

## MENGA 3.1: User's Notes

### INTRODUCTION

MENGA (Multimodal Environment for Neuroimaging and Genomic Analysis) is a software platform that allows the investigation of the correlation patterns between neuroimaging data of any sort (both functional and structural) with mRNA gene expression profiles derived from the Allen Brain Atlas (ABA) database at high resolution. MENGA is written for Matlab 2012a version and above (The Mathworks Inc., MA, USA) and freely available. MENGA is operative system independent.

### 1. PREVIOUS RELEASES

If you have already installed a previous release of MENGA, do not worry: the heavy database has remained the same!

You just need to download code\_MENGA\_31.zip from the website, unzip it in MENGA directory and substitute the old 'code' folder and all the other main files with the new ones.

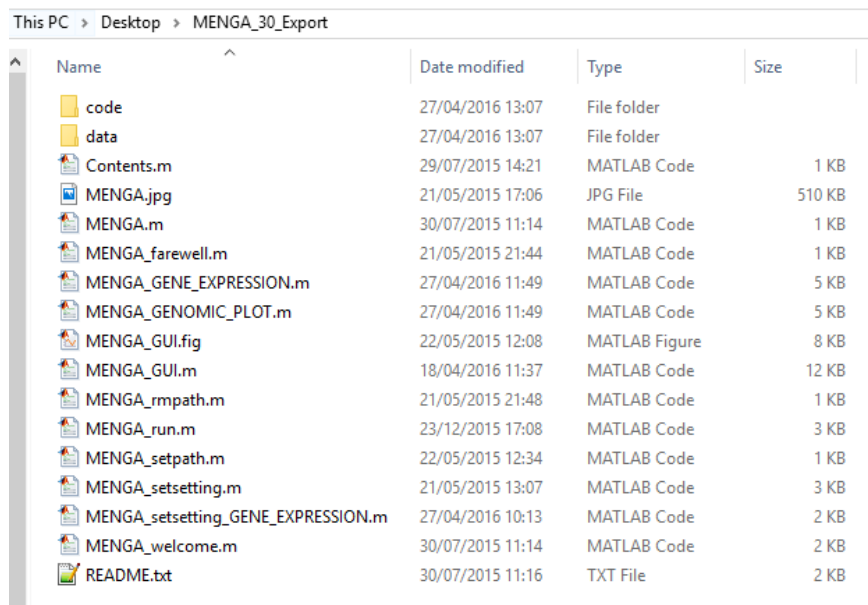
### HIGHLIGHTS

- We have developed two adds-on: *MENGA\_GENE\_EXPRESSION* and *MENGA\_GENOMIC\_PLOT* (see section 6).

### 2. GETTING STARTED

Download MENGA\_30\_Export.zip **and** code\_MENGA\_31.zip, and unzip them. Place the full content of code\_MENGA\_31 (1 folder and 15 files) in the folder MENGA\_30\_Export.

This is what it should look like:



Name	Date modified	Type	Size
code	27/04/2016 13:07	File folder	
data	27/04/2016 13:07	File folder	
Contents.m	29/07/2015 14:21	MATLAB Code	1 KB
MENGA.jpg	21/05/2015 17:06	JPG File	510 KB
MENGA.m	30/07/2015 11:14	MATLAB Code	1 KB
MENGA_farewell.m	21/05/2015 21:44	MATLAB Code	1 KB
MENGA_GENE_EXPRESSION.m	27/04/2016 11:49	MATLAB Code	5 KB
MENGA_GENOMIC_PLOT.m	27/04/2016 11:49	MATLAB Code	5 KB
MENGA_GUI.fig	22/05/2015 12:08	MATLAB Figure	8 KB
MENGA_GUI.m	18/04/2016 11:37	MATLAB Code	12 KB
MENGA_rmpath.m	21/05/2015 21:48	MATLAB Code	1 KB
MENGA_run.m	23/12/2015 17:08	MATLAB Code	3 KB
MENGA_setpath.m	22/05/2015 12:34	MATLAB Code	1 KB
MENGA_setsetting.m	21/05/2015 13:07	MATLAB Code	3 KB
MENGA_setsetting_GENE_EXPRESSION.m	27/04/2016 10:13	MATLAB Code	2 KB
MENGA_welcome.m	30/07/2015 11:14	MATLAB Code	2 KB
README.txt	30/07/2015 11:16	TXT File	2 KB

Once the files are all in the same directory, start Matlab. In 'set path' add the directory MENG\_30\_Export to the path and save.

