Chemical Exchange Saturation Transfer MRI: Sensitive to Intracellular pH Change Over Time in a Rat Model of Brain Cancer

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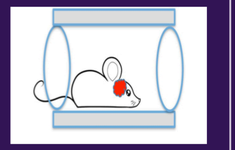
- In biological systems, robust maintenance of pH is a fundamental homeostatic process.
- The regulation of pH gradient across the cell membrane is crucial for many cellular functions and that is altered in cancer cells.



OBJECTIVE

To examine how pH_i changes over time in the brain of a rat C6 glioma model using CEST-MRI

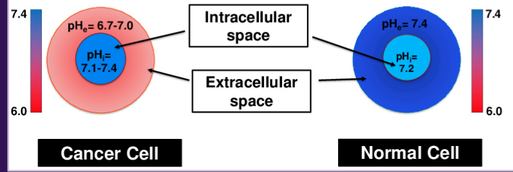


Fig.1: Schematic of intracellular pH (pH_i) and extracellular pH (pH_e) in normal and cancers cells

CHEMICAL EXCHANGE SATURATION TRANSFER (CEST)

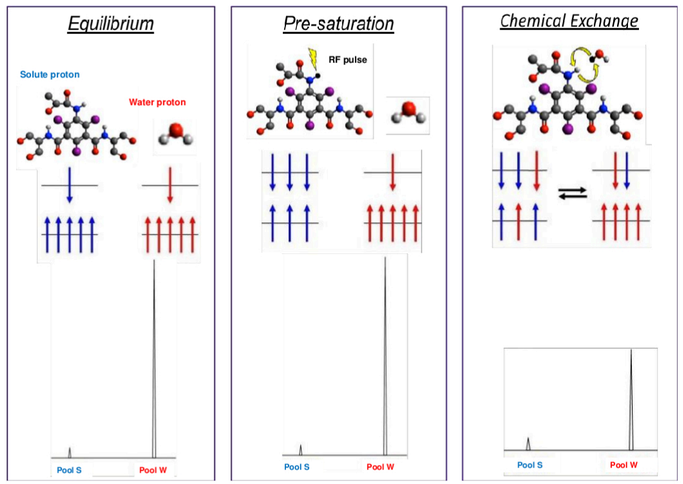
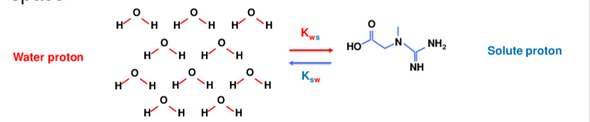


Fig.2: Chemical Exchange Saturation Transfer

- In CEST-MRI, contrast is produced by selectively exciting exchangeable protons and detecting this exchange through the transfer of magnetization to bulk water.
- To observe the effect of saturation on a pool of dilute protons, the protons in these systems must be in exchange with one another and have a distinct chemical shift difference.
- These exchangeable protons are commonly found in amide and amine groups on mobile proteins within the intracellular space.



- The magnitude of CEST contrast depends on the proton exchange rate which is pH-dependent for amine and amide protons.

$$K_{ex} = K_0 + K_a[H^+] + K_b[OH^-] = K_0 + K_a\ 10^{-pH} + K_b\ 10^{pH-pK_w}$$

$$K_{ex} = K_b\ 10^{pH-pK_w}$$

$$K_w = [H^+][OH^-]$$
$$pK_w = -log(K_0)$$

K_0: rate constant of the spontaneous proton exchange reaction between water and the amide nitrogen nucleus. K_a: rate constant of the acid-catalyzed protonation of the amide nitrogen nucleus. K_b: rate constant of the base-catalyzed proton exchange reaction between hydroxyl ion and the amide nitrogen nucleus. pK_w : ionization constant of water (15.4 at 37 °C). K_w: dissociation constant of water.

