Vessel size, Blood volume and Diffusion MR Imaging of different glioma models in rats

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Synopsis Vessel size index (VSI) and blood volume (BV) MRI was previously validated in healthy and in C6 glioma bearing rats. Significant differences of the microvascular status were observed between tumor and healthy brain tissue. Here, 5 other glioma rat models with identical volumes were examined using VSI, BV and diffusion MRI. Morphometric parameters of the tumor microvasculature and ADC values of tumor tissue were related to the chemosensitivity of each glioma model to BCNU.

Introduction

The response of gliomas to chemotherapy varies widely. Although some genetic alterations have been identified(1), there is no clear understanding of the mechanisms underlying the chemosensitivity of these tumors. In previous *in-vitro* and *in-vivo* studies (2), we showed that the sensitivity to BCNU differed between glioma models. The sensitivity to BCNU may depend on the intrinsic sensitivity of the cells and on functional properties of the tumor microvasculature.. The vessel size index (VSI) and Blood Volume(BV) magnetic resonance imaging technique provides non-invasive quantitative insight on the microvascular architecture. We applied here VSI, BV and Diffusion imaging to characterize the vascular and tumor tissue status of a panel of six different glioma models in the rat brain.

Materials and Methods

Murine glioma cells (GV1A1, C6 and 9L) and human glioma cells (CGL3, CGL9 and U87-MG) were inoculated by stereotactic injection in the right caudate nucleus of 20 rats per glioma model (BDIX, Wistar and Fisher rats for the murine gliomas and Nude rats for the human gliomas). For each model, multislice T2-weighted MRI (2.35T) was performed on 10 rats to determine the tumor volume and compose groups of 5 or 6 rats bearing gliomas of identical volumes(50-75 mm^3). The rats were anesthetized with a mixture of isoflurane in air and O_2. Body temperature was maintained constant throughout the MRI experiment. The MGESE imaging sequence (3) was applied and repeated 4 minutes after injection of the intravascular contrast agent Sinerem®(Guerbet, France, 200µmol Fe/kg) in the tail vein. VSI maps were derived from water apparent diffusion coefficient (ADC), ΔR2 and ΔR2* maps while BV maps are proportional to ΔR2* maps only. ADC maps were obtained using a Stejskal-Tanner MRI sequence with a diffusion gradient applied along X, Y and Z successively. Regions of interest (ROIs) were drawn manually over the tumor (3 slices at the center) and the contralateral area in the same slices. The slices were transverse, 1mm thick and spanned a 30-mm FOV (Matrix size= 128x64 interpolated to 128x128). VSI and BV histogram analysis were performed. The animals were sacrificed after the MRI examination, 1 minute after injection of a FITC-dextran and a Hoechst saline solution for histological analysis by fluorescence microscopy of the functional microvasculature. The 6 to 10 remaining rats per glioma model were randomized on the same day the VSI imaging was performed and kept for survival follow-up: 3-5 of them received 2 IV bolus injections of 10 mg/kg BCNU at 2 weeks interval. The mean and median survival time and increased life span(ILS) were calculated.

Results

Significant increases in ADC between the tumor and the contralateral area were found for the 9L, CGL3, C6 and GV1A1 tumor models ($p < 6.22 \times 10^{-4}$, paired t-test). The CGL9 tumor model shows no significant difference in ADC with its contralateral area but significant ADC differences with all the other tumor models ($p<0.02$, t-test) (Fig2). The distribution of VSI values in all the tumor models were comparable(Fig1). Maximum VSI values of 80µm in the tumor ROIs were found whereas contralateral areas show on average a maximum VSI of 20µm. All the contralateral distributions of ΔR2* values are Gaussian-like. However, U87MG and 9L tumors present non-normal ΔR2* distributions compared to the other tumor models: GV1A1, C6, CGL3 and CGL9. Blood Volumes are higher in the contralateral area with a higher density of pixels with values 25-95s^{-1} compared to the gliomas (Fig3). Following BCNU treatment, rats bearing CGL9 and GV1A1 gliomas had the highest increase life span (ILS>220 % and 123% respectively). On the contrary, U87MG and 9L-bearing rats treated with BCNU died at the same time as their controls (ILS=0 and 6.7% respectively).