Then type

>> MENGA

The menubar and the logo should appear.

### 3. DATA FORMATS

### Region list (.mat):

The regions of interest are loaded through a .mat file, with the list of ROIs as ABA labels. Libraries are available.

### Mask (.mat):

The brain mask to be applied to the image data is in a .mat file, in MNI space. Libraries are available (from FSL brain mask).

### Images:

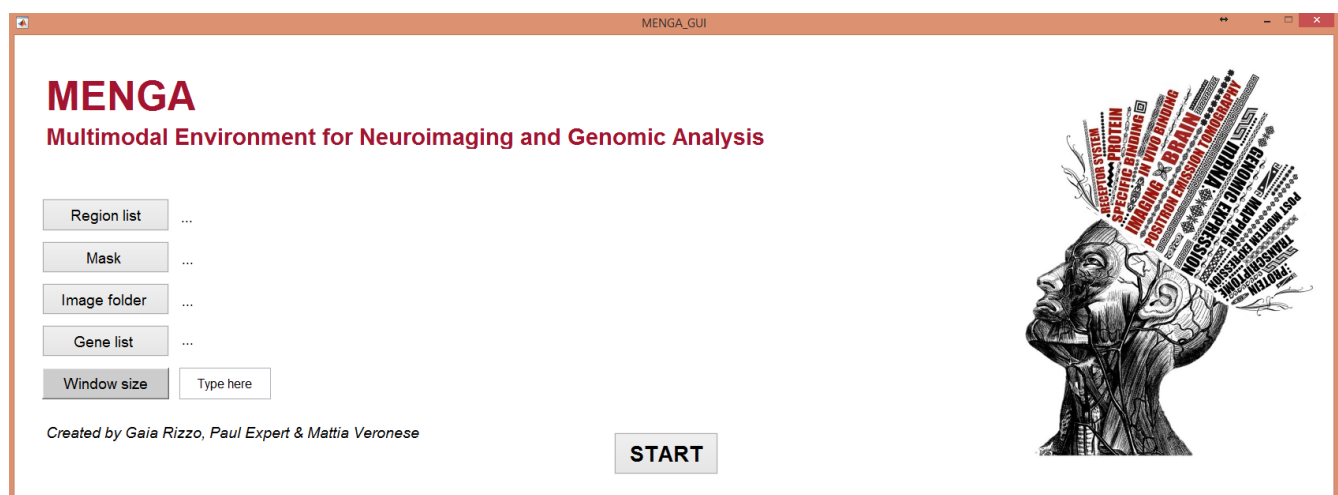
The images to be analysed must be in Analyze (.hdr/.img) or NIfTI (.nii) format. The images must be 3D, already in MNI space and oriented according to the neurological convention.

MENGA accepts images both in MNI ICBM152 coordinates (image size: 181x217x181 voxels) and FSL MNI coordinates (image size: 182x218x182) and converts them into the MNI ICBM152 convention (voxel size 1mm x 1mm x 1mm).

**Gene list (.txt):**

The list of genes must be a .txt file, with each gene in a new line, with no spaces. The genes are identified with their ABA abbreviation (see Allen Human Brain database or MENGA internal database). An example of gene file is available.

#### 4. MAIN MENU & ANALYSIS



MENGA requires the user to simply fill the five fields in the GUI to run the analysis:

**Region list:**

It determines the level of resolution to use for the analysis. The user can choose from the already implemented solutions or extend the libraries him/herself.

MENGA already includes several default lists of regions, derived from the structure (169 ROIs) and coarse level (26 ROIs) defined in the ABA. We also included in MENGA a simplified version of the coarse level, with 15 ROIs. A summary of ABA labels is reported in the Excel file included in the manual zip package.

If no region list is selected, MENGA uses the simplified coarse list as default.

