Robust Diffusion-Weighted Single-Shot MRI Can Resolve Major Mice Placental Compartments

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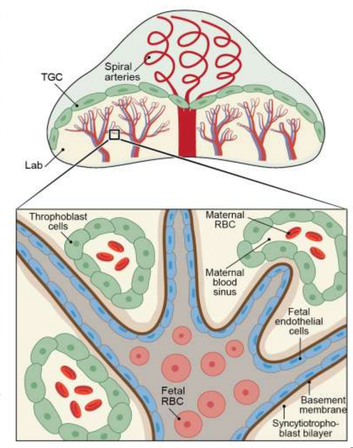
Kimmel Prize for Innovative Research

1. Introduction

The placenta is an essential and versatile organ providing life-supporting functions for the developing fetus. Further understanding placental functions and their relation to the organization of its intervening blood vessels and cells, requires characterizing the movement of fluids within and between key compartments making up the placental structures. Novel diffusion-based MRI methodologies relying on SPatio-temporal ENcoding (SPEN) were developed [1], and used to uncover in-vivo aspects about the diffusion of fluids in mouse placenta. SPEN MRI studies were carried out in the presence and absence of high Mw contrast agents (b-BSA-GdDTPA) to fractionate different placental compartments, at various gestation periods of pregnant mice. These in vivo MRI analyses were also aided by histological and fluorescence microscopy validations.

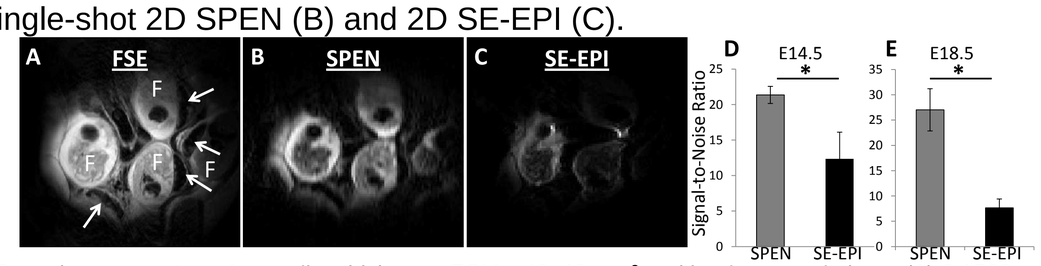
2. Placental compartmentalization

Placentas are heterogeneous organs containing two independent blood circulation systems, fetal and maternal, which are in a close proximity in the labyrinth area. Fetal blood flows within the placenta through a vascular bed with a tree-like hierarchy of vessel sizes, the smallest of which are µm-sized. These are inter-dispersed among maternal blood lacunae, which do not travel in capillaries but collect in pools in the intervillous spaces, thus ensuring an exchange between the two blood systems [2,3]. Fetal cells are surrounded by endothelial cells and membranes; fetal capillaries are surrounded by maternal blood sinuses, which conduct red blood cells (RBC) and are lined by fetal trophoblast giant cells (TGC).



3. Anatomical images of a pregnant mouse

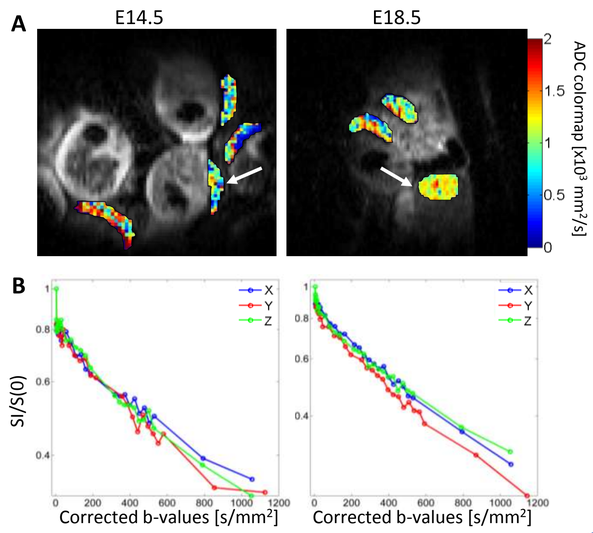
To evaluate the diffusion information that can be obtained from placental structures in mice, the imaging performances of single-shot SE-EPI and SPEN were first compared. Almost in all cases SPEN succeeded to provide significant better agreement with the multi-scan results. Anatomical images were collected on pregnant mice at gestation days E14.5 (n=2 mice; 5 placentas) and E18.5 (n=3 mice; 7 placentas), using 2D fast spin-echo as anatomical reference (A), single-shot 2D SPEN (B) and 2D SE-EPI (C).



