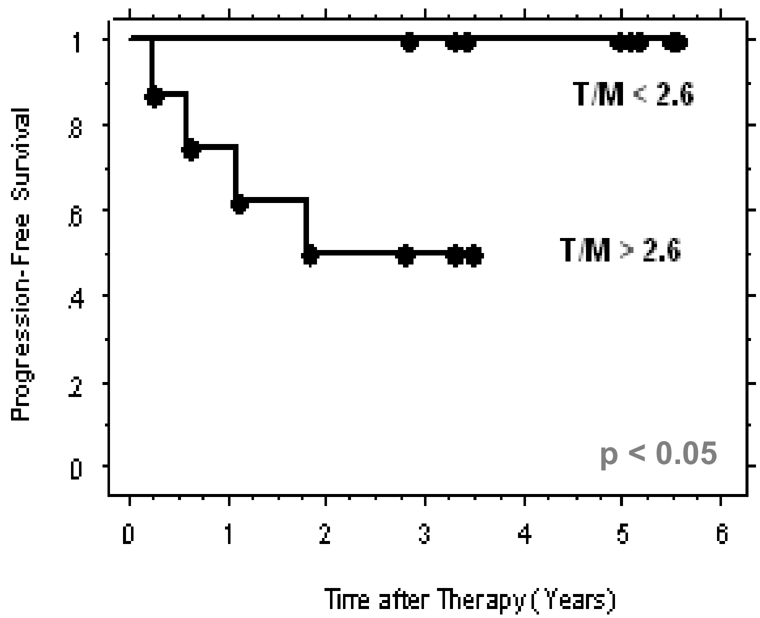
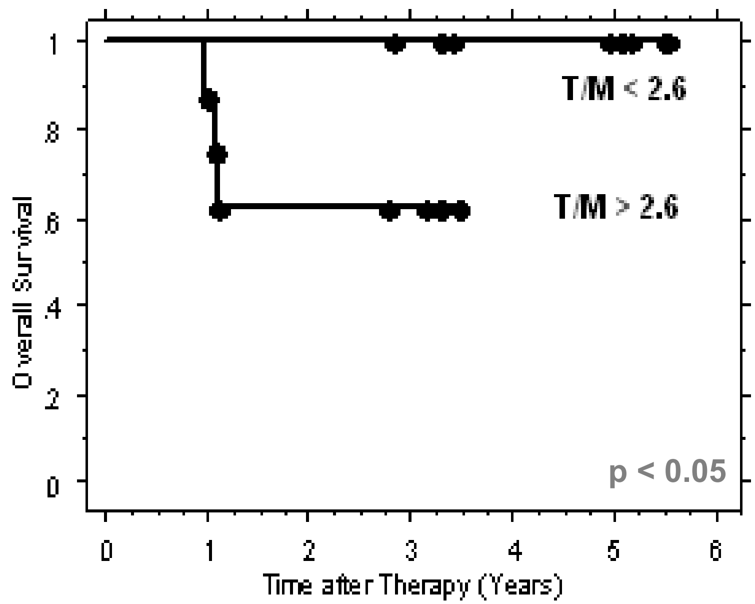
Progression-Free Survival