**Mask:**

The user has the choice to select an image mask, i.e. a binary 3D image to limit the analysis in the areas of interest removing the background. The voxels outside the mask are set to "not-a-number" (NaN) and then discarded from further analysis. Mask definition is user-dependent, and some libraries are available.

If no mask is selected, MENGA uses the FSL brain mask (limited to the left hemisphere) as default.

**Image folder:**

The user needs to select the folder containing the images to be analysed. All the images contained in the folder will be analysed separately. In the same folder, a new folder will be created for each image (named as the image itself) containing the MENGA results. See Output section.

If no image folder is selected, MENGA exits.

**Gene list:**

The user loads the gene list. There is no limit on the number of genes that can be included in the analysis, however the user needs to consider the computational load required by an analysis including hundreds of genes.

If no gene list is selected, MENGA exits.

**Window size:**

This integer represents the size (in mm) of the window used for the image import. This should reflect the effective spatial resolution of the image being analysed.

If no value is inserted, MENGA uses 5 as default.

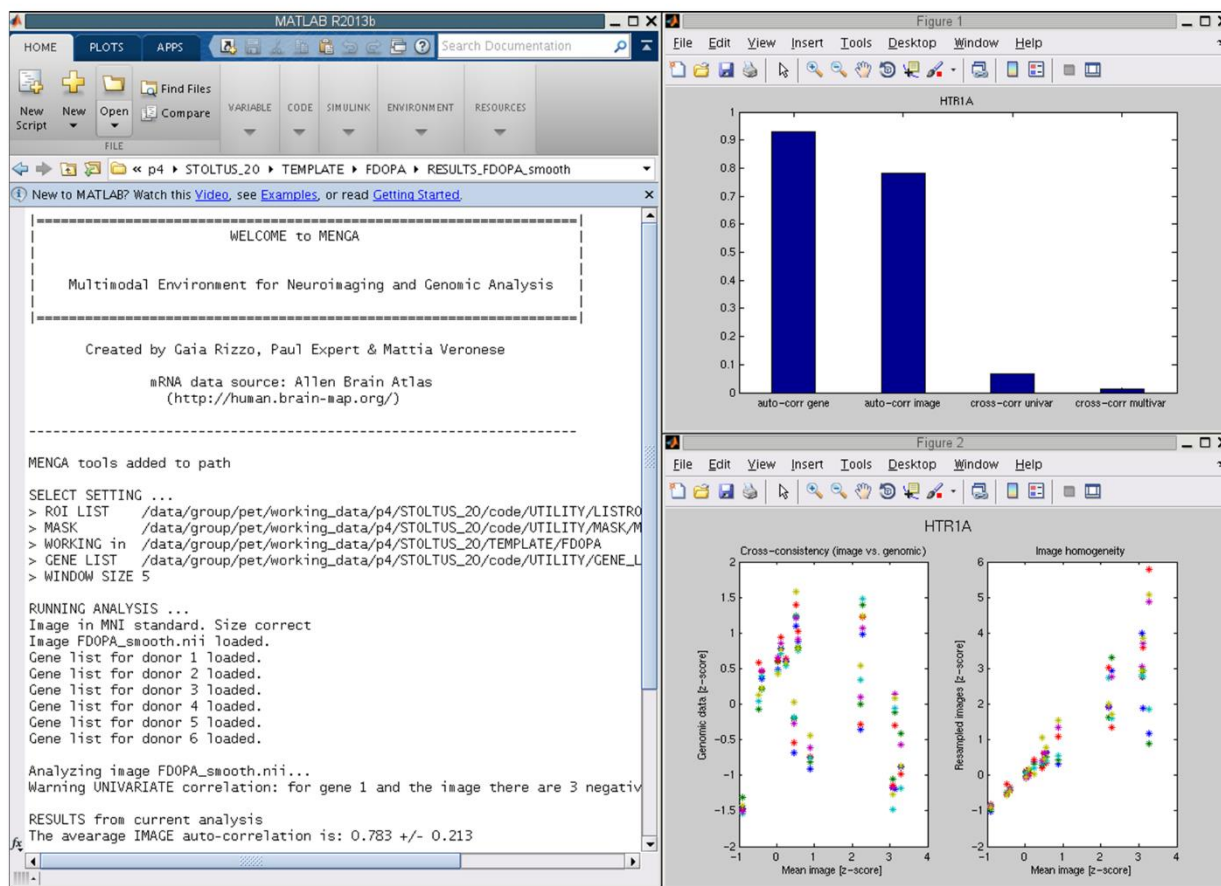
**Now just START the analysis.**

## 5. OUTPUT

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After the analysis a report with all the correlation statistics is displayed in the Matlab workspace and saved as text file (.txt) in the result folder (left panel in the figure below). The file includes also the setting information used for the analysis and a copy of the raw genomic data (in z-score).

