# *fMRI Analysis Techniques*

***NIAG***
***Tehran University of Medical Science***

# Outline

- An overview on the whole procedure of data analysis in fMRI
- General fMRI Preprocessing steps
- statistical analysis Methods (GLM & ICA)

# Overview

fMRI Data → **Pre-Processing** → **Statistical Analysis** → **Post-Processing**

Correction of non-task related variability

Deriving the statistical map of activity

Parametric (Model Based)

Non-parametric (Exploratory)

Registration to standard Atlas

Visualization

Interpretation

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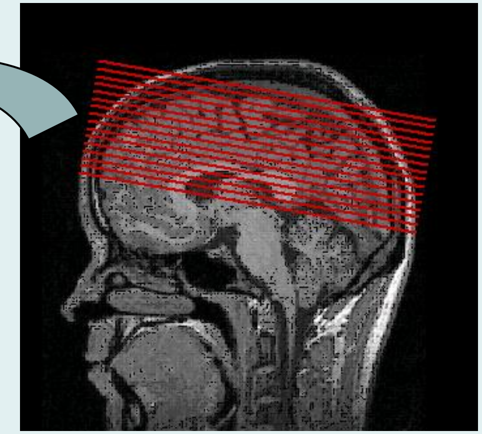
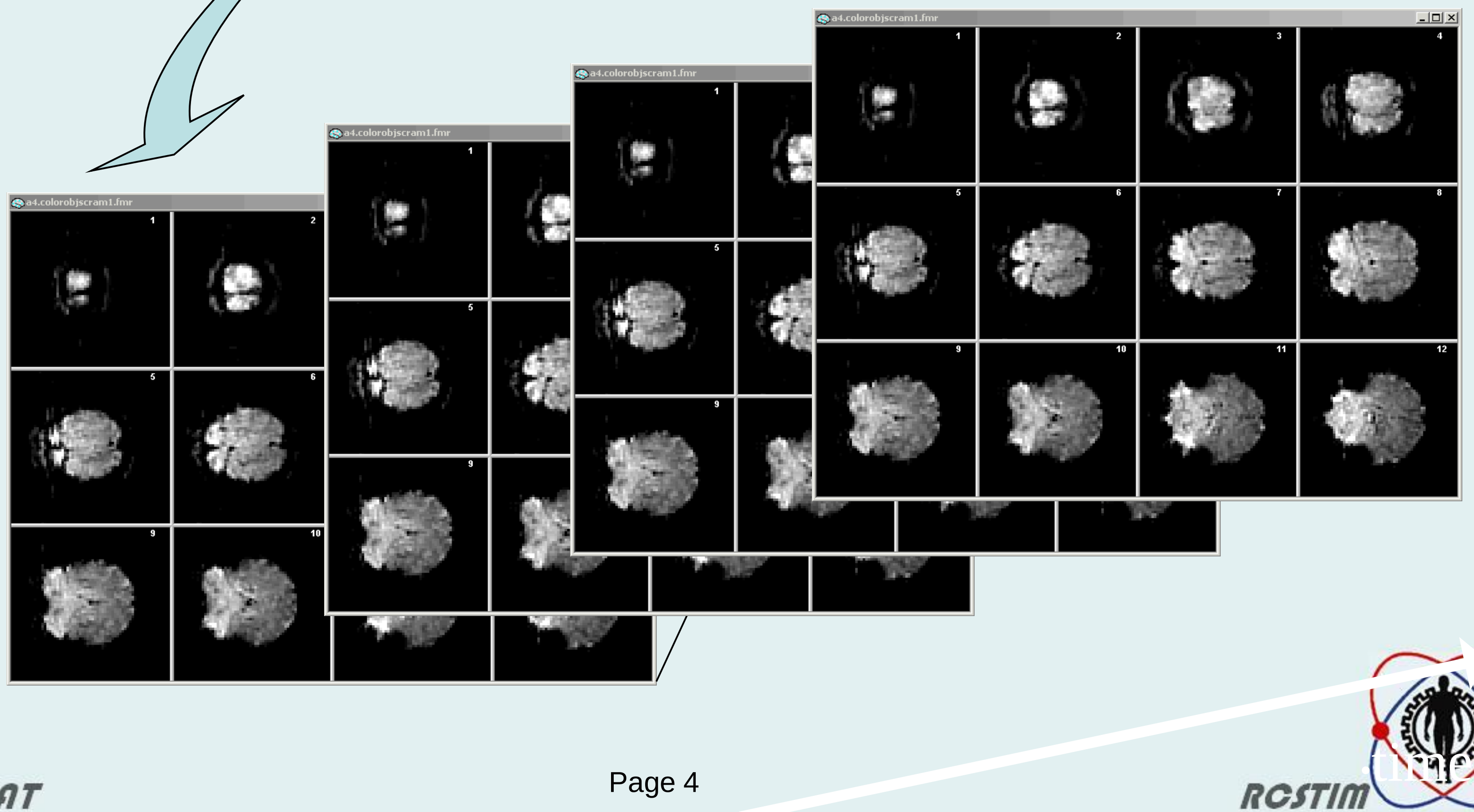
# How is a fMRI Data?



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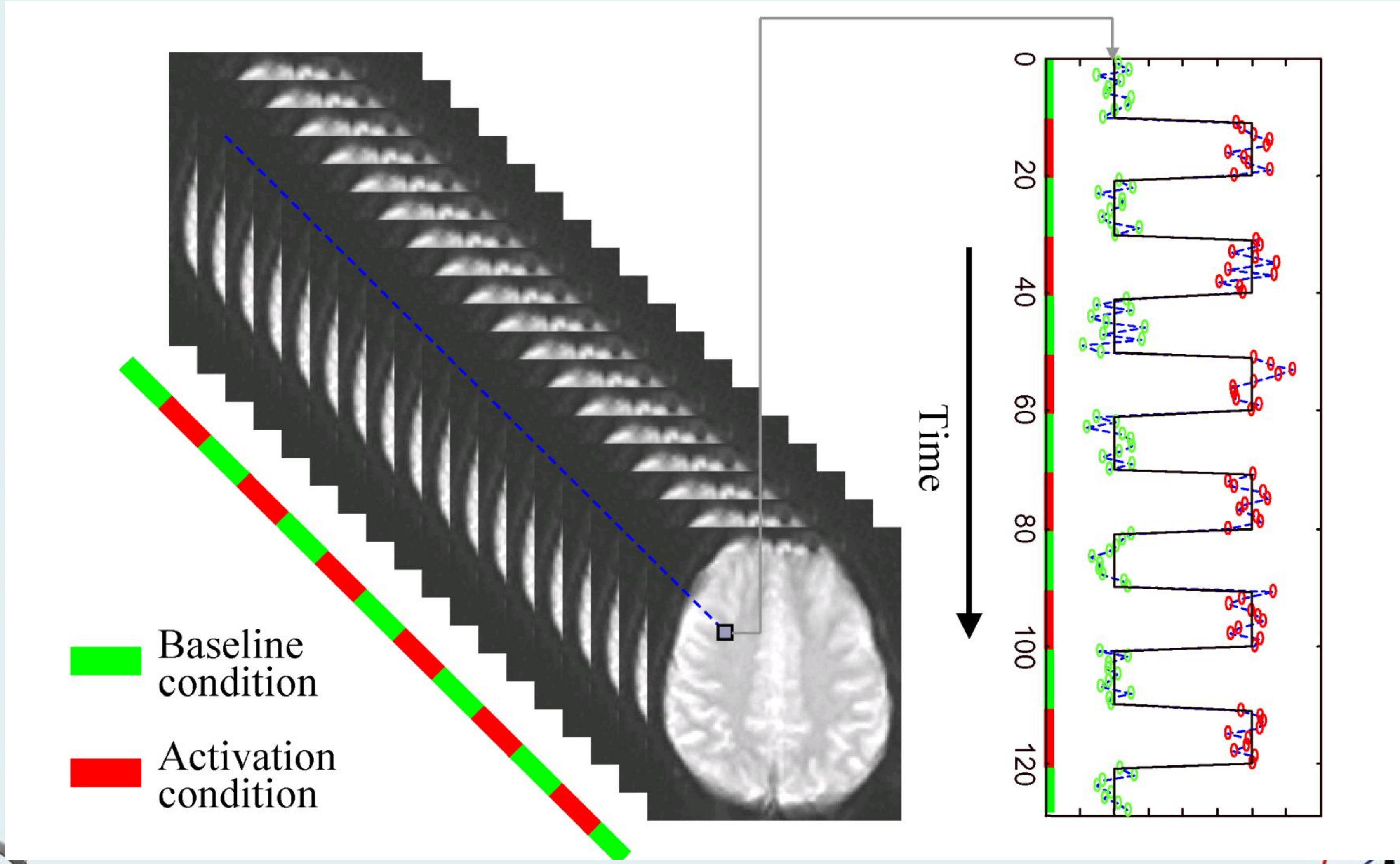
# fMRI time series



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# fMRI Image Preprocessing

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# What is preprocessing?

**Correcting for non-task-related variability in experimental data**

- Usually done without consideration of experimental design; thus, pre-analysis

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# Preprocessing Steps

- Data Transfer, and format Conversion
- Removal of first few volumes
- Motion Correction
- Slice Timing Correction
- Spatial Smoothing
- Filtering (Spatial & Temporal)
- Global intensity normalization

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# Data Format exchange

- **Conventional formats in different MRI system:**
  - ANALYZE
    - A file with extension *.hdr containing header information
    - A file with extension *.img containing image raw data
  - NIFTI
    - Usually a *.nii file containing 4D information
  - AFNI
    - Header information in *.HEAD
    - Image raw data in *.BRIK

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# Removal of first few volumes

- Usually image volumes corresponding to first 10-12 seconds are removed from the sequence
- This is to make sure that spin system is in the steady state condition in acquired images
- Thus unwanted changes in image weight are removed

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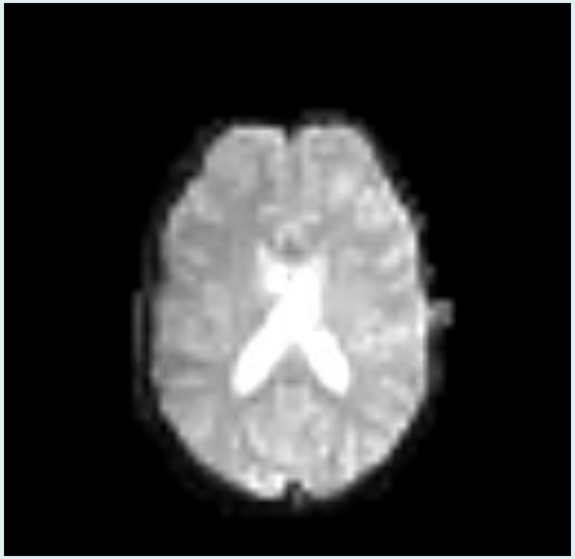
# Head Motion Correction:



- Even with padding around the
- head people move in the scanner

- Motion correction realigns all images to a common reference
- 3-12 parameters: 3 translation, 3 rotation, Skewness
- Minimization of some cost function

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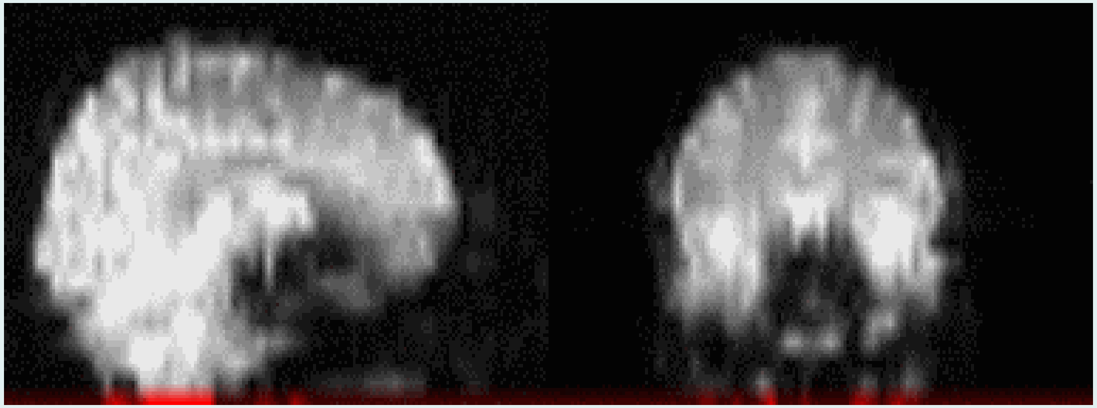
# Slice Timing Correction



- Each slice is scanned at a slightly different time

- Slice timing correction shifts the *data*

  (each voxel's time series) as if whole volume was acquired at exactly the same time

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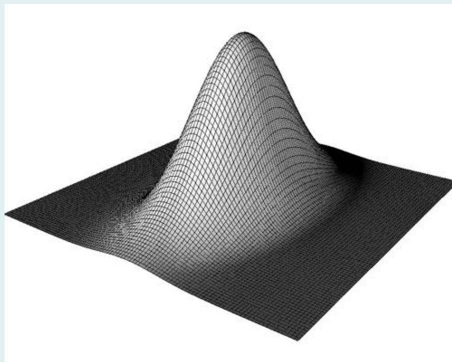
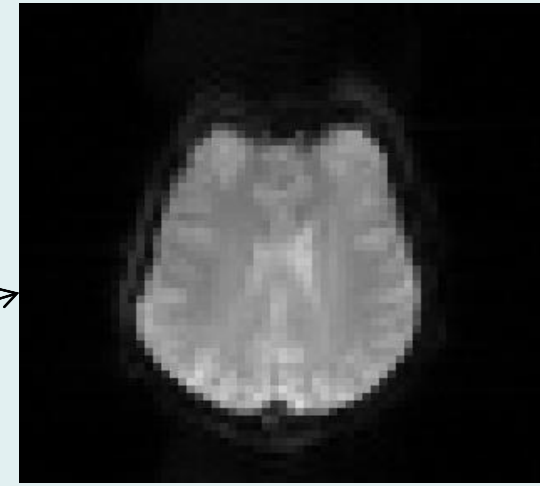
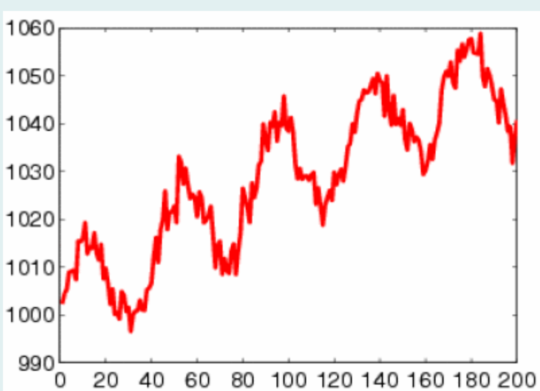
# Spatial & Temporal Filtering:

## Spatial Filtering

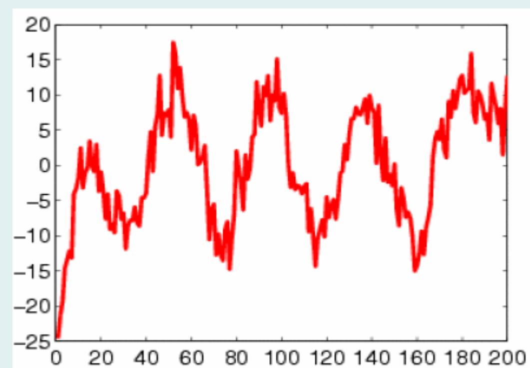




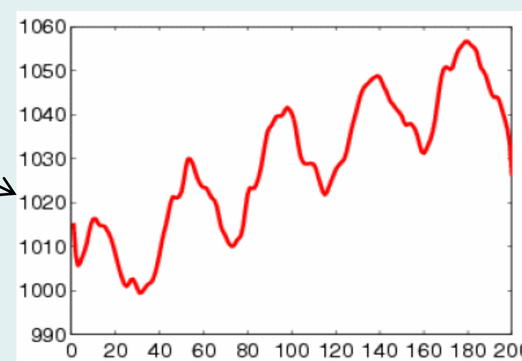
## Temporal filtering




HP




LP

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# Global intensity normalization

**Why:**

- variation in the intensity between different volumes

- How: compute 4D average and make it the same across subjects/sessions

# Statistical Analysis: General Linear Model

# Outline

**Aim:**

- detecting the status of each voxel (active rest)?

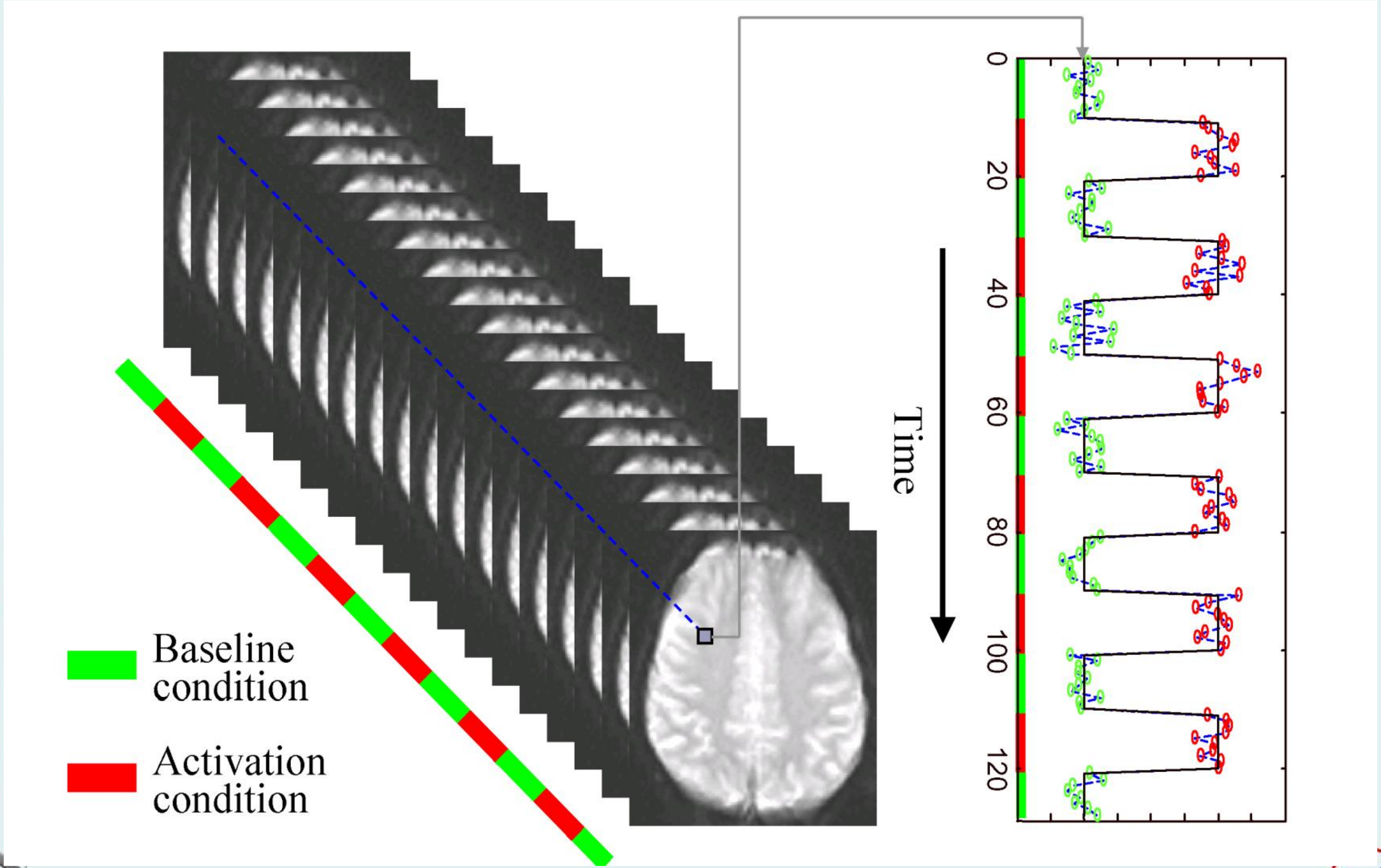
**Method:**

- General Linear Model (GLM)

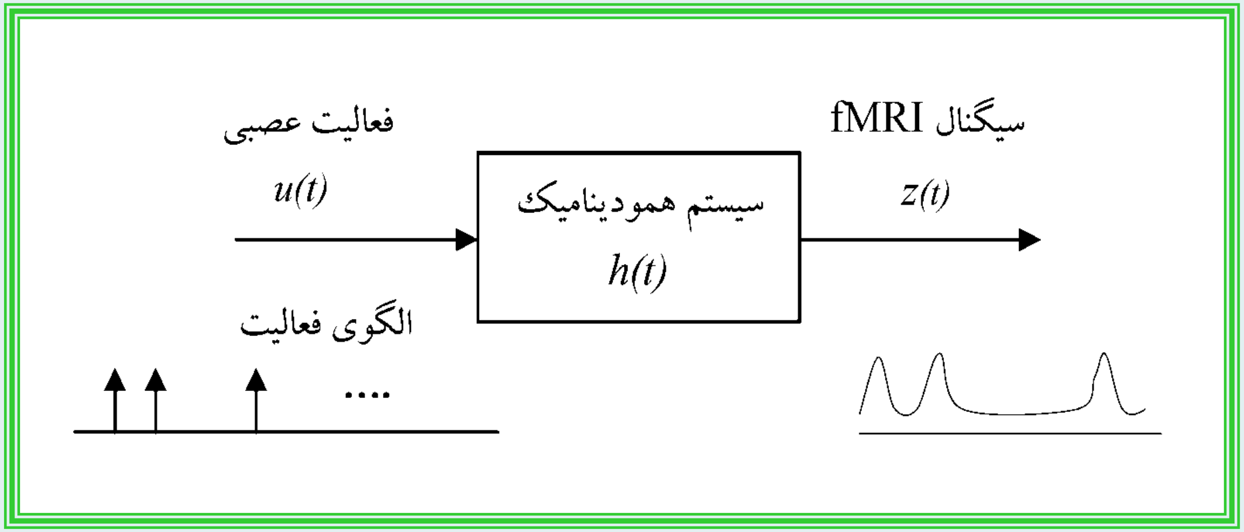
**Statistical Inference:**

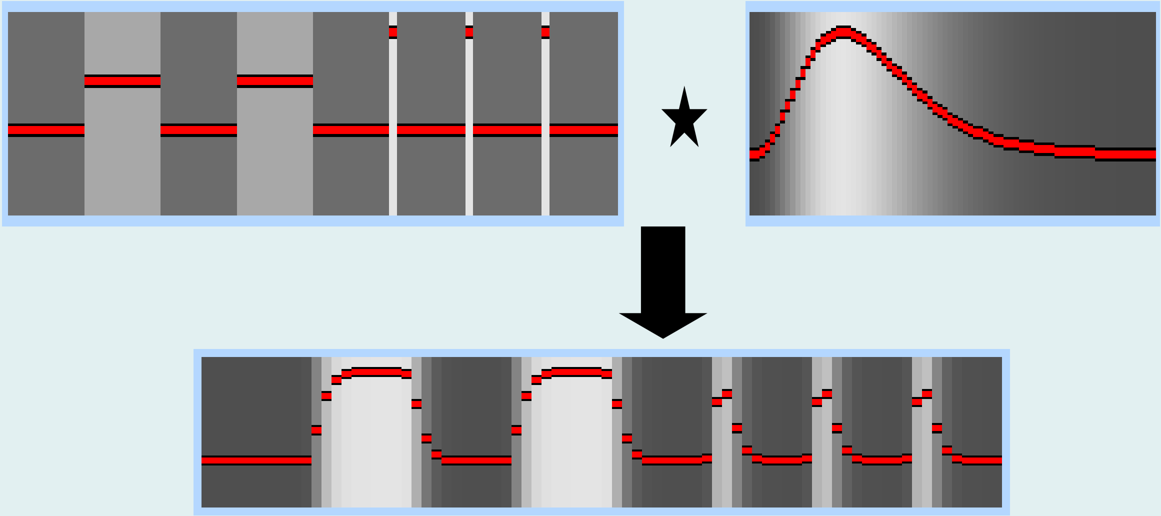
- Finding the Brain Activity and thresholding

