REX MANUAL

INTRODUCTION

While exploring experimental questions based on prior anatomical hypotheses, it is often useful to restrict the application of statistical models to specific brain regions. In service of this goal, SPM provides a wide range of methods for exporting image data values from image volumes at various stages of processing or statistical modeling. The *rex* tool is designed to extend those capabilities to permit the efficient extraction of image values and time series from single voxels, voxel clusters and cluster collections. In addition to data extraction the *rex* tool performs ROI-based analyses of functional data complementing SPM voxel-based analyses.

While *rex* is part of the larger BIT toolbox, it is a single MATLAB file that may be installed and used separately. It provides capabilities to extract image data from either a single image file or multiple volumes such as a times-series of images. The extraction volume can be either a single ROI or a collection of spatially disjoint ROIs. *rex* allows broad flexibility in the form of the extracted data, as it can return the values of a single voxel, a single cluster of voxels or a disjoint set of clusters. Various options are provided to generate descriptive statistics from clusters of voxels including mean, median, voxel-weighted mean, and one or more eigenvariates. To allow examination of experimental effects in units of percent signal change, the resulting extracted values can be scaled with respect to either the global brain mean, or the cluster time-series mean.

rex can be accessed as a MATLAB command-line function or through a graphical user interface. Extracted data can be saved as text and matlab data files or visualized with several data analysis and plotting options that allow convenient data exploration and hypothesis testing.

INSTALLATION

1) If it does not already exist, create a directory named *rex.*

2) Copy the *rex.m* file (and optionally the additional TD.* files) into the *rex* directory

3) Start MATLAB

4) To add the *rex* directory to the MATLAB search path, from the File menu at the top of the MATLAB window go to **File->Set Path->Add Folder** and then select the *rex* directory.
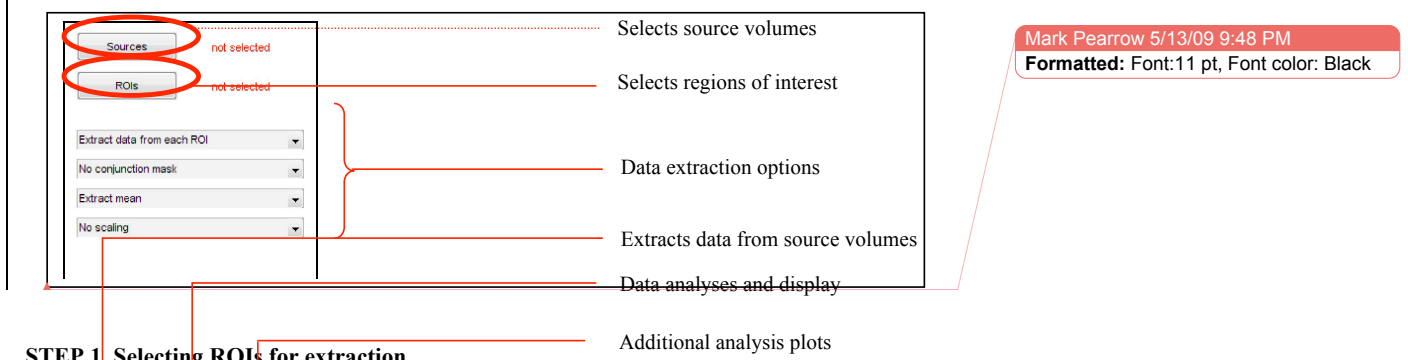
5) Click **Save** and then **Close**.

The rex program is now at the beginning of the MATLAB search path.

STEP BY STEP instructions using the *rex* GUI

To start the ROI extraction process, at the MATLAB prompt type:

```
>> rex
```

The *rex* GUI should now appear on the screen.