Overall Survival

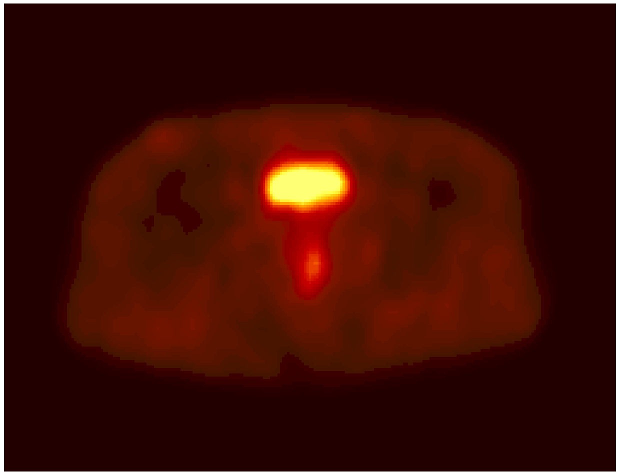


Dietz et al. Dis Colon Rectum 2008; 51:1641-8

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

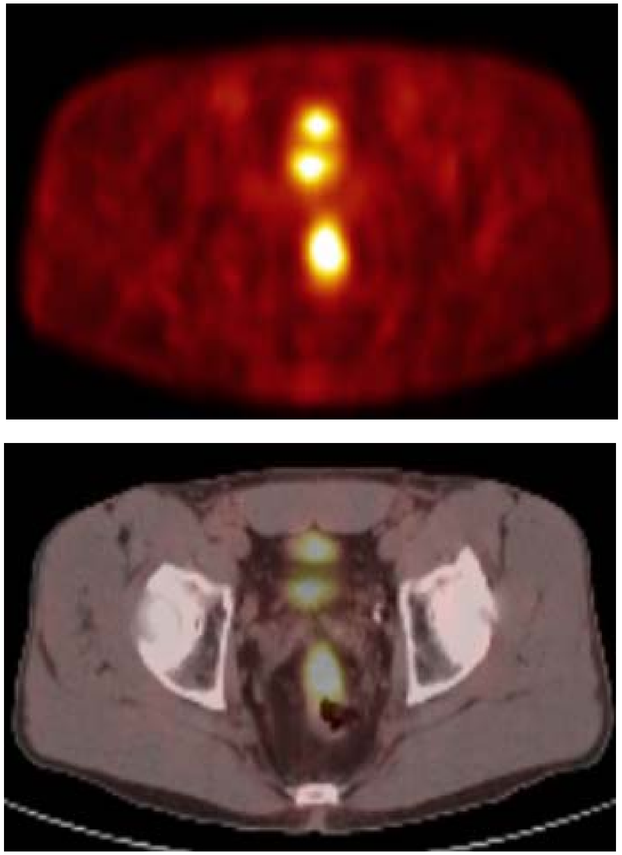
Responder

^{60}Cu-ATSM-PET



T/M = 2.3

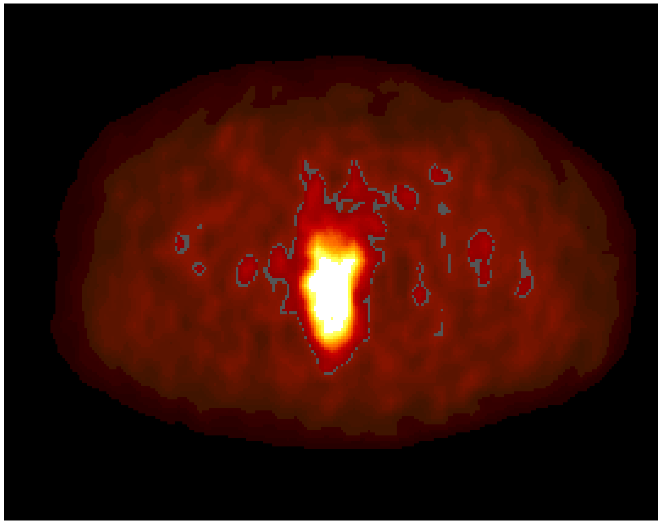
FDG-PET/CT



Measurement of Hypoxia with ^{60}Cu-ATSM-PET

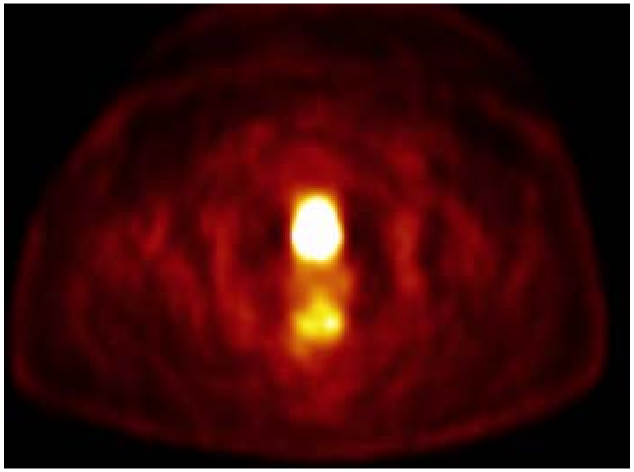
Non-Responder

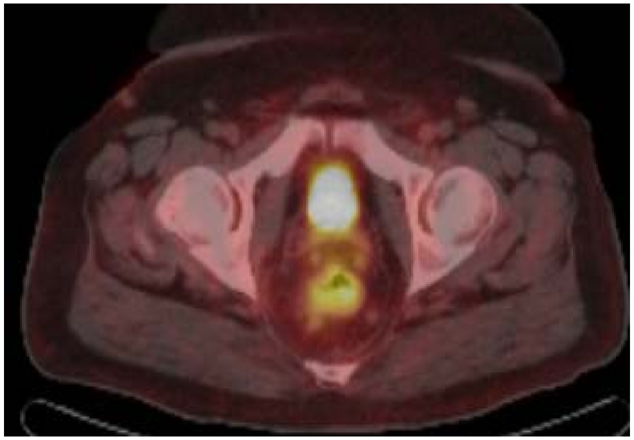
^{60}Cu-ATSM-PET



T/M = 3.1

FDG-PET/CT





Measurement of Hypoxia with ^{60}Cu-ATSM-PET