# Basic rule for detecting the status of each voxel (active rest)?

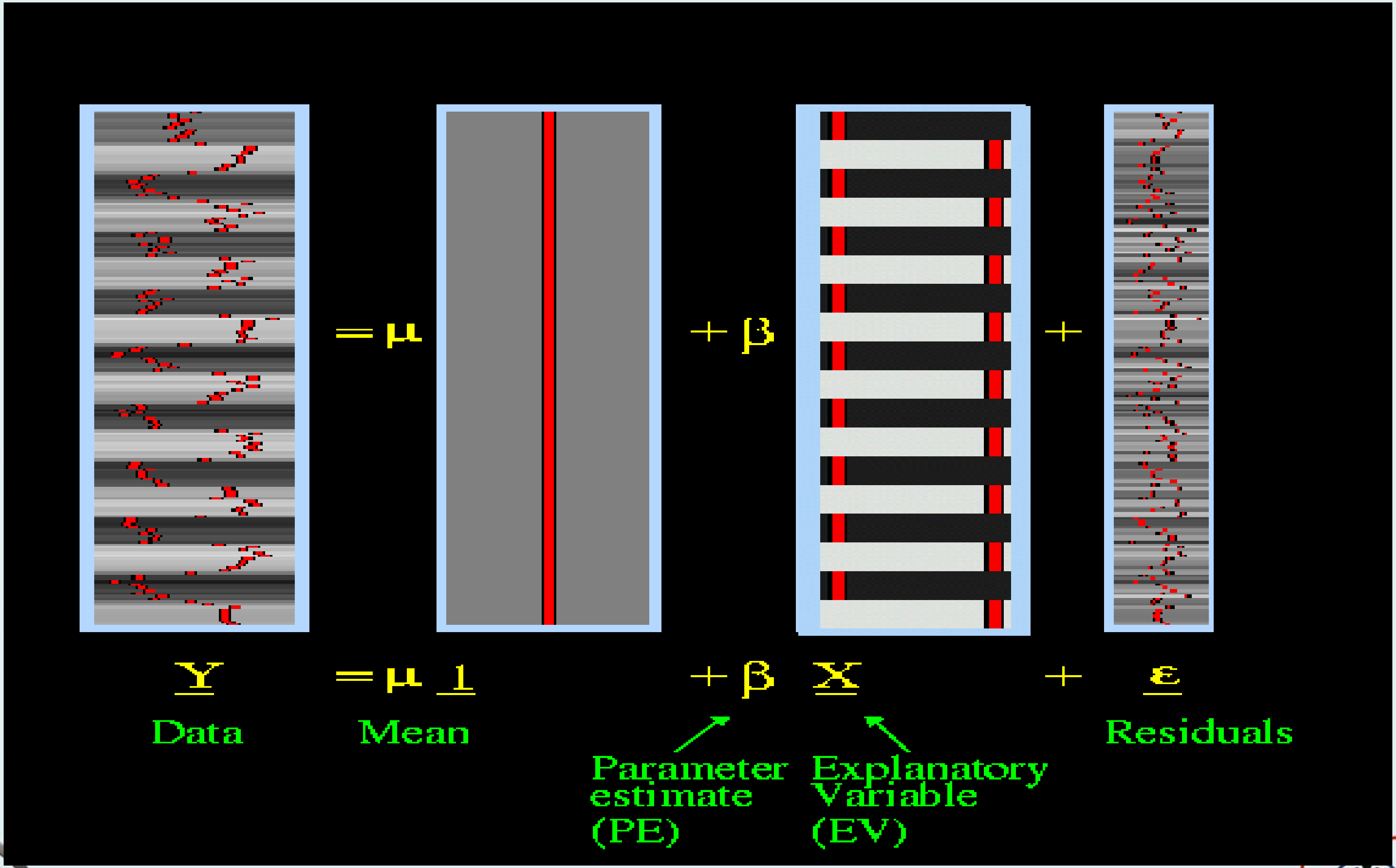


# The Role of Hemodynamic Response Function (HRF)



# GLM: General Linear Model (A simple view)



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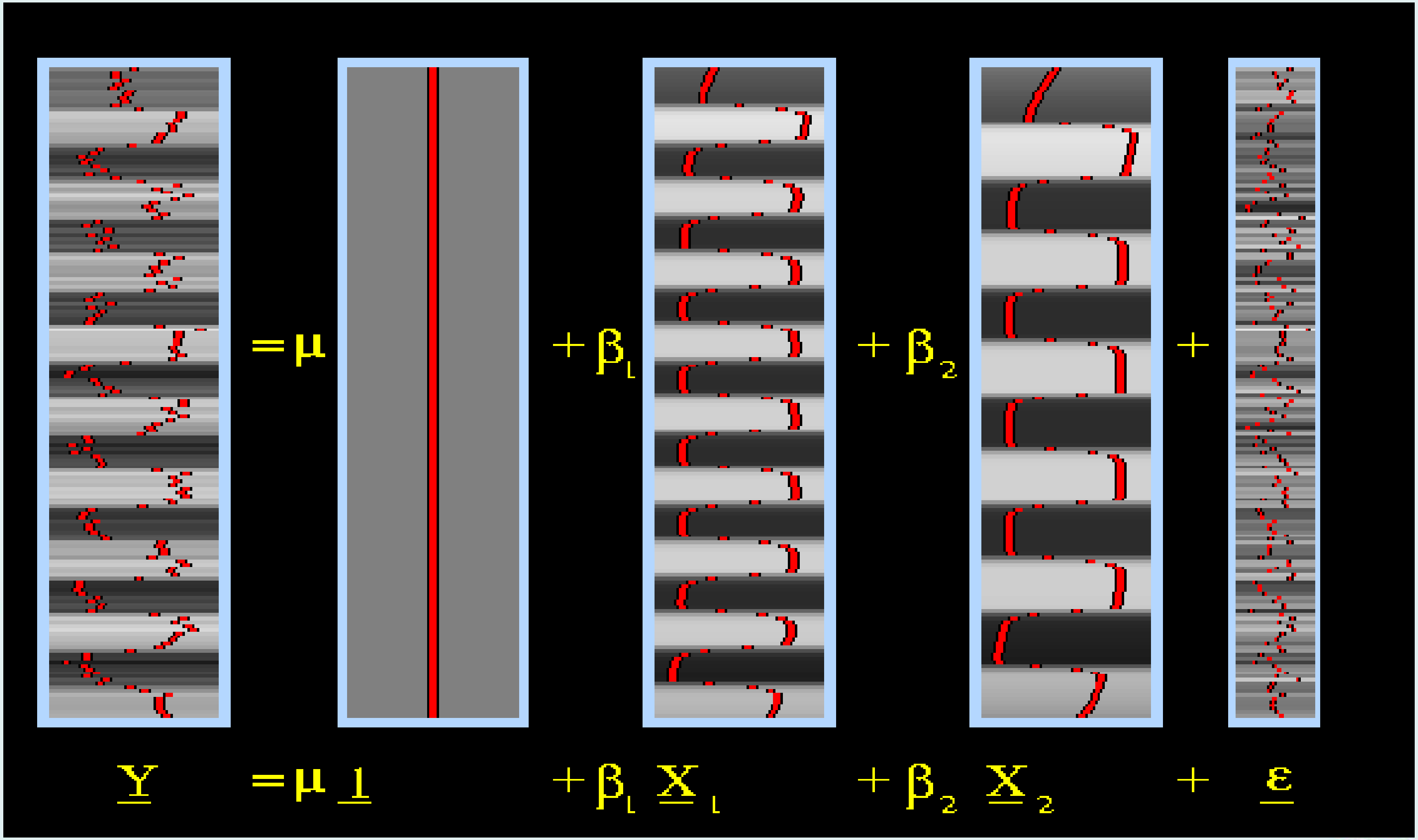
# GLM: General Linear Model

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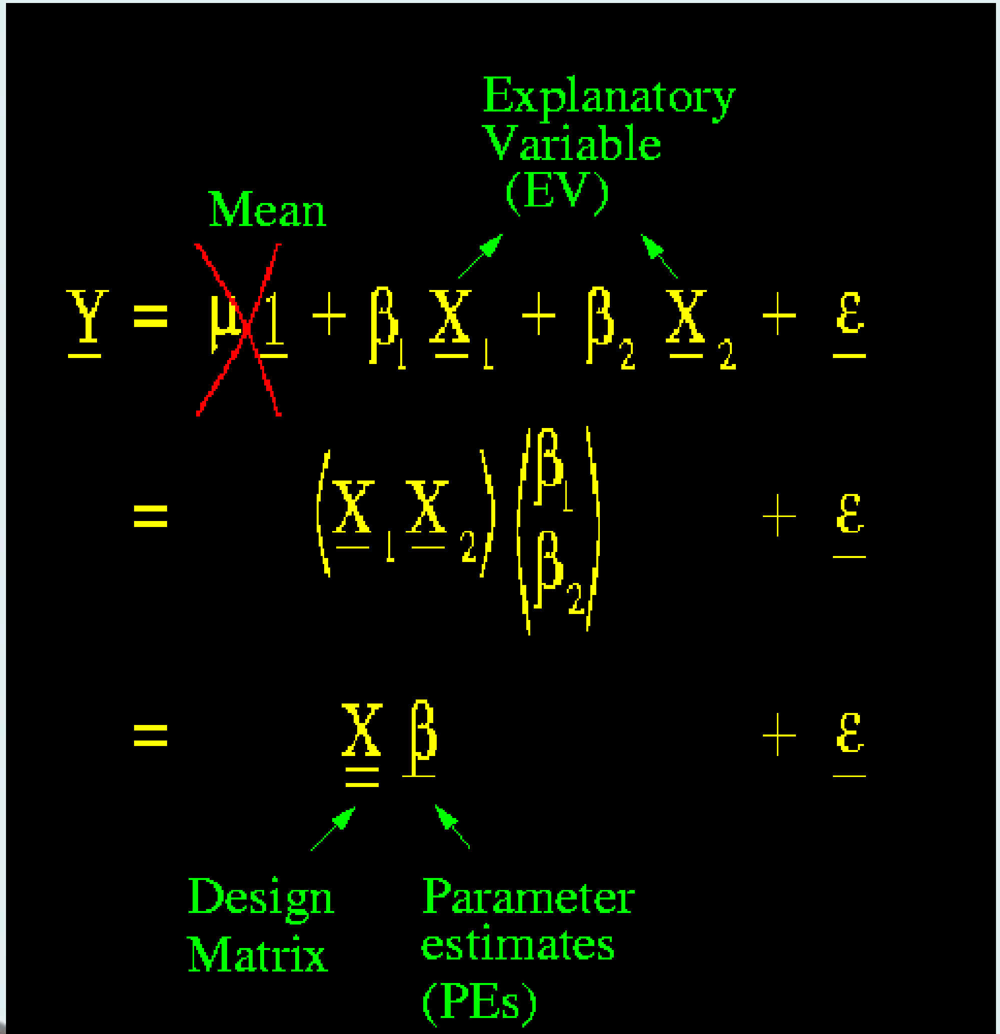
# Finding the Brain Activity




$$\hat{\beta} = (X^T X)^{-1} X^T Y$$

Contrast=[ 1    0]

COPE = $1\times\beta_1 + 0\times\beta_2$

T-stat= COPE / STD(COPE)

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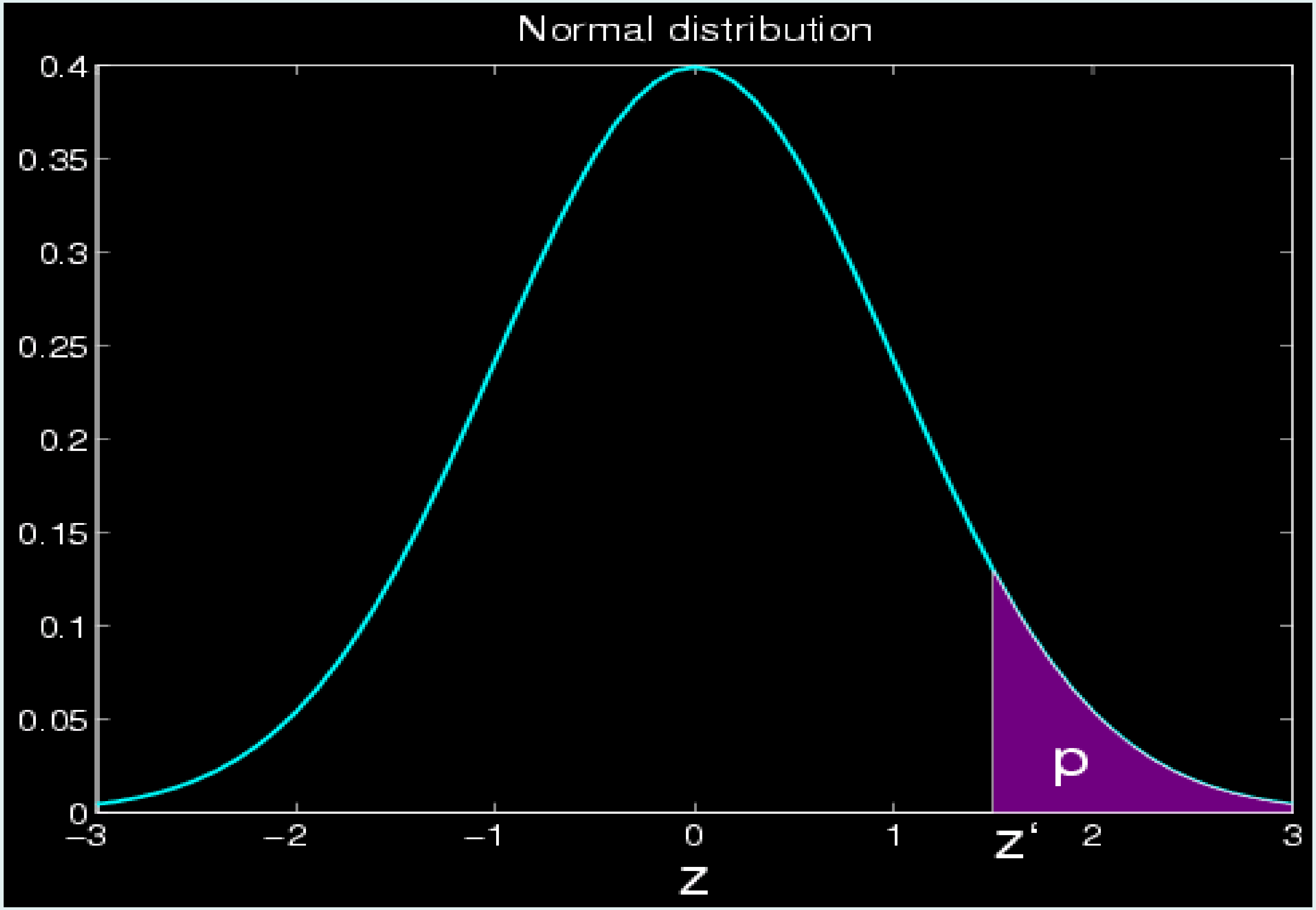
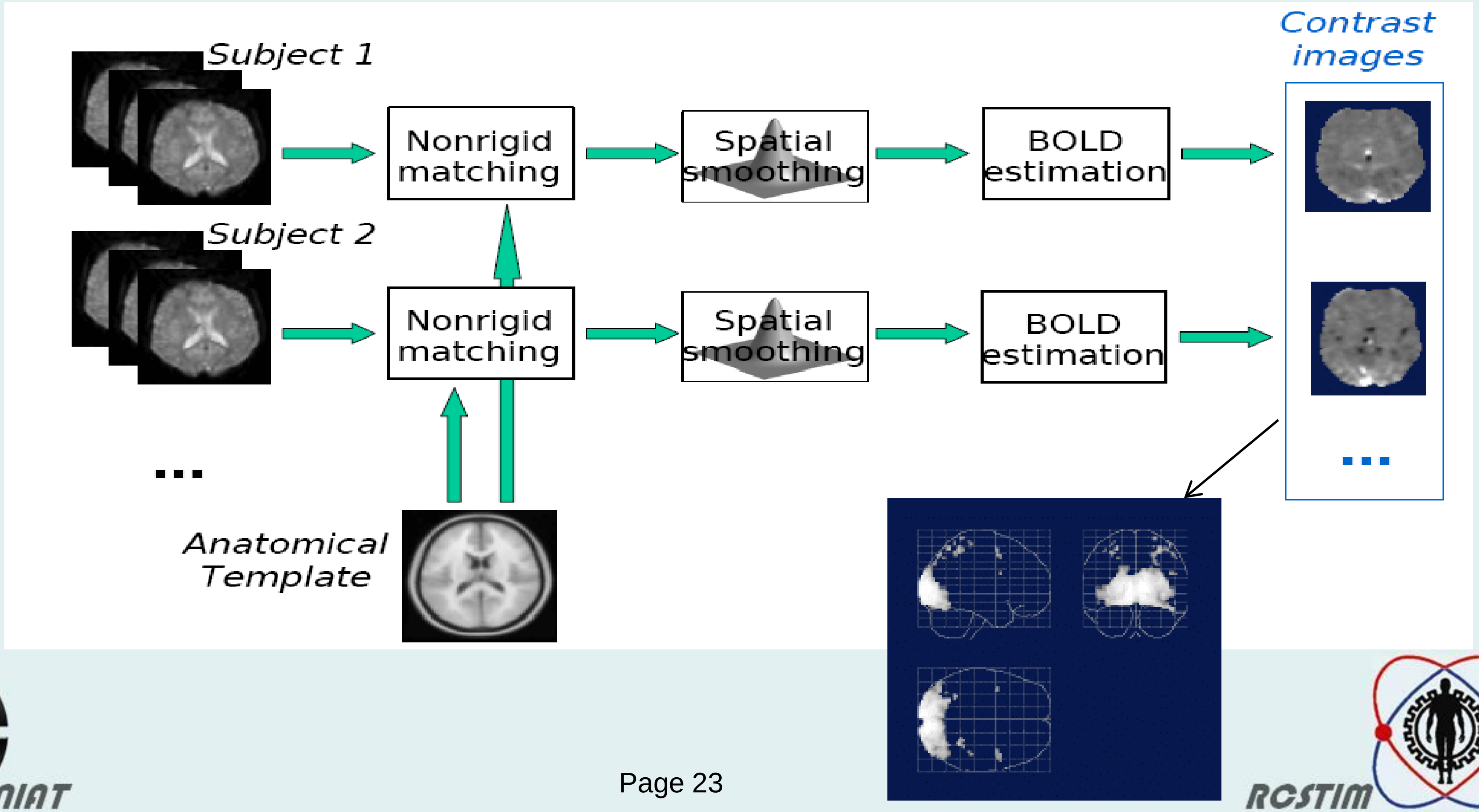
# Thresholding

- Z (Gaussian Distributed) , t (T-student), or F (Fisher) statistics



# Group fMRI Analysis:

- Which brain regions are consistently activated in a population in response to a given contrast of experimental conditions?



# Group fMRI Analysis:

Contrast images

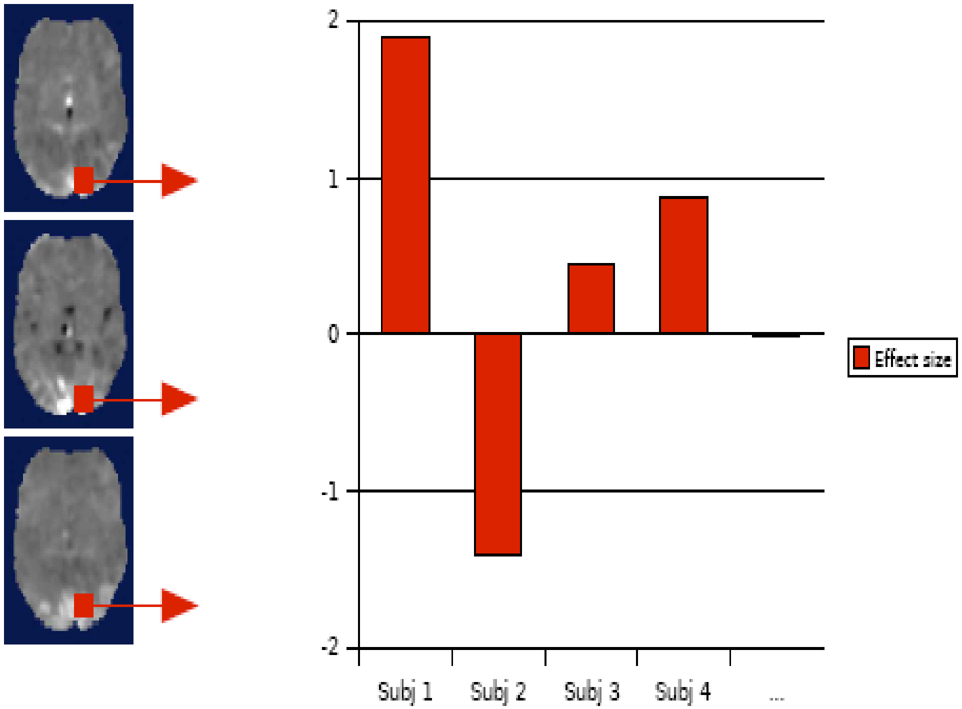
Observations in one voxel

Subj 1

Subj 2

Subj 3

...



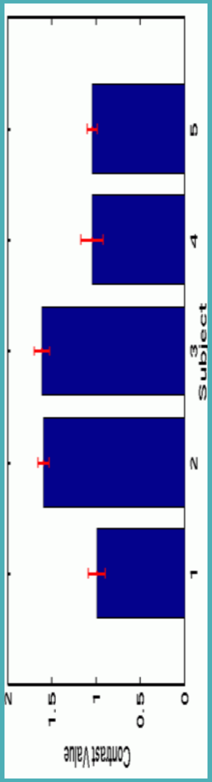

Significant *mean* effect?

# Higher Level GLM Analysis

## One-Sample Group Mean (OSGM)

$$y \quad = \quad X \; * \; \beta$$

Observations
(Low-Level Contrasts)




Data from
one voxel

$$= \begin{bmatrix} 1 \\ 1 \\ 1 \\ 1 \\ 1 \end{bmatrix} \begin{bmatrix} \beta_G \end{bmatrix}$$

Design Matrix
(Regressors)

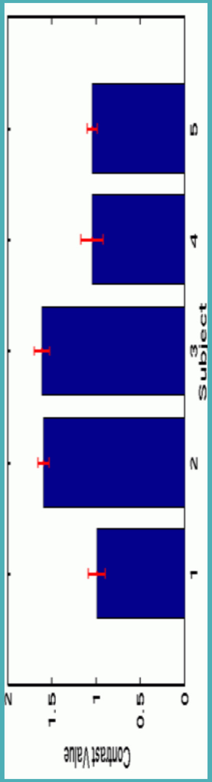
Vector of
Regression
Coefficients
("Betas")

Contrast Matrix:
C = [1]
Contrast = C*$\beta$ = $\beta_G$

# Two Groups GLM Analysis

$$y = X * \beta$$

Observations
(Low-Level Contrasts)




Data from
one voxel

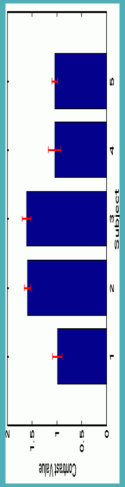
$$= \begin{bmatrix} 1 & 0 \\ 1 & 0 \\ 1 & 0 \\ 0 & 1 \\ 0 & 1 \end{bmatrix} \begin{bmatrix} \beta_{G1} \\ \beta_{G2} \end{bmatrix}$$

# Contrasts: Two Groups GLM Analysis

1. Does Group 1 by itself differ from 0?
C = [1 0], Contrast = $C^*\beta = \beta_{G1}$

2. Does Group 2 by itself differ from 0?
C = [0 1], Contrast = $C^*\beta = \beta_{G2}$

3. Does Group 1 differ from Group 2?
C = [1 -1], Contrast = $C^*\beta = \beta_{G1} - \beta_{G2}$



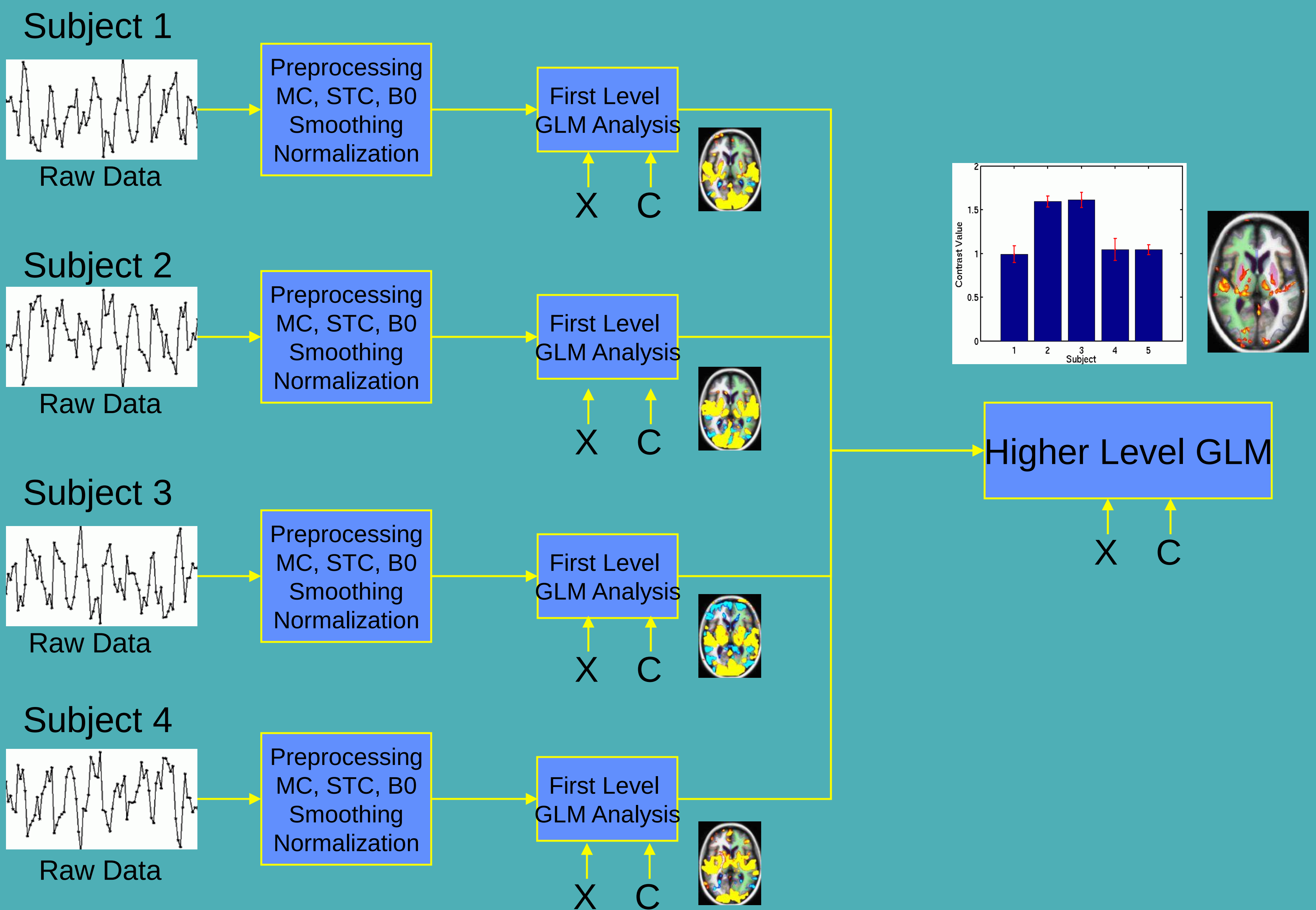
$$= \begin{bmatrix} 1 & 0 \\ 1 & 0 \\ 1 & 0 \\ 0 & 1 \\ 0 & 1 \end{bmatrix} \begin{bmatrix} \beta_{G1} \\ \beta_{G2} \end{bmatrix}$$