Sources	not selected	.... Selects source volumes
ROIs	not selected	Selects regions of interest
Extract data from each ROI ▾		
No conjunction mask ▾		Data extraction options
Extract mean ▾		
No scaling ▾		Extracts data from source volumes

Data analyses and display

Additional analysis plots

STEP 1. Selecting ROIs for extraction

rex assumes prior creation of the regions of interest as NIFTI-1 image mask files (*.img or *.nii formats) or text files (*.tal format). For example, if the goal is to use anatomical ROIs to guide the functional data extraction process, you can use *wfu_pickatlas* to create anatomically-defined ROIs that can be saved as *.img mask files. Note that *rex* permits the use of labels within mask files in order to define multiple ROIs with a single *.img file). Alternative methods to create functionally-defined ROIs are outlined in Appendix I.

To select one or more ROIs:

1) In the *rex* GUI click ROIs

2) Select one or more ROI files. The supported file types are *.nii, *.img, and *.tal files. When using NIFTI-1 image files (.nii or .img), ROIs will be defined by the location of those voxels where the value of the ROI image is greater than zero. In addition ROI NIFTI-1 files containing multiple labels can also be used (the REX tool is provided with a sample Talairach Daemon ROI file defining 55 anatomical areas in normalized space). In contrast, * .tal files are simply text files containing the spatial coordinates (in mm) of the voxels comprising an ROI (the x,y,z coordinates are in columns, and each voxel is a separate row; use normalized coordinates -e.g. MNI- if the source volumes are normalized, or subject coordinates if the source volumes are in native space).

STEP 2. Selecting image data sources

To define the source image files:

1) Click **Sources**

2) Select one or more NIFTI image volume files to extract data from these volumes

or

2) Select one SPM.mat file to extract data from volumes specified in a SPM design, or to repeat one SPM analysis for the selected ROIs.

Selecting a single SPM.mat file instead of the NIFTI format files is equivalent to selecting all of the image files that have been defined as the original data in the analyses specified in the SPM.mat file (more specifically the volumes listed in the SPM.xY structure). Any valid SPM.mat file can be used as a source, including ones resulting from either first-level or second-level analyses. For example, if you select the first-level analysis SPM.mat file for a given subject, the data sources will be assumed to be all of the functional data files for this subject, with one volume for each time point across all sessions included in the SPM.mat file. In this case *rex* will effectively extract all of the functional time series at the specified ROIs for this subject and allow you to perform first-level analyses on the resulting time-series. If you select instead a second-level analysis SPM.mat file, the data sources will be assumed to be the beta (or con) images specified in this analysis (one volume per regressor per subject; e.g. one contrast volume per subject for a standard second-level t-test analysis). In this case *rex* will effectively extract the beta/contrast values at the specified ROIs for each subject, and allow you to perform second-level analyses on the resulting data.

STEP 3. Specify additional output options

By default *rex* will extract the average value within each ROI, without scaling, and without any additional masking. Additional options include:

a) **Data-level options (ROI/cluster/voxel)**:

Each ROI file selected in step 1 characterizes a complete region of interest (ROI). Each ROI can be in turn formed by one or multiple clusters (disjoint sets of voxels), for example when using a *wfu_pickatlas*-ROI containing multiple labels, or when using a functionally-defined ROI composed of several clusters of activation. Last, each cluster can be formed by one or multiple voxels. *Rex* allows you to extact data separately for each voxel (voxel-level data), to extract data collapsed separately across all voxels within each cluster (cluster-level data), or to extract data collapsed across all of the voxels within the entire ROI (ROI-level data).

a. Use "**extract data from each ROI**" to extract data separately from each ROI using the selected summary measure. The default measure is the mean, collapsed across multiple voxels. A single output text file will be created for each ROI and it will contain the ROI-level data for each source file in rows.

b. Using "**extract data from selected clusters**" is similar to extracting data from each ROI, but if the ROI mask contains a disconnected set multiple clusters *rex* will allow the user to specify a subset of clusters and the ROI-level summary measure will only include voxels within the selected clusters.

c. Use "**extract data from each cluster**" to extract data separately from each cluster within each ROI. A separate output file will be created for each cluster and each ROI, containing the cluster-level data for each source file (rows).

