Biomarcatori e Tecniche di Diagnostica per Immagini nella Valutazione dell'Infiammazione in Oncologia

RUOLO DELL'IMAGING PET PER L'IDENTIFICAZIONE DELL'IPOSSIA TUMORALE

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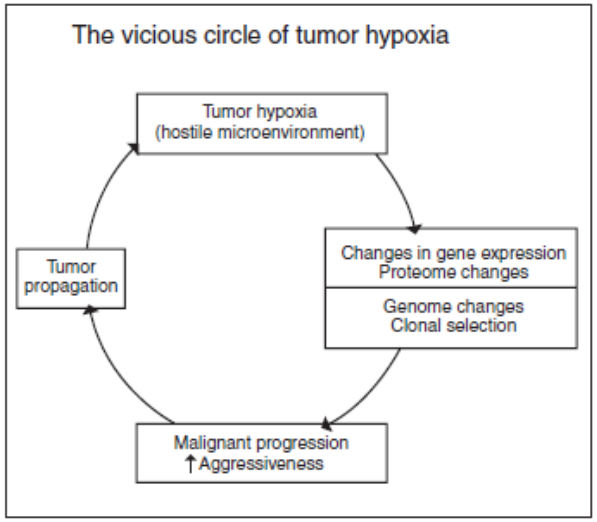
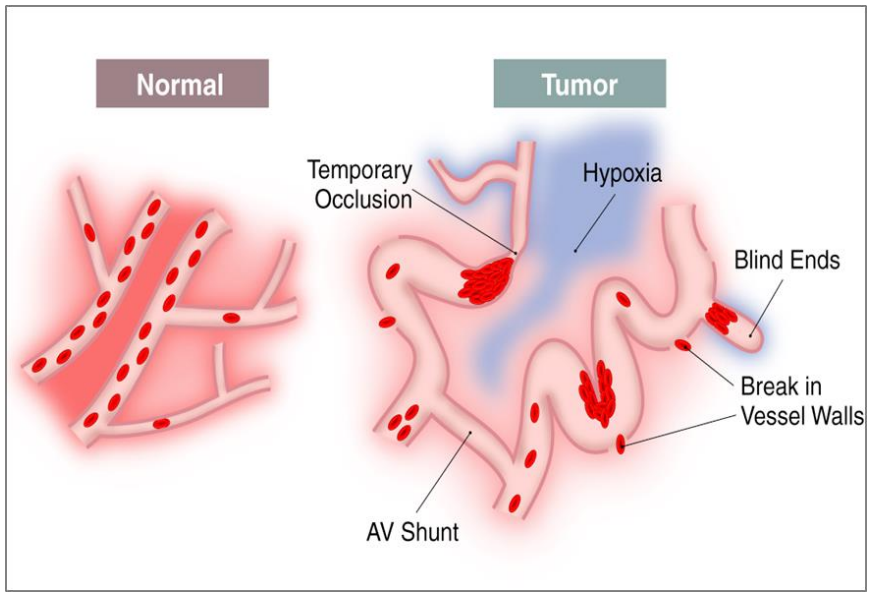
22 febbraio 2019

Tumour Hypoxia: needs and priorities

Negative prognostic factor

- Aggressive tumour phenotype
- Resistance to therapy





(Brown J.M. et al. Nature Reviews Cancer 2004)

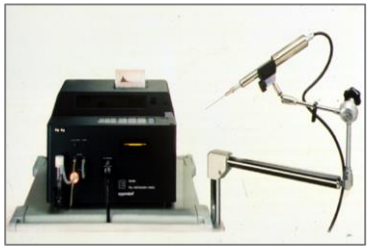
The heterogeneity of cancer and the lack of a universal hypoxia detection tracer/technique represent a challenge for the correlation of hypoxia with treatment planning and prognosis

Why investigating tumour hypoxia?

- To characterize **tumor heterogeneity**
- To evaluate **tumor prognosis**
- To predict **treatment response**
- To **select patients** who might benefit from "hypoxia-directed therapies" or intensive treatment approach (i.e RT with boost on hypoxic areas; hypoxia as therapeutic target)

How to investigate tumor hypoxia?

Direct oxygenation measurement



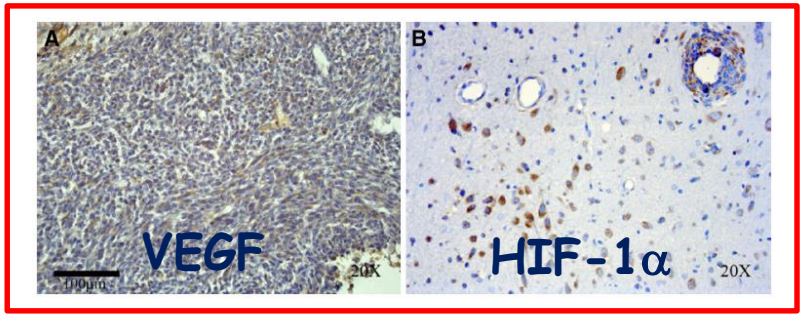
Polarographic oxygen (Eppendorf) electrodes

Limits of direct oxygenation measurements:

- Invasiveness
- Feasibility to only superficially assessable tumors
- Oxygenation status solely in a particular region
- Cannot reliably monitor hypoxia levels over time

How to investigate tumor hypoxia?

Hypoxia biomarkers: exogenous (pimonidazole) and endogenous (Hypoxia inducible factor -HIF1α, Carbonic anhydrase IX -CA-IX, Vascular endothelial growth factor -VEGF and Glucose transporter1 -GLUT-1) hypoxia markers



Equivocal results regarding the correlation between expression of hypoxia biomarkers and patient outcome (differential expression of these biomarkers in specific tumor microenvironment)

In vivo Imaging

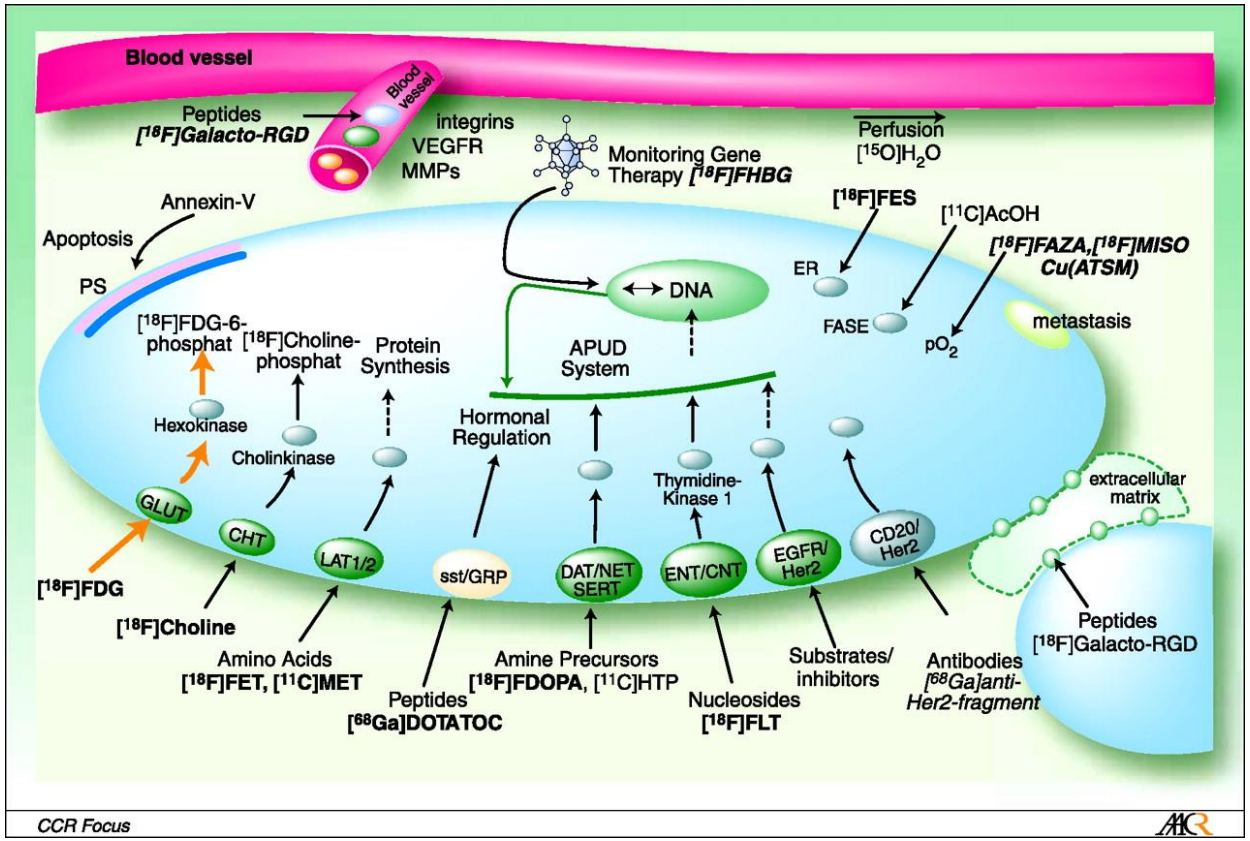
Available bioimaging modalities: principal properties and applications