4. Does either Group 1 or Group 2 differ from 0?

$$C = \begin{bmatrix} 1 & 0 \\ 0 & 1 \end{bmatrix}$$

C has two rows: F-test (vs t-test)
Concatenation of contrasts #1 and #2

# Higher Level fMRI Analysis Overview



# Fixed versus Random Effect Analysis

- Fixed effect Analysis
  - all subjects treated as a single subject (fixed effect)
  - Huge areas of activation
  - Not generalizable beyond <u>sample</u>.
- Random Effect Analysis
  - Model Subjects as a Random Effect
  - Variance comes from a single source
  - Generalizable
- Mixed Effect Analysis
  - Model Subjects as a Random Effect and var comes from both first level and subjects
  - More complicated to compute

# Fixed effect vs. Random effect

**Do we want to make an inference about**

- The particular group we sampled? (Fixed Effect)
- The population they were sampled from? (Random Efect)

**Two effect exist:**

- Intra-subject variability:

there is some uncertainty on each subject's estimated effect due to "noise" in the fMRI data

- Inter-subject variability:

different subjects may have intrinsically different effects

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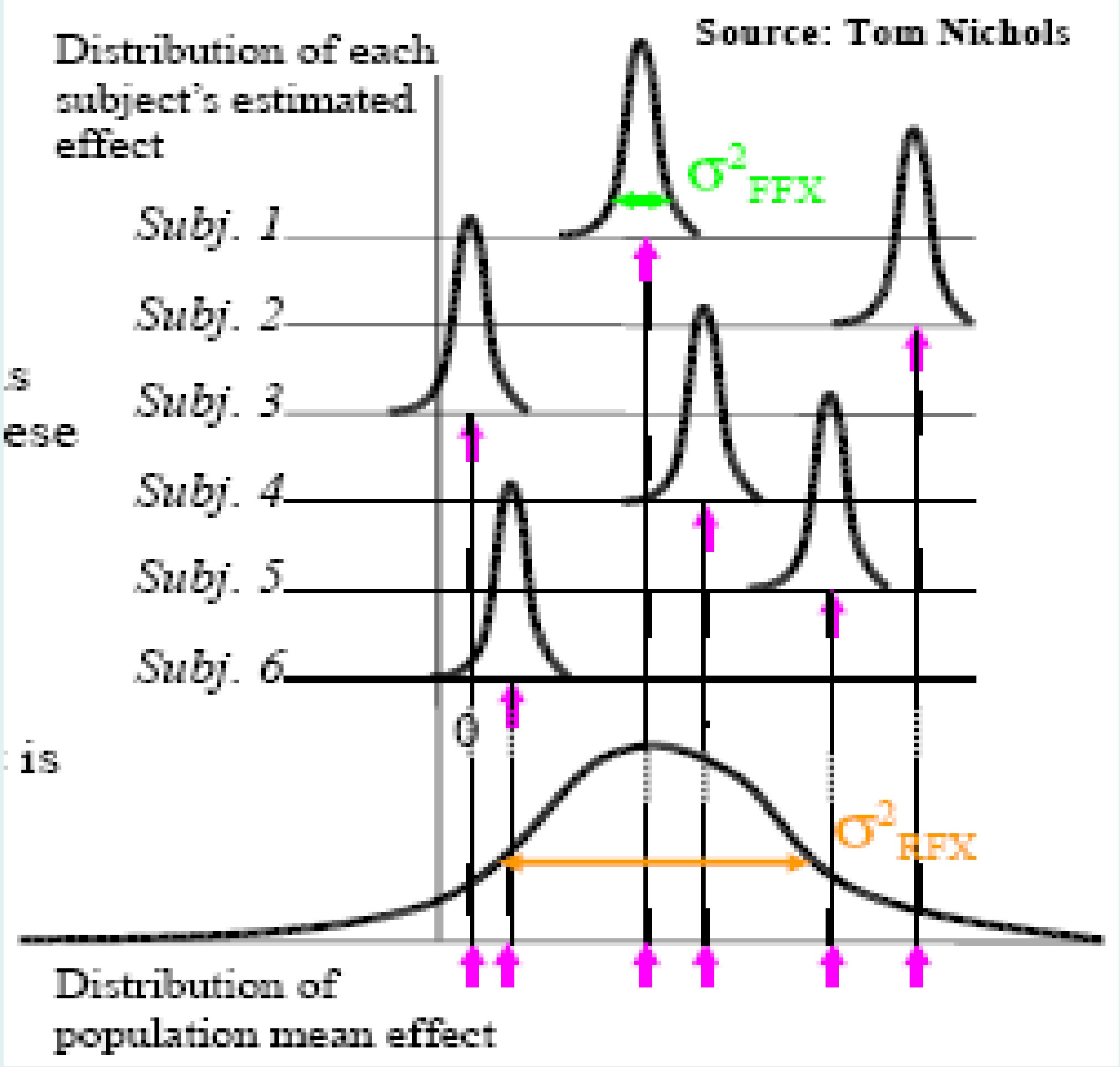
# Fixed effect vs. Random effect

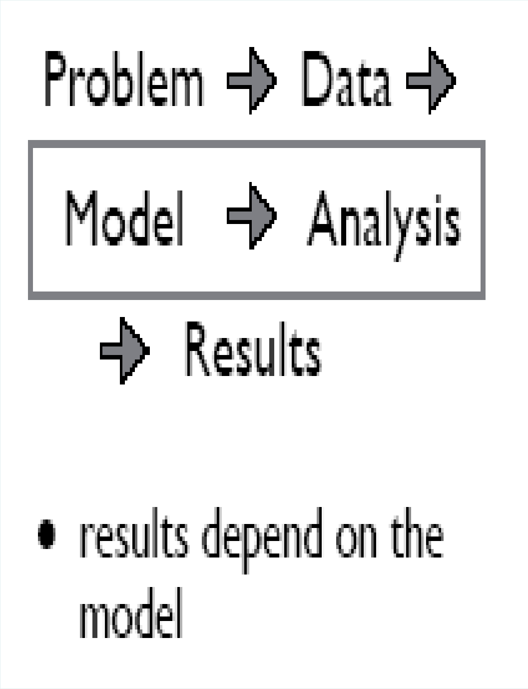


# Statistical Analysis: Independent Component Analysis

# Different Kinds of Data Analysis:

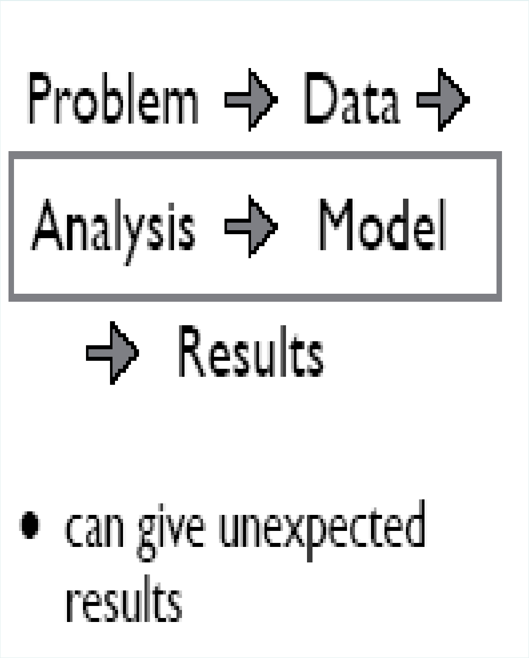
***Classical Data Analysis***

How well does my model Fit to the data?




***Exploratory Data Analysis***

Is there any interesting thing in the data?



# Why We need ICA in fMRI analysis?

- **Experiment:**

  suboptimal event timing, non-efficient design, etc.

- **Physiology**:

  secondary activation, ill-defined baseline, resting-fluctuations

- **Analysis:**

  filtering & sampling artifacts, design misspecification.

- **MR Physics Analysis**:

  MR noise, field inhomogeneity ,MR artifacts etc.

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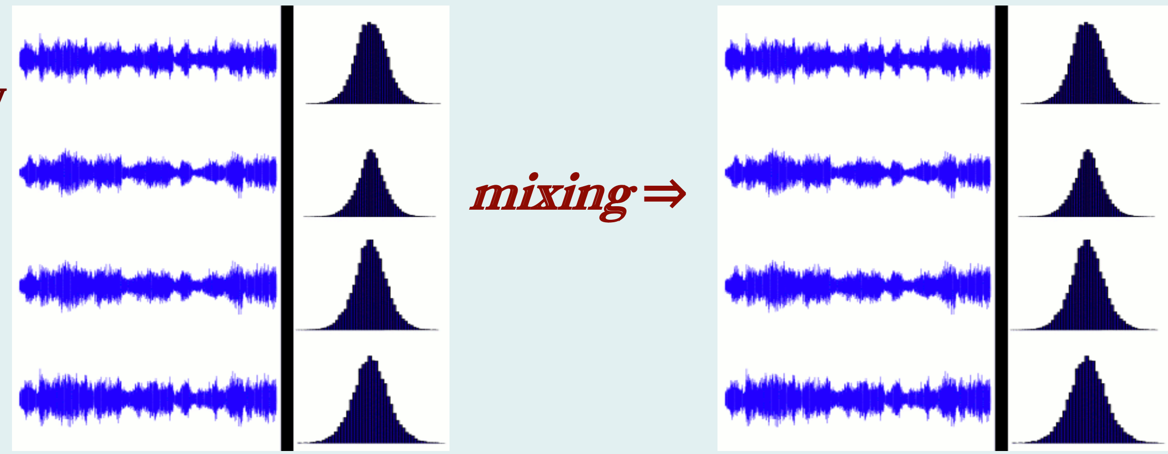
# *ICA Basis:*

- **Independency** among the sources (Spatial & Temporal)

- **Non-Gaussianity**

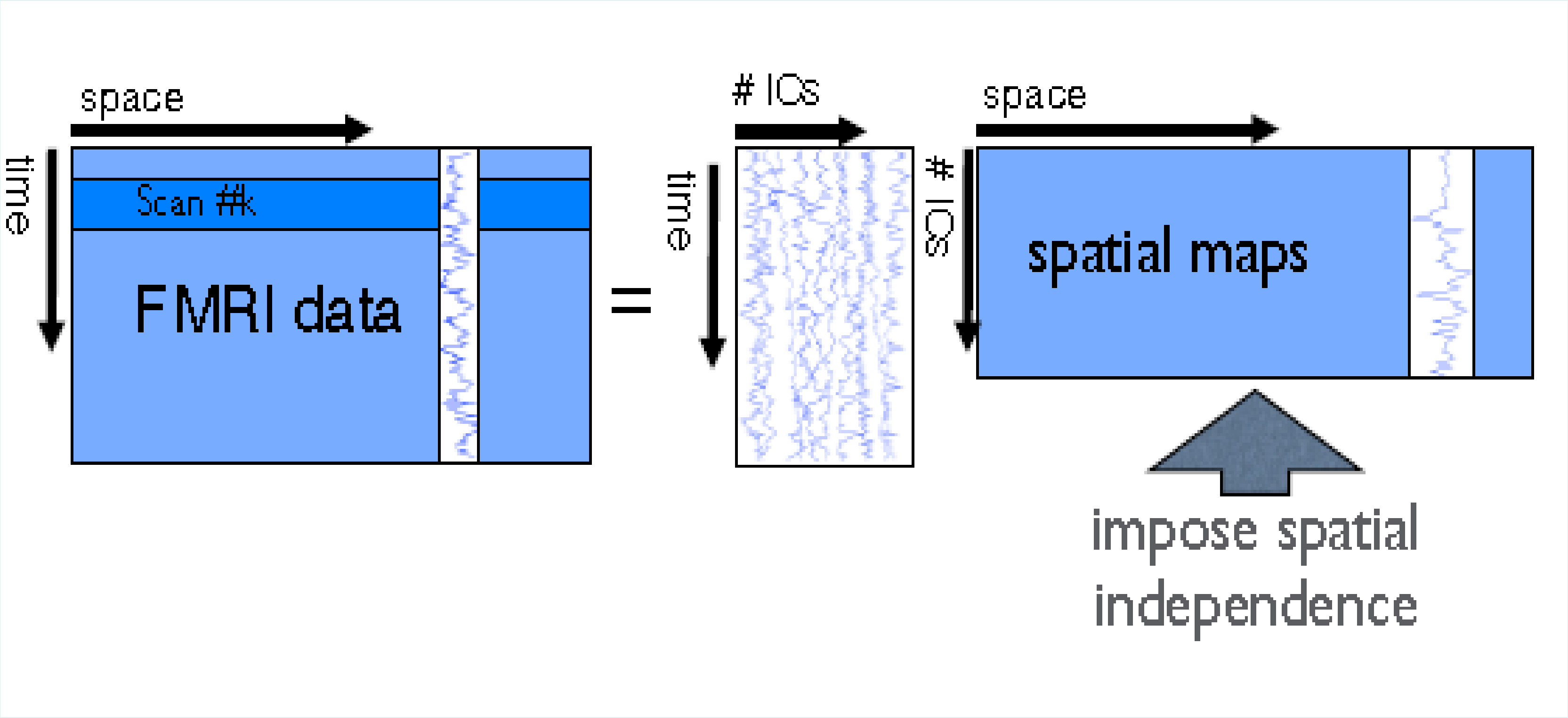



**ICA Algorithms:**

**objective function +optimization technique**

- maximum entropy & gradient descent (InfoMax)
- kurtosis or cumulates & gradient descent (Jade)
- negentropy & fixed point iteration (FastICA)

# Spatial /Tempral ICA in FMRI



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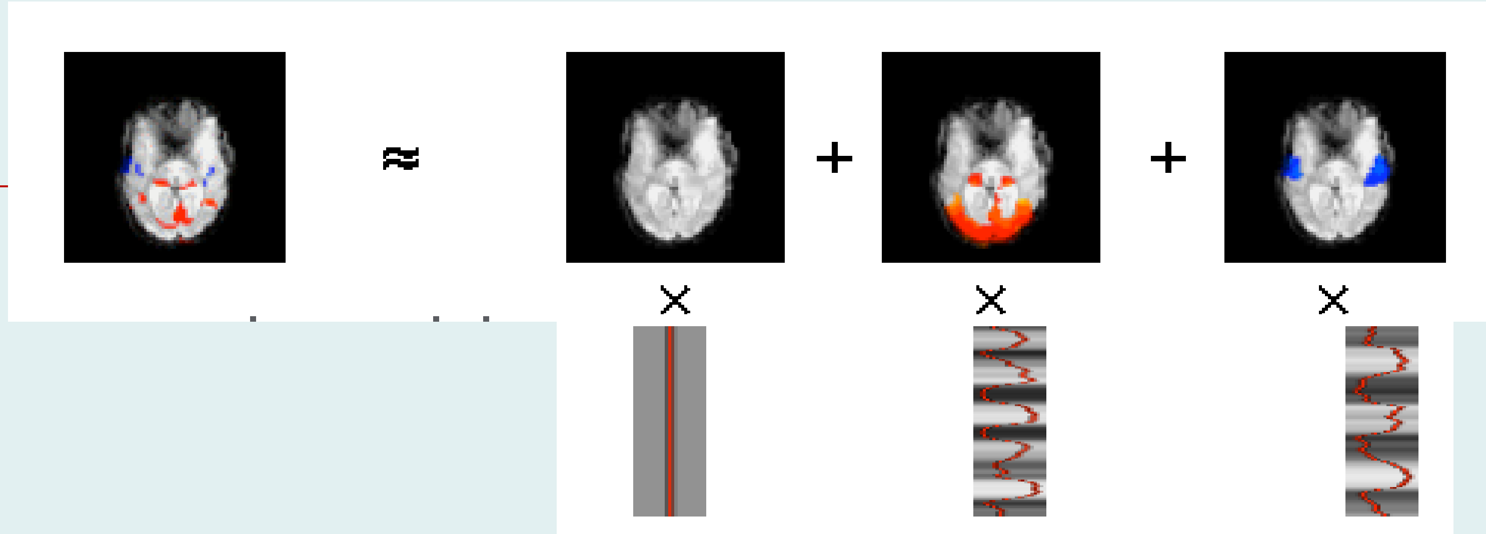

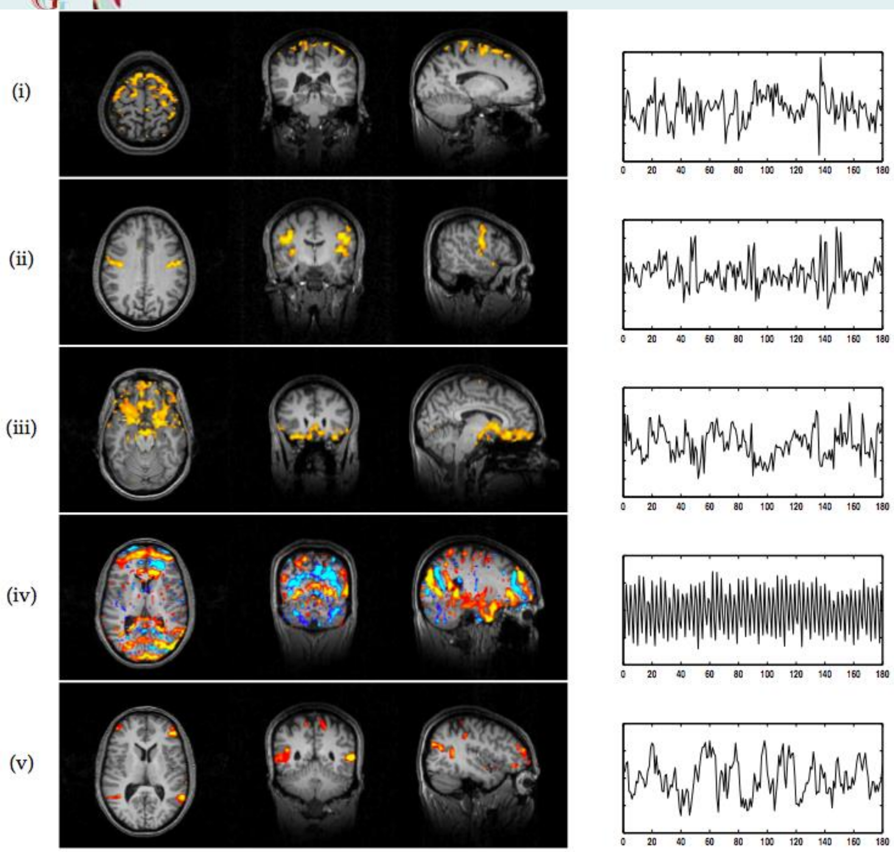


# *What are components in fMRI?*

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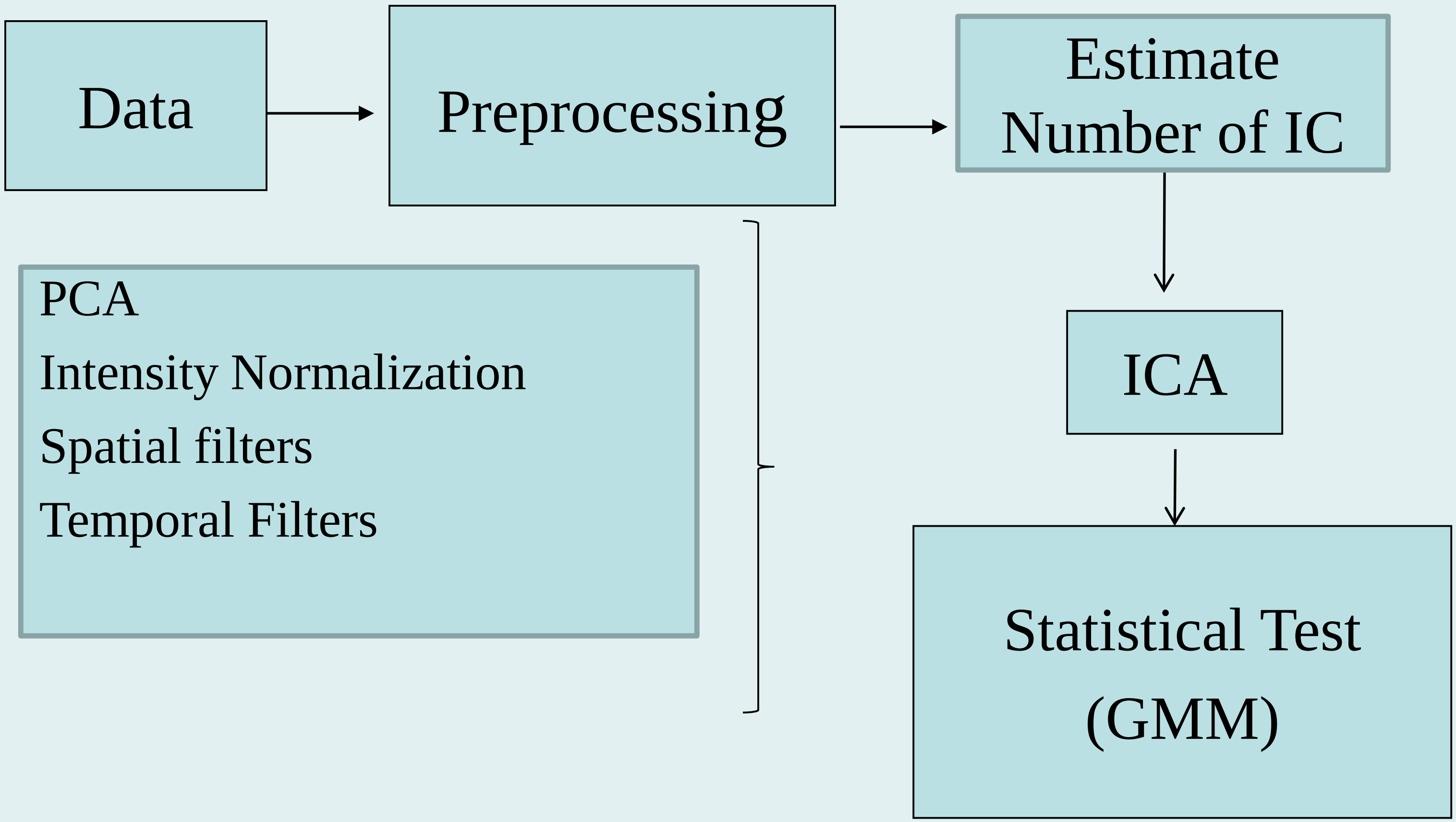
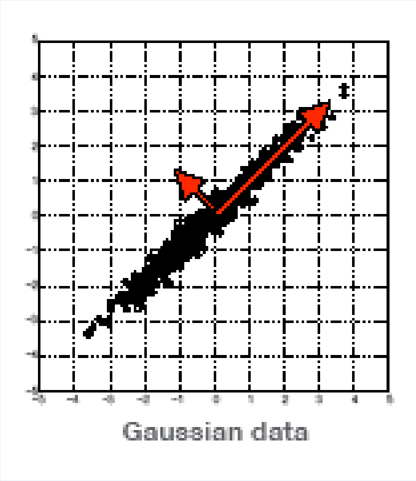
# *Chart of fMRI Analysis by ICA:*



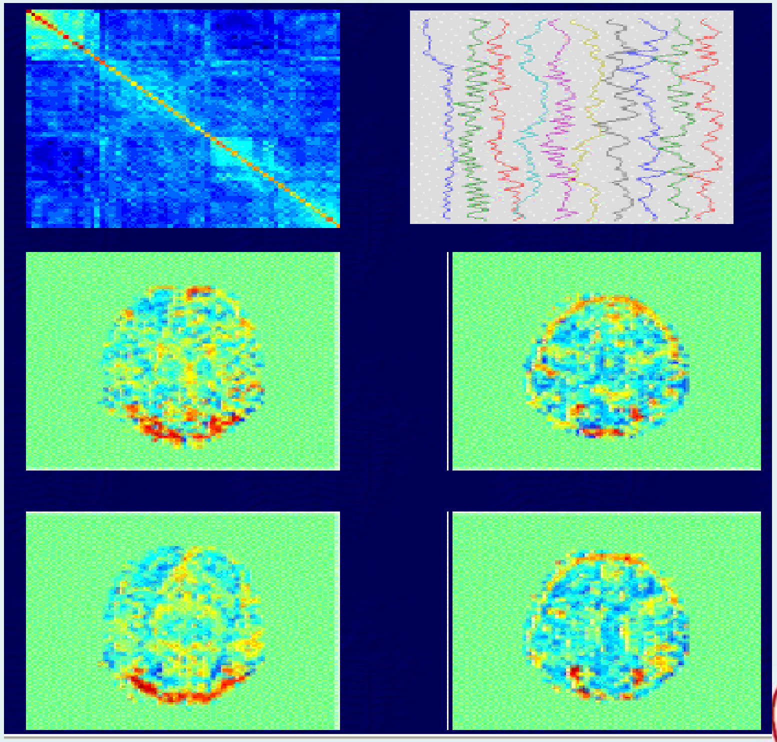
# Preprocessing:PCA

- **PCA** finds projections of maximum amount of variance in Gaussian data.




