In vivo Visualization of Cytoarchitecture to Define Boundaries in Cortex, Thalamic Nuclei and Superior Colliculus using Manganese Enhanced MRI.

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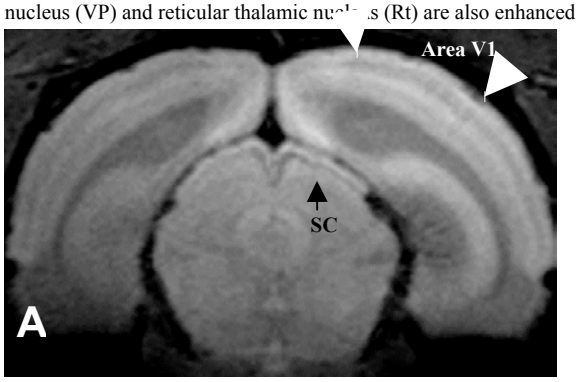
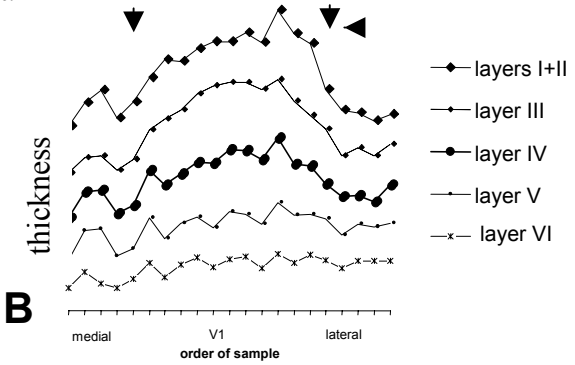
Introduction The cerebral cortex is composed of cells whose packing allows them to be separated into layers. The description of these layers is known as lamination. Cortical layers serves as a reference within brain because different layers have different input-output circuitry and therefore serve different functions. Moreover, differences in lamination have been used to differentiate cortical areas from one another. Parcelation based on cellular differences in different layers is known as cytoarchitectonics. Based on the thickness of cortical layers and neuronal density, the cortex can be divided into different cytoarchitectonic areas, each with unique functions. For instance, primary visual (V1) and primary somatosensory (S1) areas, have been identified due to the presence of densely packed cells in a thick layer IV which delineates the borders with adjacent cortical areas. Recent studies based on converging evidence using modern neuroscience methods have successfully validated cytoarchitectonics for delineation of a variety of cortical regions in many species (1-3). A shortcoming of present cytoarchitectonic techniques is they require histology on fixed and sectioned tissue. Manganese-enhanced MRI (MEMRI) has proven useful for visualizing anatomical structures, neuronal connections and activity in the brain *in vivo* (4). Previous studies have successfully demonstrated that systemic administration of $MnCl_2$ leads to excellent MRI contrast of brain anatomy including layers in the frontal and parietal cortex, hippocampus, cerebellum and olfactory bulb (5, 6). The primary goal of this study was to examine the application of MEMRI for delineating cortical areas,in addition we examined other subcortical structures to test if MEMRI can be used to identify thalamic nuclei and laminar structures in the superior colliculus.

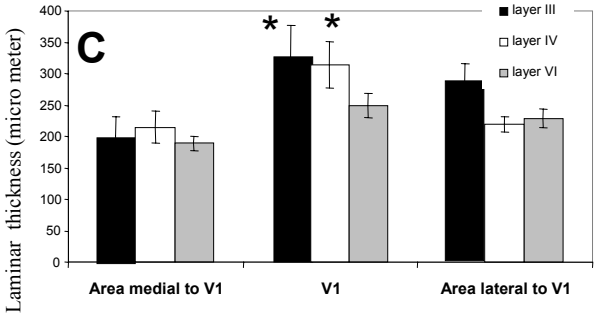
Materials and Methods Five S-D rats were used for this study. Animals were anesthetized with isoflurane (initial induction 5%, maintenance 2%). $MnCl_2$ was slowly delivered (0.2 µl/min) into the right lateral ventricle in 3 animals (15mM, 25 µl) and 3rd ventricle in the other 2 animals (25mM, 20 µl or 30mM, 30 µl) through a Hamilton microsyringe. Two animals were imaged at 20, 36, 58, and 72 hrs post-injection, and another three were imaged 72 hrs post-injection. One animal was followed up with 8 hour postmortem imaging after the last survival scan. MEMRI was performed at 11.7T, 31-cm bore magnet interfaced to a Bruker Avance console. An in-house made 15-mm-diameter receiving surface coil and a 90-mm-diameter birdcage transmitter coil were used to improve RF homogeneity and sensitivity. T1-weighted spin echo 3D images were obtained with the following parameters: TR/TE=11.3/330 ms; matrix size=256 x 256 x 180; FOV=25.6 x 25.6 x 18 mm. This yields a high-resolution 100 µm isotropic voxel size. The total acquisition time for each scan was 115 minutes. Image reconstruction and analysis were performed using ParaVision (Bruker Medical GmbH), and Photoshop (Adobe System). Laminar thickness of cortical layers in the occipital lobe was measured in the coronal plane. Results from MEMRI were later correlated with rat brain atlas (Paxinos and Watson, '98) for further validation.

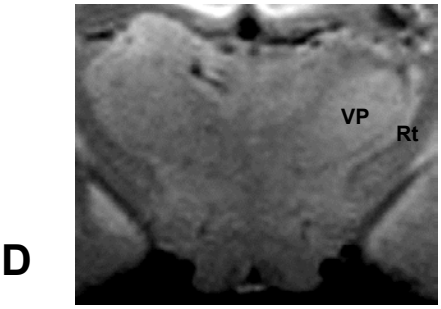
Results

A) MEMRI reflects manganese distribution in rat occipital cortex in the coronal view 72 hrs post-injection. The cell dense layers II and IV are enhanced greatly, in contrast to the less enhanced layers III, V, and VI. The thickness of layer III, and VI in contrast to the lower layers reveals clear area V1 boundaries (arrow heads) with adjacent cortical areas. Also note laminar structures in the superior colliculis (SC) are also enhanced. **B)** Line chart showing the thickness of layers I+II, III and IV increase within area V1, compared to cortical regions located medially or laterally. Arrow heads indicate borders of area V1 delineated by MEMRI. **C)** Bar plot showing (group data) laminar thickness in layers III and IV of area V1 is significantly higher than its adjacent regions. **D)** Thalamic nuclei, such as ventroposterior nucleus (VP) and reticular thalamic nucleus (Rt) are also enhanced by manganese.









Conclusions MEMRI can *in vivo* precisely delineate areal boundaries of the cortex and nuclei/lamination of the subcortical structures. The implication of the study enables noninvasive imaging of the brain to visualize cortical areas and subcortical structures, and will also be useful to study the normal and pathological brains. Furthermore, it will be valuable for localizing the site and extent of lesions without postmortem histological staining, useful for evaluating cortical functions mediating behavioral deficits or physiological testings.

References

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