Technique / Feature	Optical	MRI	PET	SPECT	X-ray CT
EM radiation	Visible / NIR	Radiowaves	High energy γ rays	Lower energy γ rays	X-ray
Spatial resolution	15-1000 mm	4-100 µm 1 mm fMRI	1-2 mm	1-2 mm	12-50 µm 50-200 µm
Depth	< 1cm	No limit	No limit	No limit	No limit
Sensitivity to probe	µg / mg	µg / mg	ng	ng	-
Key use	Visualization of cells	Anatomical / functional brain imaging	Metabolic imaging		Lung and bone tumor imaging

Mirabello et al. Frontiers in Chemistry, 2018

Molecular Imaging (PET)

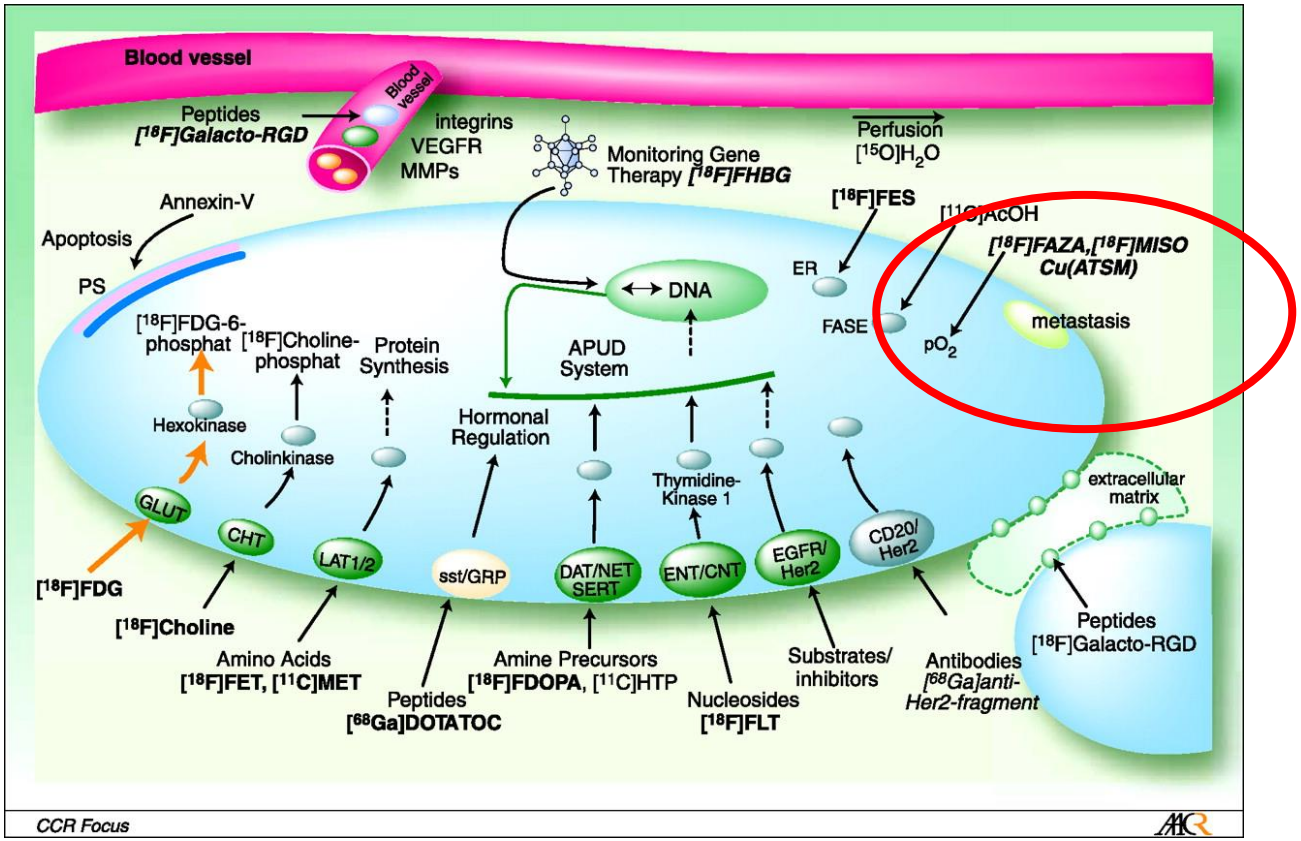
PET tracers



Clinical Cancer Research
AACR American Association for Cancer Research

Molecular Imaging (PET)

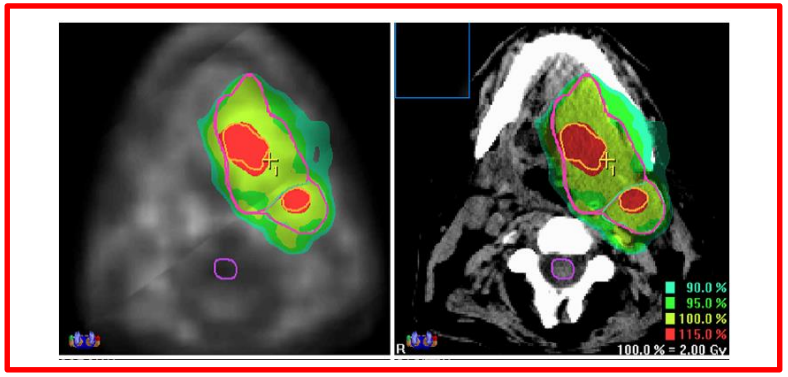
PET tracers



Clinical Cancer Research
AACR American Association for Cancer Research

PET to investigate tumor hypoxia

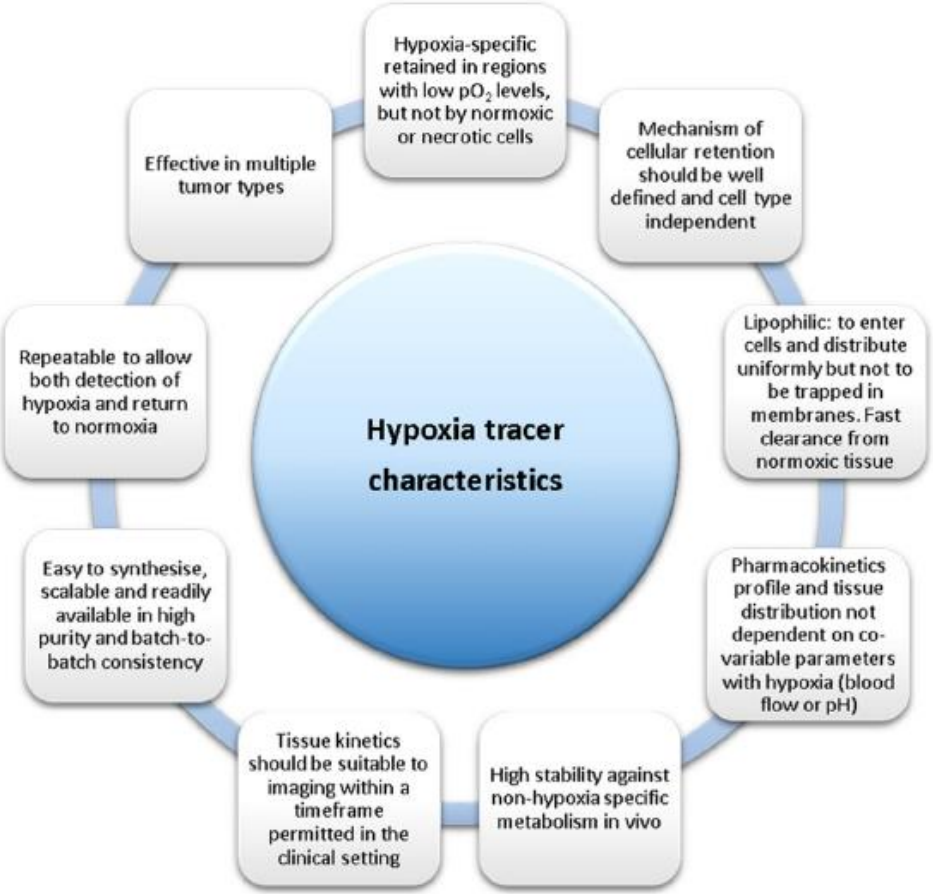
PET Imaging



- Non invasive (Clinically feasible)
- Identification and quantification of regional tumor tissue hypoxia in superficial and deep tumors
- Representative of global tumor heterogeneity

In vivo Imaging

Necessary criteria to be considered in the development of a hypoxia tracer. Ideal hypoxia tracer characteristics



Mirabello et al. Frontiers in Chemistry, 2018

PET to investigate tumor hypoxia

TABLE 1 | Summary of clinical imaging findings and recommendations for the use of most common hypoxia tracers.

