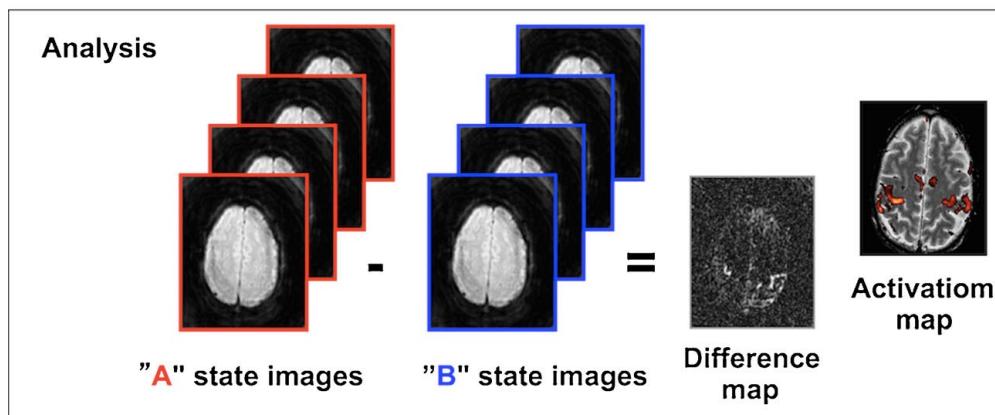
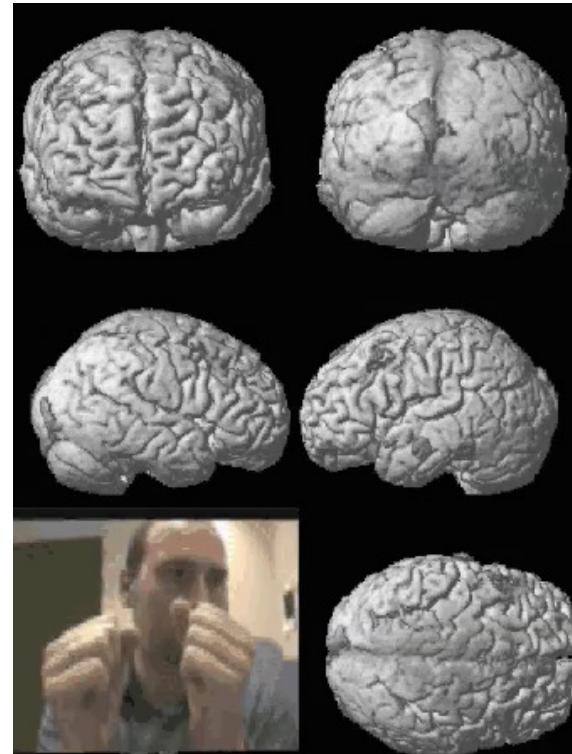
