Imaging for Neuroscience

Homework – Group 8

The goal of this homework is to investigate the relationship between resting-state functional MRI and diffusion MRI data. The following scans, collected in Verona, were acquired with a 3T MR scanner on a healthy subject.

DATASET

The provided data are organized as follows in the **Data** folder.

- Subject's **Atlas** folder:
  - Hammers_2_T1_2mm_int.nii.gz = anatomical atlas, already resampled in the T1w space image.
  - Hammers_labels.pdf = file containing the labels of atlas anatomical regions. For example, regions 1 and 2 represent the right and left hippocampus, respectively.

- Subject's **DMRI** folder:
  - diffusion_volumes.nii = the diffusion weighted volumes of the diffusion acquisition, resampled in the T1w space image.
  - diffusion_brain_mask.nii = the binary mask delineating the volume occupied by the brain in the diffusion scans.
  - bvals = txt file containing the bvalue with which each diffusion volume was acquired.
  - bvecs = txt file containing the gradient direction with which each diffusion volume was acquired.

- Subject's **FMRI** folder:
  - FMRI_2_T1_2mm.nii.gz = 4D resting-state functional data (TR=2.6sec). These data have been already processed with the following procedures: slice timing, distortion correction and motion correction (using a rigid spatial transformation) and resampling in the T1w space image.
  - MOCOparams.mat = estimated motion parameters (matrix 225x6).
  - FDparams.mat = framewise displacement parameters (matrix 225x2).

- Subject's **Structural** MRI folder:
  - T1_2mm.nii = T1w structural image.

The **Matlab_tools** folder contains useful tools:

- Nifti_tools
- betweenness_wei.m
- fdr_bh.m

AIMs of the study:

1. To perform some pre-processing steps on functional data.
2. To compute the subject's functional connectivity (FC) matrix and derive graph measures.
3. To fit the diffusion tensor and derive its most important metrics
4. To investigate the impact of various strategies of signal normalization on the computation of the DTI metrics (FA and MD)
5. To investigate at the ROI level whether there is a relationship between the DTI parameters and the computed connectivity measures.

Resting state fMRI analysis:

1. Data Preprocessing

    a. Use SPM software (https://www.fil.ion.ucl.ac.uk/spm/software/spm12/) to segment the T1w structural image into GM, WM and CSF tissues, obtaining the tissues probability maps. (This step may take a few minutes)
    b. Threshold the three tissue probability maps and create for each tissue a binary mask. Erode the masks (*imerode* matlab function) and compute the mean fMRI signal of WM and CSF.
    Provide a justification of the employed thresholds and of the erosion parameters.
    c. Create the sumEPI image by summing the EPI volumes in the 4th dimension. Create a binary mask for the sumEPI image and provide a justification of the employed threshold.
    d. Mask the Hammers atlas with two masks: the GM binary mask and the sumEPI mask.
    e. ROI time activity curve extraction: for each masked ROI of the atlas, extract the mean fMRI signal. Using the *Hammers_labels.pdf* file, discard for the following analyses the masked ROIs with less than 15 voxels and those belonging from: amygdala, cerebellum, brainstem, corpus callosum, substantia nigra, ventricles.

2. Data Denoising

    a. Noise regression:
    For each ROI, remove the non-neural undesired fluctuations from its temporal dynamic using a linear regression approach. Perform the regression using 14 regressors: the six parameters estimated during the motion correction step (provided in the *MOCOparams.mat* file), their six temporal derivatives (look for a reasonable method to compute them in matlab), and the mean signals of white matter and cerebrospinal fluid. Before being used, each regressor must be converted to Z scores. (Matlab functions: *zscore* and *lscov*)
    Visualize the regression matrix.
    b. Temporal filtering:

To take the slow and very high-frequency components out, filter the signals obtained at point 2.a. with a band pass filter choosing reasonable cut-off frequencies. (Matlab functions: *buttord*, *butter* and *filtfilt*). Set the upper-cutoff lower than 0.2 Hz. Provide a justification of the selected filtering frequency cut-offs.

3. Volume censoring

   Discard the volumes that are affected by motion artefacts. These volumes have a framewise displacement greater than 0.2 mm (the FD values in mm are reported in the first column of the Fdparams.mat matrix). Since motion artefacts could be present also in the neighbouring volumes, discard also one volume before and two after the affected volume.

4. Check of preprocessing step

   Plot the original time-course of the left thalamus region and what was obtained after each denoising step. Do you see a drift in the original signal? If so, is the denoising able to remove it?