Tumor / Tracer	[^{18}F]FMISO	[^{18}F]F-HX4	[^{18}F]FAZA	[^{18}F]FETNIM	[^{18}F]F-EF5	[^{18}F]F-RP170	^{64}CuCu(ATSM)
Brain							
Head and neck							
Breast							
Sarcoma							
Lung							
Lymphoma							
Renal							
Liver							
Colorectal							
Bladder							
Cervical							
Prostate							

Adapted from Fleming et al. (2014).

Yes, good clinical data obtained.

Recommended favorable preclinical/metabolic data.

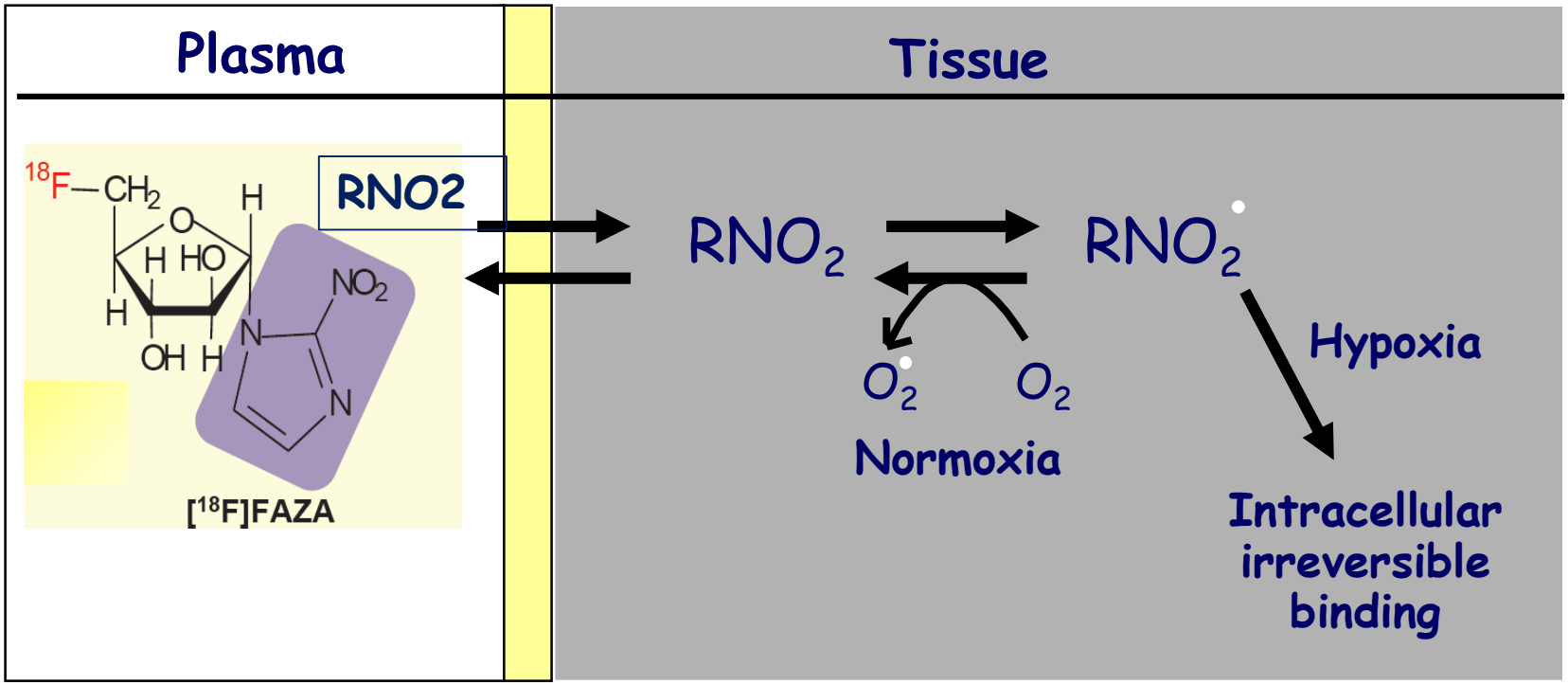
Not recommended, unfavorable preclinical/metabolic data.

No, poor clinical data.

Mirabello et al. Frontiers in Chemistry, 2018

Tumour hypoxia PET Imaging: 18F-FAZA

Nitroimidazoles (fluoroazomycin arabinoside)



RNO2: Nitro functional group

Bioreductive metabolism of nitroimidazoles: accumulation due to reduction

18F-FAZA hypoxia PET Imaging: Human Studies

Publication	No. Patients	Tumor Site (n)	Definition of Hypoxic Volume	Percentage of Patients With Increased ^{18}F-FAZA Uptake (ie, hypoxia)
Grosu et al[16] and Souvatzoglou et al[43]	18	Head and neck (18)	T/M ≥1.5*	83
Postema et al[44]	50	Head and neck (9)	Visual inspection and T/B ratio ≥1.2	66
		Lymphoma (21)		14
		High-grade glioma (7)		100
		Lung (13)		54
Schuetz et al[45]	15	Cervix (15)	T/M ≥1.2†	33
Shi et al[46]	5	Head and neck (5)	Different kinetic models	80
Garcia-Parra et al[47]	14	Prostate (14)	T/B ratio‡	0
Mortensen et al[48]	40	Head and neck (40)	T/M ≥1.4§	63
Bollineni et al[49]	11	Lung (11)	T/B ratio ≥1.2 and T/B ratio ≥1.4	100

Halmos et al. Clin Nuc Med 2014

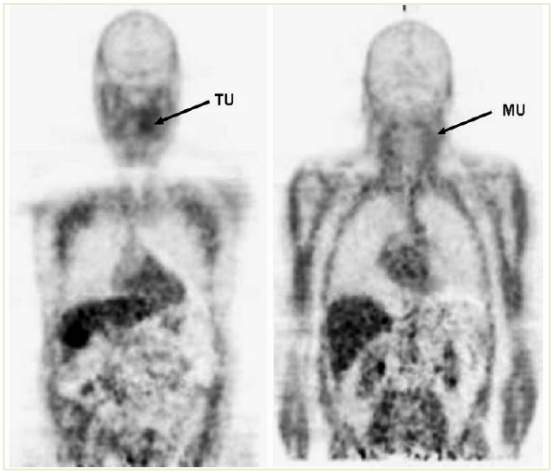
- Savi A et al. First evaluation of PET based **human biodistribution** and dosimetry of 18F-FAZA, a tracer for imaging tumor hypoxia. J Nucl Med. **2017**;58:1224-1229.
- Mapelli P et al. Concomitant **Lung Cancer** and **Gastrointestinal Stromal Tumor**: First Report of Hypoxia Imaging With 18F-FAZA PET/CT. Clin Nucl Med. **2017**.
- Mapelli P et al. Hypoxia 18F-FAZA PET/CT imaging in **lung cancer** and **high-grade glioma**: open issues in clinical application. Clin Transl Imaging **2017**
- Mapelli P et al. 18F-FAZA PET/CT Hypoxia Imaging of **High-Grade Glioma** Before and After Radiotherapy. Clinical Nuclear Medicine **2017**
- Mapelli P et al. 18F-FAZA PET/CT in the preoperative evaluation of **NSCLC**: comparison with 18F-FDG and immunohistochemistry. Curr Radiopharm. **2018**

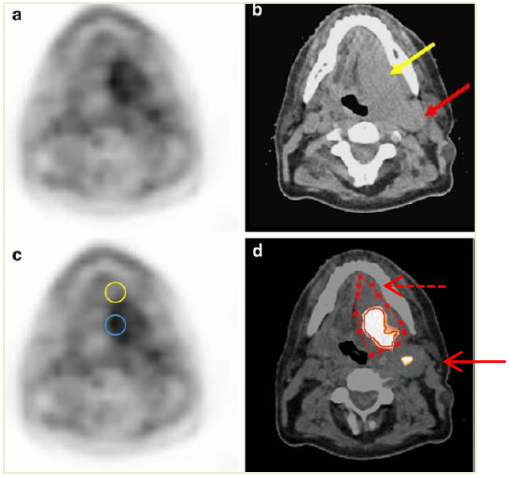
Eur J Nucl Med Mol Imaging
DOI 10.1007/s00259-007-0424-3

ORIGINAL ARTICLE

Tumour hypoxia imaging with [^{18}F]FAZA PET in head and neck cancer patients: a pilot study

M. Souvatzoglou · A. L. Grosu · B. Röper · B. J. Krause · R. Beck · G. Reischl · M. Picchio · H.-J. Machulla · H.-J. Wester · M. Piert

