**Brief Description of the template set**

**MRI Sample:**

Thirty (30) Wistar rats were used, distributed in five groups of six rats each (6, 7, 8, 9, 10 weeks old, body weight ranges of 214 ± 19 g, 275 ± 21 g, 312 ± 9 g, 331 ± 17 g, 364 ± 18 g, respectively). All procedures and protocols were performed in agreement with the policies established by the “Animal Care Committee” at Tohoku University, Sendai, Japan.

MRI data were acquired using a 7.0 T Bruker PharmaScan system (Bruker Biospin, Ettlingen, Germany) with a 38-mm-diameter birdcage coil. A T2-weighted image for each rat was obtained using a respiratory-gated 2D TurboRARE sequence with fat suppression under the following parameters: TR = 10971 ms, TEff = 30 ms, RARE factor = 4, effective spectral bandwidth = 100 kHz, flip angle = 90 degree, field of view = 32 x 32 mm², matrix size = 256 x 256, in-plane resolution = 125 x 125 μm², number of slices = 128, slice thickness = 0.3 mm, slice gap = 0 mm and number of averages = 16.

**Average template construction:**

All T2 MRIs were intensity corrected from coil inhomogeneities and intensity equalized. The brain of each T2-weighted MRI was extracted and non-rigidly registered to the median size one using elastix ([http://elastix.isi.uu.nl](http://elastix.isi.uu.nl)) (Klein et al., 2010) in a hierarchical multigrid and multiresolution approach. A transformation to the average unbiased centroid space was assessed by inverting the average transformation. For each MRI, the pyramidal transformation scheme plus the transformation to the average centroid space were concatenated to resample all MRIs to the centroid space. All warped MRIs were averaged to construct the template image. This methodology was previously adopted in (Kovacevic et al., 2005).

**Tissue spatial priors:**

Each brain MRI was filtered with an anisotropic smoothing filter. Its intensity histogram was fitted with four Gaussian curves, representing surrounding low intensity non-brain tissue, white matter, gray matter and cerebrospinal fluid, and three non-parametric curves representing partial volumes. The Gaussian curves provided preliminary segmentations into gray matter, white matter and cerebrospinal fluid which were used as tissue spatial priors in SPM5 to obtain the definitive ones. Finally, these were
warped with the nonlinear transformations to the centroid space and averaged to obtain the tissue spatial priors of gray matter, white matter and cerebrospinal fluid.

**Labeled atlas of the cortex:**

Forty-eight (48) cortical structures of the 6th edition of the Paxinos and Watson (Paxinos and Watson, 2007) atlas were digitalized using Adobe Illustrator and T2-CARB (www.neuronic.sa). A set of 40 landmarks were identified in this space and in the average template. Then, the digitalized atlas was non-linearly registered to the centroid space by means of the Approximate Thin Plate Splines registration method (Rohr et al., 2001), based on matching corresponding landmarks points in the landmark sets. This registration was refined using points in the gray/white matter interface. The final warped atlas was masked with the gray matter segmentation and manually edited for minor corrections.

**Image description:**

All images of the template set are stored in NIFTI format (float type for grayscale images and unsigned 8-byte integer type for the digital atlas). To avoid conflict with SPM, voxel size was multiplied by 10, thus 1mm actually means 100μm. The X, Y and Z directions lie approximately along the left-to-lateral, ventral-to-dorsal, and caudal-to-rostral directions respectively. Note that this is not the convention of the Paxinos & Watson space, but more likely the usual position of the rat inside the scanner.

**Template set uses:**

Using any version of SPM, an individual image can be normalized using the average template, or segmented using the spatial tissue priors. Both results can also be obtained using the unified segmentation method (Ashburner and Friston, 2005). The resulting inverse transformation of this method can be used to label the MRI in the individual space, as usually done for humans with IBASPM (Alemán-Gómez et al., 2006) and other software. Other registration methods are also applicable (Klein et al., 2009), for example “elastix” (Klein et al., 2010), which encloses a wide variety of metrics, samplers, interpolators and transformations types, can be used as well. In this case, providing the brain mask of the template might be recommendable. Before running any method, just take care about setting the anterior commissure (AC) as the origin; flip images to have left-to-right orientation; and align (if desired) the anterior posterior direction parallel to the line joining the AC with the dorsal part of the reticular nuclei conglomerate. This will reduce computation time and avoid reaching local minima in the optimization of the cost functions.
This template set can be used for functional MRI anatomical interpretation, diffusion tensor tractography, ROI-to-ROI-based anatomical and morphological connectivity and Morphometry.

Reference List