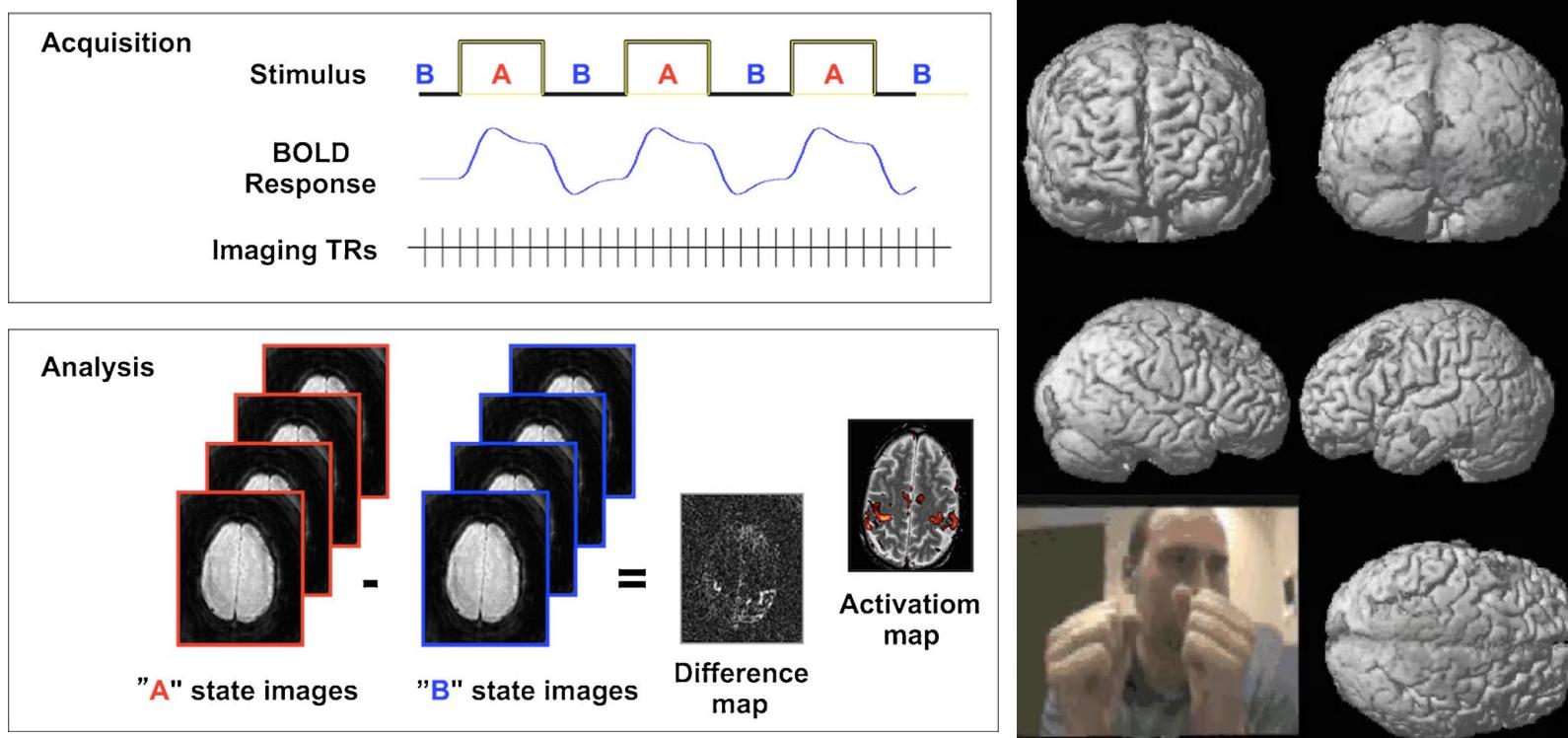