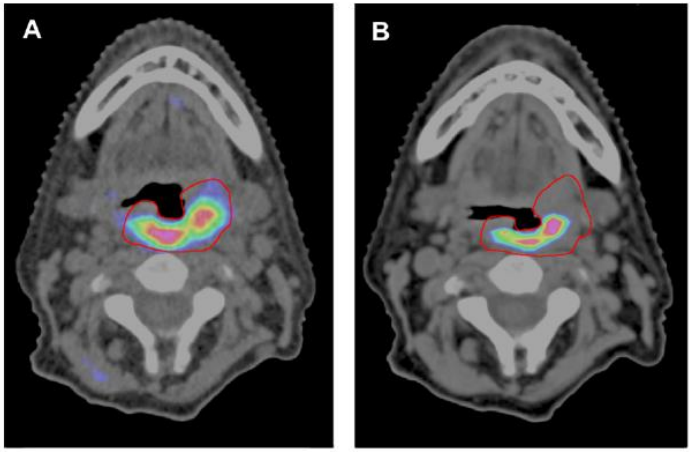




Feasible
Adequate image quality
Evaluation of hypoxic subvolume

Souvatzoglou et al. EJNMMI 2007

FAZA PET/CT hypoxia imaging in patients with squamous cell carcinoma of the head and neck treated with radiotherapy: Results from the DAHANCA 24 trial



Pre-RT GTV: 30.9cm3

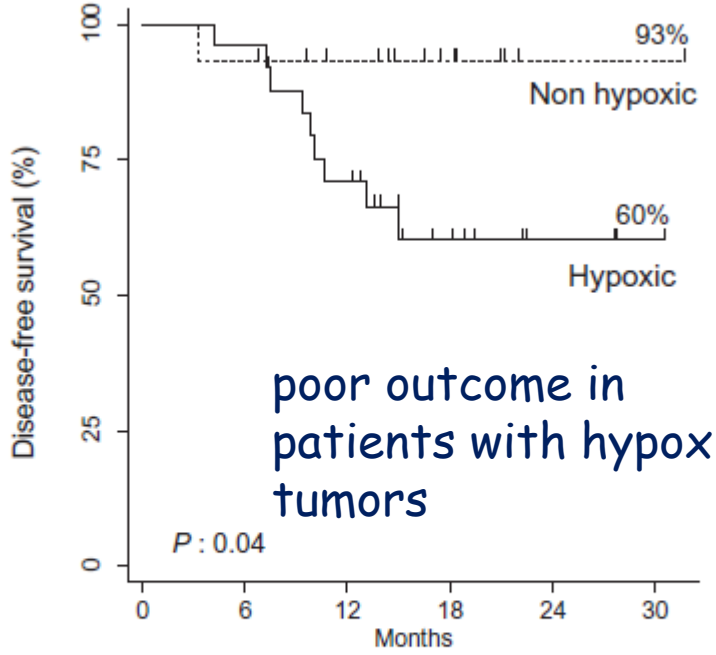
After 12 Gy GTV: 13.7cm3

stable localization

Large inter-tumor variability

FAZA PET/CT imaging as a suitable assay with prognostic potential for detection of hypoxia in HNSCC

40 HNSCC cancer pts pre and during RT

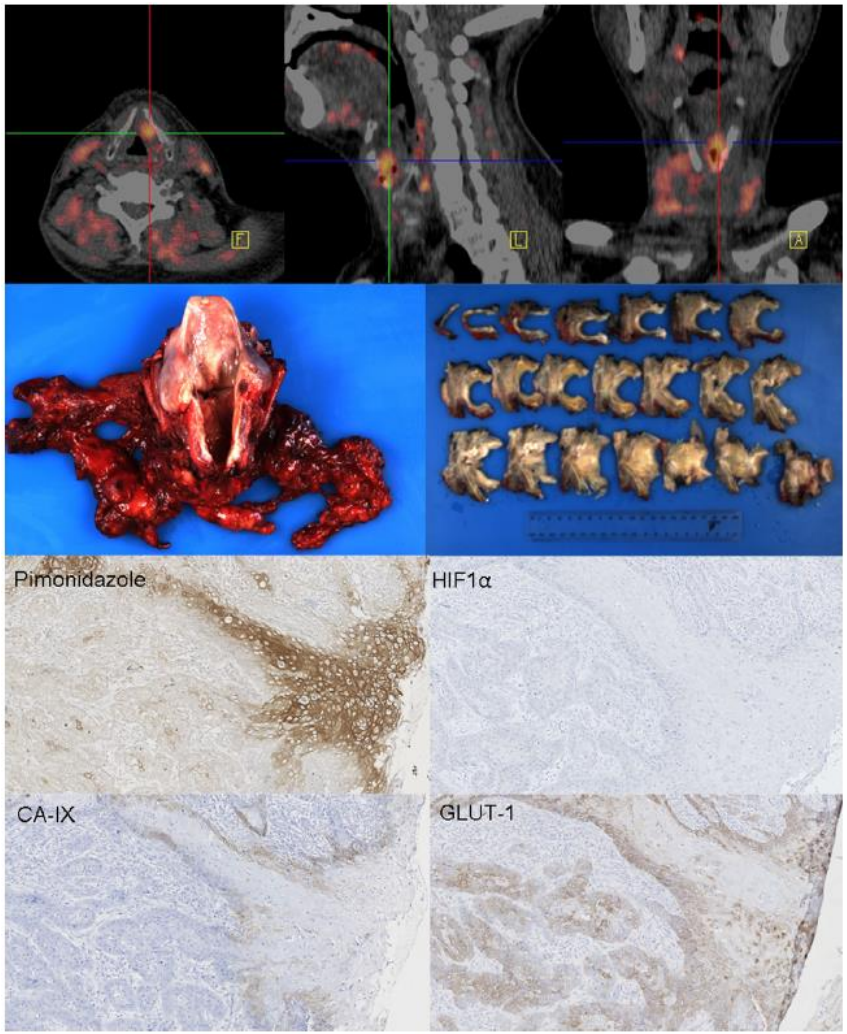


poor outcome in patients with hypoxic tumors

Fig. 4. Disease free survival of patients with hypoxic versus non hypoxic tumors as assessed by FAZA PET.

Mortensen et al. Radiother Oncol 2012

Assessment of hypoxic subvolumes in laryngeal cancer with ^{18}F-fluoroazomycinarabinoside (^{18}F-FAZA)-PET/CT scanning and immunohistochemistry



11 Laringeal cancer pts pre Surgery

FAZA-PET vs immunohistochemical examination for exogenous (pimonidazole) and endogenous (HIF1a, CA-IX and GLUT-1) hypoxia markers

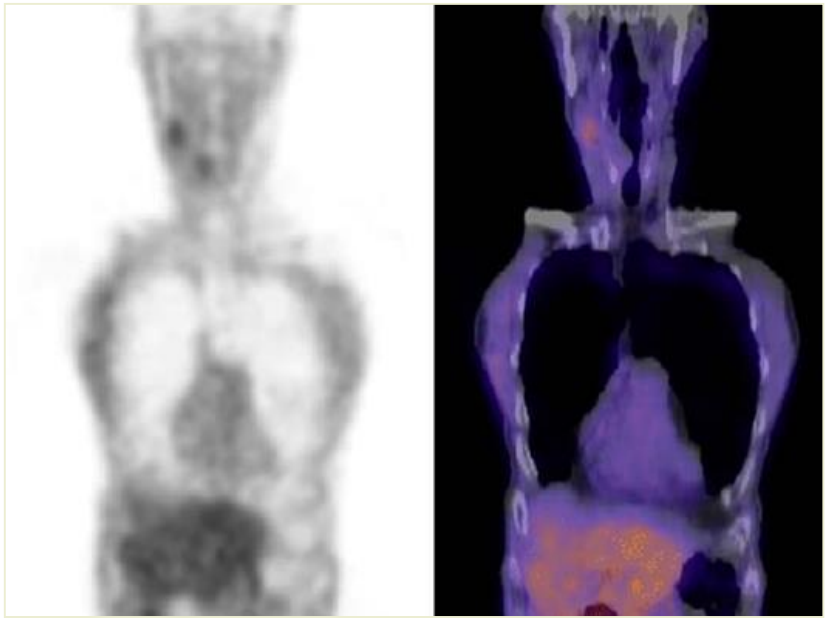
No clear association between FAZA and Hypoxia markers

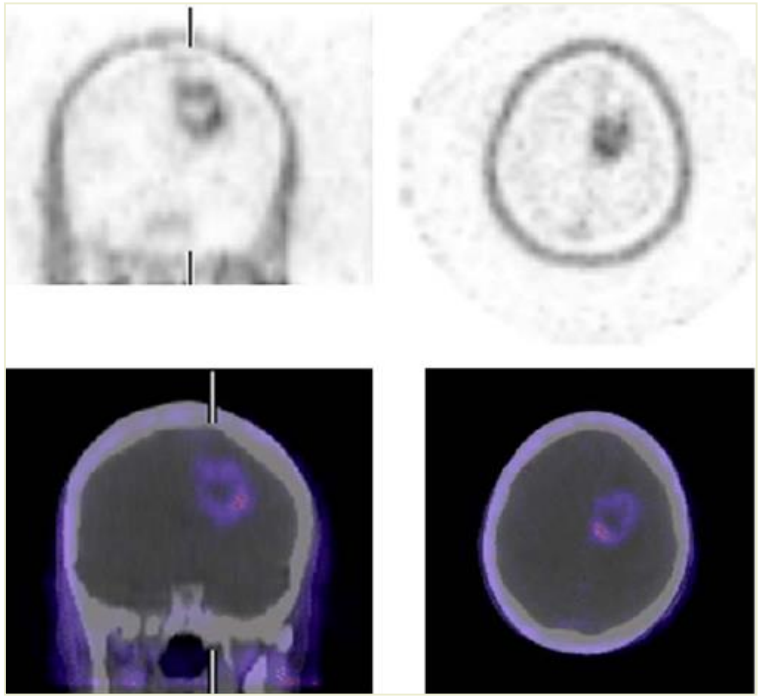
Further study is required to investigate the benefit of 18F-FAZA-PET imaging in RT planning

