MRI of Cell Layers in Mouse Brain *in Vivo* Using Intra- and Extra-cellular Contrast Agents

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Target Audience Anyone who is interested in high-resolution MRI of the brain of mammals ranging from genetically modified mice to human.

Purpose The aim of this study using the brain of living mice was to examine the effect of (i) systemic administration of manganese[1] (Mn) on T_1-weighted (T1W) and T_2-weighted (T2W) MRI and (ii) intracranial administration of Gd-DTPA on T1W MRI.

Methods Four mice received a subcutaneous injection of $MnCl_2$ (0.12 mmol/kg). Another four mice received an injection (5.0 µL) of 100 mM Gd-DTPA into a lateral ventricle as well as injections (0.3 µL into each side) of 50 mM Gd-DTPA into the olfactory bulbs. At 9.4 T, data were acquired at 20—210 min or 2—3 days after Gd-DTPA or Mn injection, respectively. T1W 3D FLASH (TR/TE = 22/7.6 ms, α 25°, measuring time 12, 48, 72, or 96 min) and/or T2W 3D FSE (TR/TE = 4200/48 for sagittal and 4200/80 ms for horizontal sections, measuring time 161 min) were performed. T_1 was determined using spin-echo MRI with TR/TE = 200—10000/10 ms.

Results Mn improved the contrast not only in T1W but also in T2W MRI. In general, however, Gd-enhanced T1W MRI provided the best contrast-to-noise ratio between layers. In Mn-enhanced T1W (12 min), T2W (161 min), or Gd-enhanced T1W (12 min) MRI of the hippocampus at 30×30×300 µm^3, the mean CNR was 5.6, 5.2, or 6.1, respectively. Gd shortened the mean T_1 of the olfactory bulb and the hippocampus from 1.48 s and 1.59 s to 0.43 s and 0.42 s, respectively.

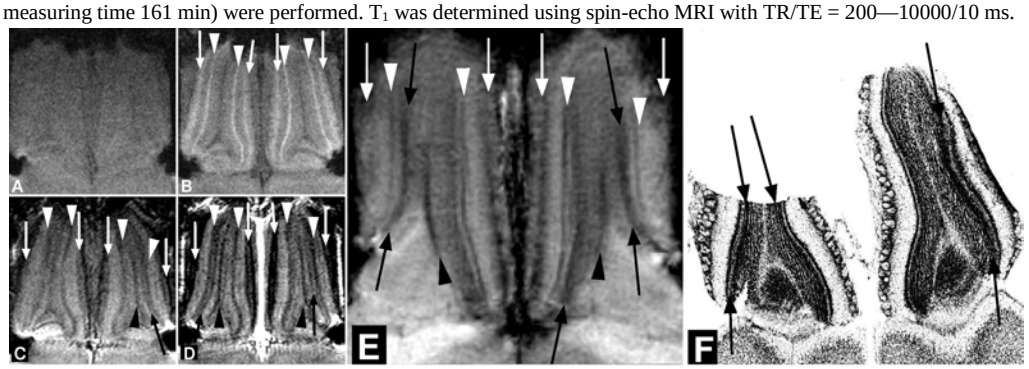


Fig. 1 (right) shows the olfactory bulb in horizontal section in (A and C) T1W (12 min) and T2W (161 min) MRI at 30×30×300 µm^3 before or (B and D) after Mn injection, respectively. Mn improves the delineation of the glomerular layer (white arrows) and mitral cell layer (white arrowheads). (E) T1W MRI after Gd-DTPA injection (72 min, 25×25×250 µm^3) shows a contrast similar to T2W MRI and to (F) a histological image[2] (neurons are stained) except the fiber tracts (black arrowheads). The mitral cell layer can be distinguished from densely packed granule cell layer (black arrows) due to a predominant Gd-DTPA accumulation in the internal plexiform layer between them.

