Segmentation of the rat hippocampal mossy fiber network from MEMRI under inhomogenous B1 field

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INTRODUCTION:
Manganese Enhanced Magnetic Resonance Imaging (MEMRI) has been successfully used in the visualization of the hippocampal mossy fiber networks in the rat brain[1]. Although the hyperintensity of mossy fiber in the T_1-weighted image can be delineated manually, automatic segmentation is often difficult in the presence of varying signal intensity across the image caused by small surface coils used to obtain high signal to noise ratio (SNR). A robust segmentation framework would facilitate the study of the morphology of this network. This study investigated methods to reduce the intensity inhomogeneity and then automatically segment the area of interest.

METHODS:
The local IACUC committee approved all experiments conducted in this study. **MRI acquisition:** Wistar rats (n=5, ~300g) were used in this study. 100mM $MnCl_2$ solution was infused at a dosage of 80mg/kg and rate of 1.5ml/hr. Rats were then scanned 24 hrs after infusion. A Varian 9.4T MRI with a volume coil for transmit and a 12mm surface coil for receive was used to acquire T1 weighted MPRAGE images with the following parameters: $T_E/T_R/T_I$=3.90/8.01/1.00 ms, flip angle=10°, FOV=25.6x25.6x12.8 mm^3, matrix=256x256x128, NEX=6. Three slices in the rostral region of the hippocampus containing the mossy fiber network were selected as the volume of interest. **Inhomogeneity correction:** Two methods were compared. The first was the N3 inhomogeneity correction algorithm[2] implemented in MIPAV (NIH, USA). The second method consisted of high-pass filtering. The original image was passed through a 7x7x7 mean filter twice and then divided by the original image. **Segmentation:** Automatic segmentation was performed using a multi-level (3 classes) Otsu thresholding method[3] to first extract voxels belonging to the class with the highest intensity. Clusters of pixels with area of less than 10 pixels were then removed to produce the final segmented ROI. Manual segmentation was performed on the original image by a tester who was blinded to the results of the B1 correction and automatic segmentation.

RESULTS AND DISCUSSIONS:
N3 is widely used for correcting intensity non-uniformity due to its non-assumption of restrictive models and non-biaseness towards actual anatomy[2]. The N3 correction results in a homogeneous intensity profile across the image (Fig 1b), in which the mossy fiber network is not well distinguishable from other regions (Fig 1e). On the other hand, the high-pass filtering improved the intensity profile (Fig 1c) and enhanced the mossy fiber network (Fig 1f). The strong dependence on actual anatomy (i.e. local signal intensity) often makes high pass filtering inhomogeneity correction undesirable but this can be used to enhance both the intensity and sharp edges of the mossy fiber network. Comparison of the segmentation protocol that uses high-pass filtering to homogenize the original image followed by multi-level Otsu thresholding and removal of smaller unconnected regions with manual segmentation gave a t-value of 0.390>0.005 and an average true positive (TP) rate of 91.8 % (table 1).

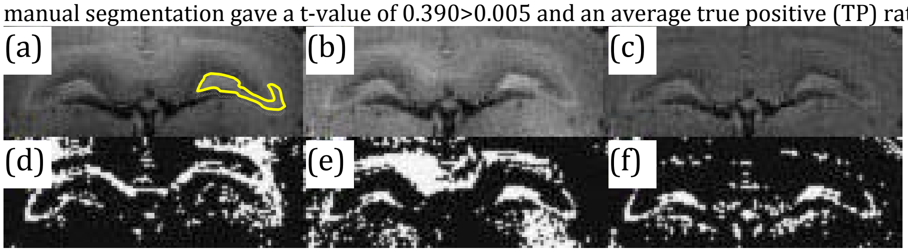

Figure 1. Comparision of the 2 methods of intensity inhomogeneity correction. (a)Original image. Mossy fiber network is outlined in yellow.(b) N3 corrected image. (c) High-pass filtered image. (d) Thresholded original image. (e) Thresholded N3 corrected image. (f) Thresholded high-pass filtered image.

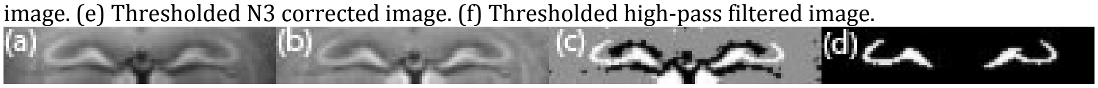

Figure 2. Segmentation protocol. (a) Original image. (b) High pass filtered image. (c) Muti-level Otsu thresholding. (d) Final segmented image after removal of unconnected regions.

Table1: Comparison between manual and automatic segmentation

Subject	Volume / Number of voxels		TP rate = $\frac{MS \cap AS}{MS} \times 100\%$
	Manual Segmentation MS	Automatic Segmentation AS	
S 1	466	428	91.8
S 2	482	532	90.6
S 3	545	495	90.8
S 4	557	562	99.1
S 5	535	464	86.7
	T-TEST=0.390>0.005		Average=91.8

References: (1)Immonen RJ et al. NeuroImage 40(2008)1718-1730. (2) Sled JG et al. Lect. notes in Comp. Sci. 1230(1997)459-464. (3) Otsu N. IEEE Trans. Syst. Man Cybern. 9(1979)62-66.