Functional MRI

Dr. Peter Van Schuerbeek

From class 1

fMRI experiment



Task paradigm: Audiovisual Valence Congruence

What you see has the same valence (emotion) as what you hear (congruent)

Versus

What you see has the opposite valence (emotion) as what you hear (incongruent)
for positive versus negative stimuli



2x2 design: (congruent - incongruent) x (positive - negative)

Visual stimuli (video) + Auditory stimuli (music)

Stimulation as individual events (3s long)

4 conditions:

1. Visual positive + Auditory positive
2. Visual positive + Auditory Negative
3. Visual negative + Auditory positive
4. Visual negative + Auditory negative

Congruent trials

Incongruent trials

12 trials / run / condition -> 48 trials / run

8 catch trials (no stimulus, but responses recorded -> attention test)

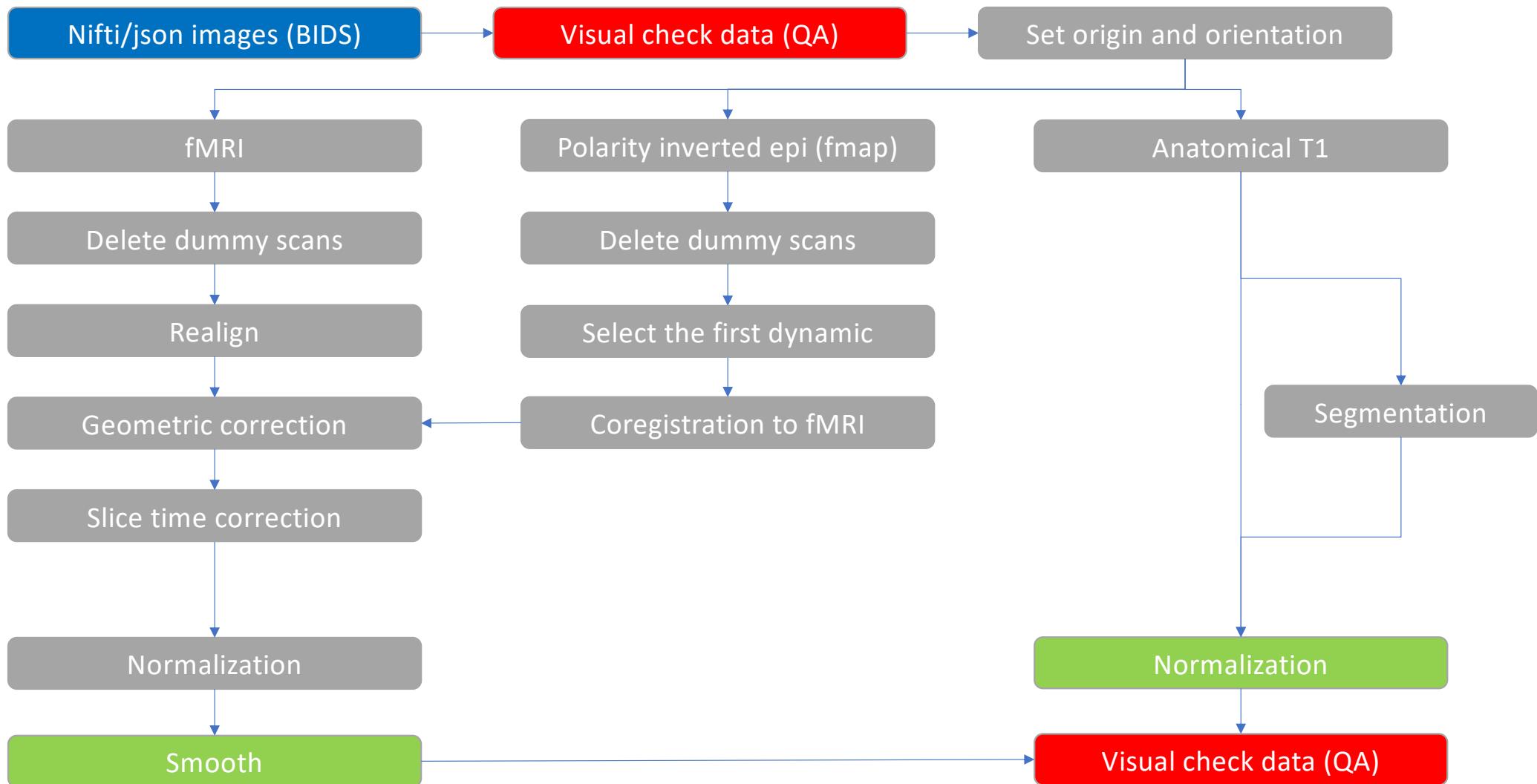
20 subjects (all healthy adult volunteers)