De Bruin et al. Radiother Oncol 2015

Initial results of hypoxia imaging using 1-α-D-(5-deoxy-5-[^{18}F]-fluoroarabinofuranosyl)-2-nitroimidazole (^{18}F-FAZA)

50 pts: Primary or supected metastatic HNSCC, SCLC or NSCLC, malignant lymphoma, high-grade gliomas





18F-FAZA PET/CT is feasible, no side effects, very useful to image hypoxia (especially in gliomas)
Very promising considering its T/B ratio

Postema et al. EJNMMI 2009

Rationale

- Lack of studies matching hypoxia in specific hypoxic subvolumes of whole tumour specimen (heterogeneity of tumor hypoxia within the tumour mass)
- No consensus over the interpretation and analysis of hypoxia-positive areas

Personalized Image-guided treatment

OSR Funded Grants on Hypoxia PET Molecular Imaging (18F-FAZA)

Research Grants

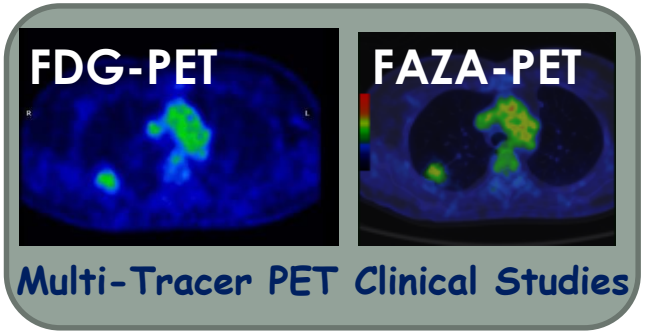
Respiratory gated PET/CT technique and FAZA for the evaluation of hypoxia in **NSCLC (FAZA-lung)**
Ricerca Finalizzata GR-1575612 - PI: M. Picchio

Prognostic value of FAZA PET/CT in **glioma patients** referred to chemo-radiation therapy: comparison with MRI and correlation with molecular markers of hypoxia **(FAZA-glioma)**
AIRC IG 2014 Id.1524 - PI: M. Picchio

The role of 18F-FAZA PET Imaging technique in detecting LN metastases in Renal cell carcinoma pts **(FAZA RCC LNI)**
GR-2013-02357486 - Group Leader: M. Picchio

Project FAZA-lung cancer

Phase 1 clinical Study



- Biodistribution and dosimetry evaluation
- Evaluation of tumor characterization and heterogeneity by comparing FDG and FAZA-PET/CT with immunoistochemical hypoxia markers of the surgical specimen
- Imaging Protocol optimisation

First Evaluation of PET-Based Human Biodistribution and Dosimetry of ^{18}F-FAZA, a Tracer for Imaging Tumor Hypoxia

Annarita Savi[1], Elena Incerti[1], Federico Fallanca[1], Valentino Bettinardi[1], Francesca Rossetti[2], Cristina Monterisi[3], Antonia Compierchio[1], Giampiero Negri[2], Piero Zannini[2], Luigi Gianolli[1], and Maria Picchio[1]

[1]Nuclear Medicine Department, IRCCS San Raffaele Scientific Institute, Milan, Italy; [2]Thoracic Surgery Department, IRCCS San Raffaele Scientific Institute, Milan, Italy; and [3]University of Milano–Bicocca, Milan, Italy

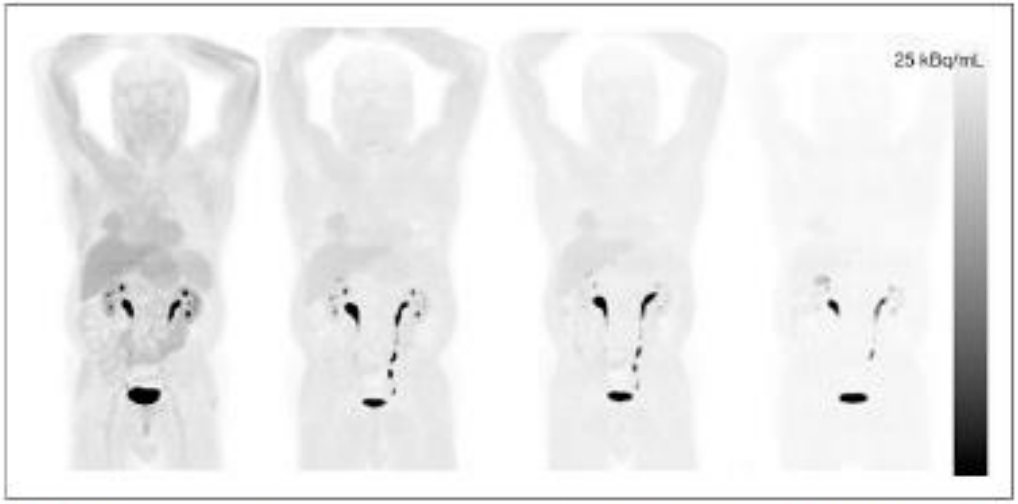


FIGURE 1. Coronal images of representative patient at 10, 60, 120, and 240 min (from left to right) after injection of ^{18}F-FAZA.

The biodistribution and internal dosimetry profiles for ^{18}F-FAZA in humans indicate a favorable radiation risk profile, thus making the use of whole-body ^{18}F-FAZA PET/CT feasible for evaluating clinical hypoxia and safe for consecutive studies when clinically required.

TABLE 2
Residence Times of ^{18}F-FAZA in Measured Source Organs

Source organ	Time
Brain	0.014 ± 0.002
Gallbladder content	0.019 ± 0.001
Intestine	0.013 ± 0.005
Heart content	0.018 ± 0.002
Kidneys	0.025 ± 0.003
Liver	0.110 ± 0.019
Lungs	0.036 ± 0.011
Muscle	1.090 ± 0.180
Red marrow	0.034 ± 0.005
Spleen	0.011 ± 0.003
Urinary bladder content at 2 h	0.055 ± 0.009
Urinary bladder content at 4 h	0.081 ± 0.008
Remainder of body at 2 h	1.130 ± 0.320
Remainder of body at 4 h	1.100 ± 0.310

Data are hours (mean ± SD; n = 5 patients).

Savi et al. J Nucl Med 2017

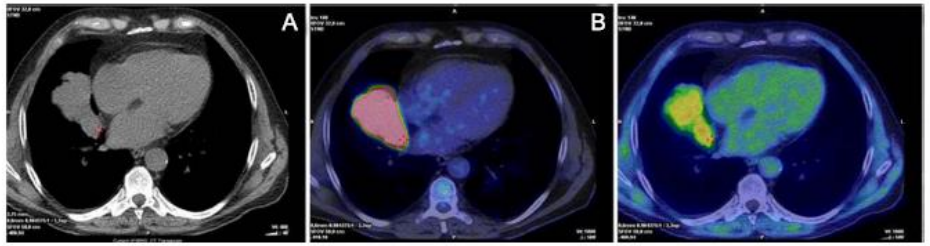
Current Radiopharmaceuticals, 2018, *11*, 50-57

RESEARCH ARTICLE

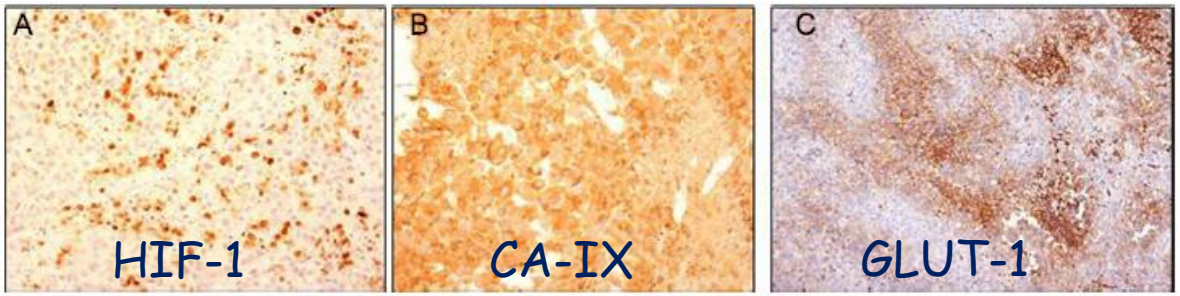
BENTHAM SCIENCE

18F-FAZA PET/CT in the Preoperative Evaluation of NSCLC: Comparison with 18F-FDG and Immunohistochemistry

