Imaging hypoxia in glioblastoma multiforme with pet

Hypoxia plays a crucial role in the progression of glioblastoma multiforme (GBM) promoting angiogenesis, genetic mutations, switch to a more aggressive behaviour and ote important consequences. Many diagnostic methods have been investigated and today PET and MRI appear to be the more attractive for the evaluation of the heterogeneous hypoxia in GBM. (Mendichovszky and Jackson 2011, Vartanian, Singh et al. 2014)

Hirata first documented the utility of hypoxic tracers ([^{18} F]FMISO in this study) in patients with differentiating GBM from lower grade gliomas based on the level of tumour hypoxia.(Hirata, Terasaka et al. 2012) Hypoxia assessment by PET imaging seems to provide complementary information to MRI within the complex relationship existing between hypoxia and angiogenesis in GBM. This was confirmed in a study of Swanson et al, where the authors documented a strong correlation between the hypoxic burden, determined with [^{18} F]FMISO, and altered vasculature documented on gadolinium-enhanced T1-weighted MRI sequences.(Swanson, Chakraborty et al. 2009)

As for other tumours, the prognostic capability of [^{18} F]FMISO has been confirmed also in GBM, in a study evaluating the correlation between hypoxic volume, intensity of hypoxia an survival in 22 patients with GBM who underwent PET scan before biopsy or between resection and radiation therapy (RT).(Spence, Muzi et al. 2008) The heterogeneous distribution of hypoxia within GBM cannot be fully investigated by [^{18} F]FMISO PET imaging, but the tumour–blood ratio provide acceptable data on the different levels of hypoxia within the tumour.(Padhani, Krohn et al. 2007)

[^{18} F]FAZA is another radiotracer tracer, which has showed promising results. The biggest study ever published, evaluating the utility of [^{18} F]FAZA in 50 patients with different types of tumours, documented increased uptake of the tracer in all gliomas, with a tumour-to-background (T/B) ratio range of 1. 9-15. 6, which is higher compared to that of [^{18} F]FMISO.(Postema, McEwan et al. 2009) However, as already said most of literature on the use of [^{18} F]FAZA in the brain is based in preclinical setting (seeTab. X)

According to the group of Wiebe, one important point in favour of [^{18} F]FAZA for the evaluation of hypoxia in brain tumours is the absence of uptake in normal brain tissue, while [^{18} F]FMISO shows, although limited, non-specific uptake in the brain.(Wiebe 2004) Recently, also Belloli and colleagues investigated the combined use of [^{18} F]FAZA and [^{18} F]FDG PET and MRI to follow the biological modification of specific line of glioma cells during the tumour progression in animal models of GBM (rats with implanted glioma F98 cells). The authors observed that [^{18} F]FAZA and [^{18} F]FDG were taken up respectively in the core and in external areas of the tumoyr, with partial overlap and remodelling during disease progression, suggesting that necrotic regions, defined on the basis of [^{18} F]FDG uptake reduction, may include hypoxic clusters of vital tumour tissue identified with ^{18} F]FAZA.(Belloli, Brioschi et al. 2013)

BOLD-MRI is an advance MRI technique, particulary suitable for the evaluation of hypoxia, which evaluate the changes in oxygen concentration and ratio between oxyhemoglobin and deoxyhemoglobin within vessels. In

contrast to oxyhaemoglobin, deoxyhaemoglobin is paramagnetic and determines an increase of transverse relaxation rate (R2*) of water in blood and surrounding tissues.(Mendichovszky and Jackson 2011) Unfortunately BOLD-MRI signal is sensible also to other tissue factors, such as blood flow, carbon dioxide tension, haematocrit, pH. Decoupling the effects of flow from deoxyhaemoglobin and static components it is essential to measure R2* and be obtained using multi-echo GRE sequences.(Padhani, Krohn et al. 2007)

T1-weighted oxygen-enhanced MRI (OE-MRI) has been proposed as an alternative imaging technique for the evaluation of hypoxia.(Zaharchuk, Busse et al. 2006) Dissolved oxygen in blood and plasma influences MRI signal by increasing the longitudinal relaxation rate of protons (R1). OE-MRI has already been employed in the evaluation of oxygen in healthy tissues and in tumours, but not in the evaluation of hypoxia in GBM, except in a preclinical study by Linnik et al. (Linnik, Scott et al. 2014) In an animal study, Wu et al. used a mechanically ventilation with 100% oxygen at the rate of 8 l/min to investigate hypoxia in brain of rats and showed close agreement between R2* and R1 changes in white and grey matter in response to oxygen inhalation.(Wu, Gao et al. 2012) In the study of Wu and colleagues, the T1 values decreased prominently in the cortical grey matter but also, with a lower extent, in the subcortical gray matter and in white matter, where the decrease was the least significant. Instead the T2 values showed an increase in response to the oxygen inhalation in all the regions examined in the following order: white matter> subcortical gray matter> cortical gray matter. Similarly, the T2* values increased with more evident change in the cortical gray matte and white matter and with a less extent in subcortical

gray matter.(Wu, Gao et al. 2012) These observations support the use of oxygen-enhanced imaging as a biomarker for tumour oxygenation, although the relationship between the signal changes resulting from variations in dissolved oxygen pressure and true tumour hypoxaemia remain to be elucidated.