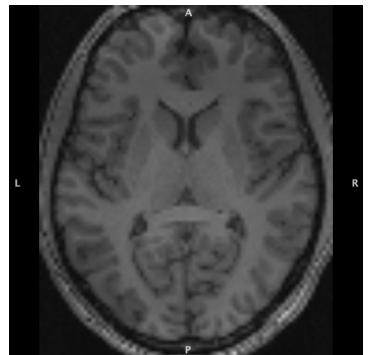
Positive valence



Negative valence



The preprocessing results



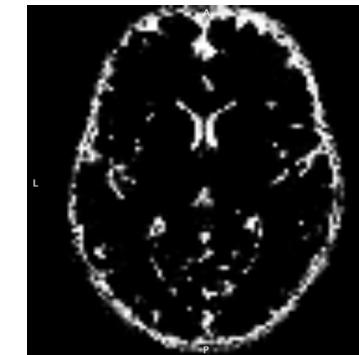
wosub-01_T1w



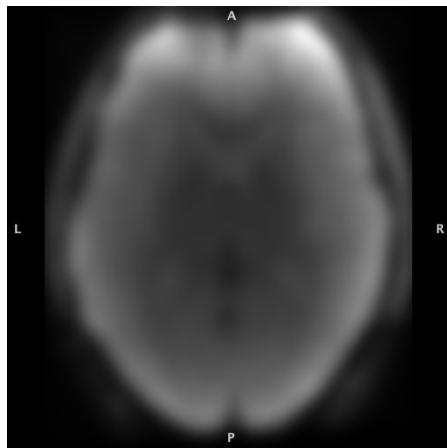
wc1osub-01_T1w



wc2osub-01_T1w



wc3osub-01_T1w



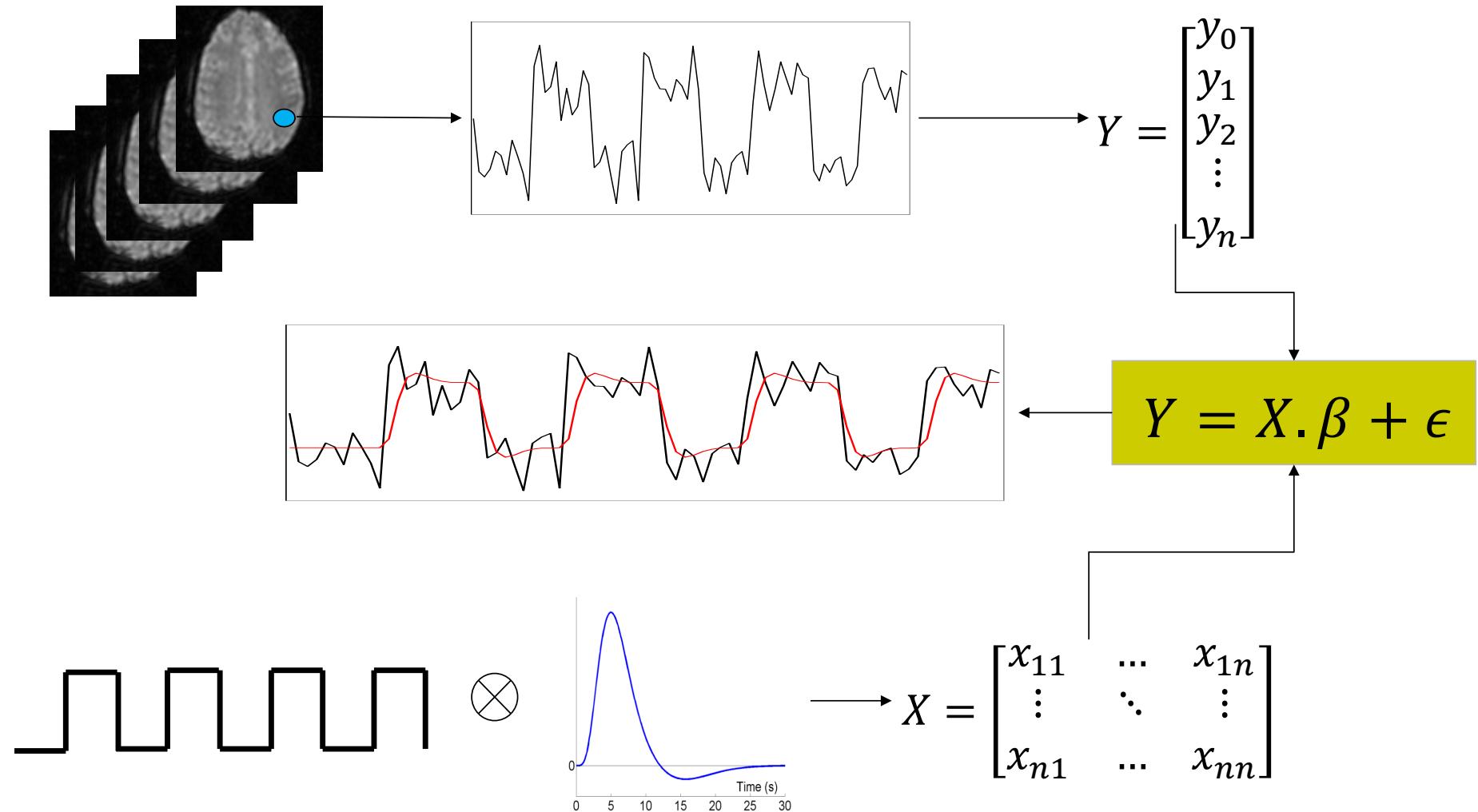
4d_swuraosub-01_task-affect-run-1_bold.nii