Paola Mapelli[1,2], Valentino Bettinardi[1], Federico Fallanca[1], Elena Incerti[1], Antonia Compierchio[1], Francesca Rossetti[3], Angela Coliva[1], Annarita Savi[1], Claudio Doglioni[2,4], Giampiero Negri[2,3], Luigi Gianolli[1] and Maria Picchio[1,2,*]

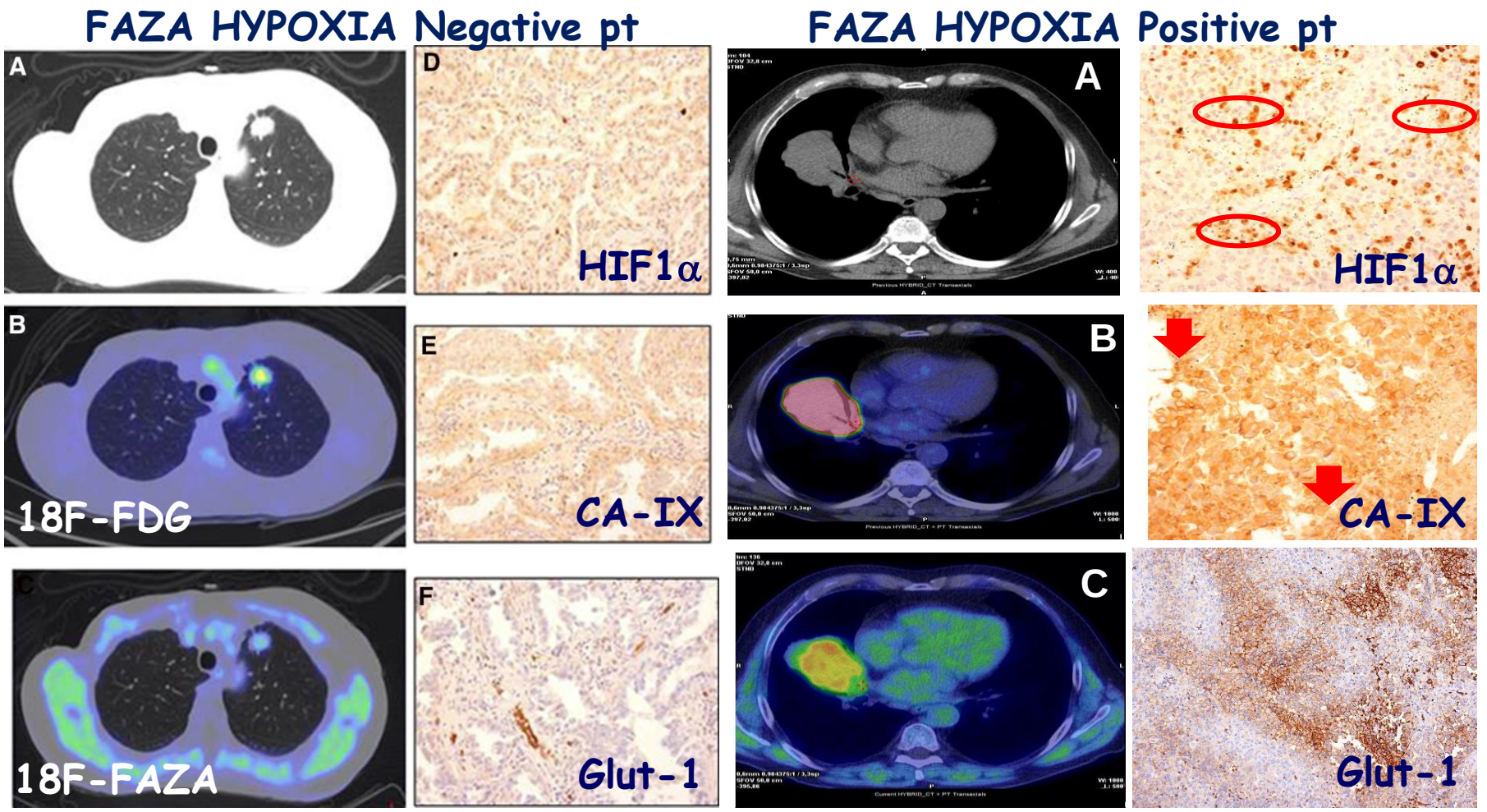


Immunohistochemical analysis supported the presence of hypoxia as seen on 18F-FAZA PET/CT images



Moderate nuclear reactivity in 20% of neoplastic cells for HIF-1 along with expression by numerous intratumoural, inflammatory cells, mainly macrophages (*A*); **CA-IX** stained 70% of neoplastic cells (B) with moderate intensity and **GLUT-1** showed intense staining in 30% of neoplastic cells (*C*).

Tumor characterization and heterogeneity
FDG-PET - FAZA-PET - IHC



Adapted from Mapelli P, et al. Clin and Transl Imaging, 2017

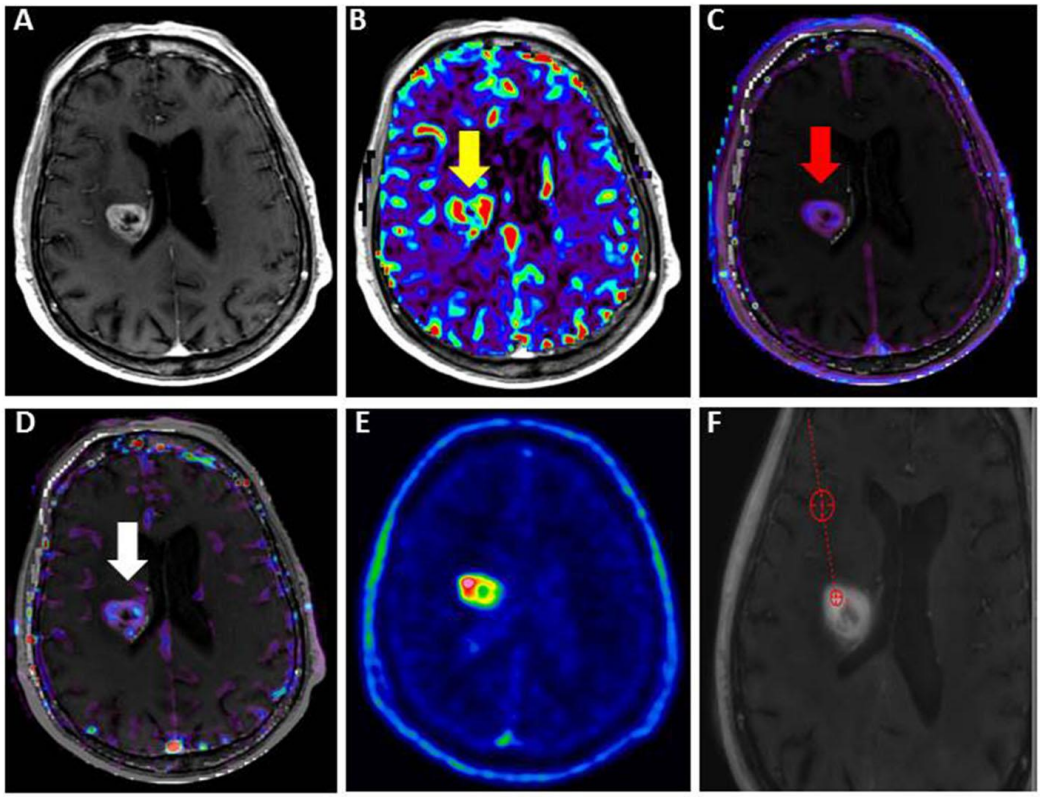
Hypoxia PET Imaging in lung cancer

- Safe dosimetry and adequate biodistribution for clinical studies
- Good correlation with immunohistochemistry
- Potential role to adopt hypoxia-directed trp approaches guided by non invasive PET Imaging methods

Project FAZA-glioma

- Guiding tumour sampling (comparison with standard MRI-guided sampling)
- Planning personalized radiation treatment (comparison with standard MRI-based treatment planning)
- Defining the spatial concordance between disease pseudoprogression/radionecrosis and hypoxia
- Predicting patient outcome

Project FAZA-glioma
FAZA-PET/MR to guide biopsy



A: MRI (T1-post contrast)

B: High values of relative Cerebral Blood Volume (rCBV)

C: Transfer Constant (Ktrans)

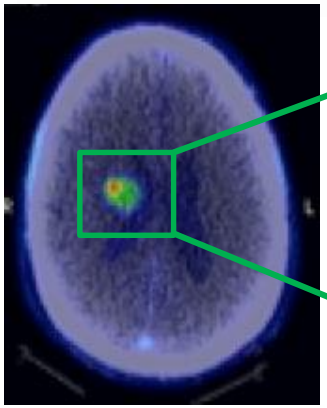
D: Fractional Plasma Volume (Vp)