***In fMRI data:***

- calculate the data covariance matrix
- calculate the full set of Eigenvectors
- calculate the principal components by projecting the data onto the Eigenvectors



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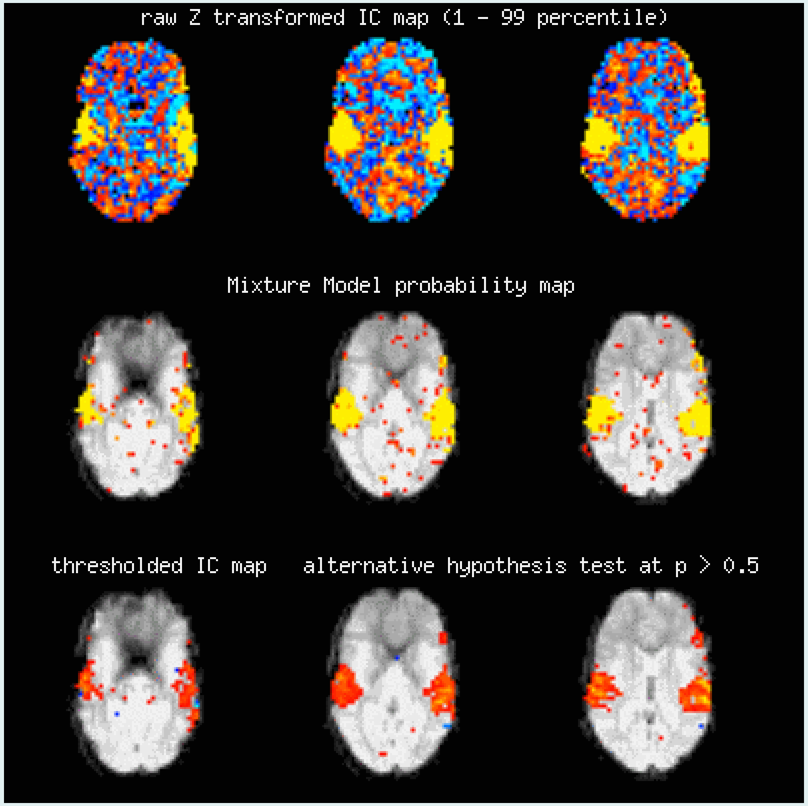
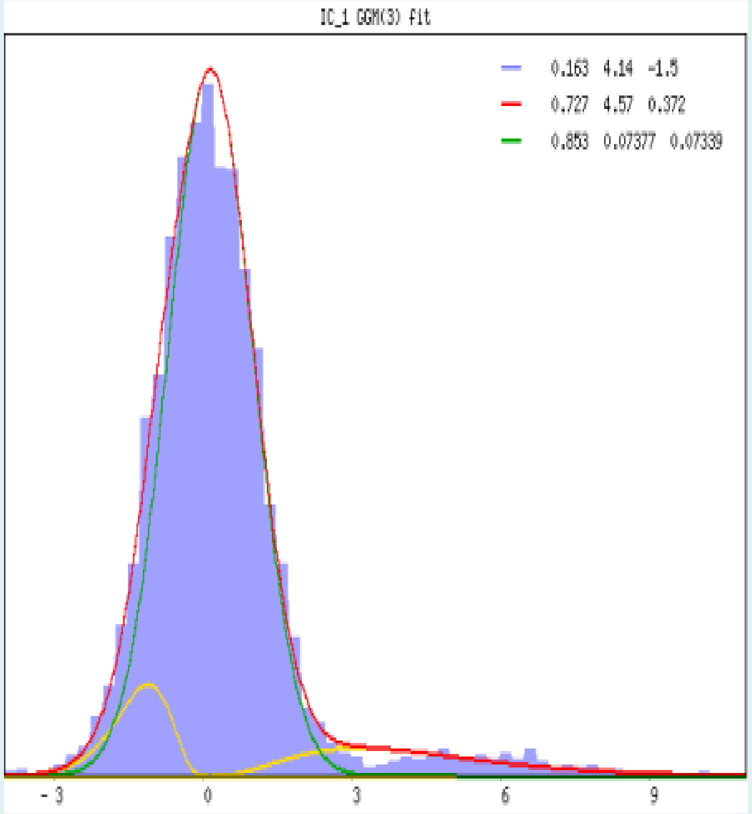
# Alternative hypothesis test

use a Gaussian / mixture model fitted to the histogram of intensity values (using EM)



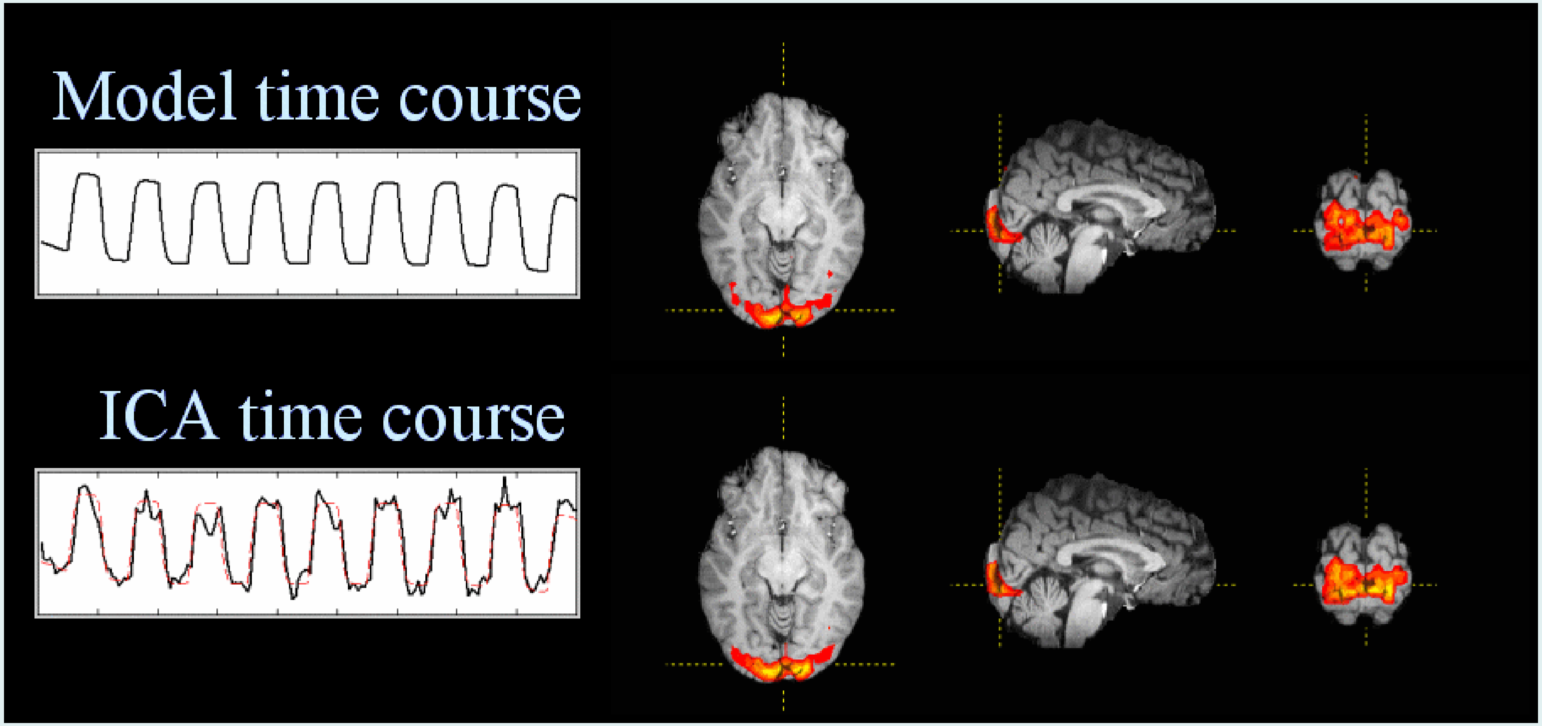



# Applications

- MELODIC can be useful to investigate the BOLD for finding areas of 'activation' which respond in a more complex way to an external stimulus
- estimate artifacts in the data
- analyze data for which no model of the BOLD response is available

# Investigating the BOLD response

- audio-visual stimulation
- 30s on/off reversing checkerboard with colour: GLM results and primary PICA map



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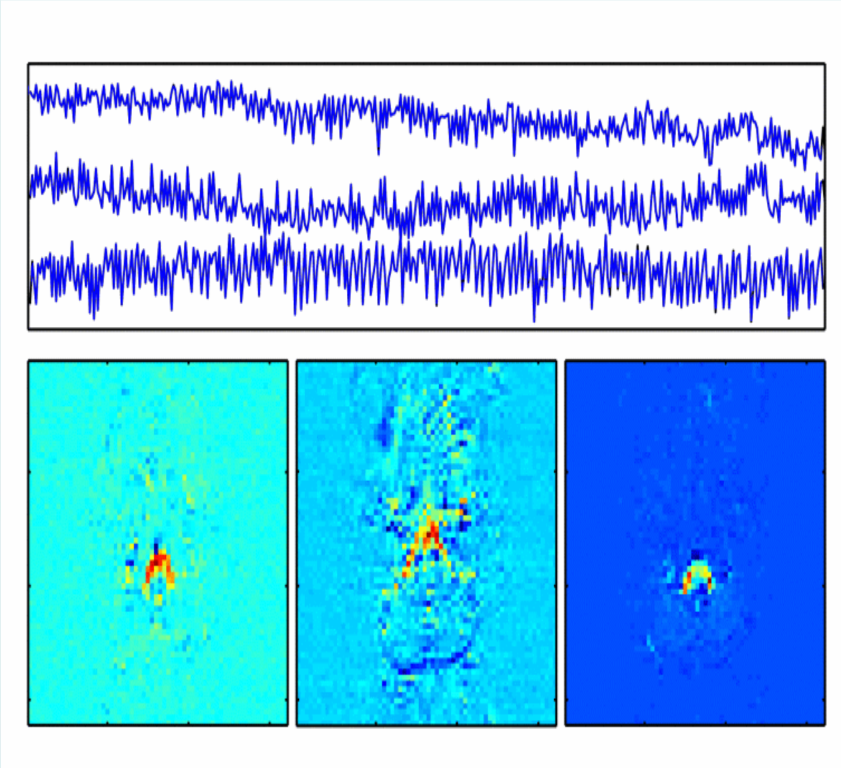
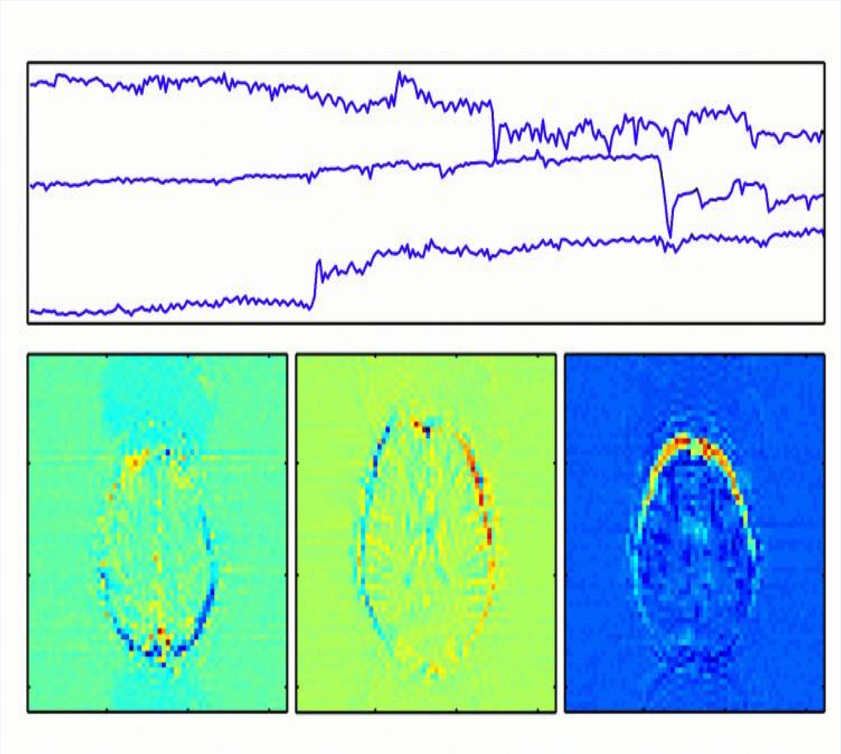
# Different kinds of Artifact

**high-frequency noise**



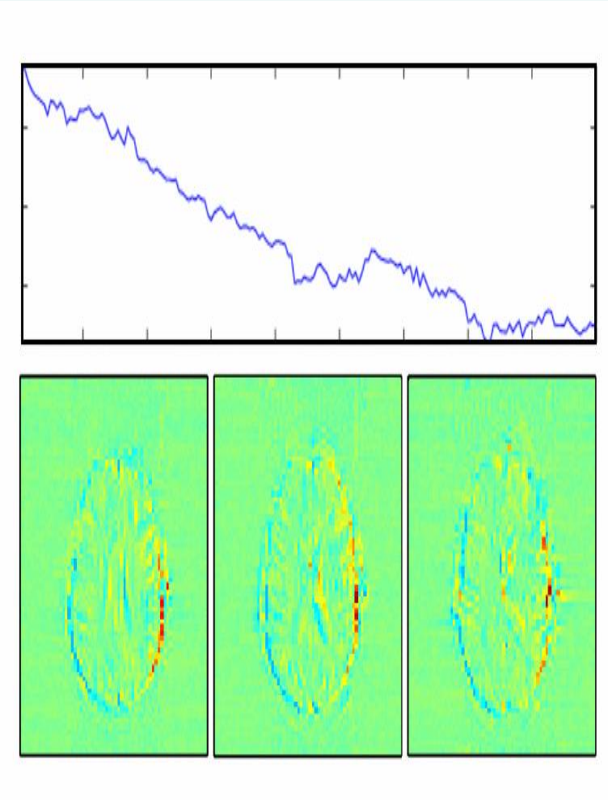
**abrupt head motion**



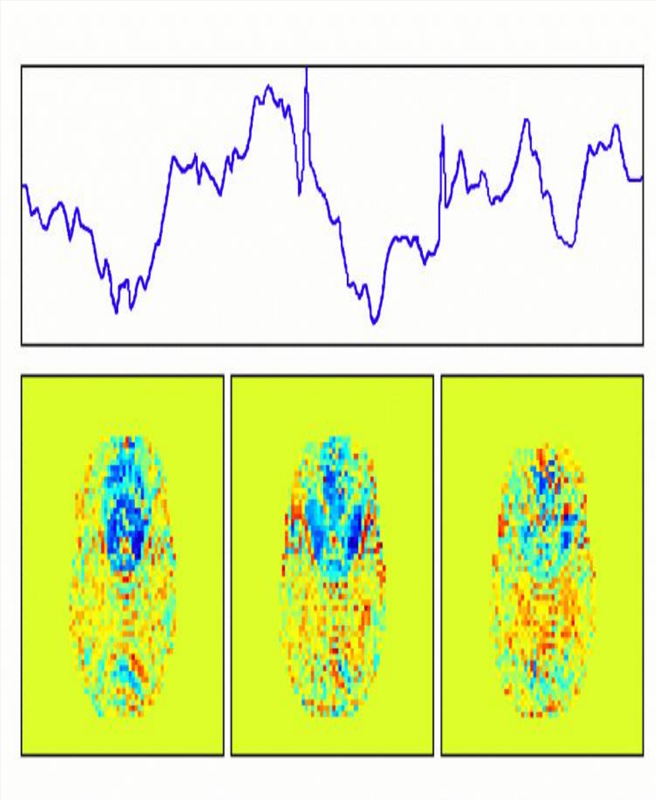
# Different kinds of Artifact



**slow head motion**

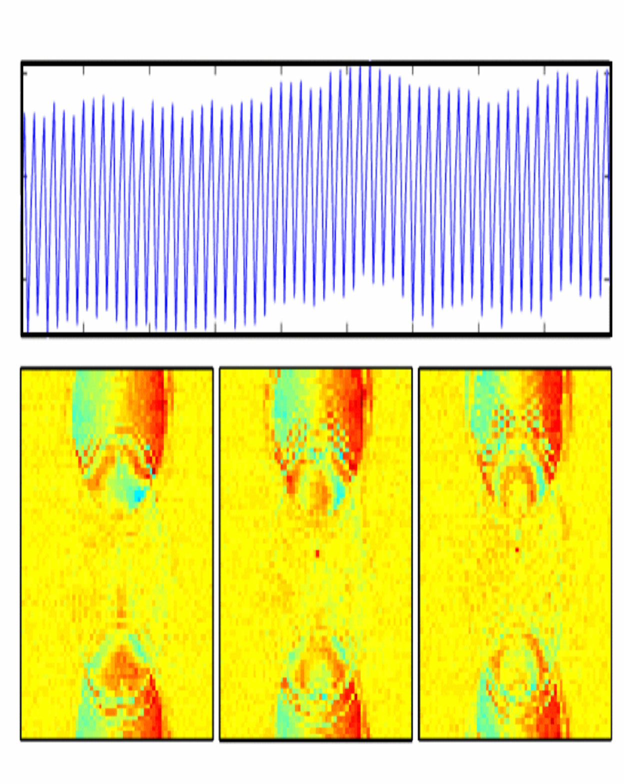


**signal loss due to B0 inhomogeneity**

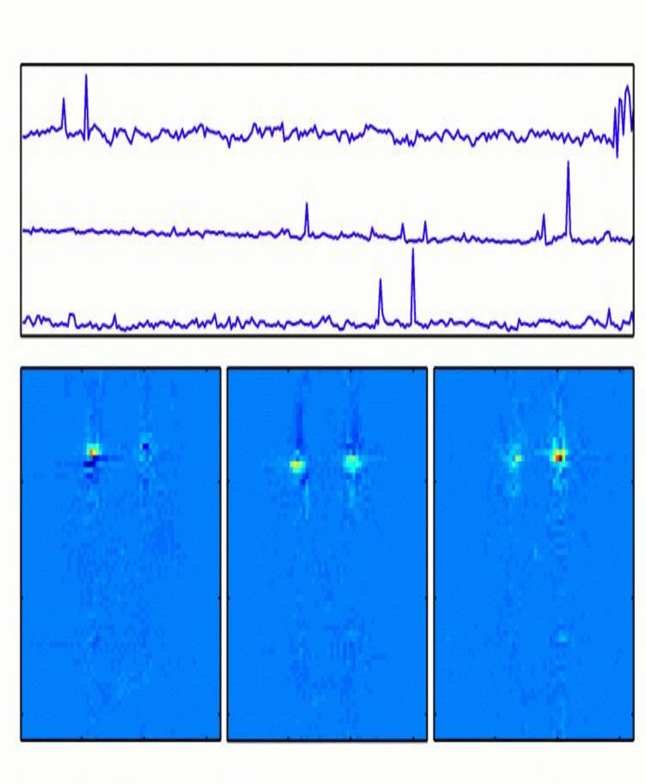


# Different kinds of Artifact

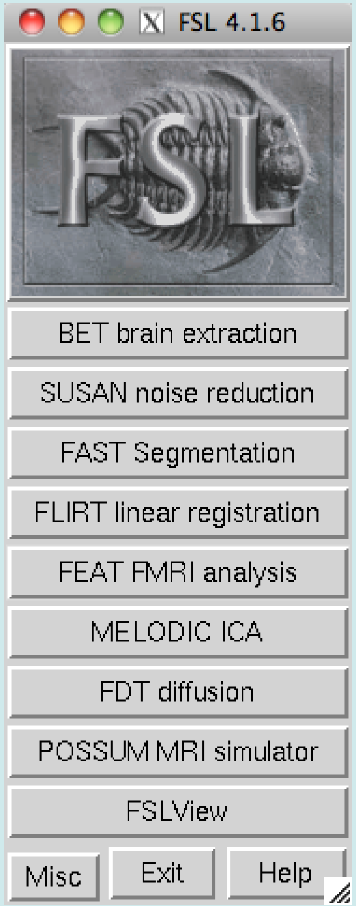
**EPI (N/2) ghosting**



**Eye-related artefacts**



# fMRI Analysis
# Processing with FSL




Presented By:
Nafiseh Hasani

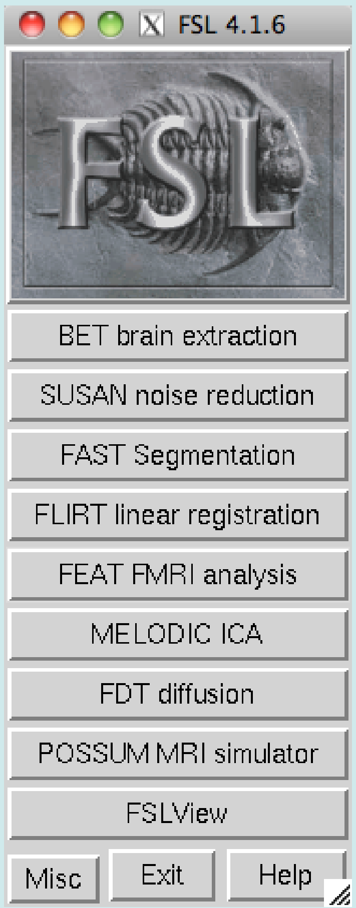
# Preparing Data for fMRI Analysis

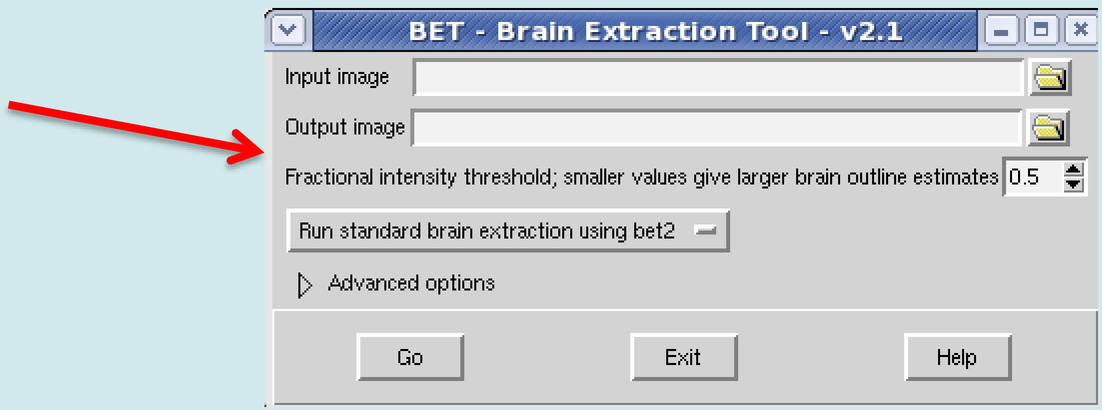
- With Dicomworks and DCM2Nii software convert functional and anatomical data into correct format (nifti ,4D)

