**Recommended level-2 analysis procedures to replicate level-1 PiB data:**

1. Download PET (50-70min) and MRI data for the 34 YC-0 and 45 AD-100 subjects from the GAAIN website.

* We recommend downloading and using the NIFTI images provided rather than converting from DICOM.

1. Manually reorient the subject data to match the Montreal Neurological Institute (MNI) template provided with the SPM8 software (in [your SPM8 path]/canonical/avg152T1.nii)
   * Use "**Display**" module in SPM8 (see p.143 in SPM8 manual)
   * Reorient the data so that the mm coordinate of the Anterior Commissure is within about 3cm from [0 0 0] and the orientation is within about 15o of MNI space.
   * A few PET images need to be flipped along the y-axis for the correct orientation. Do so by putting "-1" in the "resize (y)" box. (Do not reorient these images by putting "-3.14" in the "yaw (rad)" box as it will result in a left-right flip.)
2. Coregister subject MRI to MNI template
   * Use “**Coregister: Estimate**” module
   * Reference Image: avg152T1.nii (in [your SPM8 path]/canonical/avg152T1.nii)
   * Source Image: subject MRI
   * Keep all default parameters
3. QC subject MRI to MNI template coregistration
   * Use "**Check Reg**" function in SPM8
   * \*The goal of this registration is to bring subject MRI in rough alignment with the MNI template. It is acceptable if the two images are not perfectly aligned\*
4. Coregister subject PET to subject MRI
   * Use “**Coregister: Estimate**” module
   * Reference Image: subject MRI
   * Source Image: subject PET
   * Keep all default parameters
5. QC subject PET to MRI coregistration
   * Use "**Check Reg**" function in SPM8
   * Ensure that subject PET image is well-aligned to subject MRI
6. Apply unified segmentation to subject MRI
   * Use "**Segment**" module in SPM8
   * Keep all default parameters
   * If you do not wish to save any output image files, under "Output Files", you can change "Grey Matter" and "White Matter" to "None", and "Bias Corrected" to "Don't Save Corrected".
7. Apply normalization parameters to transform subject PET and subject MRI into MNI-space
   * Use “**Normalise: Write**” module
   * Parameter File: \*\_seg\_sn.mat (forward transformation parameter)
   * Images to Write: subject PET and subject MRI
   * Change "Bounding box" to [NaN NaN NaN; NaN NaN NaN]
     + This tells SPM to use the same bounding box as the template.
8. QC subject PET to MNI-template transformation
   * Use "**Check Reg**" function in SPM8
9. Download the standard VOIs from the GAAIN website

* Download the 2mm resolution VOIs if you normalized to the avg152T1.nii template as described above (to match the 2mm resolution of that template).
* We recommend downloading and using the NIFTI images provided rather than converting from DICOM.

1. Extract PiB SUV values from normalized PET images (e.g. **w**AD01\_PiB\_5070.nii) using standard VOIs
   * One example program to use is **fslstats** from the FSL Toolbox
2. Re-calculate individual CTX SUVr values for each reference VOI of interest
3. QC individual CTX SUVr values
   * For each subject, calculate the percent difference from the CTX SUVr values published in the Centiloid paper (**Supplementary Table 1**).
   * If any individual difference is **greater than 5%**, re-examine the processing steps and the quality of registrations and normalizations for that subject. A difference of that magnitude likely indicates a processing error, with image registration being the most common source of problems.
4. Re-calculate mean CTX SUVr values of the 34 YC-0 subjects and 45 AD-100 subjects
5. QC group mean CTX SUVr values
   * For each group (YC-0 and AD-100), calculate the percent difference from the mean values reported in **Table 1** of the Centiloid paper.
   * This difference should be **less than 2%** to pass the QC.
6. Convert individual CTX SUVr values into Centiloids, using equation 2.2.1 in the Centiloid paper.
   * **CL = 100×(PiBSUVrIND\* – PiBSUVrYC-0\*) ⁄ (PiBSUVrAD-100\* – PiBSUVrYC-0\*)**
7. QC re-calculated Centiloid values
   * Perform a linear correlation of the re-calculated Centiloid values to the Centiloid values reported in the paper (**Supplementary Table 1**).
   * The expectation is that the **slope will be between 0.98 and 1.02**, the **intercept will be between −2 and 2 Centiloid** and the **R2 will be >0.98**.
   * This should be included as supplemental data in the first publication with the slope, intercept and R2 reported in the manuscript.