d. Use "**extract data from each voxel**" to extract the data separately from each voxel. A separate output text file will be created for each ROI containing the voxel-level data across all source files (rows) and voxels (columns). In this case the summary measure is disregarded and there is no collapsing across multiple voxels.

b) **Summary measure (mean/median/weighted mean/eigenvariates)**

Note: Summary measures only apply to ROI- or cluster-level extraction

a. Use "**mean**" or "**median**" to obtain the mean or median of the data across the selected voxels

b. Use "**weighted mean**" to obtain a voxel-weighted mean across the selected voxels (not available when ROIs are defined using *.tal files). The values of the ROI mask file at each voxel will be taken as the weights to be used when computing a weighted average across all selected voxels. The mask values are normalized to sum to 1 and a weighted sum is computed.

c. Use "**eigenvariate**" with a chosen number of eigenvariates to summarize the data across voxels in terms of a singular value decomposition of the time series. When choosing multiple eigenvariates the output text file (for each ROI/cluster) will contain each eigenvariable as a column (and source files as rows as usual). Eigenvariates are extracted using a Singular Value Decomposition (SVD) of the time series across all the voxels within each ROI/cluster. Each eigenvariate can

be interpreted as a separate weighted mean of the data, where the voxel weights are chosen to sequentially capture the maximum signal variance. For example, the first eigenvariate represents the weighted mean of the ROI data that results in the time series with maximum possible variance (any other weighted mean will result in a combined signal with smaller variance). Multiple eigenvariate extraction is useful as a data-reduction technique for multivariate analyses of ROI data, characterizing the time series within an ROI in terms of a small number of components that best capture the variability of responses across all of the voxels within this ROI.

c) **Scaling options (global scaling / within-ROI scaling / none)**

If extracting time-series data (e.g. you chose a first-level SPM.mat file as source) typically you would want to scale the original data within-sessions to increase the interpretability of the data (units in percent signal change)

a. Use "**global scaling**" to scale the output data based on the global intracerebral mean (SPM session-specific grand-mean scaling) of the data averaged across all source files (or within-sessions, when a SPM.mat file is selected as source). Use this option, for example, if you wish to extract a time-series of functional data in units of percent signal change referenced to the SPM default intracerebral mean of 100.

b. Use "**within-ROI scaling**" to scale the output data based on the local mean (within-ROI) of the data averaged across all source files (or within-sessions, when a SPM.mat file selected as source). Use this option if you wish to extract time-series functional data in units of percent signal change referenced to the mean value of each ROI)

d) **Conjunction mask**. Optionally you can define an additional NIFTI format conjunction mask file. Data will only be extracted from voxels contained in this global conjunction mask. For example, the *Mask.img* file generated in SPM after estimating a model can be used to restrict all ROI data extraction to voxels within the analysis mask.

STEP 4. Extract and explore the data

Click "**Extract**" to extract the data from the source image files at the voxels inside the specified regions of interest. A text message-box will be displayed containing the paths to the newly created data files and a results window will display the extracted data. In addition to the output data files (*.rex.txt* files) containing the extracted data (one file per ROI/cluster), *rex* will create mask files (*.rex.tal* files) indicating the locations (in mm) of the voxels corresponding to the ROI/cluster associated with each data file.

Sample GUI:
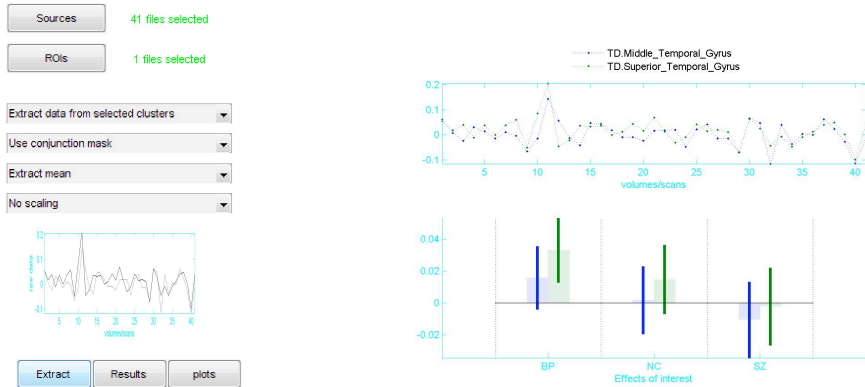
Sample output files:

