Probabilistic atlas of the C57BL/6J mouse cerebellum

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Introduction: The cerebellum is a major brain structure central for the voluntary control of the motor system and is implicated in a range of movement disorders. Mice are a well validated models for studying the human cerebellum and creation of digital atlases that can capture biological variability in control and diseased mice are critical for understanding the underlying basis of human neurological disease[1,2]. Paper based atlases, based on cytoarchitectural delineations, do not capture anatomical variability in the cerebellum and are of limited use in revealing disease mechanisms. Magnetic resonance imaging (MRI), which permits the registration and statistical analysis of multiple data sets, can be used to create digital atlases to assess anatomical variability[3-5]. A current shortcoming is that existing MRI-based atlases lack sufficient details of cerebellar architecture to enable the necessary statistical and computational analysis. In this study we developed 1) a detailed protocol for segmenting the *ex vivo* C57BL/6J cerebellum on high-resolution MR images and 2) a probabilistic atlas of the C57BL/6J cerebellum.

Method: Eighteen animals were perfused and fixed with 4% paraformaldehyde and 0.1% Magnevist®. Brains were extracted and incubated in 0.1% Magnevist/PB for 4 days, placed in Fomblin and imaged on a 16.4T (89mm) Bruker micro-imaging system using a 15 mm SAW coil (M2M Imaging, USA). MRI data were acquired using a 3D gradient echo sequence with TR/TE/FA= 50ms/12ms/30°, 82 KHz spectral bandwidth, and 8 excitations with an acquisition time of 5h 15mins to produce T_1/T_2*-weighted images at 30μm^3 isotropic resolution.

Images were placed in the stereotaxic Waxholm space[6] and a symmetric model was created using a recursive non-linear hierarchical fitting strategy similar to Fonov *et al*.[7]. The final fitting step used a nonlinear transformation with a step size of 30μm. This resulted in a model with double the resolution of the original input data (15μm^3 vs. 30μm^3). The components of the cerebellum were delineated on the basis of differences in signal intensity and/or location relative to cerebral fissures, and partitioned using vector-based segmentation (Wacom Cintiq). Surface smoothed three-dimensional surface reconstructions were created in Amira (Visage Imaging, Inc.).

A probabilistic model was created using the same method as the ICBM152 model[8]. Namely the traced structures were nonlinearly transformed back to native space before a lower order nonlinear native space to model space transform was applied. In our case this was a grid transform with a step size of 4 times that of the voxel size (60um).

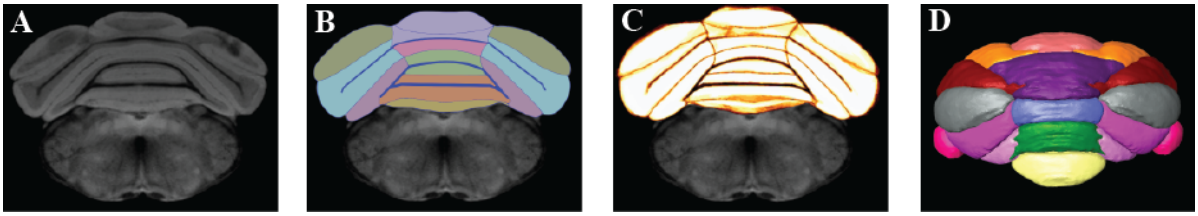

Figure 1: (A) Axial image from average model. (B) Axial image from labelling of cerebellum. (C) Axial image from label boundary variability map of cerebellum. (D) Three-dimensional surface reconstruction of cerebellum.

Results: This study developed a detailed parcellation scheme for segmenting the C57BL/6J mouse cerebellum. It is based on reproducible MRI landmarks, made visible as a consequence of the higher signal-to-noise ratio achieved during group averaging and the inter-subject anatomical reproducibility of structures. The segmentation protocol creates standardized structural delineations that can be applied to future anatomical studies in the mouse. In addition, a digital atlas was generated based on T_2*-weighted signal intensities containing over 35 structures with average region volumes and probability maps for each structure, to demonstrate the confidence range for anatomical mapping.

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