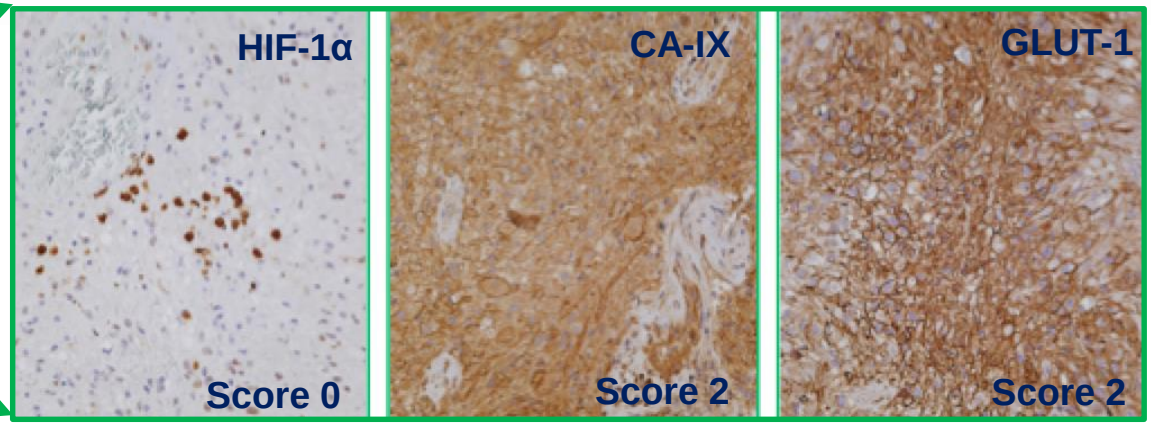
E: 18F-FAZA PET uptake

F: Image co-registration to guide biopsy

Mapelli P, et al. Clin and Transl Imaging, 2017

Project FAZA-glioma
FAZA-PET vs IHC





Patients	HIF-1α	CA-IX	KI67	GLUT-1	Mean number of vessels/3HPF	IHC CD31
1	1	3	5%	4	11	1
2	1	1	20%	2	32	2
3	1	1	5%	3	19	1
4	1	1	10%	2	16	1
5	1	3	25%	3	12	1
6	1	2	15%	3	21	2
7	1	1	20%	2	7	1
8	1	1	15%	2	15	1
9	1	2	40%	4	44	3
10	1	3	20%	3	42	3
11	1	0	25%	3	25	2
12	1	1	3%	1	13	1
13	2	1	30%	2	39	2
14	1	0	10%	0	13	1
15	0	0	15%	1	27	2
16	1	1	35%	2	13	1
17	1	4	7%	3	11	1

IHC score system:

0=0-25%
1=25-50%
2=50-75%
3=75-100%

Project FAZA-glioma
Correlation FAZA-Hystopathology

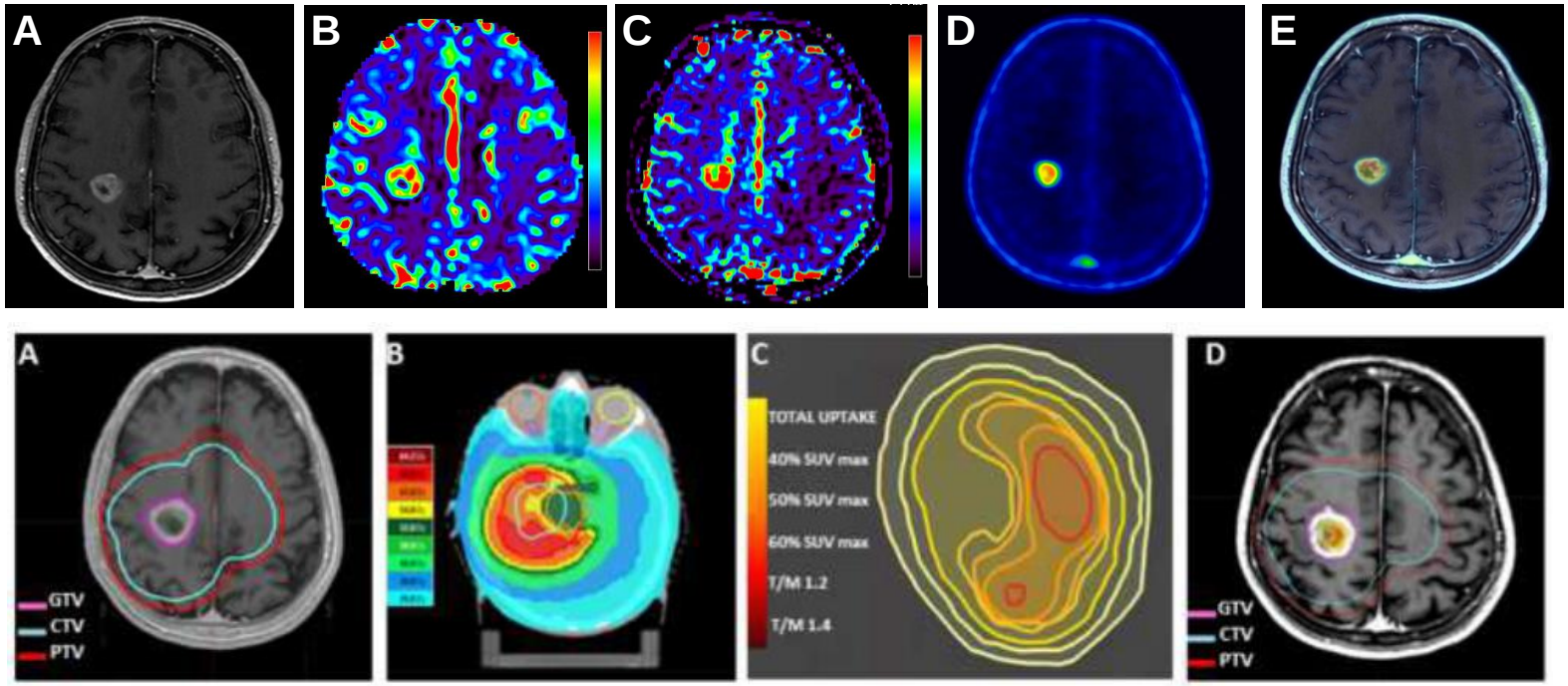
Surgical subgroup (n=7)

	Correlation with PET-derived parameters					
	SUVmax	SUVmean40	SUVmean50	SUVmean60	MTV40-50-60	HV 1.2-1.3-1.4
CA-IX	p=0.0002	p=0.0058	p=0.009	p=0.0153	P=0.0424	p=0.0058
HIF-1a	ns	ns	ns	ns	ns	ns
GLUT1	ns	ns	ns	ns	ns	ns
Ki-67	ns	ns	ns	ns	ns	ns
CD31	ns	ns	ns	ns	ns	ns

Biopsy subgroup (n=10)

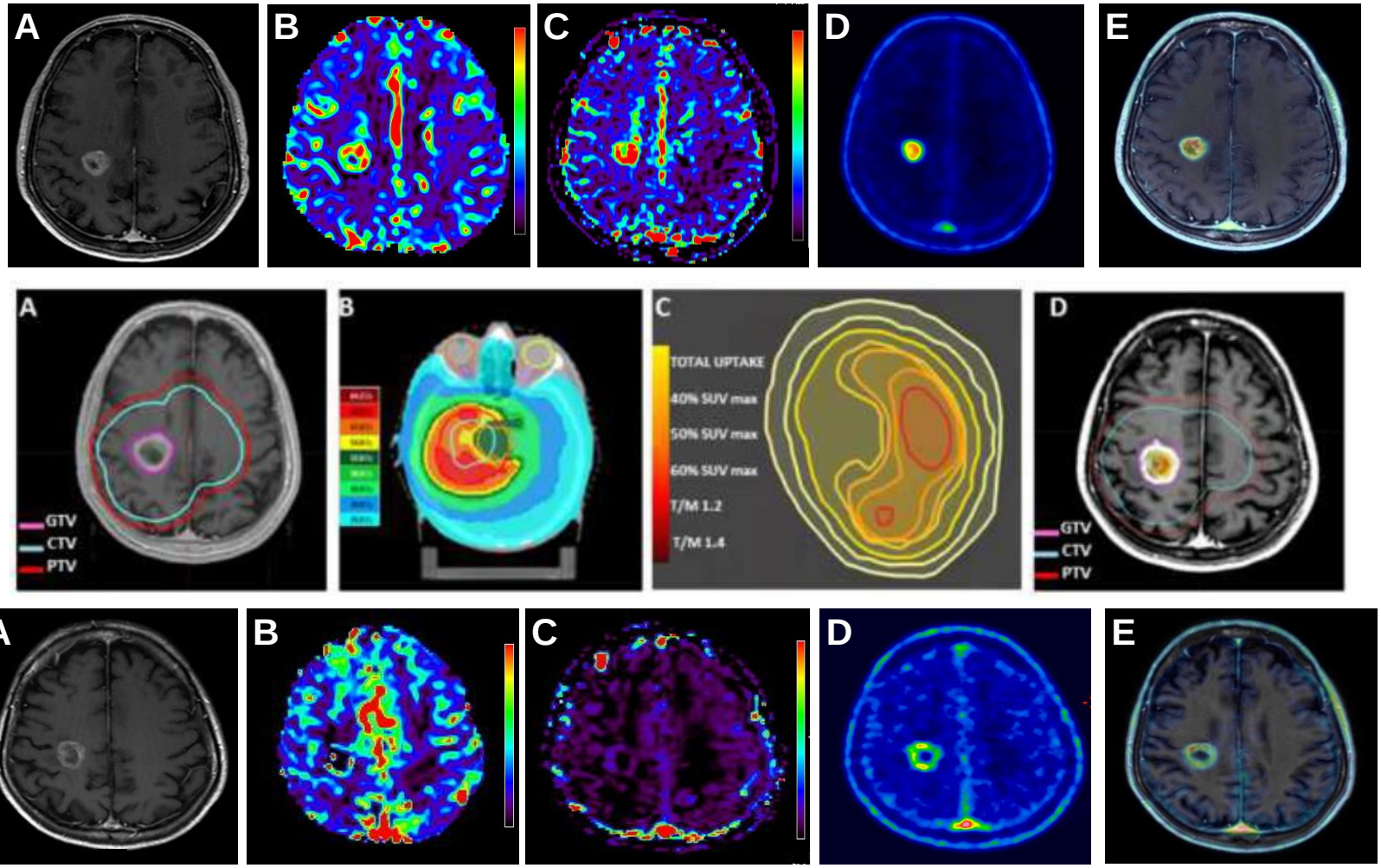
	Correlation with PET-derived parameters					
	SUVmax	SUVmean40	SUVmean50	SUVmean60	MTV40-50-60	HV 1.2-1.3-1.4
CA-IX	ns	ns	ns	ns	ns	ns
HIF-1a	ns	ns	ns	ns	ns	ns
GLUT1	ns	ns	ns	ns	ns	ns
Ki-67	ns	ns	ns	ns	ns	ns
CD31	p=0.0094	p=0.0107	p=0.0094	p=0.0154	ns	ns