# FSL: Single Subject

## BET

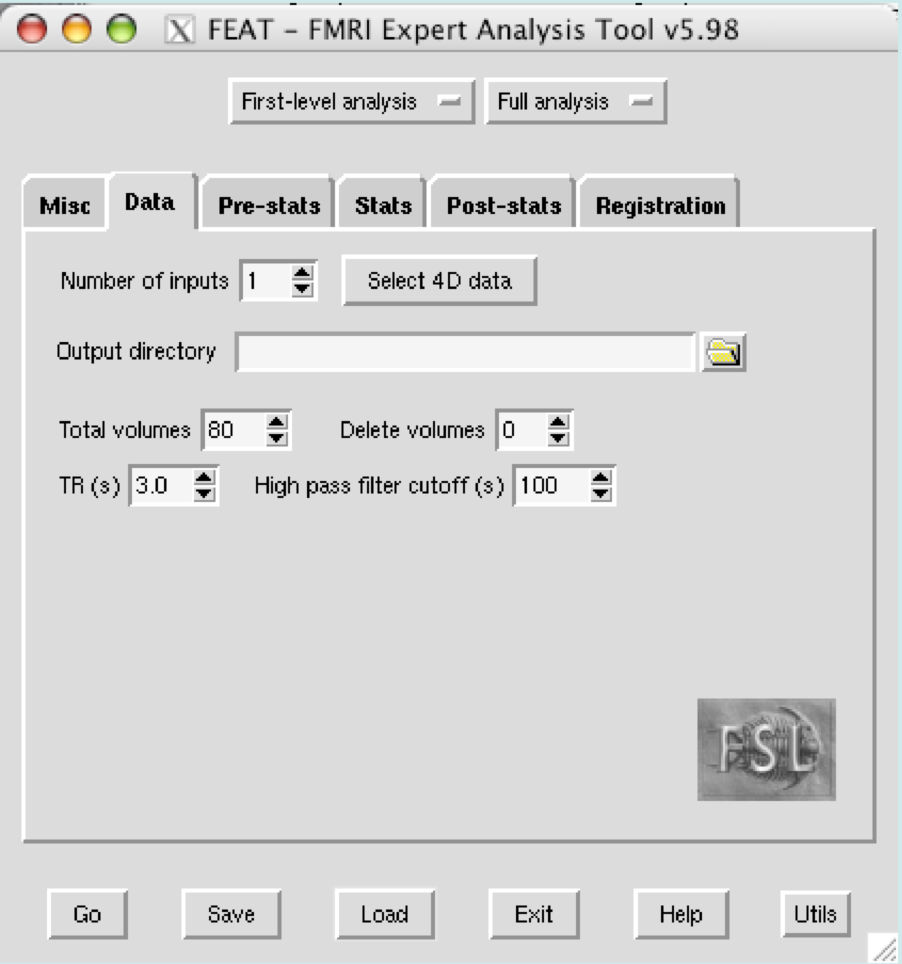
- All anatomical images have to be run through BET before using FEAT



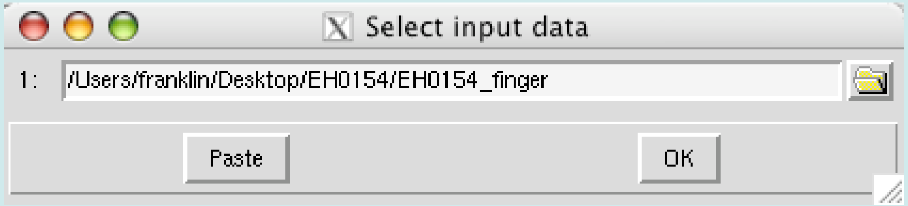



# FEAT FMRI Analysis

## Data




- Number of analyses
- Total Volumes
- Delete Volumes
- TR
- High Pass Filter Cutoff



# FEAT FMRI Analysis

## Delete Volumes

- The Number of initial FMRI volumes to delete before any further processing
- Typically two or three volumes
- These volumes are deleted as soon as the analysis is started

# FEAT FMRI Analysis

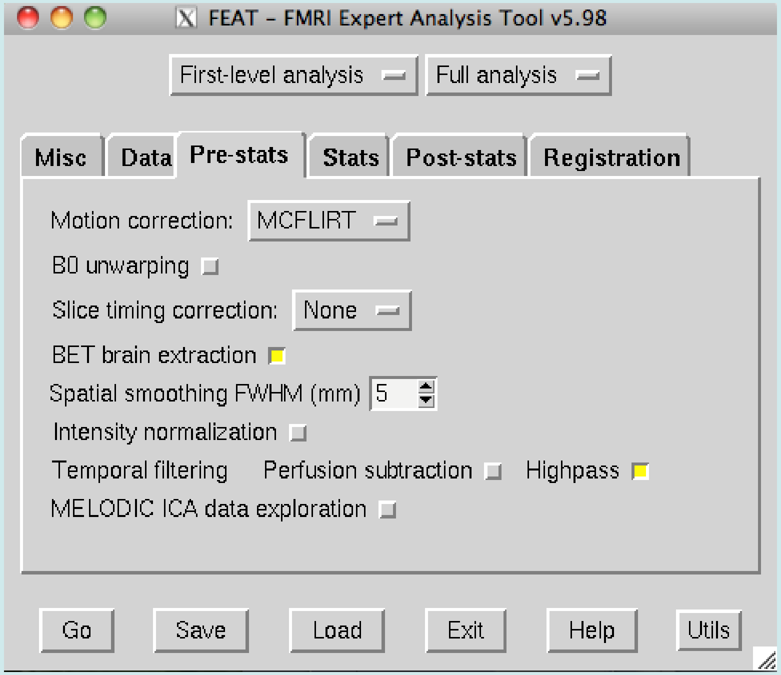
## High Pass Filter Cutoff

- A sensible setting in the case of an rArA or rArB type block design, is the r+A or r+A+r+B ,total cycle time
- For event-related designs the rule is not so simple. But in general the cut off can typically be reduced at least to 50s

# FEAT FMRI Analysis

## Pre-stats

- ❑ **Motion Correction**
- ❑ **BET Brain Extraction**
- ❑ **Slice timing correction**
- ❑ **Spatial Smoothing FWHM (mm)**
- ❑ **Temporal Filtering**



# FEAT FMRI Analysis

## Slice timing correction

- If slices were acquired from the bottom of the brain to the top select **Regular up**
- If slices were acquired from the top of the brain to the bottom select **Regular down**
- If the slices were acquired with interleaved order (0, 2, 4 ... 1, 3, 5 ...) the choose the **Interleaved** option.

# FEAT FMRI Analysis

## Spatial Smoothing FWHM

- This is intended to reduce noise without reducing valid activation; this is successful as long as the underlying activation area is larger than the extent of the smoothing

- if you are looking for very small activation areas then you should maybe reduce smoothing from the default of 5mm, and if you are looking for larger areas, you can increase it

# FEAT FMRI Analysis

## Temporal Filtering

- Highpass temporal filtering : remove low frequency artifacts

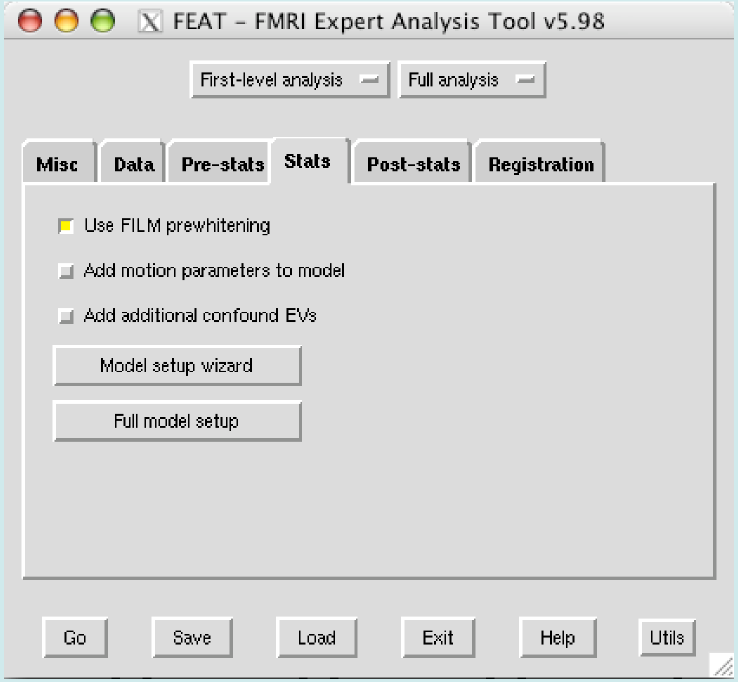
- Lowpass temporal filtering:  reduces high frequency noise by  Gaussian smoothing (sigma=2.8s), but also reduces the strength of the signal of interest. so is turned off by default.

# FEAT FMRI Analysis

**Model Setup:**

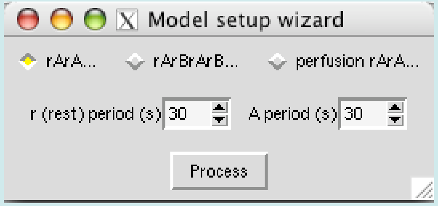
## 1. Model Setup Wizard

- Allows you to setup simple experimental designs.



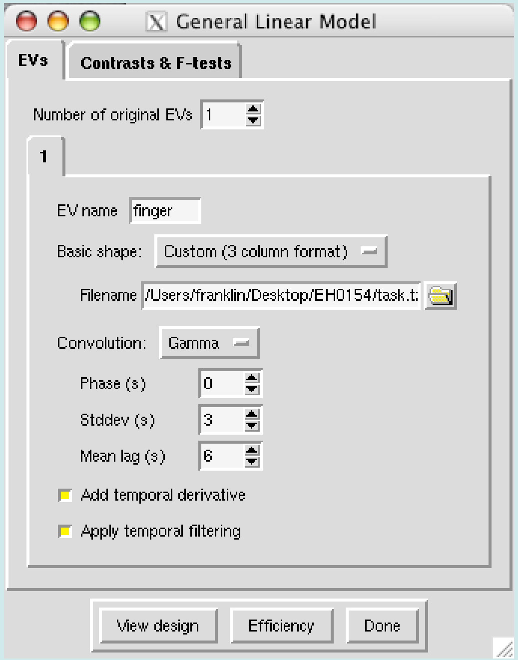

## 2. Full Model Setup

- You will need to give a text file containing ones and zeros for each explanatory variable.



# FEAT FMRI Analysis

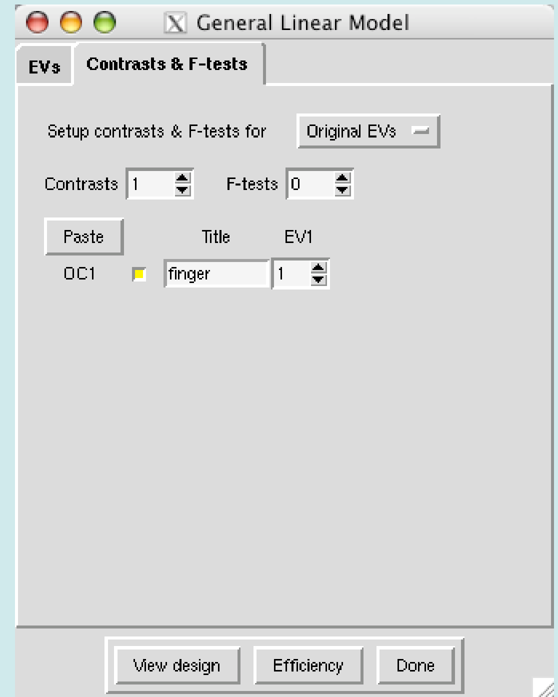
## Contrasts




- ❑ Original EVs- Explanatory Variables
- ❑ Basic Shape- design matrix Custom (3 Column Format)

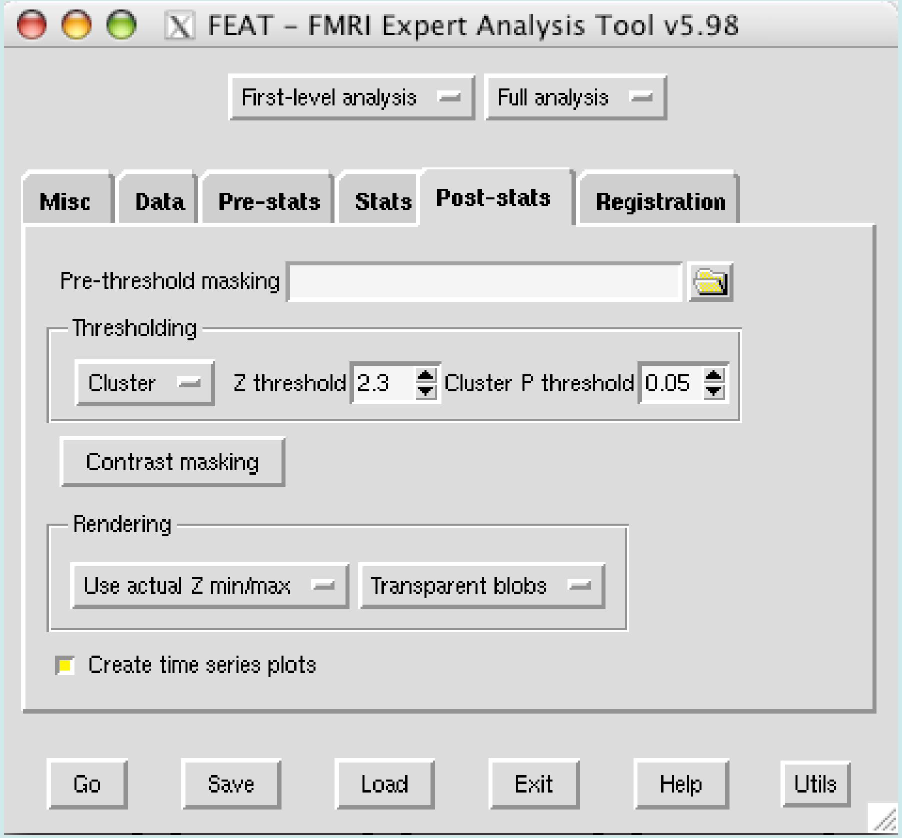
| | | |
|---|---|---|
| 0 | 30 | 1 |
| 30 | 30 | 0 |
| 60 | 30 | 1 |
| 90 | 30 | 0 |
| 120 | 30 | 1 |
| 150 | 30 | 0 |
| 180 | 30 | 1 |
| 210 | 30 | 0 |

3-Column Text File



# FEAT FMRI Analysis

## Post-stats



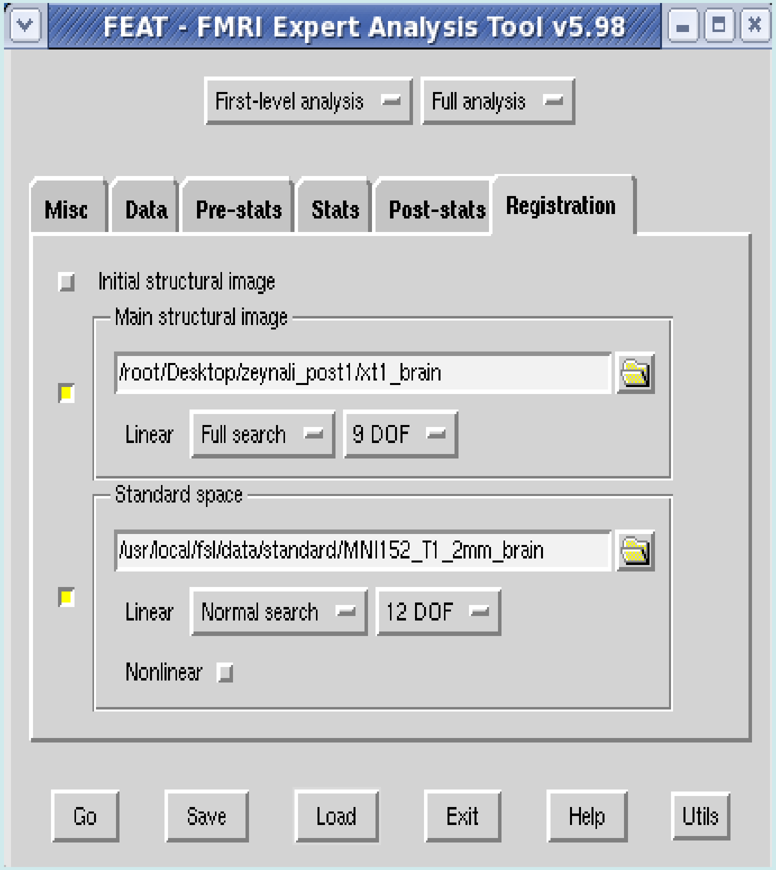

- The data is usually run with the default threshold of $Z > 2.3$.

# FEAT FMRI Analysis

## Registration

- **Main Structure –** 3D anatomical image.

- **Standard Space –** Should be an image in Talairach space



# FEAT FMRI Analysis

## Results

- For each analyses ran through FEAT an output directory is created with the extension **“.feat”**
  - This folder you will contain all statistical images and files.
    - The results can be viewed by clicking on the **report.html**.
  - This will included the Pre-stats, Stats, Post-Stats, Registration, and Log.

# FEAT FMRI Analysis

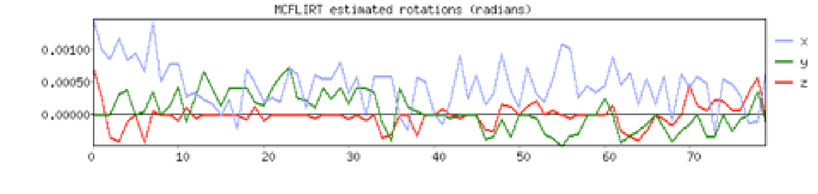
## Results

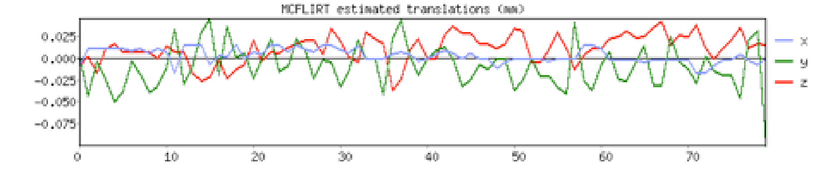
### Pre-stats

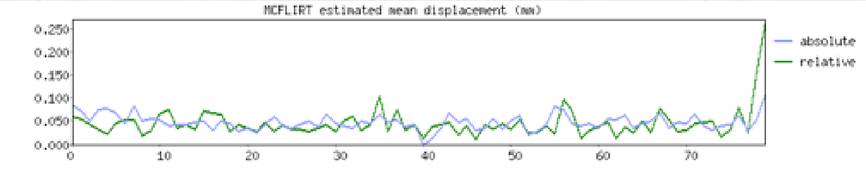
**Analysis methods**
FMRI data processing was carried out using FEAT (FMRI Expert Analysis Tool) Version 5.98, part of FSL (FMRIB's Software Library, www.fmrib.ox.ac.uk/fsl). The following pre-statistics processing was applied; motion correction using MCFLIRT [Jenkinson 2002]; non-brain removal using BET [Smith 2002]; spatial smoothing using a Gaussian kernel of FWHM 5mm; grand-mean intensity normalisation of the entire 4D dataset by a single multiplicative factor; highpass temporal filtering (Gaussian-weighted least-squares straight line fitting, with sigma=50.0s).

**References**
[Jenkinson 2002] M. Jenkinson and P. Bannister and M. Brady and S. Smith. Improved optimisation for the robust and accurate linear registration and motion correction of brain images. NeuroImage 17:2(825-841) 2002.
[Smith 2002] S. Smith. Fast Robust Automated Brain Extraction. Human Brain Mapping 17:3(143-155) 2002.

**MCFLIRT Motion correction**
Mean displacements: absolute=0.04mm, relative=0.04mm



# FEAT FMRI Analysis

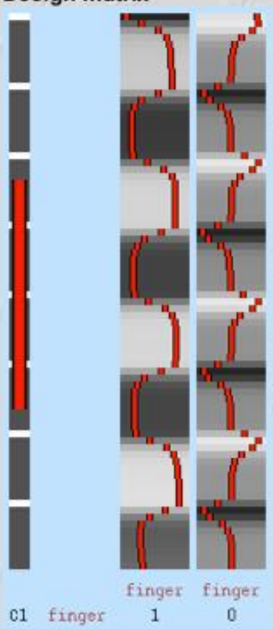
## Results

### Stats

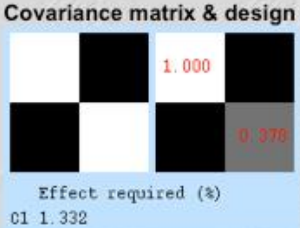
**Analysis methods**
FMRI data processing was carried out using FEAT (FMRI Expert Analysis Tool) Version 5.98, part of FSL (FMRIB's Software Library, www.fmrib.ox.ac.uk/fsl). Time-series statistical analysis was carried out using FILM with local autocorrelation correction [Woolrich 2001].

**References**
[Woolrich 2001] M.W. Woolrich, B.D. Ripley, J.M. Brady and S.M. Smith. Temporal Autocorrelation in Univariate Linear Modelling of FMRI Data. NeuroImage 14:6(1370-1386) 2001.

**Design matrix**




**Covariance matrix & design efficiency**



# FEAT FMRI Analysis

## Results

### Post-stats

**Analysis methods**

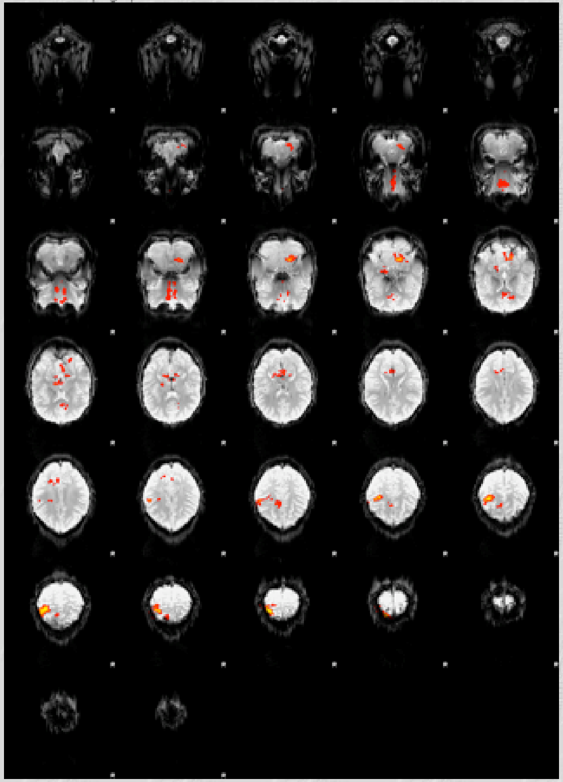
FMRI data processing was carried out using FEAT (FMRI Expert Analysis Tool) Version 5.98, part of FSL (FMRIB's Software Library, www.fmrib.ox.ac.uk/fsl). Z (Gaussianised T/F) statistic images were thresholded using clusters determined by Z>2.3 and a (corrected) cluster significance threshold of P=0.05 [Worsley 2001].

**References**

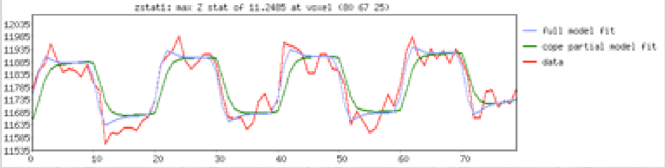

[Worsley 2001] K.J. Worsley. Statistical analysis of activation images. Ch 14, in Functional MRI: An Introduction to Methods, eds. P. Jezzard, P.M. Matthews and S.M. Smith. OUP, 2001.