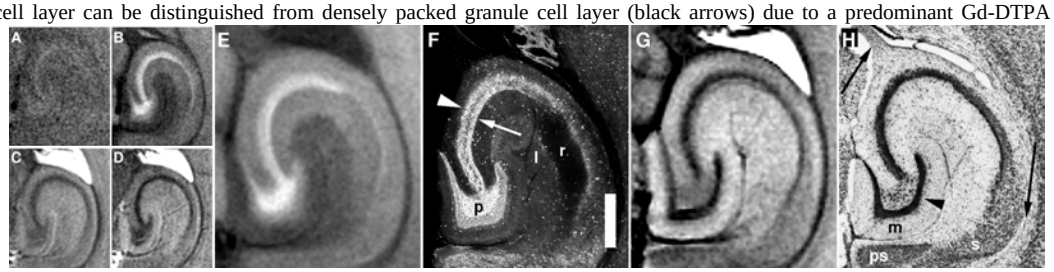
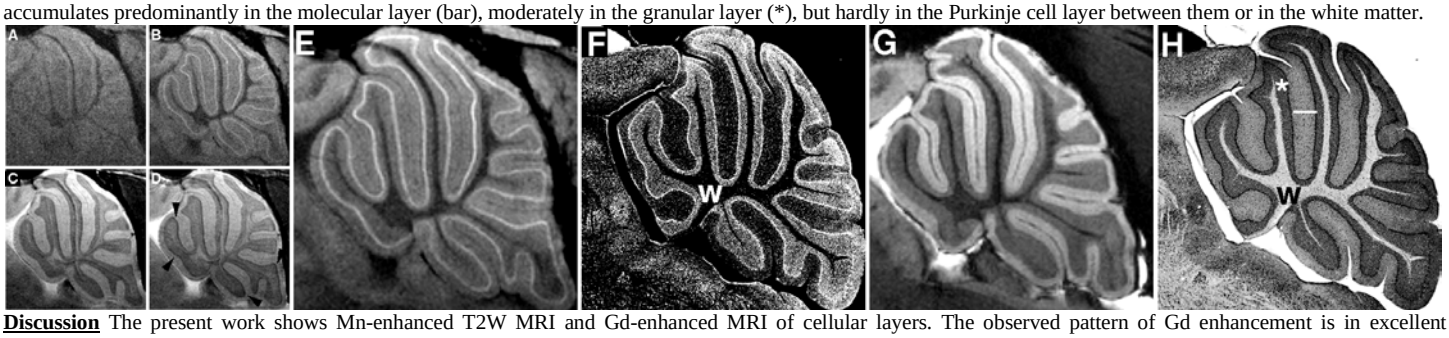


Fig. 2 (right) shows the hippocampal formation in horizontal section in (A and C) T1W (12 min) and T2W MRI (161 min) at 30×30×300 µm^3 before or (B and D) after Mn injection, respectively. Mn improves the delineation of cellular layers. (E) T1W MRI (96 min, 25×25×250 µm^3) after Mn injection shows a contrast similar to a (F) histological image[3] (neurons and mossy fibers are highlighted, scale bar = 400 µm). Mn enhances the polymorphic layer (p) of the dentate gyrus most clearly. Mn-enhanced MRI also distinguishes the layer of the mossy fiber (arrowhead) partly from the pyramidal layer (white arrowhead). Mn enhances the stratum lacunosum (l) less than the stratum radiatum (r). (G) T1W MRI (48 min, 25×25×250 µm^3) after Gd injection shows a contrast similar to T2W MRI and to (H) a histological image[2] (neurons are stained) except the mossy fibers (white arrow in F) and myelinated fibers (black arrows). Gd accumulates hardly in the granular layer (black arrowhead) but moderately in the polymorphic layer (p in F) and predominantly in the molecular layer (m). Gd also accumulates hardly in the pyramidal layer (white arrowhead in F) but moderately in the layer of mossy fiber (white arrow in F), which shows up in T1W MRI as a narrow gray band between the pyramidal layer and the most enhanced stratum radiatum and lacunosum (r and l in F). Also in the subiculum (s) and parasubiculum (ps), Gd accumulation is in inverse relation to the cell density.

Fig. 3 (below) shows the cerebellum in sagittal sections in (A and C) T1W (12 min) and T2W MRI (161 min) at 30×30×300 µm^3 before or (B and D) after Mn injection, respectively. T1W MRI shows the Mn accumulation in the Purkinje cell layer more clearly than T2W MRI, where only a slight signal reduction of the layer can be seen (black arrowheads). (E) T1W MRI (96 min, 30×30×300 µm^3) after Mn injection shows a contrast similar to (F) a histological image[2] (neurons are highlighted). Mn enhances the Purkinje cell layer clearly, the surrounding cortical layers moderately, and the white matter (w) only slightly. (G) T1W MRI (12 min, 30×30×300 µm^3) after Gd injection shows a contrast similar to T2W MRI and to (H) a histological image[2] (neurons are stained) except the white matter (w). Gd accumulates predominantly in the molecular layer (bar), moderately in the granular layer (*), but hardly in the Purkinje cell layer between them or in the white matter.



Discussion The present work shows Mn-enhanced T2W MRI and Gd-enhanced MRI of cellular layers. The observed pattern of Gd enhancement is in excellent agreement with the expected extracellular space of the tissue while the Mn enhancement partly differs from the expected intracellular space of the tissue.

Conclusion Mn- as well as Gd-enhanced MRI provides new insights in histology and radiology of the brain *in vivo*.

References 1. Koretsky AP, Silva AC. Manganese-enhanced magnetic resonance imaging (MEMRI). NMR Biomed. 2004;17:527-31. 2. Mikula S, Trotts I, Stone JM et al. Internet enabled high-resolution brain mapping and virtual microscopy. Neuroimage 2007;35:9-15. 3. Jinno S, Aika Y, Fukuda T, et al. Quantitative analysis of GABAergic neurons in mouse hippocampus with optical disector using confocal laser scanning microscopy. Brain Res. 1998;814:55-70.