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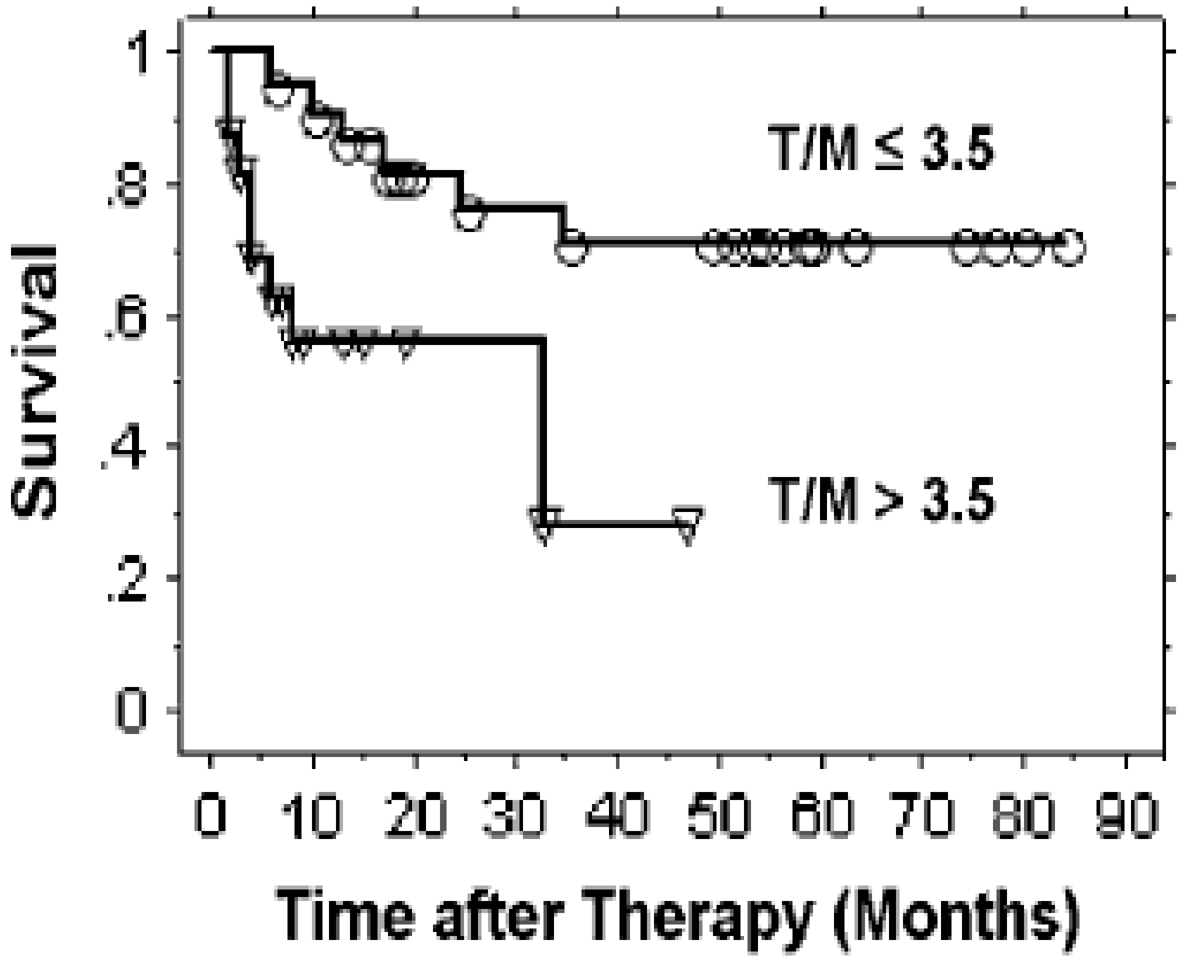
- Cervical cancer (n = 38, 1b1-IIIB)
- ^{60}Cu-ATSM uptake (T/M & peak slope index), but not FDG uptake was predictive of disease-free and overall survival
  - T/M of 3.5 or (5 %/min peak slope index) distinguished patients with better prognosis from those with poorer prognosis
  - T/M ratios and slope index revealed similar information
  - FDG uptake did not correlate with ^{60}Cu-ATSM uptake ($p = 0.9$)

