RNA-nanoparticles to enhance and track dendritic cell migration

Grippin, A.J. (UF, Medicine and Engineering); Sayour, E.J. (UF, Medicine); Wummer, B. (UF); Monsalve, A. (UF, Engineering); Wildes, T., Dyson, K. (UF, Medicine); Dobson, J. (UF, Engineering) and Mitchell, D.A. (UF, Medicine)

Introduction

Despite aggressive chemotherapy, surgical resection, and radiation therapy, glioblastoma (GBM) remains almost universally fatal. In a pilot, randomized, and blinded clinical trial, we recently demonstrated that administration of RNA-loaded DC vaccines was associated with significantly improved progression-free and overall survival in patients with GBM (Mitchell et al, *Nature* 2015). Furthermore, clinical outcomes correlated with DC migration to vaccine-site draining lymph nodes measured by Indium-111 labeling of RNA-loaded DCs and SPECT/CT imaging. While these studies demonstrated that tracking DC migration may be an important clinical biomarker for response to DC vaccination, the complexity and regulatory requirements associated with nuclear labelling to track DC migration limits widespread application of this technique. We have therefore developed RNA-loaded magnetic nanoparticles (RNA-NPs) to enhance DC migration to LNs and track that migration with a widely available imaging modality (i.e. MRI).

Experimental

Cationic liposomes were loaded with iron oxide nanoparticles with or without cholesterol. The resulting nanoparticles were complexed with RNA and used to transfect DCs *ex vivo*. RNA-NP-loaded DsRed+ DCs were then injected intradermally into mice and tracked noninvasively with T_2-weighted 11T MRI at the University of Florida AMRIS facility before excision and quantification with flow cytometry.

Results and Discussion

In vitro experiments demonstrate that iron oxide loading does not reduce RNA-NP-mediated transfection of DCs. Additionally, replacement of cationic lipids with cholesterol increased RNA-NP transfection of the DC2.4 cell line and enhanced the T cell stimulatory capacity of treated bone marrow-derived dendritic cells (BMDCs). Compared to electroporation, RNA-NPs enhanced DC migration to lymph nodes and reduced T_2 MRI intensity in DC-bearing lymph nodes.

Conclusions

This data suggests that iron oxide-loaded RNA-NPs enable noninvasive cell tracking with MRI and enhance DC migration to lymph nodes. We have further shown that inclusion of cholesterol in RNA-NPs augments the stimulatory capacity of transfected DCs. Future work will consider effects of RNA-NPs on antitumor immune responses and the utility of MRI-detected DC migration as a biomarker of vaccine efficacy.

Acknowledgements

A portion of this work was performed at the National High Magnetic Field Laboratory, which is supported by National Science Foundation Cooperative Agreement No. DMR-1157490 and the State of Florida. This work was supported in part by NIH R01 (PI: Mitchell; Grant #: R01CA175517); NIH R01 (PI: Mitchell; Grant #: R01CA195563); the Circle of Hope for Cancer Research (PI: Grippin); and the NIH/NCATS Clinical and Translational Science Award to the University of Florida (PI: McCormack; Grant #: TL1TR001428).

References

Mitchell, D.M., *et al.,* Nature, 519, 366–369 (19 March 2015).