Additionally, MENGA generates two figures (right panel in the figure below), each one saved as both .jpg and .fig files), summarizing genomic and image correlation statistics and the corresponding scatter plots.



**A Matlab file for expert users is also stored in the result directory recording all of the internal results.**

These include:

- *gen2don*: a structure with the genomic values in the ROIs for each donor (as mean, standard deviation, number of samples, etc.). Different genes are reported in different rows.
- *img2don*: a structure with the re-sampled image values in the ROIs for each donor (as mean, standard deviation, number of samples, etc.). Different images are reported in different rows. There are 6 fields because each **single** image is resampled in the genomic space of each donor (in order to have an exact 1:1 mRNA-image match).
- *genelabel*: list of genes
- *listroi*: list of ABA labels of ROIs
- *namelistroi*: list of ROI names
- *mask*: mask used
- *window size*: size of the window for image import

- *tabauto\**: matrix with the genomic and image univariate autocorrelation
- *statauto\**: matrix with the summary statistics of the genomic and image univariate autocorrelation (as mean, standard deviation, coefficient of variation, minimum and maximum)
- *tabcorr*: matrix with the genomic/image univariate crosscorrelation
- *statcorr*: matrix with the summary statistics of the genomic/image univariate crosscorrelation (as mean, standard deviation, coefficient of variation, minimum and maximum)
- *tabcorrMulti*: matrix with the value of multivariate crosscorrelation (one value for each gene)
- *pvalueMulti*: value of chance likelihood (one value for each gene)
- *direc*: matrix with information about the directionality of the univariate correlation (1 when positive correlation, -1 where negative)
- *direcMulti*: matrix with information about the directionality of the multivariate correlation (1 when positive correlation, -1 where negative)

If you need assistance just contact us!

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## 6. ADDS-ON

We have developed two independent adds-on to:

- 1) extract mRNA data in log2 values (MENGGA\_GENE\_EXPRESSION.m), in case one wants to analyse transcriptome data without having to complete the integration imaging-genomic analysis;
- 2) extract mRNA data in linear scale (MENGGA\_GENOMIC\_PLOT.m) to be used to implement the **genomic plot** (Veronese M, Zanotti-Fregonara P, Rizzo G, Bertoldo A, Innis RB, Turkheimer FE. [Measuring specific receptor binding of a PET radioligand in human brain without pharmacological blockade: The genomic plot.](#) Neuroimage. 2016 Apr 15;130:1-12. doi: 10.1016/j.neuroimage.2016.01.058. Epub 2016 Feb 2).

Both new functions exploit MENGGA database without the need to select an image folder, the only settings to define are the level of resolution (i.e. list of regions) and the gene list (i.e. the txt file with the list of genes – defined accordingly [ABA database](#)).

As results of the analysis, the adds-on reports:

- a) in the command window: mRNA values (either log2 for gene expression or linear scale for genomic plot) as mean +/- SD across donors

- b) a txt file stored in the local folder of the gene list containing the selected settings and the genomic raw data (either log2 for gene expression or linear scale for genomic plot) for all the single donors
- c) a mat file stored in the local folder of the gene list containing the selected settings and the genomic raw data (the MENGA struct gen2don, see above), 'genval' (matrix with the genomic data for all the single donors) and 'TAB\_GEN' (matrix with the mean and SD of genomic data across donors).

**NOTE:** we have developed the MENGA\_GENOMIC\_PLOT add-on to return **only** the transcriptome data in linear scale (as reported in the Neuroimage paper), since most of the time the users have already available the regions-wise PET estimates to use for the genomic plot. Image data in donor space can always be obtained using standard MENGA analysis (MENGA.m).

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