	Trans x	Trans y	Trans z	Rot x	Rot y	Rot z
rp_aosub-01_task-affect_run-1_bold_00001.txt ~	0.000000e+00	-4.7340067e-16	0.000000e+00	0.000000e+00	0.000000e+00	1.2588970e-17
	1.7513722e-02	-6.7856195e-03	6.0396937e-03	4.4362332e-04	-1.0208815e-04	3.4466021e-04
	4.8461260e-02	-3.7386199e-02	6.2362634e-02	1.4502508e-03	-4.0640802e-04	9.5685461e-04
	6.9180189e-02	-3.9871672e-02	6.4310601e-02	1.3979599e-03	-3.5460454e-04	1.4147925e-03
	5.20911583e-02	3.2066219e-02	7.7718133e-02	4.3498257e-04	-9.9844301e-05	8.9267421e-04
	5.7427248e-02	1.6258788e-02	7.6479140e-02	7.0890674e-04	1.6059046e-04	7.9906206e-04
	8.6317280e-02	-3.5120548e-02	7.2283943e-02	1.3903804e-03	-2.0507305e-05	1.3339979e-03
	1.1731055e-01	-3.0335500e-02	6.3979068e-02	1.7311331e-03	-1.9270127e-04	1.7468015e-03
	8.0937031e-02	5.0152872e-02	9.1561197e-02	6.9045654e-04	2.2436062e-04	1.1304025e-03
	6.3606665e-02	3.5731932e-02	7.7646949e-02	5.5901698e-04	1.8457663e-04	8.6840523e-04
	5.2421874e-02	-1.0953733e-02	1.0717907e-01	9.7029852e-04	-6.1821191e-05	1.0495726e-03
	7.4589833e-02	-4.0247471e-04	1.2329963e-01	7.9186069e-04	1.8275389e-05	1.1582123e-03
	1.0715947e-01	-5.0387869e-02	1.0422207e-01	7.68870931e-04	-7.4170782e-05	1.4709182e-03
	1.4121567e-01	-7.7419048e-02	1.3071721e-01	1.4501688e-03	-3.1955366e-04	1.8303812e-03
	1.7744815e-01	-6.8792732e-02	5.1087038e-03	8.2285381e-04	-7.5366066e-05	2.0172927e-03
	1.2917189e-01	1.7510493e-02	5.8319505e-02	-2.8877083e-06	-1.3831180e-05	1.5282397e-03
	1.3561558e-01	3.8867613e-03	8.8903376e-02	6.1257401e-04	-2.0483346e-04	1.6858282e-03
	1.5266451e-01	1.3644125e-02	1.3092410e-02	1.4152692e-04	3.2651009e-03	2.3670061e-03

Realignment parameters

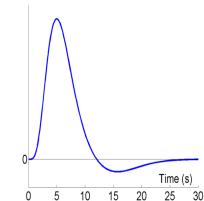
First level analysis

The general linear model (GLM)