Dehdashti et al., Int J Radiol Oncol Biol Ohys 2003; 55:1233 and J Nucl Med 2008; 49:201-5

Measurement of Hypoxia with ^{60}Cu-ATSM-PET



Dehdashti et al., JNM 2008; 49:210

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

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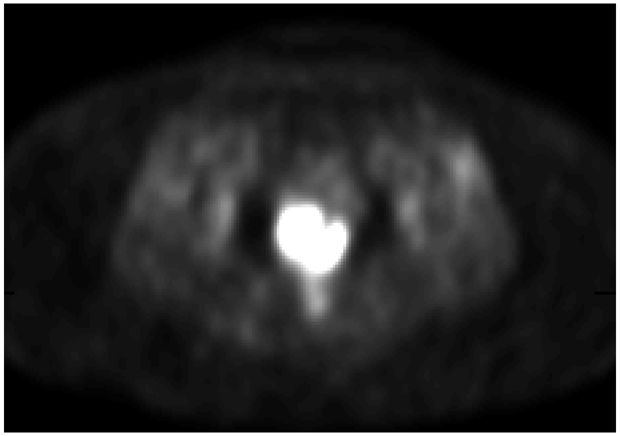
Responder

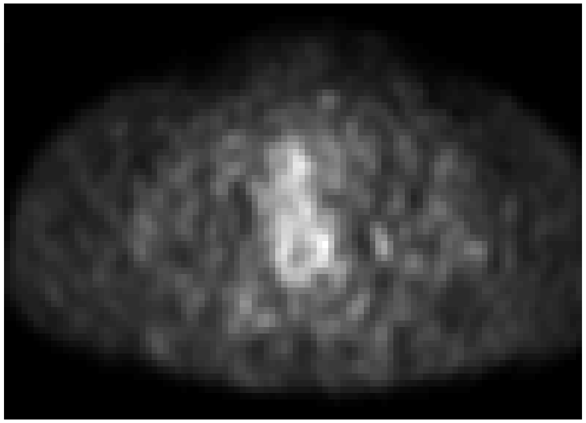
Pre-therapy

FDG-PET



SUV = 13.3

^{60}Cu-ATSM-PET

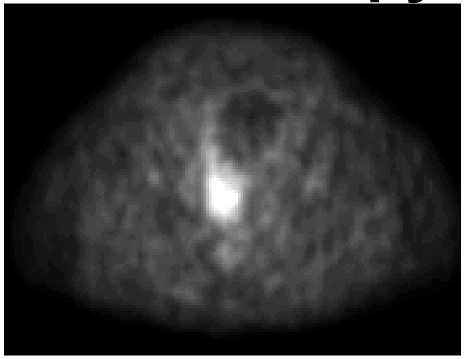


T/M = 2.3
Peak slope index = 2.2%/min

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

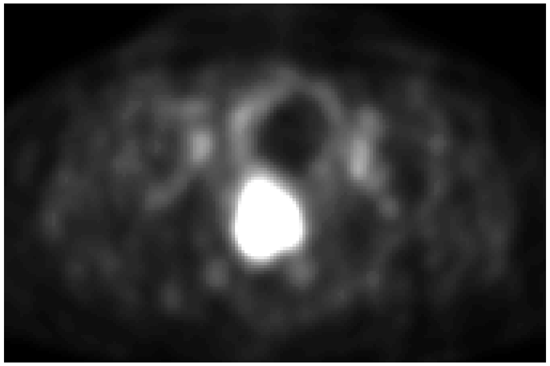
Non-Responder

^{60}Cu-ATSM-PET
Pre-therapy



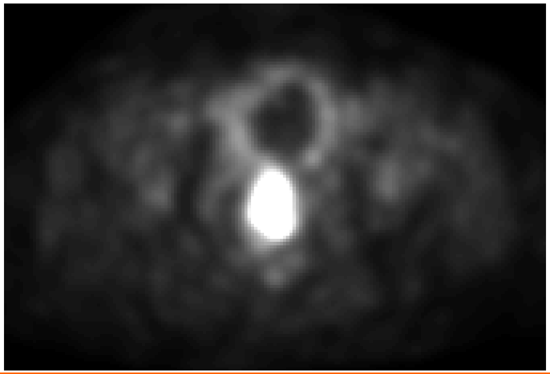
T/M = 5.1
Peak slope index = 7.1%/min

FDG-PET
Pre-therapy



SUV = 11.7

Post-therapy



Measurement of Hypoxia with ^{60}Cu-ATSM-PET

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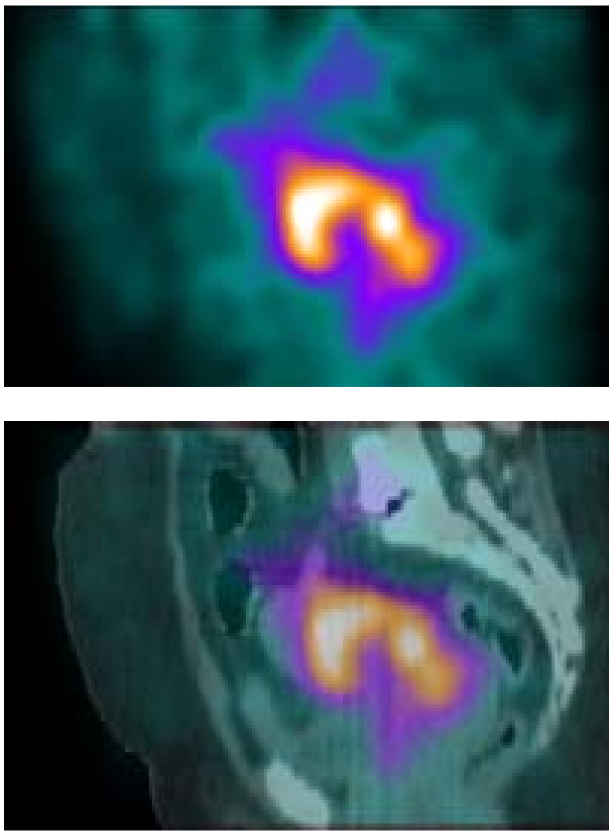
Non-Responder