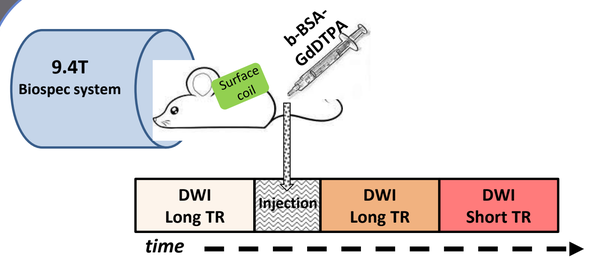
Scanning parameters: 1 mm slice thickness, FOV = 40x40 mm² and in-plane resolution = (A) 0.1563 mm, (B-C) 0.4 mm. Only 50% of the EPI images succeeded in supplying an appropriate data.

4. Changes in placental ADC during pregnancy

Two gestation days, 3 days after placental formation (E14.5; n=6 mice; 19 placentas) and towards the end of pregnancy (E18.5; n=10 mice; 39 placentas), were evaluated (A). We found no statistical difference between the placental ADC values for the two gestation ages (Mean ADC of 2.4±0.9 x 10^{-3} mm²/sec and 2.9±0.6 x 10^{-3} mm²/sec for E14.5 and 18.5 respectively; p=0.065). Corresponding diffusion semi-log plots measured at the two ages (B) (indicated by arrows) were repeated for a large array of b-values in the search for bi-exponentiality.



5. Resolving sub-voxel placental structures using a multi-component fitting



$$\frac{SI_1}{SI_1(0)} = \left(F_{mat}e^{-bADC_{mat}} + F_{fet}e^{-bADC_{fet}} + F_{TGC}e^{-bADC_{TGC}}\right) + c$$

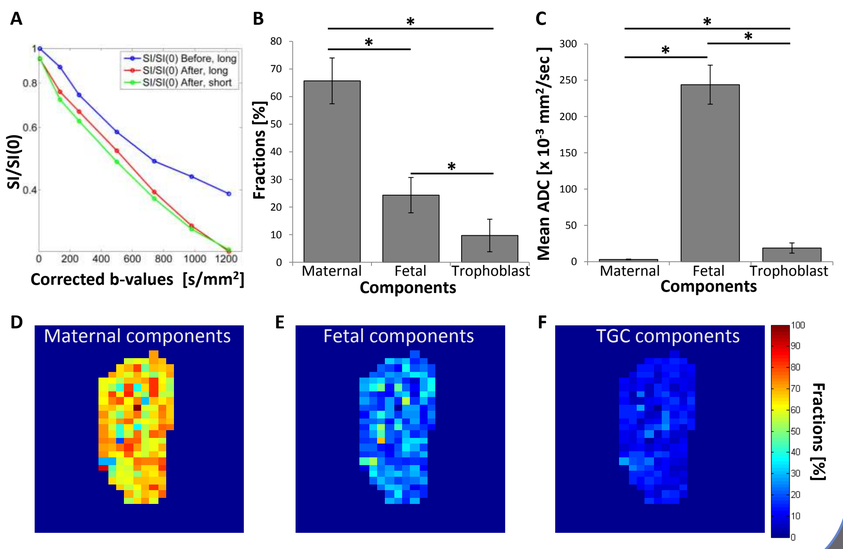
$$\frac{SI_2}{SI_2(0)} = \left(F_{mat}e^{-bADC_{mat}} + F_{fet}e^{-bADC_{fet}}\right) + c$$

$$\frac{SI_3}{SI_3(0)} = \left[F_{mat}e^{-bADC_{mat}}(1 - e^{-TR\cdot R_1})\right] + c$$

