Estimation of Sodium Triple Quantum MR Signals from 9L Glioma Cells and Growth Media

Levenson, C.W., Kynast, N. (FSU, Biomedical Sciences & Neuroscience) and Schepkin, V.D. (NHMFL)

Introduction

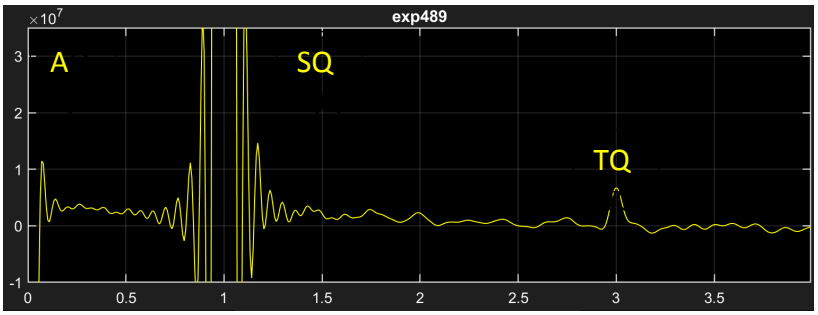
The project is devoted to the development of tumor cells evaluation using NMR compatible bioreactors. The approach has the potential to explore in more detail the differences between naïve and chemo-therapy resistant tumor cells, which were observed previously *in-vivo* by sodium MRI [1, 2]. The hypothesis is that intracellular sodium could be involved and/or be a biomarker of the alterations in tumor cells leading to changes in their resistance to chemotherapy. Sodium triple quantum signals have a possibility to detect changes of intracellular sodium in a rat heart model [3]. Such an approach can be equally extended to cells in bioreactors. Thus, the first step was to evaluate triple quantum (TQ) sodium signals from 9L cells and the media used for tumor cells growth.

Experimental

Experiments were conducted at the 21.1 T magnet in Tallahassee. The sodium single quantum (SQ) and TQ signals were detected using the triple quantum time proportional phase increment (TQTPPI) pulse sequence described previously [4]. Dulbecco's modified Eagle's medium (DMEM) was used for glioma 9L cell grow with the addition of 10% of serum as a nutrient. Media solutions and 9L cells were placed in 15 ml vials for MR experiments. Temperature of the samples was 32^{o} C.

Results and Discussion

The total amount of protein in serum is ~ 70 mg/ml. Thus, our media has a protein content of only ~ 7 mg/ml. The major part of serum protein is albumin which is electro-negative at the normal pH and, thus, capable to attract positively charged sodium ions. Such interaction is detected by the presence of the TQ sodium signals. Out of the total sodium content in media, only a minor part is bound to proteins as seen on the **Fig. 1.** The increased sensitivity for TQ signals was achieved by selecting a fixed inter-pulse delay of 25 ms in the TQTPPI pulse sequence [4].



The growth of glioma 9L cells was observed only at the walls of the NMR tubes, but not in the volume of the sample. This is specific to tumor cells. The estimated number of cells in the tubes was ~ 5 million. For estimation, the protein content for Hela cells is ~300 pg/cell. Thus, the expected protein content from cells could be ~ 1.5 mg.

The preliminary experiments with 9L cells in our samples indicate that ~ 5 million cells produce the sodium TQ signal which is either comparable or less than the background signal from the proteins in serum.

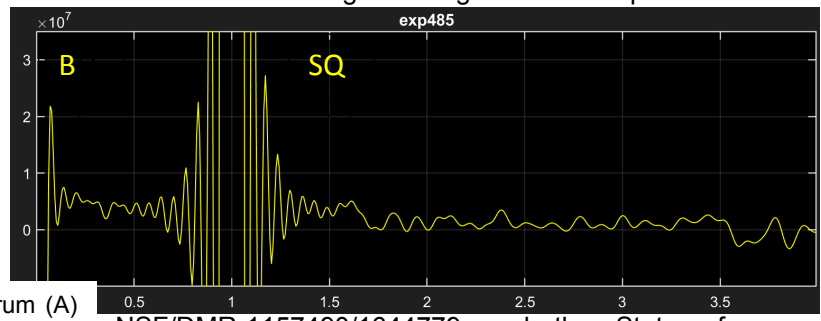


Fig.1 Sodium SQ and TQ signals in DMEM media plus 10% serum (A) and in media without serum (B). X-axis represents a phase rotation rate for the RF pulses in the pulse sequence at the fixed inter-pulse delay of 25 ms. Note that there is no TQ signal without serum (B).

Conclusion

A bioreactor design is needed allowing to support ~ 50 million cells in the volume of the NMR coil (ID = 32 mm).

Acknowledgements

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References

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[4] Schepkin, V.D., *et al.*, J Magn Reson, **277 (3)**, 162-168 (2017).