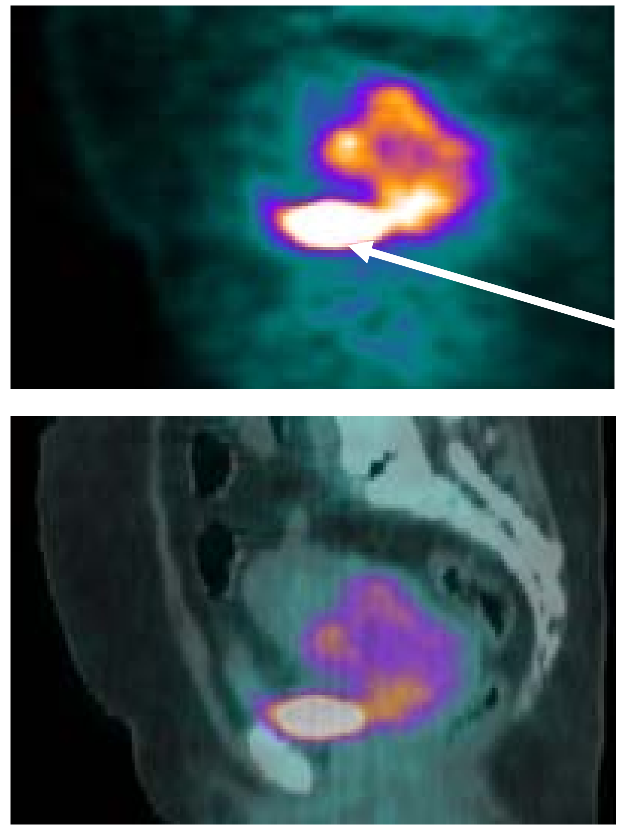
^{60}Cu-ATSM-PET/CT



T/M = 4.5

Sagittal

FDG-PET/CT



Urinary Bladder

SUV_{max} = 8.0

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

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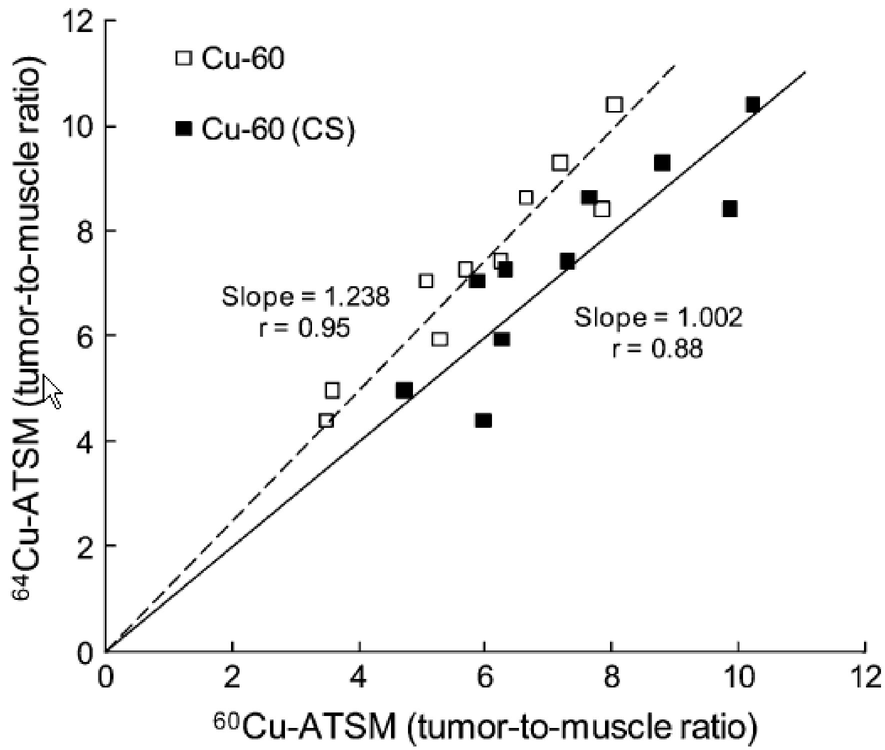
- **Assess quality of ^{60}Cu-ATSM-PET and ^{64}Cu-ATSM-PET images 10 patients with advanced cervical ca**
- **Patients studied with ^{60}Cu-ATSM-PET and ^{64}Cu-ATSM-PET in 2 separate days (range 1 - 9 days, averaged 5.8 days)**