**Thresholded activation images** 2.3 – 11.2

zstat1 - C1 (finger)



**Time series plots**



# FEAT FMRI Analysis

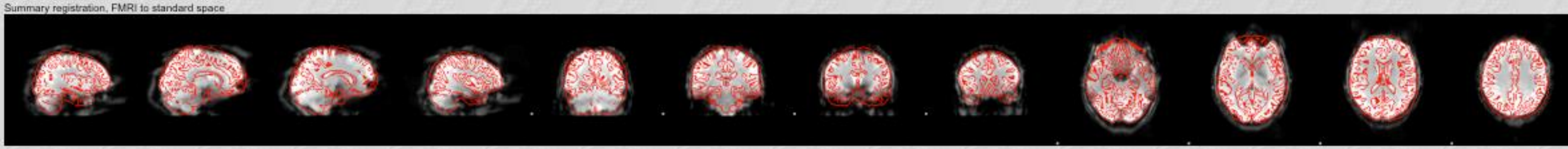
## Results

### Registration

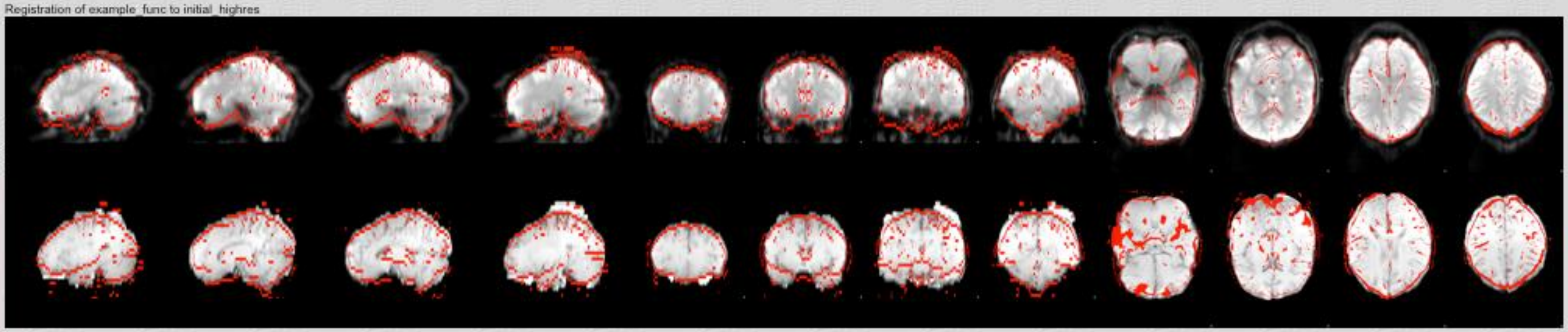
**Analysis methods**
FMRI data processing was carried out using FEAT (FMRI Expert Analysis Tool) Version 5.98, part of FSL (FMRIB's Software Library, www.fmrib.ox.ac.uk/fsl). Registration to high resolution structural and/or standard space images was carried out using FLIRT [Jenkinson 2001, 2002].

**References**
[Jenkinson 2001] M. Jenkinson and S.M. Smith. A Global Optimisation Method for Robust Affine Registration of Brain Images. Medical Image Analysis 5:2(143-156) 2001.
[Jenkinson 2002] M. Jenkinson, P. Bannister, M. Brady and S. Smith. Improved Optimisation for the Robust and Accurate Linear Registration and Motion Correction of Brain Images. NeuroImage 17:2(825-841) 2002.

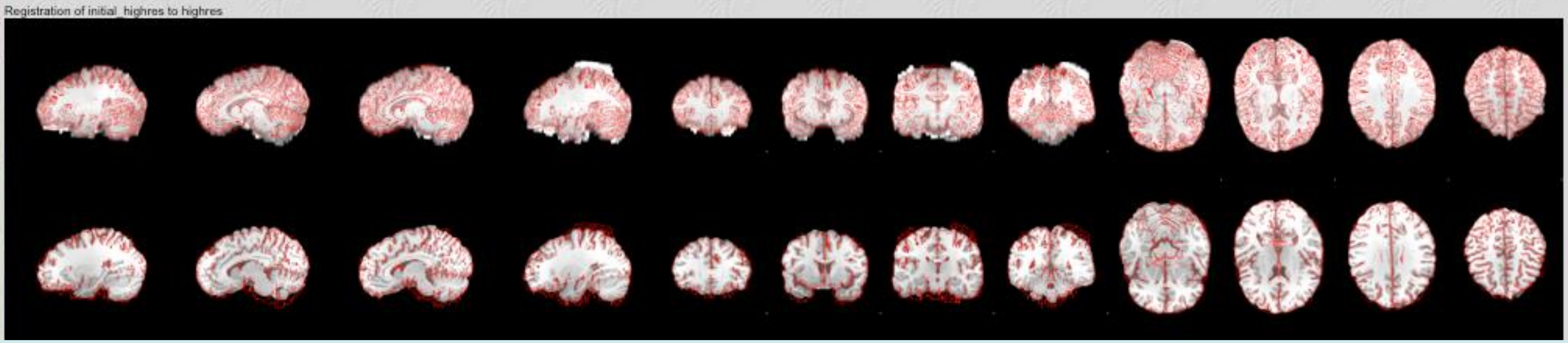
Summary registration, FMRI to standard space



Registration of example_func to initial_highres



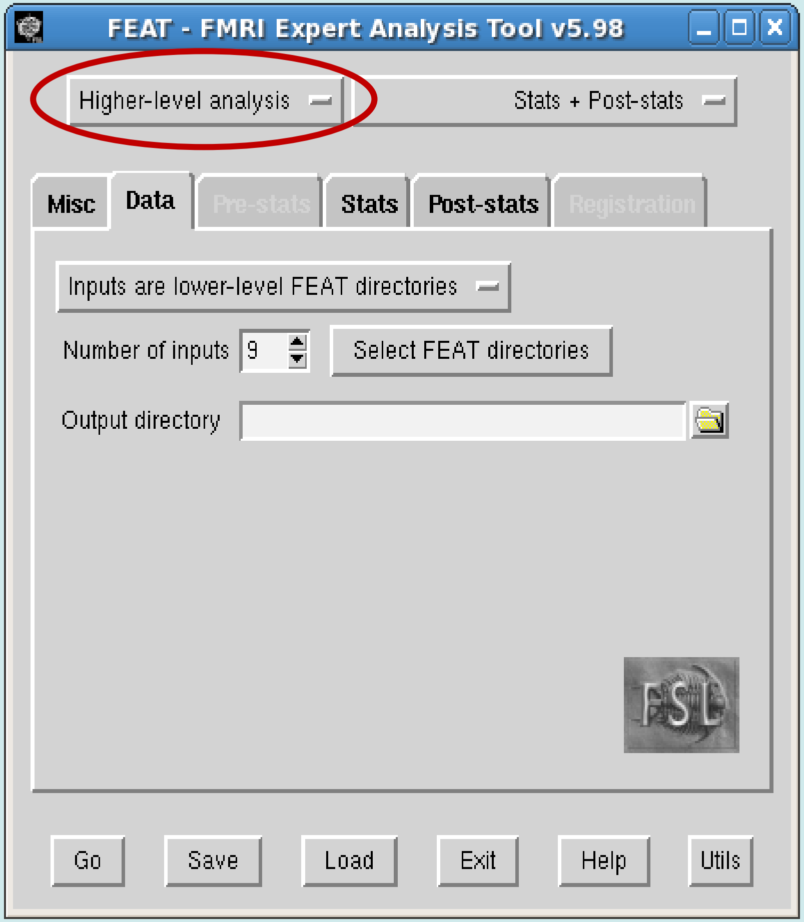
Registration of initial_highres to highres



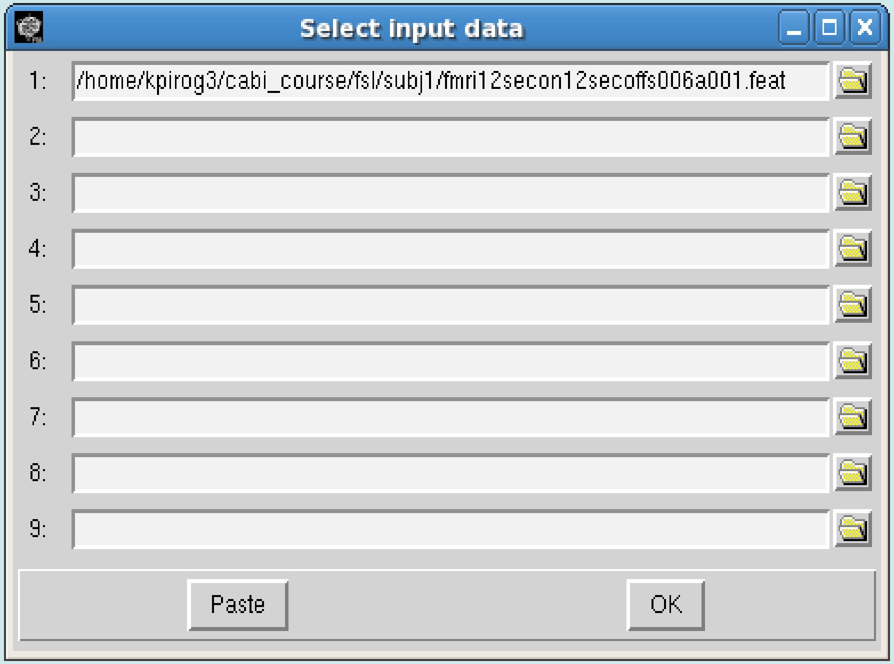
# FSL: Group Analysis

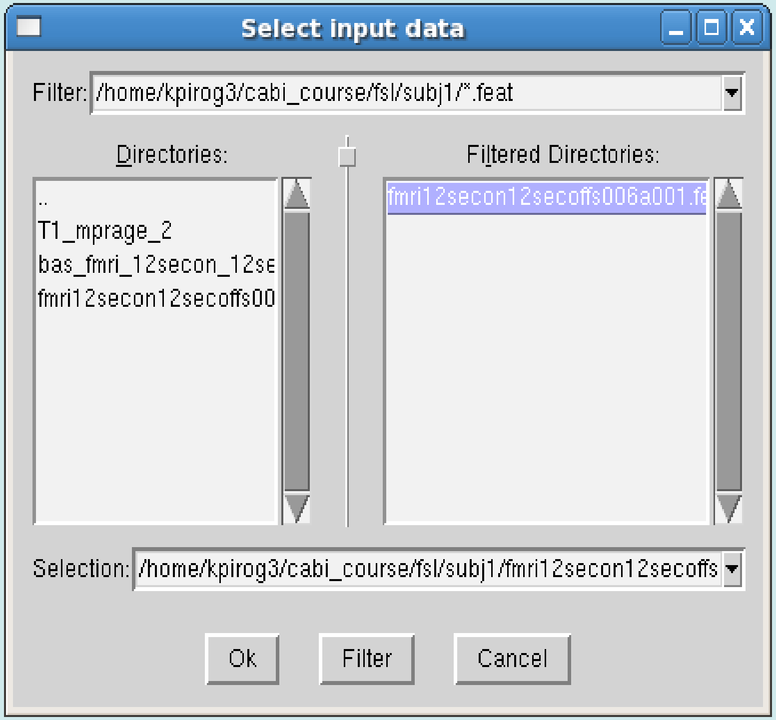
- ❑ Set tab at top left to Higher-level analysis

- ❑ Data Tab:
  - – Inputs are FEAT directories
  - – 9 inputs
  - – Output directory should be ok if you launched FSL from the directory I told you to create for yourself!



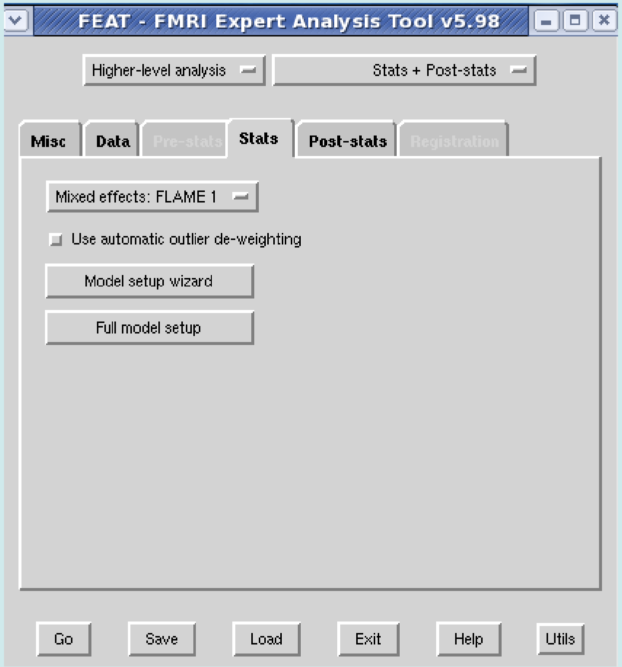
# FSL: Group Analysis




- ❑ Click on folders to get to file selection dialog and select the fmri...feat directory for each participant

# FSL: Group Analysis




- Fixed effects
- Mixed effects: FLAME1
- Mixed effects: FLAME 1+2
- Mixed effects: Simple OLS

# FSL: Group Analysis

## Fixed effects

- FE ignores cross-session/subject variance, reported activation is with respect to the group of sessions or subjects present, and not representative of the wider population

# FSL: Group Analysis

## Mixed effects: Simple OLS

- OLS (ordinary least squares) is a fast estimation technique which ignores all lower-level variance estimation and applies a very simple higher-level model. This is the least accurate of the ME options.

# FSL: Group Analysis

## Mixed effects: FLAME1

- For the most accurate estimation of higher-level activation you should use FLAME (FMRIB's Local Analysis of Mixed Effects) modeling and estimation. it allows separate modeling of the variance in different subject groups
- The first stage of FLAME is significantly more accurate than OLS, and nearly as fast

# FSL: Group Analysis

## Mixed effects: FLAME1 +2

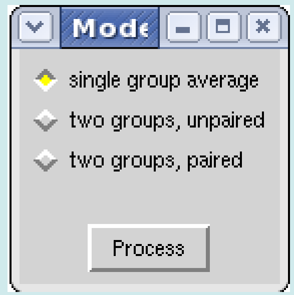
- The second stage of FLAME increases accuracy slightly over the first stage, but is quite a lot slower (typically 45-200 minutes).

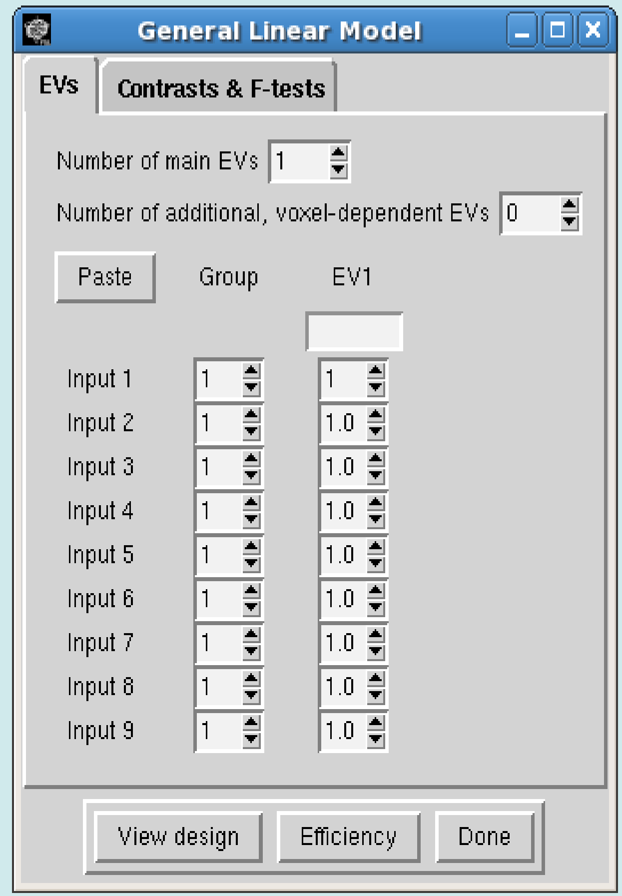
- **FLAME 1+2** is most significant in a highest-level analysis when you have a small number of subjects (say <10).

# FSL: Group Analysis

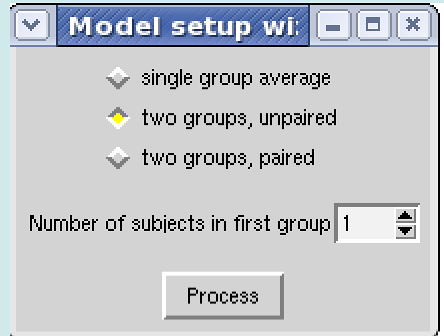
## Model Setup

- ❑ EVs Tab
  - Only 1 group of subjects
  - Want to see motor activity for all subjects, so set EV to 1 for each input