Project FAZA-glioma
FAZA-PET/MR for RT Planning



Mapelli P, et al. Clin Nucl Med. 2017

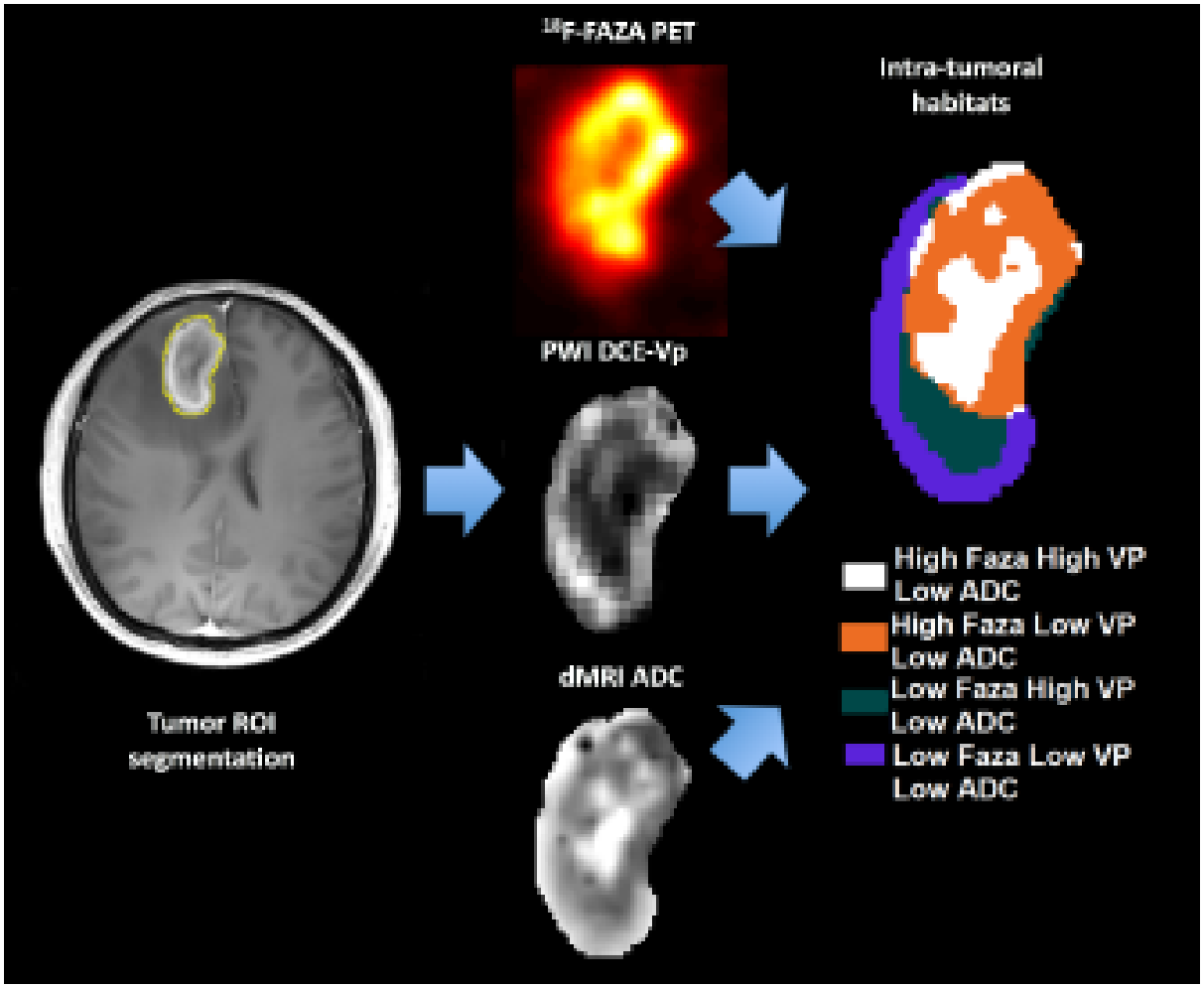
Project FAZA-glioma
FAZA-PET/MR for Treatment Response



Mapelli P, et al. Clin Nucl Med. 2017

Project FAZA-glioma
FAZA-PET/MR spatial concordance

Figure 1. Distances between the centers of mass of ^{18}F-FAZA, PWI and dMRI for each slice in the tumor ROI.



Hypoxia PET Imaging in Glioma

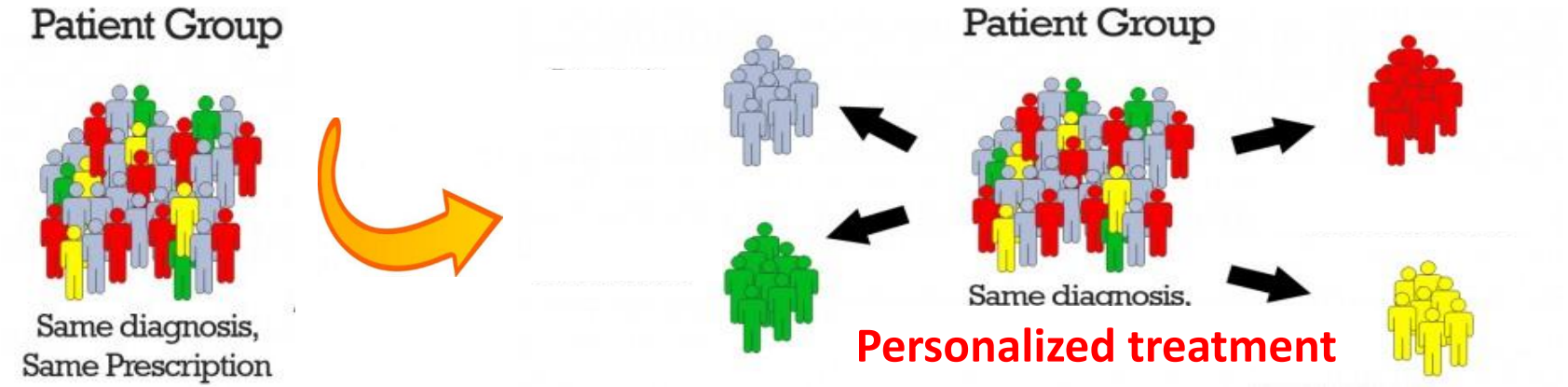
- Valuable tool for guiding stereotactic biopsy in high-grade glioma patients
- Potential role to plan RT treatement planning (radiation boost)
- Support discrimination between pseudoprogression and radionecrosis

Hypoxia PET Imaging
Conclusion and Perspectives

Imaging Biomarkers are essential for clinical development of Hypoxia-targeting treatment

- Although still necessary validation/standardisation of hypoxia Imaging to establish final clinical role

  - ❑ Knowledge on tumour heterogeneity
  - ❑ Evaluation of treatment response and tumour prognosis



Thank you