  - **comparable pattern; but, ^{64}Cu-ATSM-PET images less noisy compared with ^{60}Cu-ATSM-PET (better T/B with ^{64}Cu-ATSM-PET)**
  - **Comparable uptake: T/M of 5.9 ± 1.6 for ^{60}Cu-ATSM and 7.3 ± 1.9 for ^{64}Cu-ATSM (r = 0.95, *P* < 0.0001)**

Lewis, et al., JNM 2008; 49:1177

^{60}Cu vs. ^{64}Cu-ATSM: Cervical Cancer



Cs= cascade subtraction correction

	Half-life
^{60}Cu	23.7 min
^{64}Cu	12.7 hr

Lewis, et al., JNM 2008; 49:1177

Measurement of Hypoxia with ^{60}Cu-ATSM-PET

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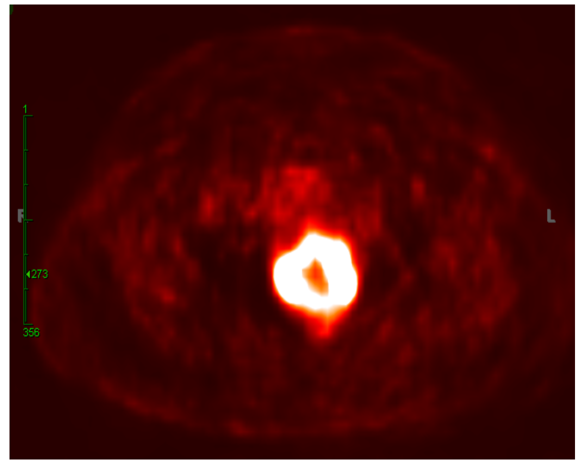
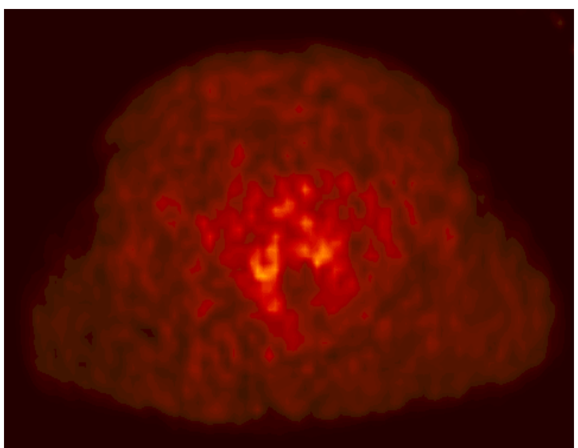
CT