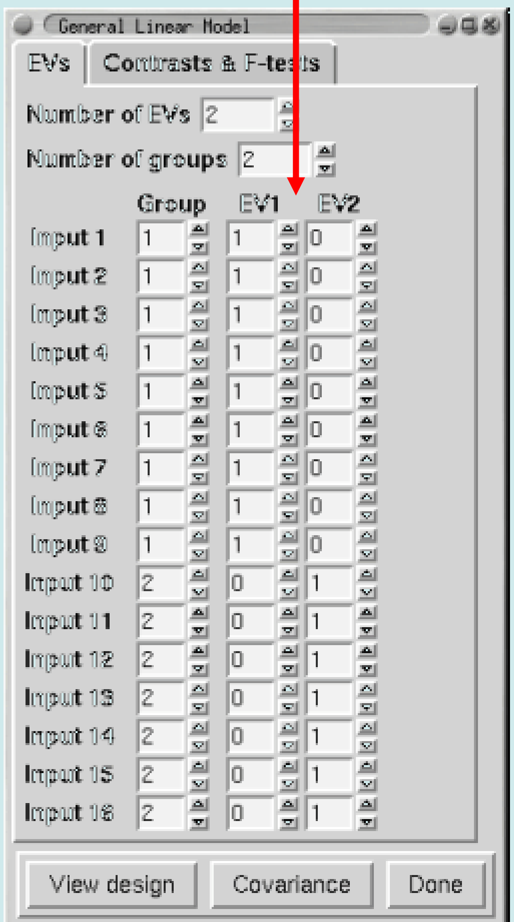
# Group Effects

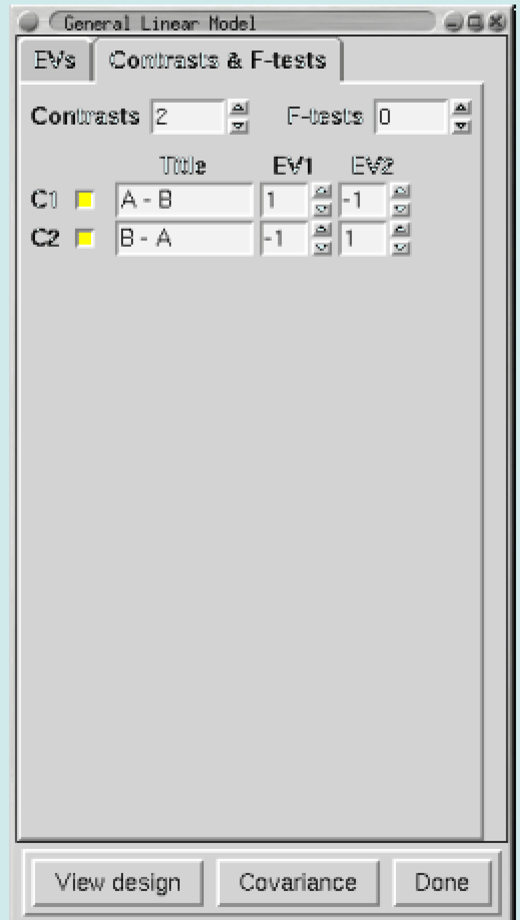


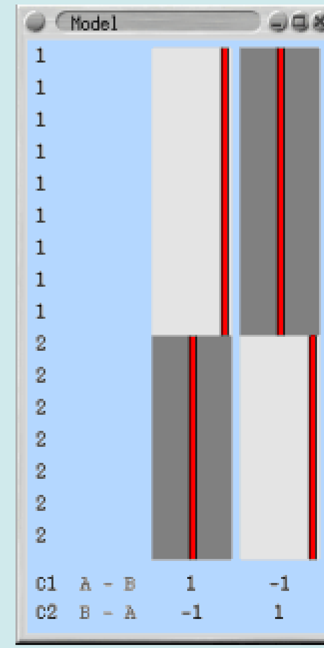

EV1 = include in group1
EV2 = include in group2

group1

group2

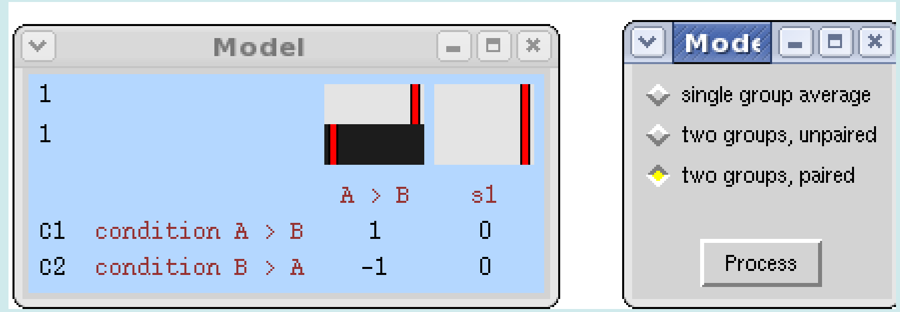


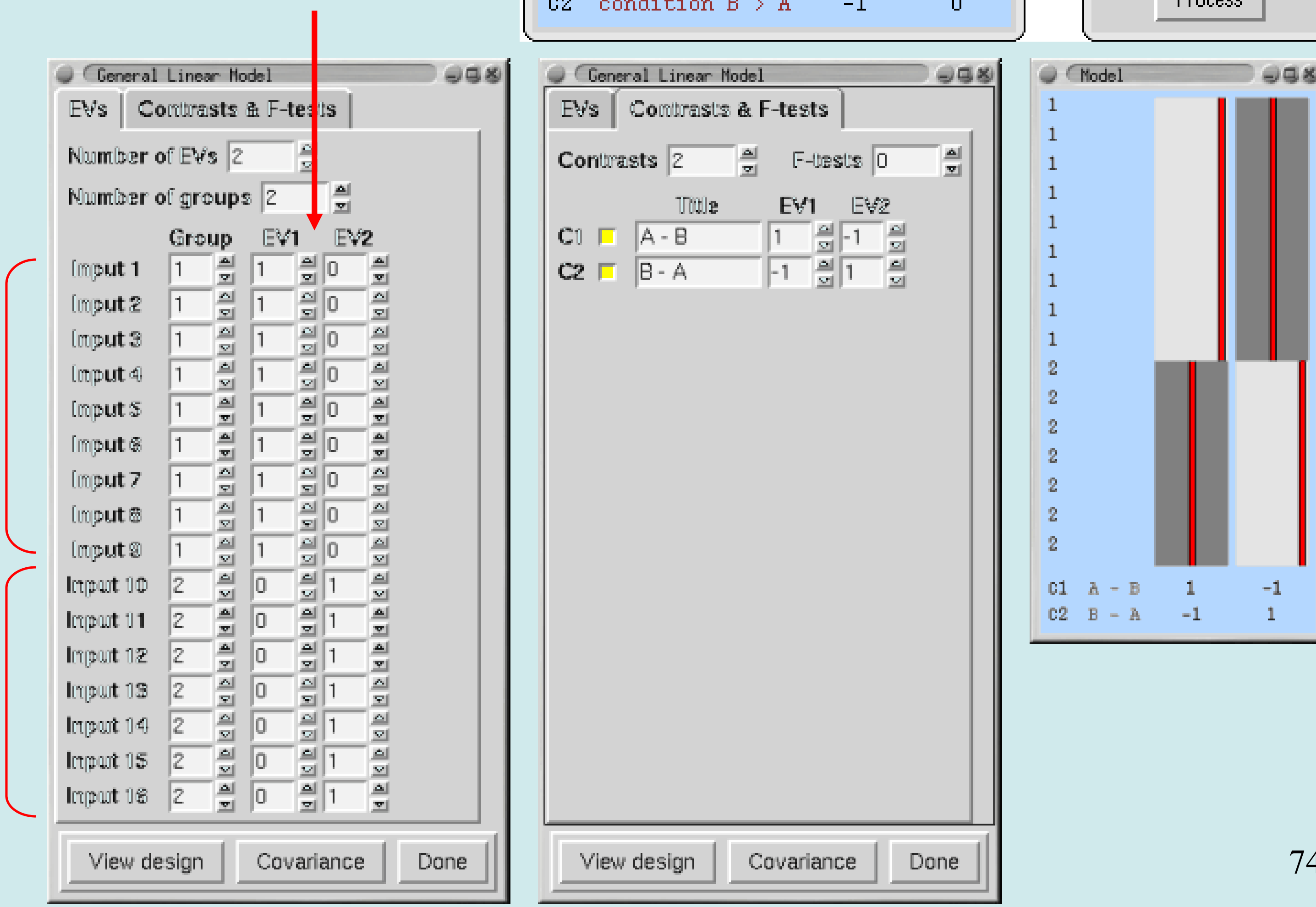
# Group Effects

EV1 = include in condition1
EV2 = include in condition2



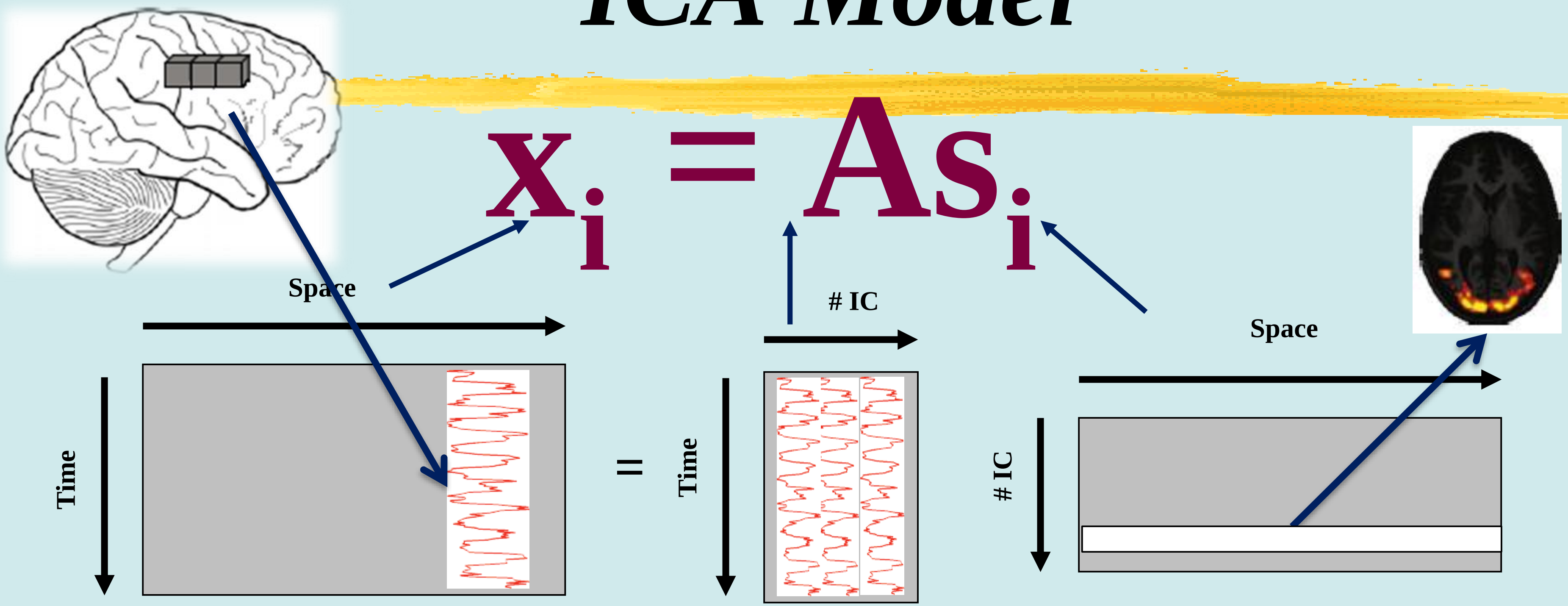

condition1

condition2



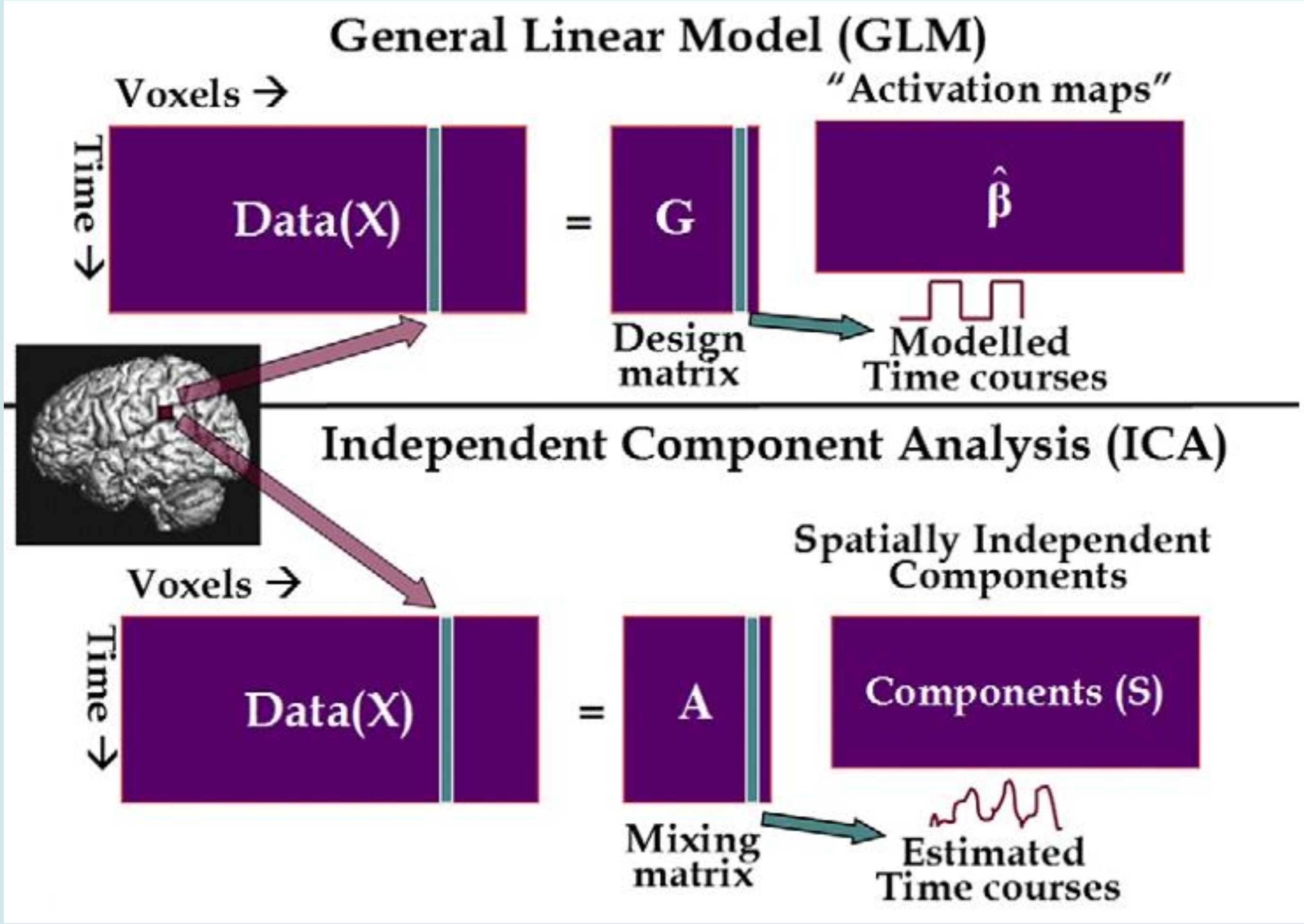
# *ICA Model*

$$x_i = As_i$$



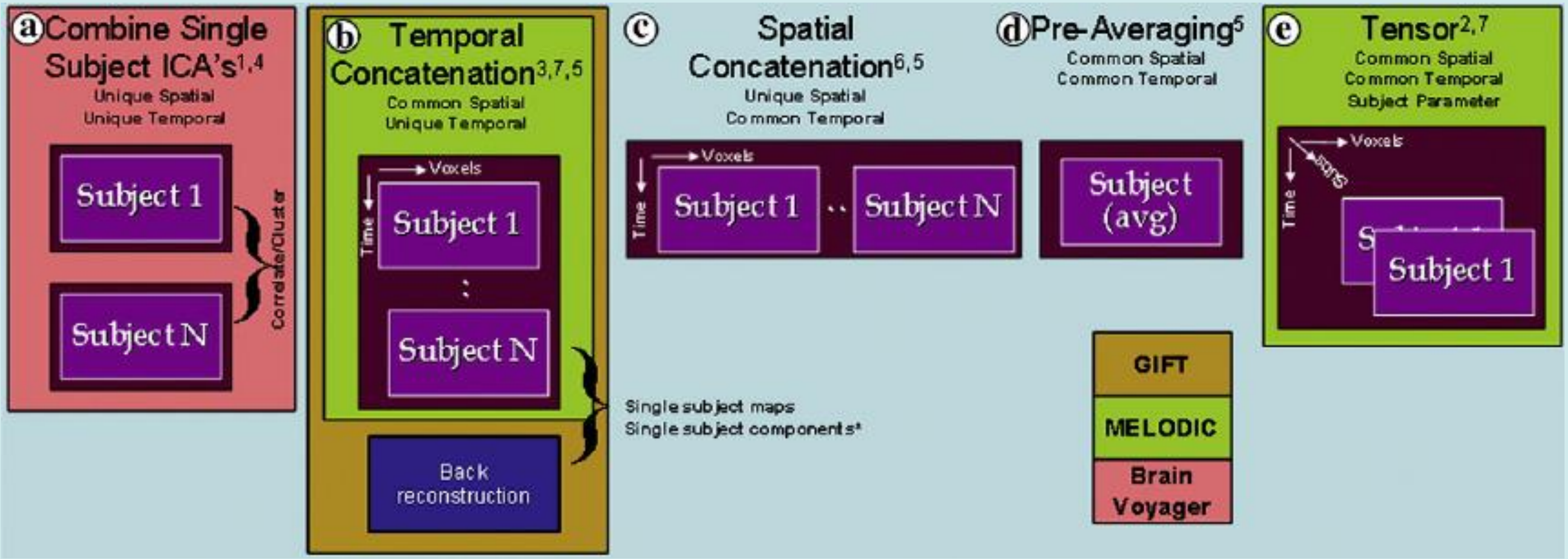

| FMRI Data | Mixing Matrix | Spatial Maps |
|---|---|---|
| P x N | P x K | K x N |
| P= Num. of Scans | P= Num. of Scans | K= Num. of IC |
| N= Num. of Voxels | K= Num. of IC | N=Num. of Voxels |
| Columns are Vectors of Observations | No Statistical Constraint (Possibly correlated) | Statistically Independent Non-Gaussian Sources (including Noise) |

# GLM vs. ICA


General Linear Model (GLM)
Voxels →
Time ↓
Data(X)
=
G
"Activation maps"
β̂
Design matrix
Modelled Time courses
Independent Component Analysis (ICA)
Voxels →
Time ↓
Data(X)
=
A
Spatially Independent Components
Components (S)
Mixing matrix
Estimated Time courses

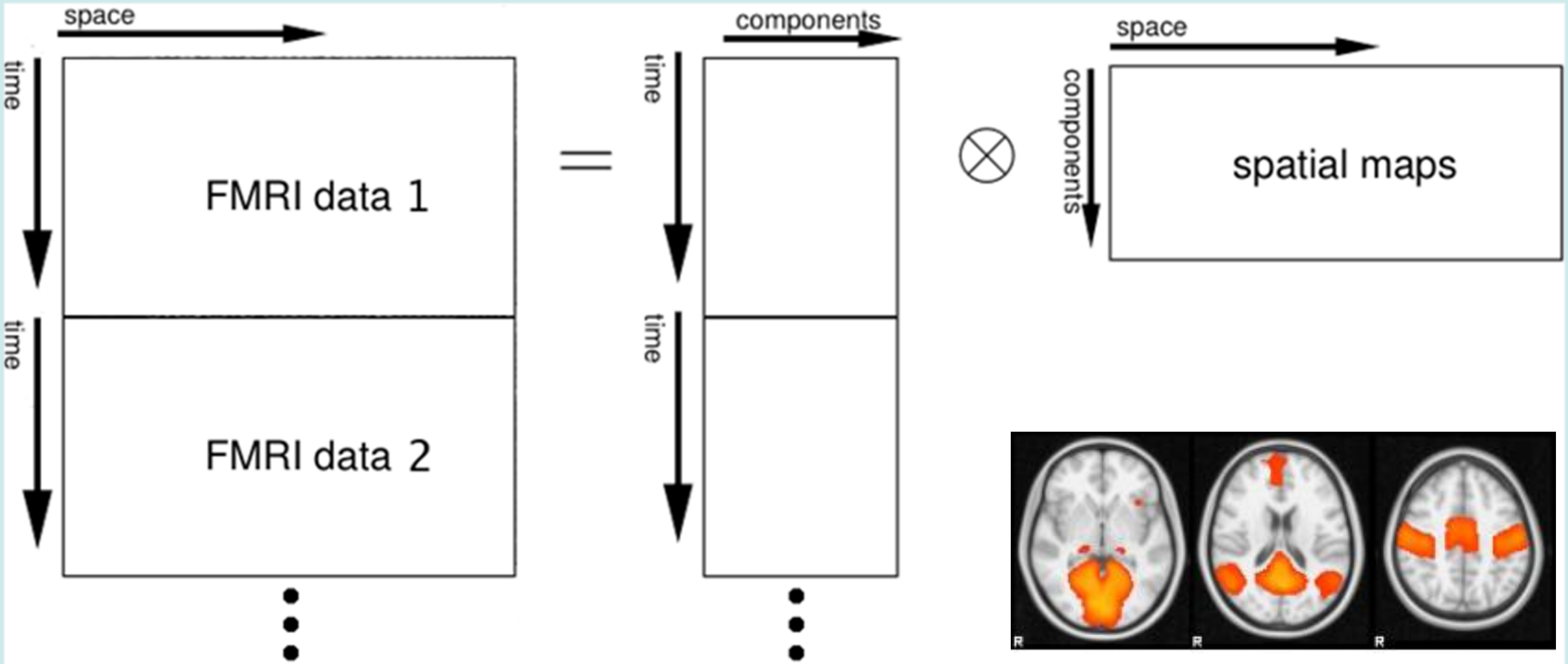
# Group ICA models

***Each Single subject analysis yields a different set of ICs***



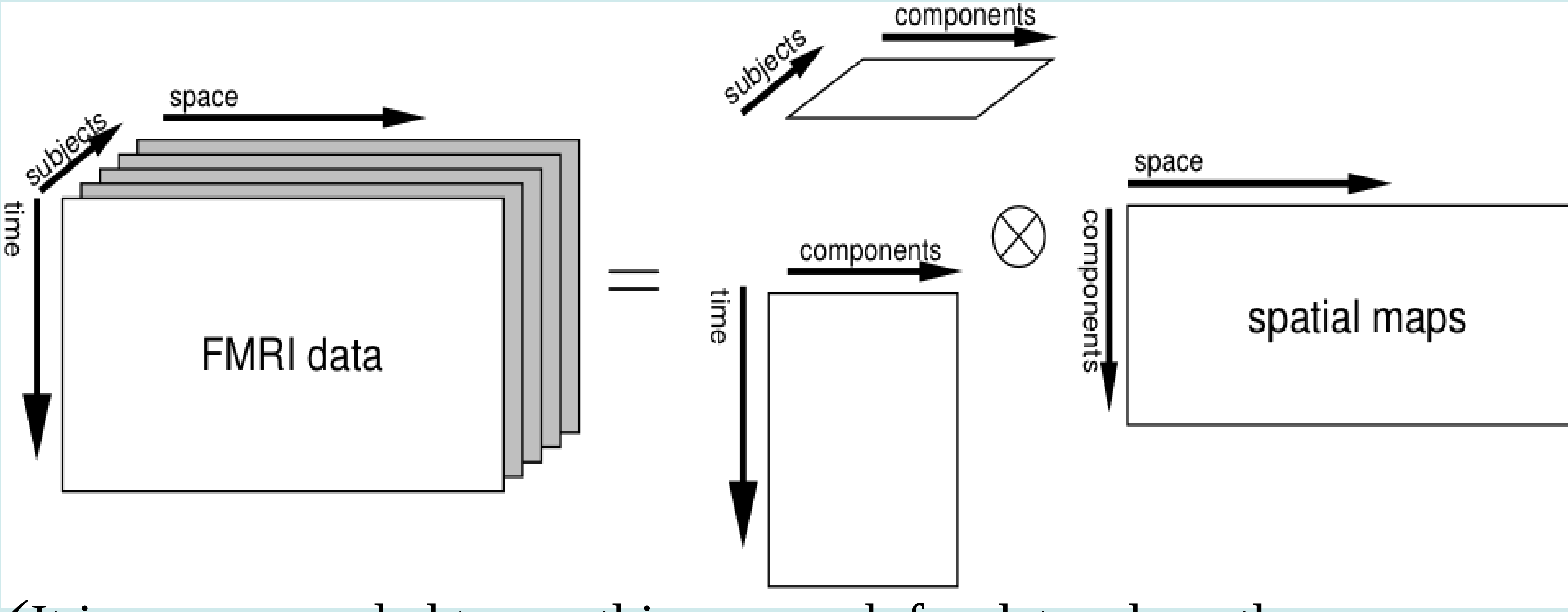

Calhoun et. al 2009

# *Temporal Concatenation*




✓ **This approach does not assume that the temporal response pattern is the same across the population.**

# *Tensor ICA*




- ✓It is recommended to use this approach for data where the stimulus paradigm is consistent between session/subjects. Tensor-ICA assumes that the temporal response pattern is the same across the population and provides a single decomposition for all original data sets.

# *Practical*

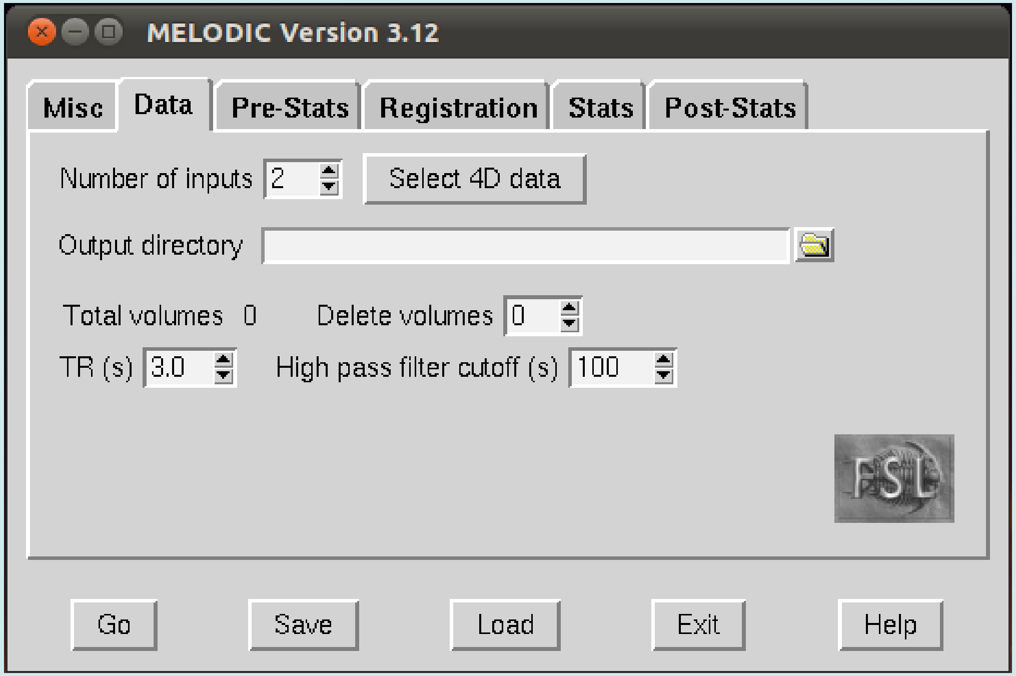
**FSL**

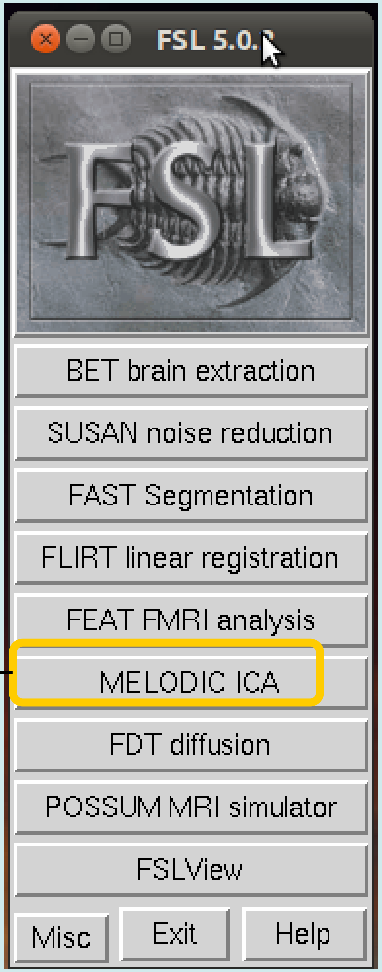
**MELODIC**

**Multivariate Exploratory Linear Optimized Decomposition into Independent Components**

# Practical-set

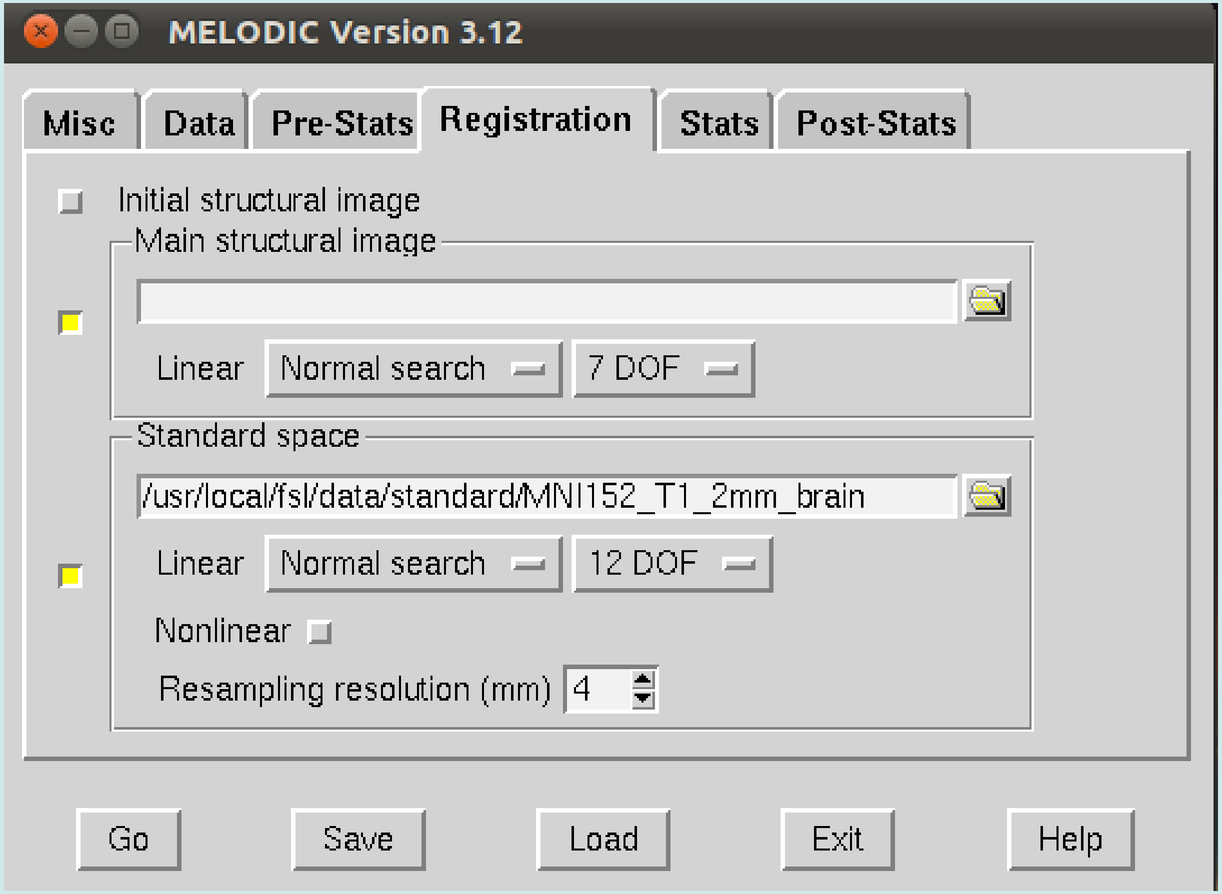
- To call the MELODIC GUI, type **Melodic** in a terminal.