DCE-MRI, using contrast agents of low molecular weight, has been proposed as an additional MRI method for identification and quantification of hypoxia in some types of tumour and some authors successfully demonstrated a correlation between perfusion parameters to oxygen tension. (Ceelen, Smeets et al. 2006) DCE-MRI parameters have been demonstrated also to indicate preoperatively areas with high hypoxia in glioma patients. In particularly Jensen et al. demonstrated that capillary transit time (tc) correlated with HIF-1 expression and VEGF expression in the histopathological examination of corresponding of active tumour regions. Other parameters, blood volume (Vb), capillary heterogeneity (a^{-1}) and k_{ep} (washout rate) also showed a correlation with biomarkers of hypoxia.(Jensen, Mumert et al. 2014) O' Connor, in a study evaluating ten patients with solid tumours, proposed that DCE may provide complementary information to OE-MRI regarding the tumour microenvironment, estimating local perfusion and extracellular–extravascular volume,(O'Connor, Naish et al. 2009) Subsequently, Linnik et al. validated the measurement of hypoxia validated OE-MRI using a murine glioma xenograft with histopathological confirmation. The study involved 5 patients, who underwent the same imaging protocol of the rats: OE-MRI and DCE-MRI and histological confirmation with reduced pimonidazole adducts and CD31 staining. Furthermore, the area under the

curve (AUC) was also calculated for the R1 curve for OE-MRI and the gadolinium concentration curve for DCE-MRI. Whereas DCE-MRI did not relate to hypoxia in the xenograft model, the authors found a strong correlation between estimation of hypoxia by means OE-MRI and histology results, supporting further research to validate also the utility of OE-MRI in the evaluation of response to therapy and prediction of prognosis (Fig.). (Linnik, Scott et al. 2014)

DWI-MRI instead has been used to clarify the mechanism of action of bevacizumab role, scanning patients with recurrent GBM before and after treatment with bevacizumab.(Rieger, Bahr et al. 2010) The mechanism of action of bevacizumab is still matter of debate. It is thought to produce damage to the endothelial cells, decreasing transport of nutrients and oxygen to the tumour cells,(Field, Jordan et al. 2014) but recently, it has been postulated an alternative theory: antiangiogenic therapy could stimulate a “ vascular normalization”, which would allow improved chemotherapy delivery and radiation effects through enhanced oxygen delivery.(Jain 2005) The study showed that bevacizumab induced stroke-like lesions with diffusion restriction and corresponding ADC decrease in 13 out of 18 patients enrolled in the study. A biopsy, performed in ADC-decreased lesion in one patient, demonstrated and nuclear hypoxia with HIF-1α up-regulation atypical necrosis but no tumour recurrence, supporting the hypothesis that bevacizumab-increases hypoxia in the tumour bed, expecially in case of prolonged treatment. Furthermore the imaging analysis revealed that regional cerebral blood flow (rCBF) and regional cerebral blood volume (rCBV) were decreased in responders with diffusion restricted

lesions.(Rieger, Bahr et al. 2010) Recently the effect of anti-angiogenic therapy has been investigated by a new technique, called vessel architectural imaging (VAI) which analyses the temporal shift in the MR signal estimating the vessel calibre and provides additional information about the microcirculation and oxygen saturation levels. From preliminary investigations, VAI seems to be a reliable MRI method to demonstrate the effect of anti-angiogenic therapy.(Emblem, Mouridsen et al. 2013) Other authors suggested AVOL, a measure of arteriovenous overlap (voxels with both arteriosus and venous perfusion characteristics), as index of abnormal tumour microvasculature and as indicator of bevacizumab therapy efficacy. (LaViolette, Cohen et al. 2013)

Barajas and colleagues (Barajas, Phillips et al. 2012) investigated histopathological and physiologic MRI features using diffusion-weighted imaging (DWI), dynamic susceptibility–weighted, and contrast enhanced perfusion imaging (DSC). Image-guided tissue specimens were taken from contrast enhanced (CE) and non-enhancing (NE) regions in GBM (93 CE and 26 NE regions from 51 patients with newly diagnosed GBM). The authors analysed variables of anatomic, imaging, and histopathological features (tumour score, cell density, proliferation, architectural disruption, hypoxia, and microvascular hyperplasia). Tissue samples from CE regions were found to have increased tumour score, cellular density, proliferation, and architectural disruption compared with NE regions.(Barajas, Phillips et al. 2012)

MRI in the evaluation of perfusion

Perfusion measurement of regional cerebral blood flow (rCBF) has been proposed as a method for identifying angiogenically active tumours. Increased angiogenesis in high-grade gliomas is correlated with higher cerebral blood volume (CBV) after contrast administration with dynamic MRI, relative to contralateral normal white matter rCBF and tumour aggressiveness. (Provenzale, York et al. 2006, Gruner, Paamand et al. 2012)

Also microvascular density (MVD) of tumour tissue has been shown to relate to tumour behaviour and prognosis. Furthermore it has been demonstrated that abnormalities in contrast agent recirculation provide independent information concerning the microcirculation and may be of value as surrogate markers in trials of antiangiogenic therapy.(Alan Jackson 2002) Early changes of rCBV, evaluated by MRI before and at weeks 1-2 and 3-4 during radiotherapy, can indicate response to treatment and correlate with survival [Cao]. Also Galban investigated the predictive impact of MRI in this setting, suggesting the use of voxel-by-voxel parametric response maps at 3 weeks after radiotherapy to predict overall survival.(Galban, Chenevert et al. 2009)

Another MRI technique which has shown promises in the assessment of the tumour microvascular environment is susceptibility weighted imaging (SWI), which aims to underline the susceptibility differences between tissues. Liu et al. demonstrated that R2*values are significantly different between high-grade gliomas, low-grade gliomas, postulating that these differences may be related to the different content of deoxyhaemoglobin.(Liu, Liao et al. 2014)