**READ ME**

* All original scripts can be found on Devon (Karl) Overson’s github: <https://github.com/devonko/MB_MUSE_new_CA>
* If you visit the github, you will see several additional scripts that are needed for the MB-MUSE diffusion analyses (FA, radiality index) that we do not need for QSM analyses.
* We focused on the Lausanne “36 ROI” parcellation, which is actually 34 ROIs.

**STEP-BY-STEP PROCESSING**

1. Prepare/copy files (*c0\_copy\_files.sh*) and run FreeSurfer for each participant
   1. Bias-correct the T1-weighted anatomical image and the T2-weighted FLAIR image: *ra0\_biasfieldcorrect.sh*
      1. Estimated time: 10-15 minutes
      2. Note: I copied T1\_bfc.nii.gz for VisAtten.05 controls, but had to redo the T2\_bfc.nii.gz because Karl is using the diffusion T2 file (*b*0)
   2. Run FreeSurfer on the T1-weighted image: *ra1\_submit\_freesurfer.sh*
      1. Estimated time: ~4 hours
   3. Copy other relevant files over: *copy\_files.sh.*
      1. This copies and bias-corrects the FLAIR T2 for VisAtten.05, and copies all QSM images aligned to T1 space
      2. To obtain QSM image: see QSM/Documentation/SWAN\_processing.docx
      3. Estimated time: 15-20 minutes (longest part is the bias-correct step)
   4. Run FreeSurfer on the FLAIR image: *ra2\_t2\_bbreg.sh*
      1. This script runs Freesurfer on the T2 image to improve the accuracy of the pial surface estimation.
      2. Estimated time: ~3 hours
      3. Note: Here is where I started running all QSM.02 and VisAtten.05 participants in batch together, using the new T2\_bfc files (FLAIR, not diffusion *b*0!) for VisAtten.05 controls (obtained in Step 1c).
2. Run the *c4\_lausanne.sh* script
   1. This script parcellates the anatomical images output by FreeSurfer into the 34 bilateral ROIs of interest (as well as 60, 125, and 250 ROI parcellations).
      1. Based on the Desikan-Killiany atlas (Desikan et al., 2006).
   2. Estimated time: ~ 40 minutes.
   3. *Note*: the *easy\_lausanne* function was revised to be compatible with python3.9 instead of python2.7 by Chris Petty on 04/06/2023.
   4. *Note*: the *easy\_lausanne* function requires a diffusion or bold image as the target image, but I instead used the T2\_bbregister.nii.gz image as the target.
3. Run the *c5\_surf\_calc.sh* script
   1. This script produces some intermediate files from Freesurfer that will be used in *mat6*: the lh.scanner.\* and rh.scanner.\* files
   2. It reorients the files from FreeSurfer space to scanner/RAS space.
   3. Instructions for running are at the top of the script (just copy/paste into terminal)
   4. Estimated time: < 5 minutes

1. Run the *mat6\_registration\_anat.m* script
   1. I run this script via GUI by typing “matlab” into a terminal window when connected to the Linux with graphics enabled (ssh -Y)
   2. This script generates your cortical “columns”
   3. It connects corresponding pairs of vertices from the pial and gray/white matter boundary meshes obtained using FreeSurfer on the T1-weighted anatomical image, which are obtained separately for each hemisphere.
   4. Estimated time: ~3 minutes per participant (i.e., 3\*N)
   5. The lines connecting these two vertices are then saved as mrtrix tracks (i.e., .tck format), although NOTE that they are NOT true, traditional streamlines.
2. Run the *c7\_QSM\_sample\_new\_reg.sh* script
   1. This script essentially disaggregates your previously obtained columns into 21 separate, equidistant gray matter segments
   2. Then, using the mrtrix *tckresample* function, the script will first resample the one single column into 21 segments
   3. It will then sample (estimate) QSM at each of the 21 segments for each column and save this file as an output
   4. I just run this run one by pasting directly into the terminal window
   5. Estimated time: < 10 minutes total
   6. To visualize and inspect the outputs from this step: mrview underlay\_image.nii.gz -tractography.load WholeBrainTracks\_T2/rh\_tracks\_21points\_anat.tck
3. Run the *mat9\_refined\_ROIs\_hard\_cutoff\_QSM.m* script
   1. Open matlab gui, as in as Step #4
   2. This is where you bin the 21 sampled values along the individual columns according to 1 of the 34 ROIs
   3. You have to choose the cortical thickness thresholds that you want to use
      1. Currently, using minimum thickness of 0.5mm and maximum of 6mm – seems to be the most liberal
   4. There are also different curvature thresholds, which are used to get at the crown, fundus, and bank anatomical separations (or whole brain)
   5. From this script, you should have the following outputs: 34 ROI x 4 curvature matrix, per participant.
   6. Estimated time: < 10 minutes (for n = 44)
4. Run the *mat10\_nofilter\_combine.m* script to combine the mat9 outputs across subjects
5. Open *mat11\_depth.m* and manually update / run analyses / create plots