Temozolomide and Radiotherapy antitumor efficacy evaluation with Magnetic Resonance Imaging and Proton Magnetic Resonance Spectroscopy in Human Glioma Models in Nude Rats.

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Malignant glioblastoma remain uniformly fatal despite aggressive therapeutic protocols. Validation of more predictive biomarkers of treatment efficacy in experimental human glioblastoma models would greatly benefit from the establishment of additional quantitative endpoints. The aim of this study was to validate proton Magnetic Resonance Spectroscopy (^{1}H-MRS) and Diffusion-weighted MR Imaging (DwMRI) to evaluate the anti-tumor activity of Temozolomide (TMZ) and radiotherapy (RT) in 2 human glioblastoma models.

CGL9 and U87-MG glioma cells were inoculated at D0 by stereotactic injection in the right caudate nucleus of 2 groups of 22 nude rats. Tumor-bearing rats were ranked according to body weight and randomized at D12 (U87-MG) or at D19 (CGL9) to receive either 5 administrations of 16.5 mg/kg TMZ *per os* daily or 5 tumor-localized irradiations of 2Gy daily (D12-D16 and D19-D23 for U87-MG and CGL9 respectively), or no treatment (CTL). Imaging was performed on a Bruker Pharmascan 4.7 T at D12, D13, D16, D19, D23 (U87-MG) and D19, D20, D23, D26, D33 (CGL9). Tumor volume was measured using T_2-weighted images (U87-MG) or T_1-weighted, contrast-enhanced images (CGL9). DwMRI and ^{1}H-MRS were performed at the same timepoints.
Apparent Diffusion Coefficient (ADC) maps were computed from DwMRI volumes, and distributions of ADC analyzed within regions of interest within the tumor and the contralateral lesion-free tissue. Spectroscopic data were acquired using a SVS PRESS sequence, with voxel sizes adapted to the dimensions of the glioma in order to avoid partial volume effects with normal cerebral tissue. A spectrum was also acquired on the contralateral tissue. Spectral data were analyzed using LC-Model.

TMZ increased the life span of both U87-MG and CGL9 tumor-bearing rats (ILS = 126% and >200% for U87-MG and CGL9 respectively). The TMZ-treated to CTL tumor volume ratios (T/C%) were 8 and 2% for U87-MG and CGL9 at the last imaging timepoint, respectively. Radiotherapy (RT) increased the life span of 27% of U87-MG and 10% of CGL-9 tumor-bearing rats
ADC was increased by 34% in TMZ compared to CTL group for U87-MG tumors, whereas ADC was not modified by TMZ in CGL9 tumors.
In the U87-MG CTL group, a progressive reduction in NAA and creatine was observed during the study period. The ratio of total choline and total creatine increased from 0.5±0.2 to 2.5±0.3 in the CTL group, while it decreased from 0.8±0.3 to 0.3±0.2 in the TMZ group. Analysis of MRS data on the CGL9 model and on RT group is pending**.**

Using MRI, we observed a strong inhibition of tumor growth by TMZ treatment on both models, together with increased survival. ADC is a sensitive parameter to the effect of TMZ on U87-MG, but not on CGL9 tumors. Monitoring tumor metabolism using ^{1}H-MRS is well suited to follow the growth of U87-MG tumors and allows quantification of the antitumor effect of TMZ with choline being the most obvious candidate as a pertinent biomarker.