Conclusion . CGL9 gliomas with the highest increased life span after BCNU treatment has lower ADC values compared to all the other models, synonymous of a higher cellular density. The ADC could be an appropriate parameter to predict the tumor response to BCNU treatment. VSI ansd BV values confirm their ability to distinguish differences between tumor and contalateral tissue and seem to indicate variable blood volume distributions between glioma models. More understanding is expected from functional histology.

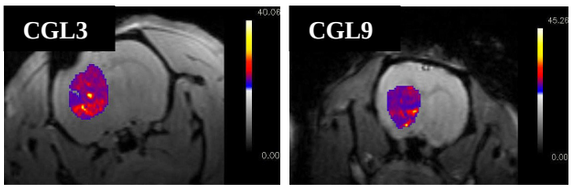

***Fig1** : Examples of VSI maps in a CGL3 (VSImax= 40µm) glioma and a CGL9 Glioma (VSImax= 45µm)*

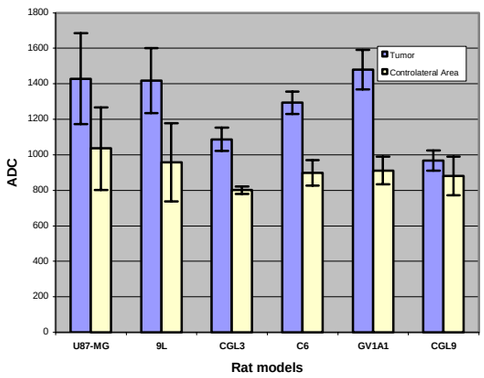

***Fig2** : Mean ADC for each tumor model and the contralat-eral area*

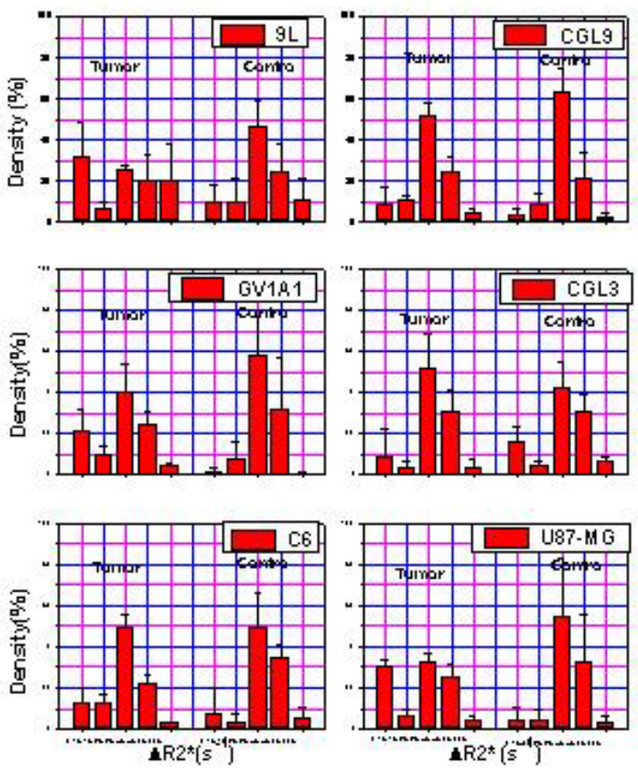

***Fig 3** : ΔR2* distribitutions of pixel (desnsity) for each model in the tumor(left) and the contralateral area(right). Values have been divided in 5 categories (0-15 s^{-1} ; 15-25 s^{-1}; 25-45 s^{-1}5; 45-95 s^{-1} and 95-255 s^{-1}). y axis scale=0-100%. 255 s^{-1} corresponds to a maximum blood volume of 17%.*

References :

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