EXPERIMENTAL METHODS

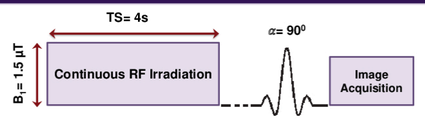
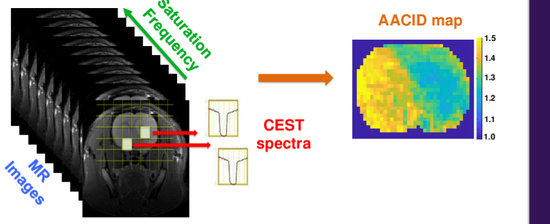


Fig.3: RF pulse sequence

- B_0: 9.4 T - Bruker
- Pulse sequences:
  EPI: TR= 7 s , TE= 25 ms
  FSE: TR= 7 s , TE= 7 ms
- FOV= 38.4×38.4 mm^2
- Cells: 8μL C6 GBM
- 8-week old male Wistar rats
- Tumour position: Right frontal lobe
- Saturation frequencies: 1.2 ppm - 6.6 ppm

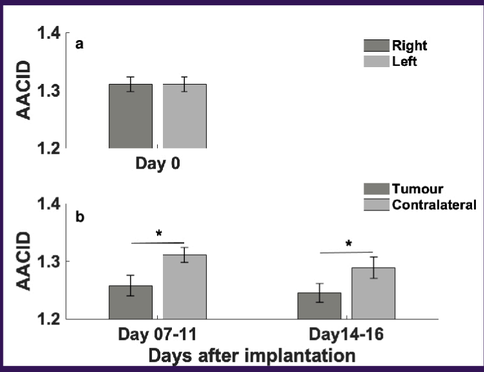
AACID : Amine and Amide Concentration-Independent Detection



$$AACID = \frac{M_z(3.50\ ppm) \times [M_z(6.00\ ppm) - M_z(2.75\ ppm)]}{M_z(2.75\ ppm) \times [M_z(6.00\ ppm) - M_z(3.50\ ppm)]}$$

Fig.5: Average AACID values (a) in right and left brain of control rats, (b) in tumor and contralateral brain of rats, measured on day 7-11, and day 14-16 post implantation of tumour.
Bars represent standard error of the mean.

RESULTS



RESULTS

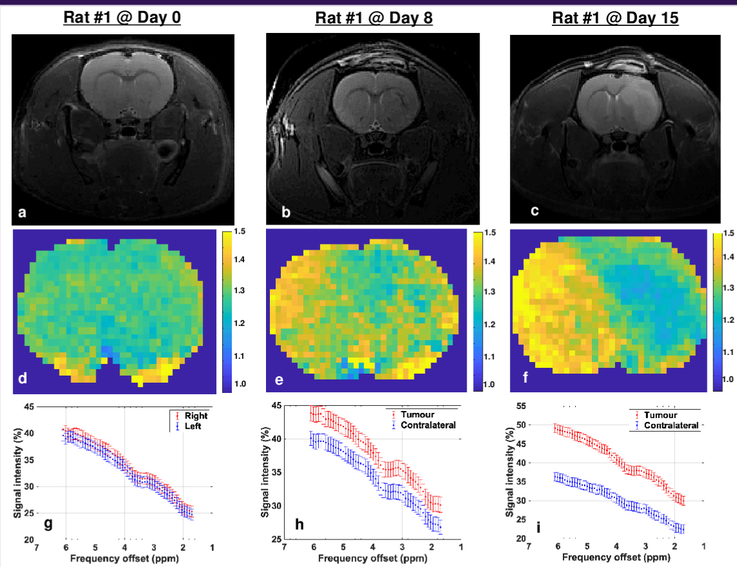


Fig.4: (a), (b), (c) T2-weighted images of rat brain; (d), (e), (f) AACID maps; (g), (h), (i) CEST spectra at day 0 and days 8 and 15 post implantation of C6 glioma cells. Purple line is the outline of the tumour region.

DISCUSSION

- No difference observed in AACID value between left and right side of the brain in control animals (N=5).
- A significantly lower average AACID value ($p<0.05$, paired t-test) was found in the tumour compared to the contralateral region at both time points.
- At day 7-11: average AACID value was ~4.0% lower in tumour indicating a 0.21 higher pH (N=14).
- At day 14-16: average AACID value was ~3.4% lower in tumour indicating a 0.17 higher pH (N=13).
- As expected, in this rat model, the difference between tumour pH and contralateral pH remains the same across time.

FUTURE STUDIES

- Measurement of pH_i by CEST following drug injection to modulate pH in C6 glioma model.
- Utilize DTI in combination with CEST to examine the effect of cellularity on pH measurements.

TAKE-HOME MESSAGE

- We successfully generated AACID maps of the brain in a rat C6 glioma model and demonstrate pH differences over time.
- This technique has the potential to monitor pH_i in humans as no exogenous contrast agent is required.

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