MRI guided drug delivery targeting glioma using inteleukin-13 conjugated liposome (IL-13-Liposome)

Xiaoli Liu[1], Achuthamangalam Madhankumar[1], Patti Miller[2], Becky Webb[1], Susan Hafenstein[3], Elias Rizk[1], James Connor[1], and Qing Yang[4]
[1]*Neurosurgery, Pennsylvania State College of Medicine, Hershey, PA, United States,* [2]*Radiology, Pennsylvania State College of Medicine, Hershey, PA, United States,* [3]*Medicine, Pennsylvania State College of Medicine, Hershey, PA, United States,* [4]*Pennsylvania State College of Medicine, Hershey, PA, United States*

Introduction

As a MRI contrast agent, Gd-DTPA (Magnevist, Bayer Healthcare), is widely used for detection of brain tumor but limited to the cases where blood brain barrier (BBB) is significantly compromised by tumors because Gd-DTPA itself is not able to cross the BBB. The tumor boundary produced by Gd-DTPA-enhanced MRI reflects only the extent where BBB is comprised instead of an actual tumor-normal tissue boundary. It is desirable to develop a new drug delivery tool that is capable of crossing intact BBB to detect and treat the glioma at its earlier stage when treatment is more effective. Previous work demonstrated that inteleukin-13 receptors alpha 2 (IL-13Rα2) are highly expressed in glioma cells [1]. IL-13 conjugated with liposomes contained doxorubicin (anticancer drug) inhibited the glioma tumor growth effectively in a mouse model [2]. In present work, we developed a MRI guided drug delivery agent, IL-13-liposomes-Gd/DTPA-doxorubicin, that delivered both Gd/DTPA and anticancer drug, doxorubicin, over the BBB and provided detection and treatment of glioma concomitantly.

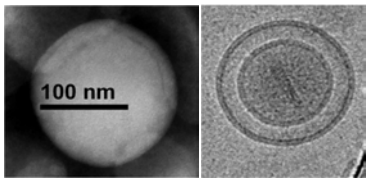

Fig.1: The morphology of the IL-13-liposomes-Gd/DTPA-

Methods

Preparation of IL-13-liposomes-Gd/DTPA-doxorubicin: Liposomes were prepared using the lipids refer to [2] and the FITC, lipophilic carbocyanine $DiOC_{18}(3)$. When the film of lipids was formed, the film was reconstituted in a saturated solution of Gd-DTPA with poly-L-lysine to form multilamellar vesicles and covalently conjugated to human IL-13 protein, then exposed to the doxorubicin and purified. The average particle size was 100-150 nm. The Gd concentration in the liposomes was in the range of 4.0-10.0 mg/L quantified using ICPAES and doxorubicin concentration is 0.2-0.5mg/mL**.** Its relaxivity was compared with free Magnevist and its uptake in glioma and stem cell was tested. Its relaxivity was 0.7mmol/L when measured at 7T MRI system (Bruker Biospin 7/20a, Ettlingen, Germany).

Results and Discussion:

1) IL-13-liposomes-Gd/DTPA-Dox presented a spheroid with an average sized 130 nm in diameter determined by Transmission Electronic microscopy (Cryo JEOL 2100) in Fig. 1.
2) The comparison of the relaxivity between the free Magnevist and IL-13-liposomes-Gd/DTPA-doxorubicin demonstrated our novel agent is capable of generating same level of contrast as free Magnevist. The relaxivity of free Magnevist is 4.7 (A) and IL-13-liposomes-Gd/DTPA-Dox 4.0 (B). Both agents have the same Gd concentration.
3) The novel IL-13-liposomes-Gd-DTPA-doxorubicin delivered Gd-DTPA and doxorubicin into the glioma and glioma stem cells.
3) Fig. 1a and 1b shows the results of MRI from an intracranial tumor enhanced by Magnevist and IL-13-Lip-Gd-FITC of the same mouse 24 hr apart. Our IL-13-Lip-Gd-FITC produced a similar contrast enhancement to Magnevist. Most excitingly, however, the MRI enhanced with our targeted liposome revealed several additional smaller tumor masses shown in Fig. 1b (arrow) that are not visible in Magnevist-enhanced image in Fig. 1a. These tumors infiltrated away from primary tumor mass (not shown) were validated with histology of the same brain slice in Fig. 1c and 1e. The blue florescent image indicated targeted liposomes that appeared to distribute around the tumor tissues along white matter tracks, typically seen in human GBM. It is likely that these tumors had not caused a significant BBB damage since Magnevist enhanced MRI did not show these tumors. Our result from H&E staining for tumor (Fig. 1c and Fig. 1e) and fluorescent image in Fig. 1d demonstrated the sensitivity and specificity of our liposome targeting infiltrating gliomas. In this study, we used anticancer drug, doxorubcin, that is known incapable of crossing BBB. Success in inducing cancer remission in our glioma animal model indicated that our liposome facilitate the drug delivery thtrough the BBB.

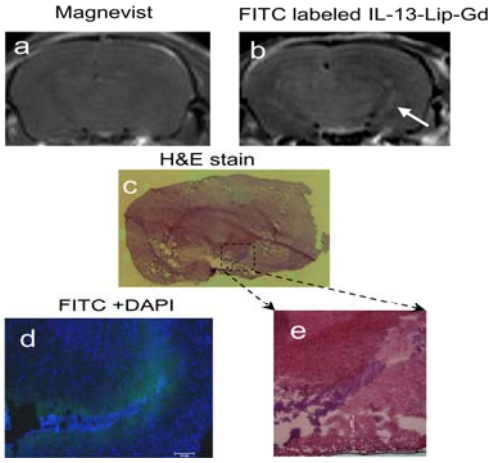

Fig. 2: MRI images enhanced with IL-13-Lip-Gd (b) identified infiltrating tumor masses (arrow) that are absent in the Magnivist enhanced MRI (a) of the same brain slice. The tumor tissues in the same brain area appear to be blue in H&E stain image (c and e), validated MRI finding. The green fluorescence of FITC+DAPI image of an adjacent slice (d) indicates targeted liposome distribution coincided with infiltrating tumors along the white matter track.

References:

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2.Madhankumar, A.B., et al., *Interleukin-13 receptor-targeted nanovesicles are a potential therapy for glioblastoma multiforme.* Mol Cancer Ther, 2006. 5(12): p. 3162-9.