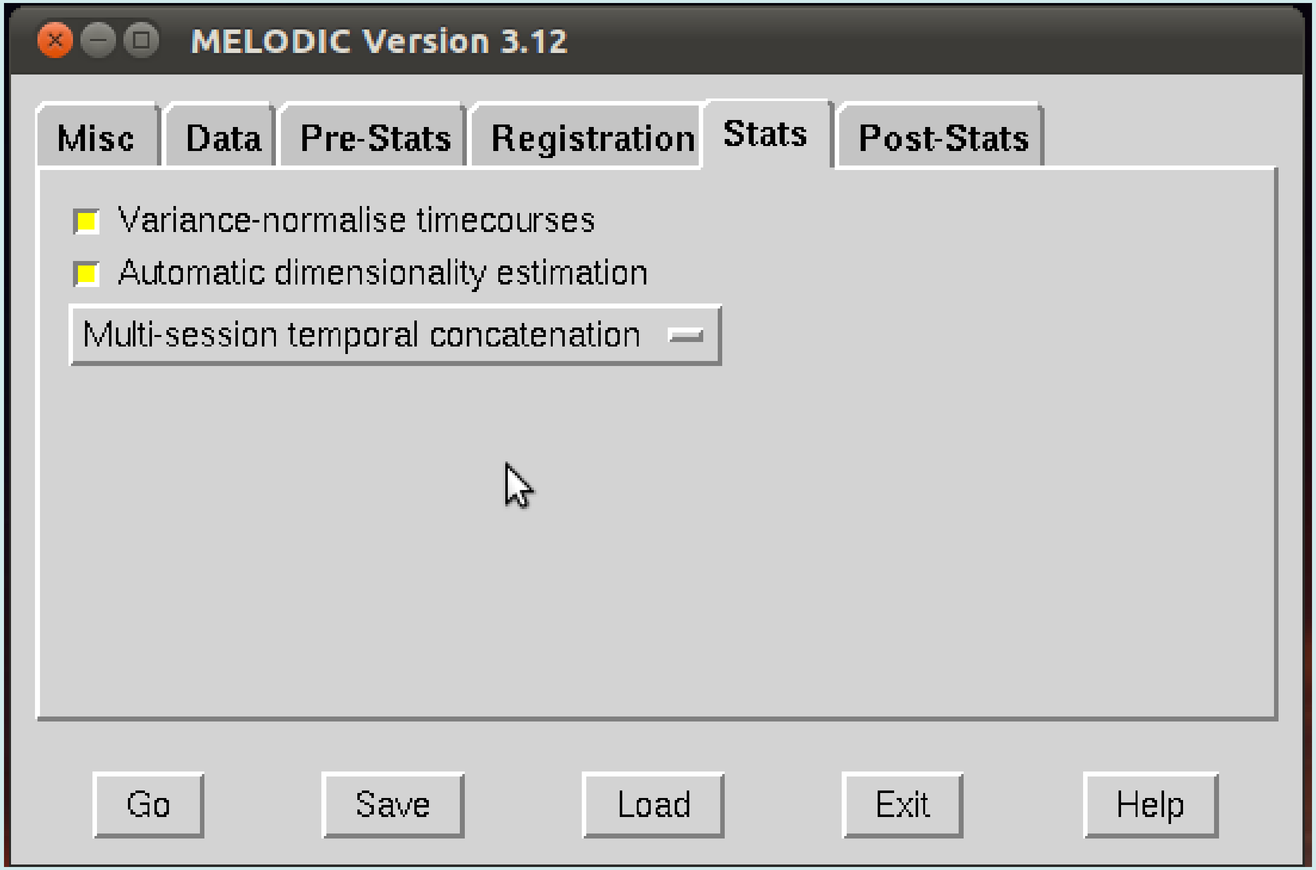




Press the **MELODIC** button

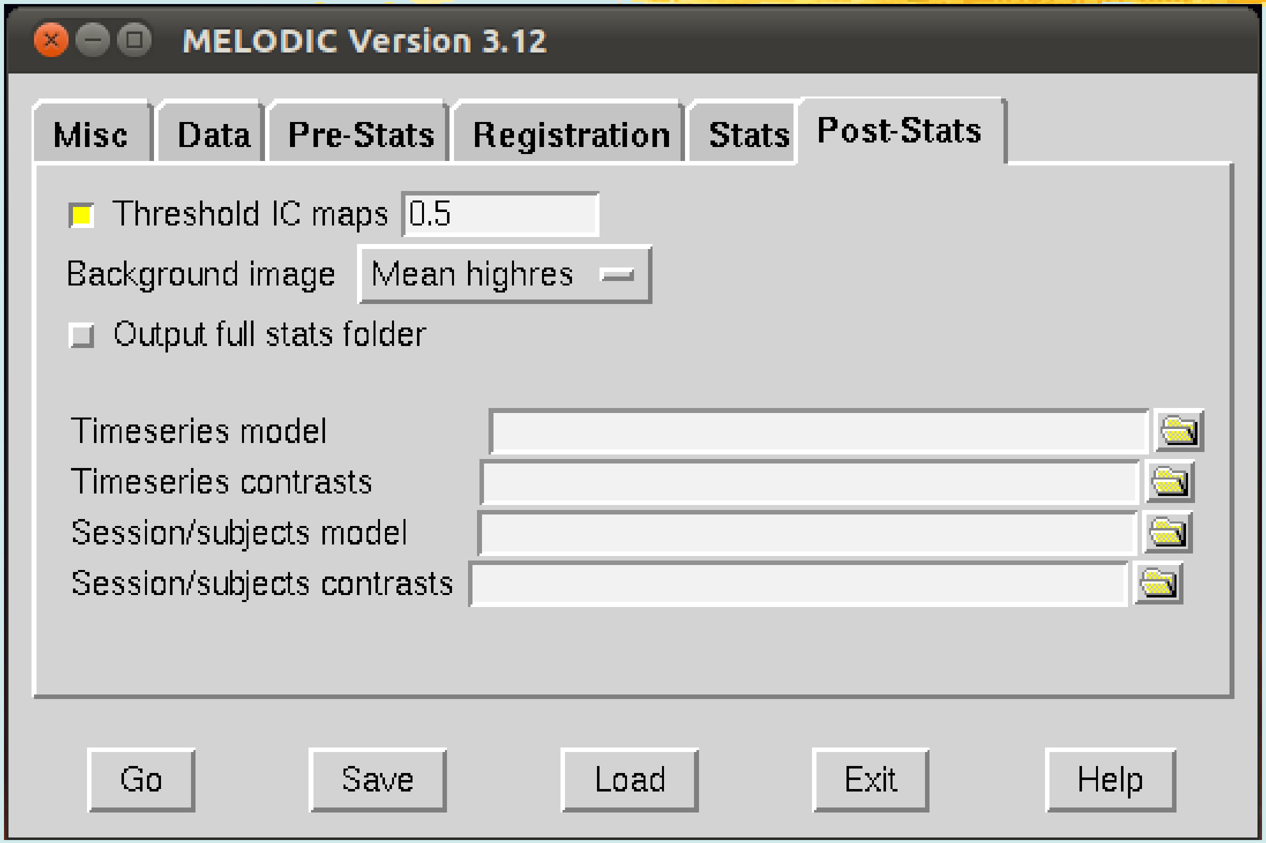
# Practical-set



# *Practical-set*



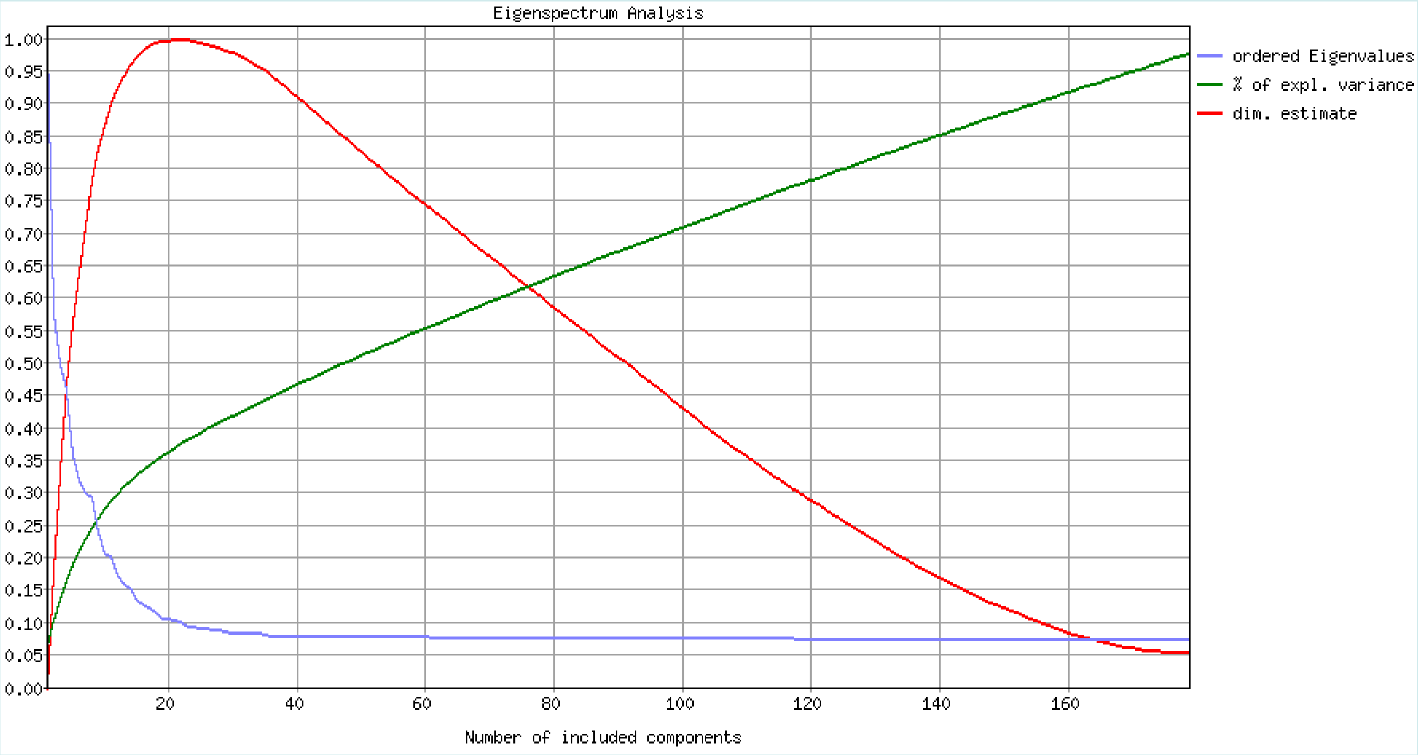
# *Practical-set*




**If a temporal design was specified in the Post-Stats section then the time series plot will also contain a plot of the total model fit**

# Practical-output

## # ICs Estimation



# *Practical-output*

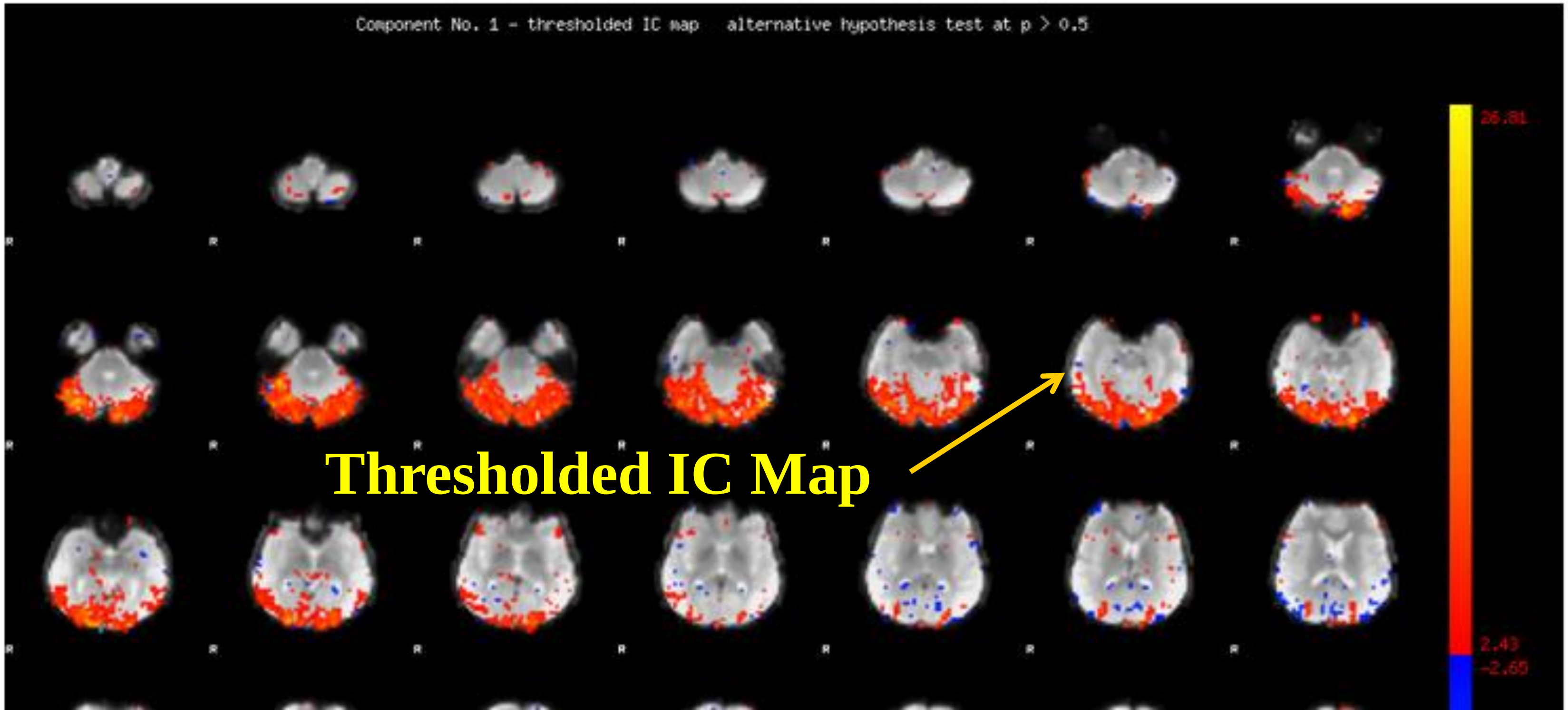
Main - Components: 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22

≤ - ≥

**MELODIC Component 1**

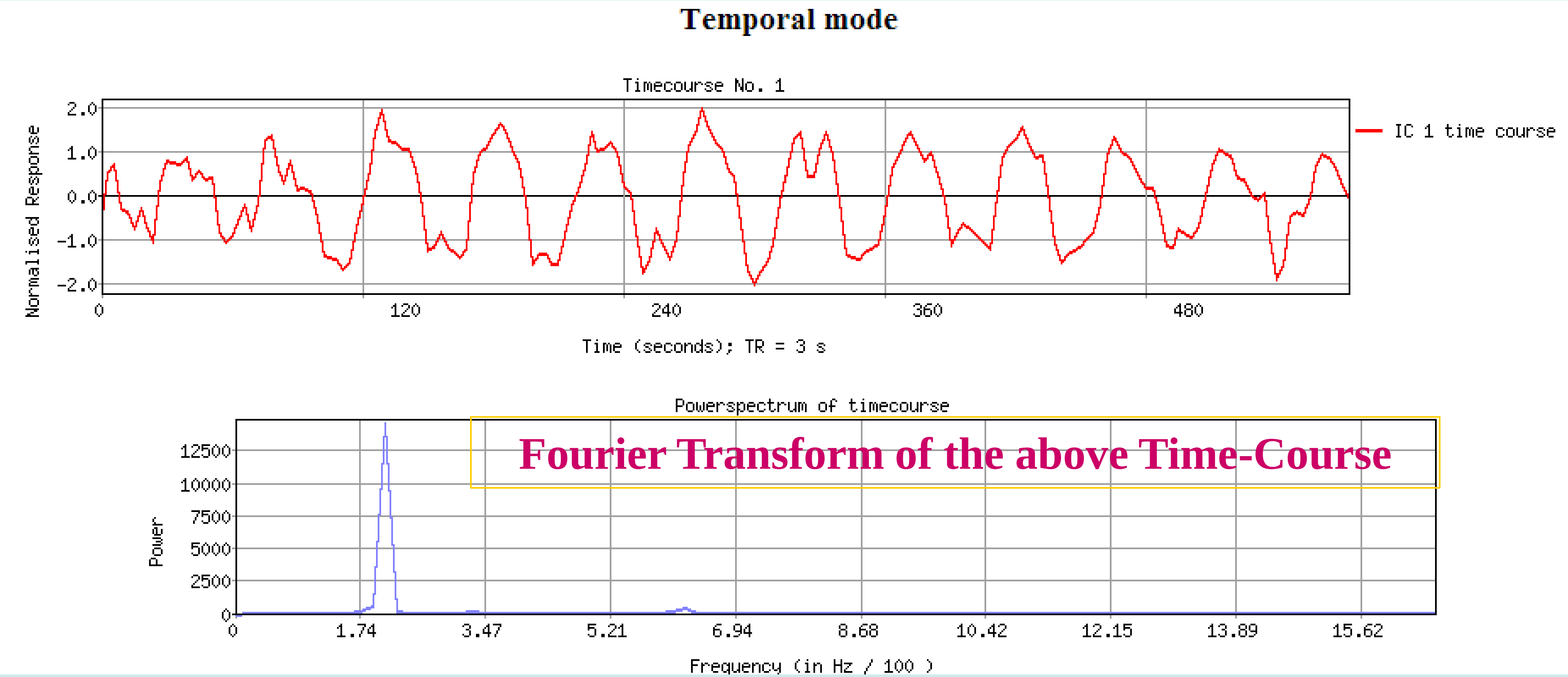
8.72 % of explained variance;   3.28 % of total variance

**22 Components**



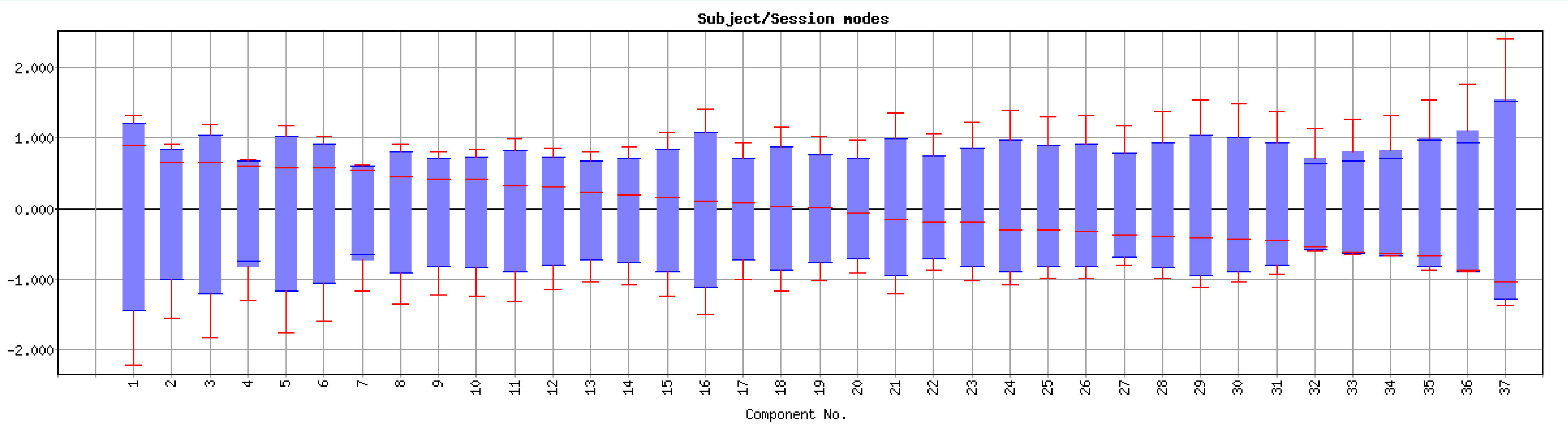
# *Practical-output*

✓**For each component the final mixing matrix melodic_mix contains the temporal response of all different data sets concatenated into a single column vector**



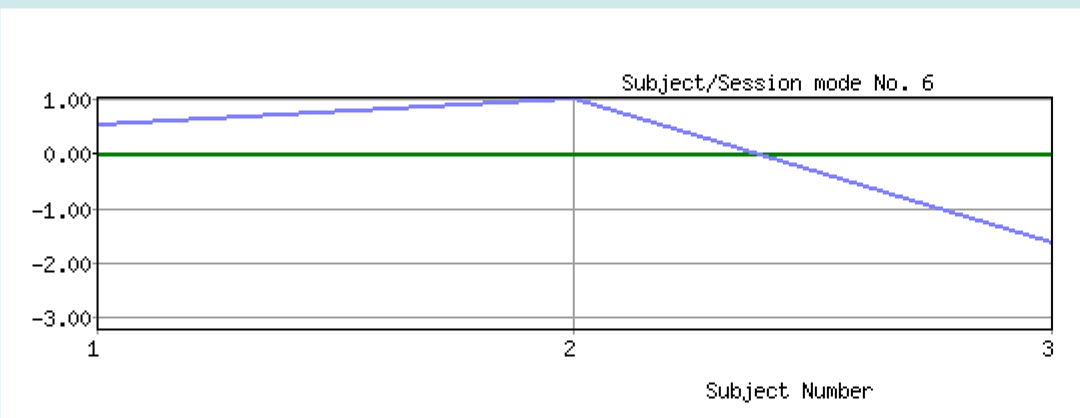
# *Practical-output*

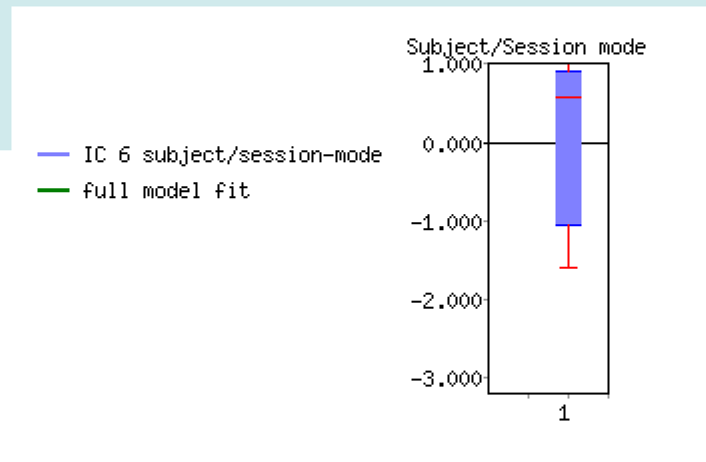
**The TICA Subject/Session modes plot contains a series of boxplots, one per estimated component. The boxplots themselves show the distribution of the relative effect sizes per subject for a given component. The components are ordered in decreasing order of median effect size.**



# *Practical-output*

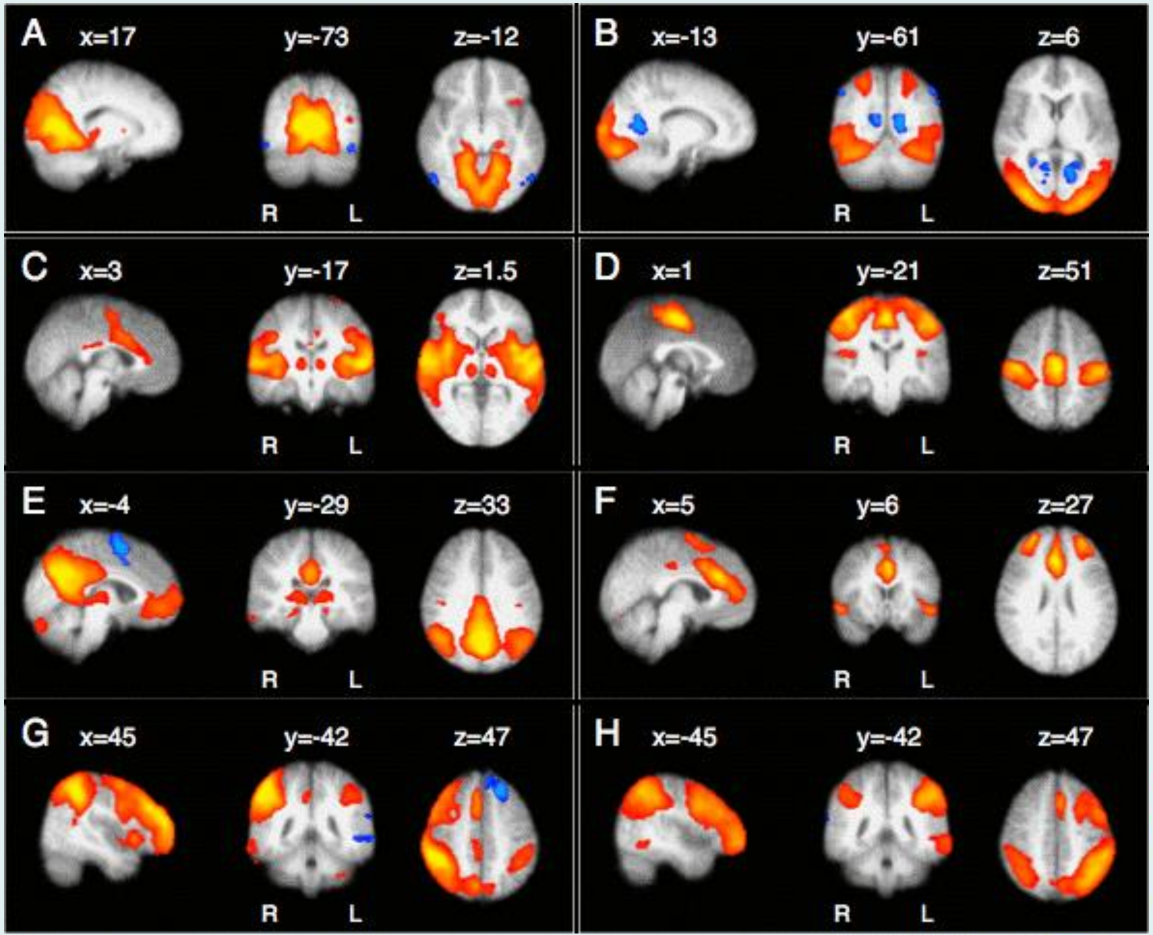
**Each component has three main parts: spatial map, time course and subject/session effect sizes.**



# ICA on resting/null data

- Group study
- (10 subjects under rest): consistent resting fluctuations
- medial (A) visual cortex
- lateral (B) visual cortex
- auditory (C)
- sensori-motor (D)
- visuo-spatial system (E)
- executive control (F)
- dorsal visual stream (G,H)



N E U R O I M A G I N G & A N A L Y S I S T E A M

# *Thanks for Your Attention*