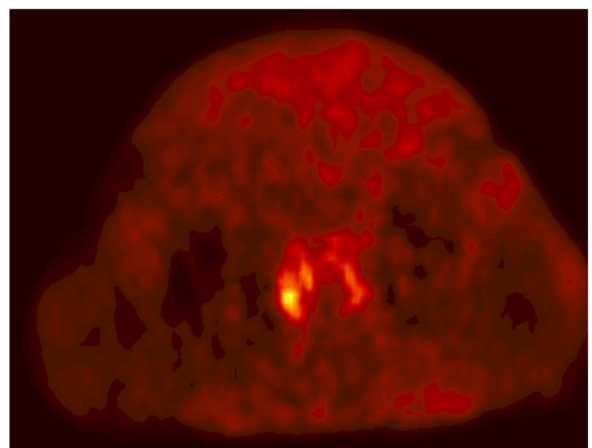
FDG-PET



^{60}Cu-ATSM-PET



^{64}Cu-ATSM-PET



Measurement of Hypoxia with ^{60}Cu-ATSM-PET

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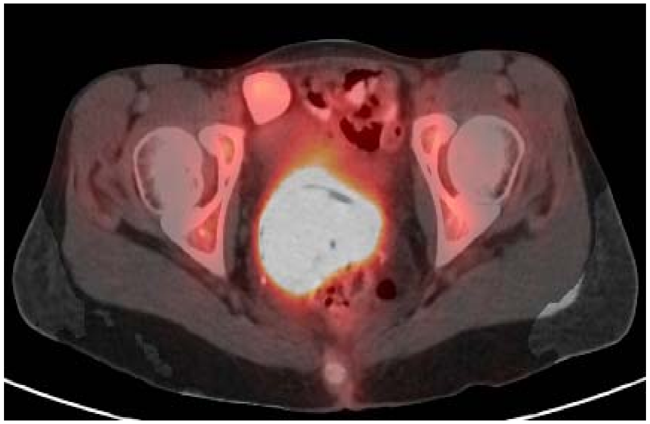
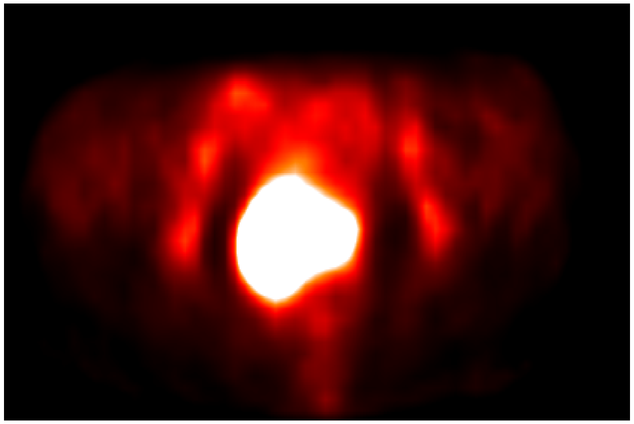
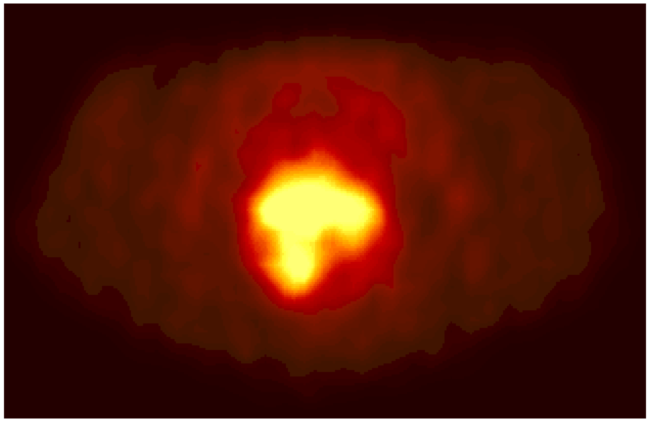
Fused FDG-PET/CT