| *TD.Middle_Temporal_G yrus.rex.txt*	*TD.Middle_Temporal_Gy rus.rex.tal*	*TD.Superior_Temporal_Gyr us.rex.txt*	*TD.Superior_Temporal_Gy rus.rex.tal*
(one row per source file)	*(one row per voxel)*	*(one row per source file)*	*(one row per voxel)*
0.0630	-36 2 -50	0.0548	-34 6 -50
0.0080	-34 2 -50	0.0191	-32 6 -50
-0.0220	-32 2 -50	0.0407	-30 6 -50
0.0326	32 2 -50	-0.0092	-28 6 -50
0.0145	34 2 -50	0.0391	-26 6 -50
-0.0135	36 2 -50	0.0002	-24 6 -50
0.0114	38 2 -50	0.0382	26 6 -50
-0.0034	-36 4 -50	0.0603	28 6 -50
-0.0641	-34 4 -50	-0.0499	30 6 -50
...	...	...	...

Example of *rex* data extraction and display performed on second-level analysis data. The source volumes are the subjects' beta images, with two anatomically-defined ROIs. The effects of interest represent three different subject groups. The extracted data (.txt files) contain the beta image values for each subject averaged across all the voxels within each ROI.

In addition, if the data sources have been defined using the SPM.mat file, you can click "**Results**" to replicate the original voxel-based SPM analyses for the extracted ROI/cluster/voxel data. As in SPM-results you will be prompted to select (or define) a contrast, and *rex* will re-estimate the model and display the statistical analysis results for all of the extracted data. For each ROI/cluster/voxel selected *rex* will display the effect size for the chosen contrast, T statistic, uncorrected p-value, and FDR-corrected p-values (multiple comparison corrections are applied to correct for multiple ROIs).
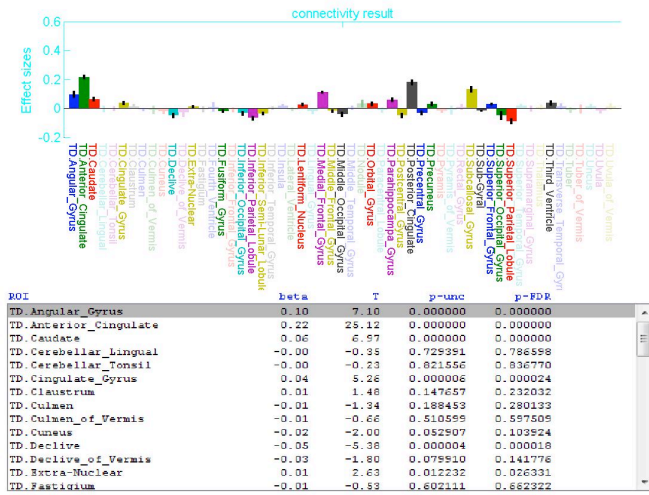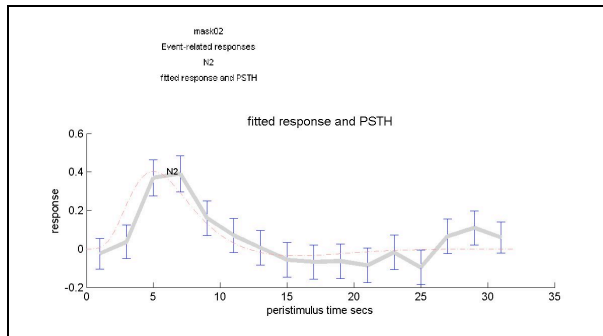
Example of the *rex* **results** function used to perform SPM second-level analyses on a complete set of anatomically defined regions (Talairach Daemon areas). Analyses are performed here on the average activation within each ROI. Corrected p-values represent a whole-brain correction for these ROI-based analyses.
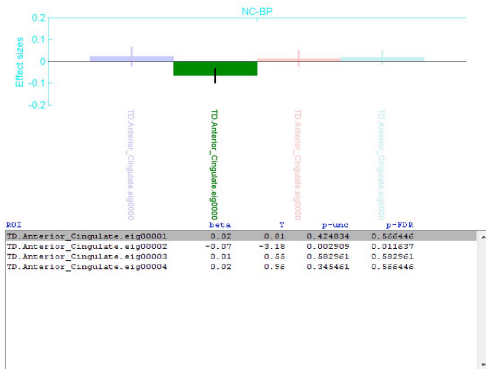
Example of the *rex* **results** function used to perform multivariate second-level analyses on one region of interest (Anterior Cingulate). Analyses are performed here on the first four eigenvariates characterizing the activation profiles within the selected ROI. Corrected p-values represent a multivariate correction of each of the individual eigenvariate results. This example illustrates the application of multivariate analyses of ROI data showing between-group differences that would be missed if only looking at the average activation within this ROI.