Maternal blood capillaries, fetal blood capillaries and trophblast giant cells (TGC) could become distinguishable by executing suitable ADC measurements before and after administration of b-BSA-GdDTPA.

We hypothesized that the placental signal attenuation observed when diffusion is measured at long TR and before the administration of the contrast agent, can be described as the sum of three contributing components (described on the left).

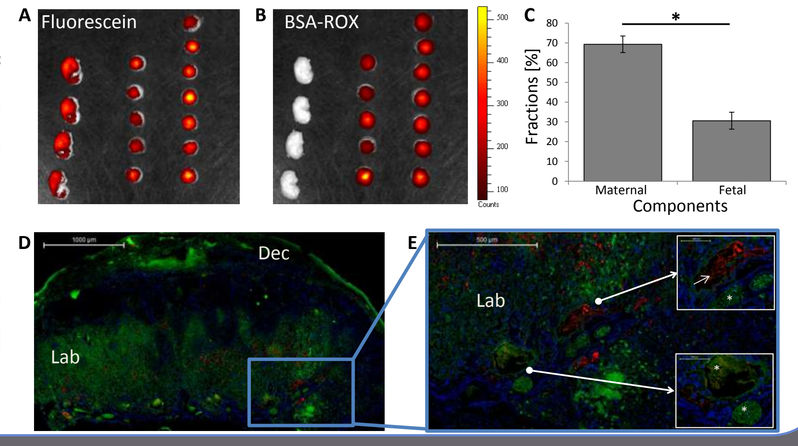
An example of the multiple relaxation curve (A). This analysis reveals that maternal blood constitutes 66±8%, fetal blood is 24±6%, and the TGC are 10±6% of the overall placental volume (B). ADCs were found to be 3.1±0.4, 244±27 and 19±7 x10^{-3} mm²/sec for maternal, fetal and TGC compartments, respectively (C). Placental fraction maps were calculated on a pixel-by-pixel basis providing a clear spatial distribution between the different compartments (D-F).



6. Optical imaging of mice placentas

To validate the *in vivo* dSPEN-based compartmental characterizations, fetal and maternal placental blood vessels were differentiated using optical *ex vivo* imaging. The analyses show the expected penetration of Fluorescein into both fetal and maternal circulation, whereas BSA-ROX remains confined in the maternal circulation within the labyrinth (A-B). The maternal and fetal fractions measured were 69±4% and 31±4% respectively (C).

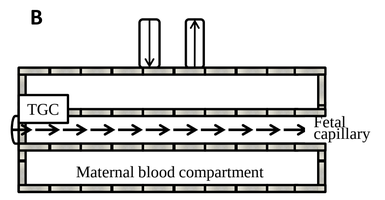
Fluorescence images of placental histology sections of BSA-ROX (red) were confined to the maternal circulation within the labyrinth, whereas fluorescein (green) also penetrated the fetal circulation (D-E).



7. Conclusions

The diffusion features found in this study are consistent with known descriptions of placental anatomy (Section #1). Maternal blood enters the labyrinth via large arterial canals; the movement of fluids associated to the maternal placental blood thus appears to be dominated by free diffusion. By contrast, the flow inside the fetal capillaries is fast and consistent with the need to assure proper nutrients transfer.

Finally, trophoblast cells line the lumen of maternal vessels and mediate fetal/maternal filtration of water, consistent with their intermediate ADC value.



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