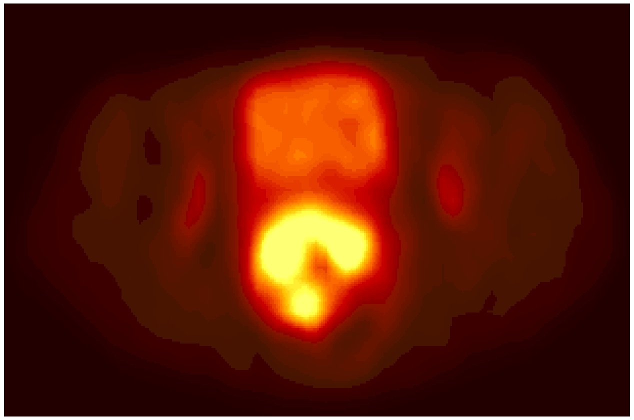
FDG-PET



^{60}Cu-ATSM-PET



^{64}Cu-ATSM-PET



Summary

- **Tumor hypoxia can successfully be measured by imaging ^{18}F-labeled nitroimidazoles hypoxic agents and ^{60}Cu-ATSM-PET**
  - **Feasible to study hypoxia of human tumors *in vivo* with ^{60}Cu-ATSM-PET and FMISO**
    - **^{60}Cu-ATSM has better image quality and higher hypoxic/normoxic activity**
  - **More clinical evidence that ^{60}Cu-ATSM-PET is:**
    - **Predictive of response to therapy**
    - **Predictive of survival**
  - **^{64}Cu-ATSM is an optimal substitute for ^{60}Cu-ATSM**

ACRIN 6682
Phase II Trial of ^{64}Cu-ATSM PET/CT in Cervical Cancer

Principal Investigator: Farrokh Dehdashti, MD

9/30/10

Background

- Tumor hypoxia is an important prognostic factor in cervical cancer and predicts for decreased overall and disease-free survival
- Hypoxic-measuring tools are needed:
  - To predict patient outcome
  - To select treatments on an individual basis
  - To evaluate early response to treatment

Study Hypotheses and Objectives

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Specific Hypotheses

- ^{64}Cu-ATSM-PET/CT distinguishes patients with poorer survival rate from those with better survival rate prior to initiation of therapy
- ^{64}Cu-ATSM-PET/CT provides unique prognostic information different from that revealed by known prognostic factors in an invasive squamous cell cervical cancer

Primary Objective

- To determine if higher ^{64}Cu-ATSM uptake is associated with lower progression-free survival

ACRIN 6682 Endpoints

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- **Primary Endpoint:** **to assess the relationship between ^{64}Cu-ATSM uptake in the primary cervical tumor and progression-free survival after chemoradiotherapy.**
- **Secondary Endpoints:** **to assess the relationship between ^{64}Cu-ATSM uptake and:**
  - **overall survival**
  - **rates of local recurrence and development of distant metastasis**
  - **frequency of complete metabolic response by FDG-PET**
  - **tumor volume and the frequency of lymph node metastasis at diagnosis**
  - **markers of tumor hypoxia assessed by immunohistochemistry on biopsy tissue from the primary tumor**

ACRIN 6682 Schema

Pre-therapy clinical whole-body FDG-PET/CT
Stages IB2 –IVA invasive squamous cell carcinoma, scheduled to undergo radiation therapy and concurrent cisplatin chemotherapy

↓

Pre-therapy pelvic ^{64}Cu-ATSM-PET/CT and analysis of tumor biopsy for hypoxic markers

↓

Concurrent chemoradiotherapy

↓

Clinical FDG-PET/CT three (3) months after completion of therapy

↓

Clinical follow-up for detection of recurrence and/or death

N=100, enrollment period=18 months

ACRIN 6682 Status

- **Protocol**
  - **Current Accrual: 7**
    - **Expecting New Sites for Accrual by late Fall 2010**
  - **Current Amendment approved by CTEP to address changes in Cu-ATSM kit formulation**
- **IND**
  - **Amendment submitted August 2010 to address changes in Cu-ATSM kit formulation and compounding process**
- **Radiopharmacy Support**
  - **Have continued work with radiopharmacy to support receiving the kit and compounding the dose for centers that do not have the pharmacy support to compound the dose (Readiness Oct-Nov 2010)**

ACRIN 6682 Status

ACRIN American College of Radiology Imaging Network

- **Site Currently Accruing:**
    - Washington University
    - University of Iowa (pending PET Qualification)
    - Weill Cornell (pending Cu-ATSM training)
- **Sites Pending Participation**
    - Dependent on 3rd party Radiopharmacy
        - Boston Medical
        - Fox Chase
        - MD Anderson
        - Clinical Radiologist
    - Able to receive Cu-ATSM in Site Pharmacy
        - City of Hope
        - Duke University
        - University of Wisconsin
        - Johns Hopkins
        - Memorial Sloan-Kettering Cancer Center
        - Wake Forest Medical Center
        - University of Southern California
        - Wayne State University
        - University of Alabama, Birmingham

Thank You