Last, if the data sources have been defined using the SPM.mat file, you can click "**plots**" to further explore the effects of interest within the extracted ROIs. The options here are the same as

those encountered when using **plots** within the SPM results window, including display contrast estimates and 90% C.I., and fitted and adjusted responses for both first-level block designs or for second-level analyses. For event-related responses you can also explore fitted responses and PSTHs, 90% C.Is., adjusted data, parametric responses and Volterra kernels.

Example of the *rex* **plots** function used on first-level analysis dataset in which the source volumes are individual scans comprising a time-series extraction. The data originate from an event-related study using famous faces (REF?) The event-related PSTH plot results for one functionally-defined ROI shown at the right.

Please report any bugs, comments and/or suggestions to: Susan Whitfield-Gabrieli, swg@mit.edu

APPENDIX I. Functional ROI Definition

In situations where anatomical landmarks do not provide accurate guidance as to the boundaries of regional functional specialization, so called "functional localizers" are sometimes used to identify brain regions specialized for a particular processing function. These localizers usually take the form of an additional task whose associated neural activity modulations are believed to be orthogonal to the effects of interest related to the target tasks. The patterns of activity detected by the localizer scan can then be thresholded to form a functional ROI that can be used to spatially constrain the analysis of the target tasks. Functional ROIs may be defined in a number of different ways:

Define a spatially localized functional ROI using the spm_VOI function

1) Copy *spm_VOI.m* to the *spm8_first* directory at the beginning of your MATLAB path. The spm_VOI.m file replaces one that is part of the core SPM8 distribution, so it needs to be in the MATLAB search path *before* the version that came with SPM8.

2) Go to SPM **Results**, select a contrast of interest and thresholds.

3) Choose **small volume**

4) Choose a **Search volume** using a sphere, box or image.

5) If you have installed the modified version of *spm_VOI.m*, an additional figure window will pop up with a new glass brain that has all of the voxels that survived the small volume correction process. The descriptive statistics for these voxels are located in the SPM graphics window. However, SPM doesn't update the original glass brain in the Graphics window.

6) You can now save the functional ROI in a *.tal file, an ASCII file containing the XYZ locations of the ROI in MNI space if you are working with images that have been spatially normalized to the MNI space..

Define a spatially localized functional ROI using xjView

1) Type >> xjview at the MATLAB command prompt

2) Navigate to the cluster of interest

3) Change the radio button from **"ALL"** to either **"Only +"** and **"Only – "** (optional)

4) Choose **"Pick Cluster"**

5) Save the ROI mask as a NIFTI format file

Define a map-wise functional ROI in SPM

1) Go to SPM **Results**, select a contrast of interest and associated mask and thresholds.

2) Choose **"save"** to save an ROI NIFTI format file that includes all the significant voxels in the thresholded contrast.