5. Static FC Matrix Computation

   Compute the pairwise Pearson's correlation (and the relative *p*-value) between the time-series of the ROIs. Visualize the FC matrix after applying the Fisher's *z*-transform to the coefficients (*atanh* matlab function).

6. Multiple Comparison Correction

   Perform a multiple comparison correction with Bonferroni or False Discovery Rate approach and a significance level alpha=0.05. If you decide to use the FDR method, use the provided *fdr_bh.m* function. Provide a justification of the chosen correction method.

7. Graph Measures

   To summarize the functional connectivity in terms of node centrality, for each ROI compute the node degree, the eigenvector centrality (with *eigenvector_centrality_und* provided function) and the normalized betweenness centrality (with *betweenness_wei.m* provided function). In the metrics computation, consider only the statistically significative functional connections obtained after the multiple comparison correction at point 6. Plot the node degree, the eigenvector centrality and the normalized betweenness centrality of the ROIs using the *stem* matlab function. Which are for each metric the 10 ROIs with the higher metrics values? Provide the indices of these regions.

Diffusion MRI analysis:

1. Diffusion signal visualization & understanding

   a. Load the diffusion volumes, the bvals file and the bvecs file.

How many different DWIs have been acquired?
Excluding b=0, how many diffusion shells does this acquisition feature? (consider a small tolerance **α**=±20 s/mm^2 in the shell definition).

b. Plot the diffusion signal of a voxel populated principally with cerebrospinal fluid (hint: you can skip steps 1a-1b and get back to them when you have computed the DTI metrics, which can aid you in the selection); is the diffusion signal ordered by its b-value? If not, sort it so that the signal points corresponding to the same shell are shown consequently (and shells are ordered in an ascending fashion).
Provide both the plot of the unsorted signal and the sorted one.
c. By visually inspecting the sorted signal, provide a brief comment both on the inter b-value and on the intra b-value variabilities. Why do these signal variations occur?

2. Diffusion tensor computation

a. From the entirety of the diffusion volumes data, create a new 4D matrix containing only the volumes corresponding to b=0 s/mm^2 and to the shell closest to b=1000 s/mm^2 identified at point 1a.
b. Fit the voxel-wise diffusion tensor (using the linear least square approach seen in class) on the whole brain diffusion data created at point 2a. When performing the log(S/S0) transformation of the signal, use as S0 the voxel-wise mean value of all b=0 volumes of the available dataset. Use the eigenvalue/eigenvector decomposition to recover the FA / MD/ RD indices.
c. provide the visualization of the FA and MD maps for the central slice, as well as its colour-encoded map weighted by FA.
d. Mask the FA and MD maps (as done for the Hammers atlas in fMRI: point 1.d), extract their mean values in each ROI (fMRI: point 1.e.).

DMRI/fMRI integration

1. Visual inspection

Visualize the scatterplot of these variables:

- ROIs node degree versus ROIs FA
- ROIs node eigenvector centrality versus ROIs FA
- ROIs node normalized betweenness centrality versus ROIs FA
- ROIs node degree versus ROIs MD
- ROIs node eigenvector centrality versus ROIs MD
- ROIs node normalized betweenness centrality versus ROIs MD

2. Quantitative results

Compute and provide the Spearman correlation between the six pairs of variables of point 1. Discuss the results: is there a statistically significant relationship between any pair of variables?

Submit your homework in the elearning page of the course in the Homeworks section. The submission MUST be completed one week before the day of the exam. You are required to do the homework in English (report, codes etc.).

In a .zip folder (named Hwnumber_Name_Surname) you are required to turn in a copy of your own commented Matlab code(s), as well as:

1) a brief and complete presentation in power point/pdf of the performed analysis (methods, problems and issues, results, discussions, figures …). Mandatory maximum 10 slides (excluding the first slide with title and your name)

2) A .mat file with the following variables:

GC : the regression matrix (fMRI: point 2.a)
a matrix with the ROIs fMRI signal after the denoising step (noise regression + temporal filtering) (fMRI: point 2.b)
zFC : the z-Fisher transformed functional connectivity matrix (fMRI: point 5)
zFC_corr : the z-Fisher transformed functional connectivity matrix after multiple comparison correction (fMRI: point 6)
MD map (DMRI: point 2.b)
FA map (DMRI: point 2.b)
RD map (DMRI: point 2.b)
DEG : a vector containing the ROIs node degree (fMRI: point 7)
EC : a vector containing the ROIs node eigenvector centrality (fMRI: point 7)
BTW_NORM : a vector containing the ROIs normalized betweenness centrality (fMRI: point 7)