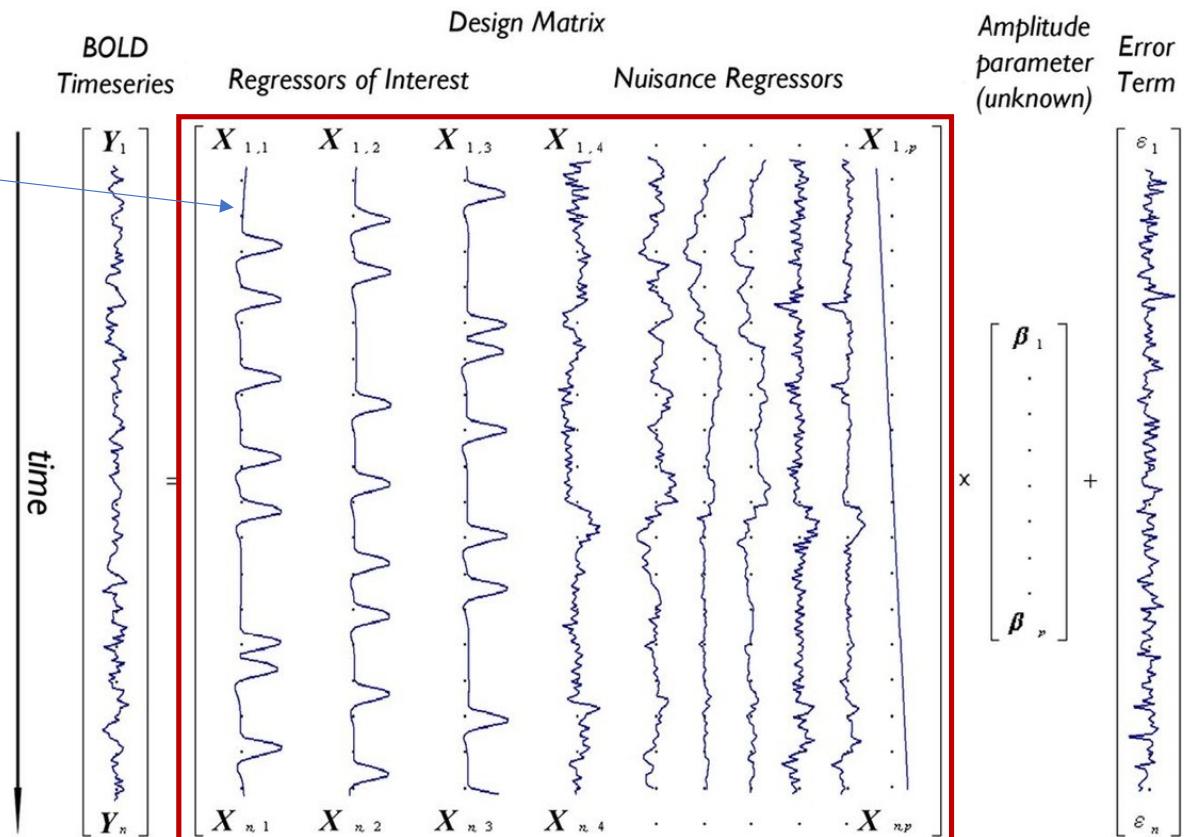


The design matrix

Onsets and duration of trials



$$Y = X \cdot \beta + \epsilon$$

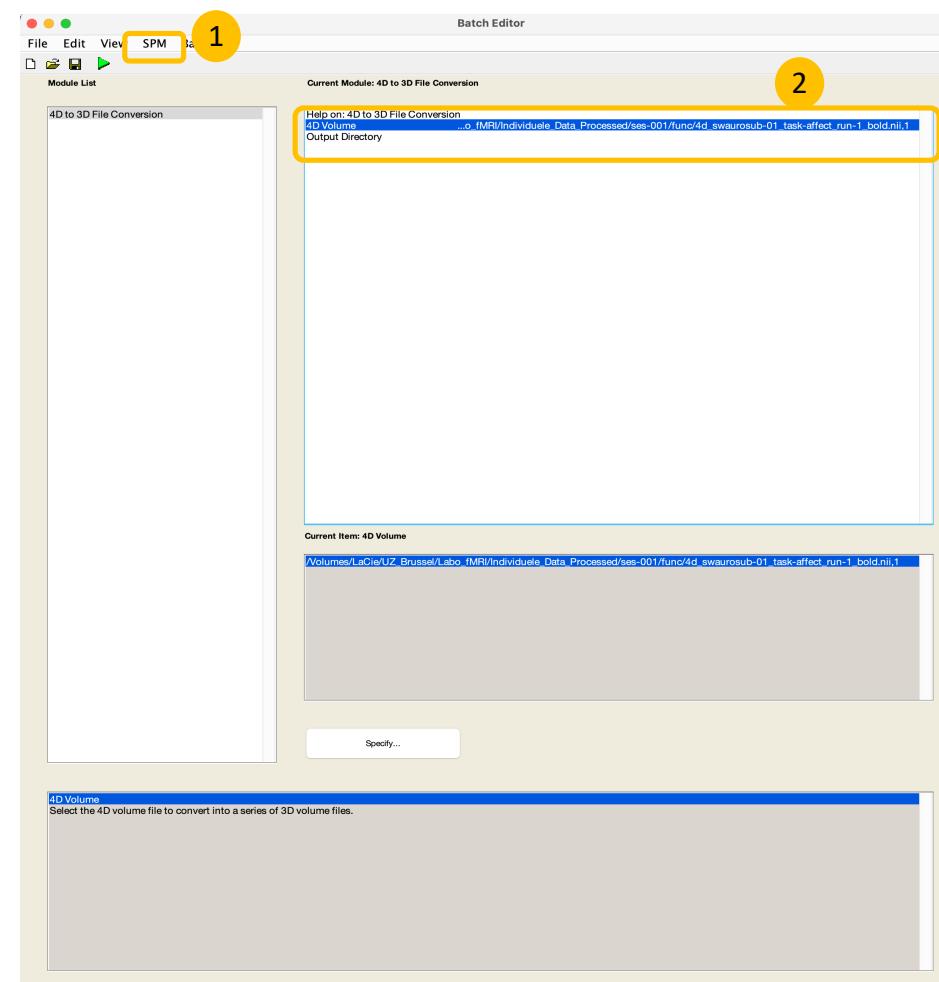


All columns should be linearly independent!

Rest blocks are not modelled explicitly

Step 1: Loading the preprocessed data

1. In the top Menus: select SPM -> Util -> 4D to 3D File Conversion
2. 4D volume (using Specify): select the preprocessed fMRI data (4d_swauro... .nii file)
3. Make a folder to save the SPM processing results



Step 2: Model specification

1. In the top Menus: select SPM -> Stats -> fMRI model specification
2. Directory (using Specify): select the output directory
3. Units for design: Seconds
4. Interscan interval: 1 (=TR)
5. Microtime resolution: 10 (=number of slice packages)
6. Microtime onset: 1 (= first slice)
7. Subject/Session -> Scans (using Dependency): 4D to 3D File Conversion: Series of 3D Volumes
8. Conditions (once for each of the 5 conditions)
9. Condition -> Name: give the name of the condition
10. Condition -> Onsets: copy the onsets for that condition from the events.tsv file
11. Condition -> Durations: copy the durations for that condition from the events.tsv file
12. Multiple regressors (using Specify): select the rp_sub..._bold_00001.txt file

Batch Editor

File Edit View **SPM** 1

Module List

4D to 3D File Conversion
fMRI model specification DEP

Help on: fMRI model specification
Directory .../l/Individuel_Data_Processed/ses-001/SPM_processing
Timing parameters
.. Units for design
.. Interscan interval
.. Microtime resolution
.. Microtime onset
Data & Design
.. Subject/Session
.. Scans
.. Conditions
.. Condition
.... Name
.... Onsets
.... Durations
.... Time Modulation
.... Parametric Modulations
.... Orthogonalise modulations
.... Condition
.... Name
.... Onsets

DEP 4D to 3D File Conversion: Series of 3D Volumes

Seconds
1
10
1

12x1 double
12x1 double
No Time Modulation

Yes

1
2
12x1 double

Current Item: Multiple regressors
/Volumes/l/aCie/17_Brussel/lab_fMRI/Individuel_Data_Processed/ses-001/func/rp_sub..._bold_00001.txt

Specify...

A Z Sort & Filter

Sort A to Z
Sort Z to A
Custom Sort...

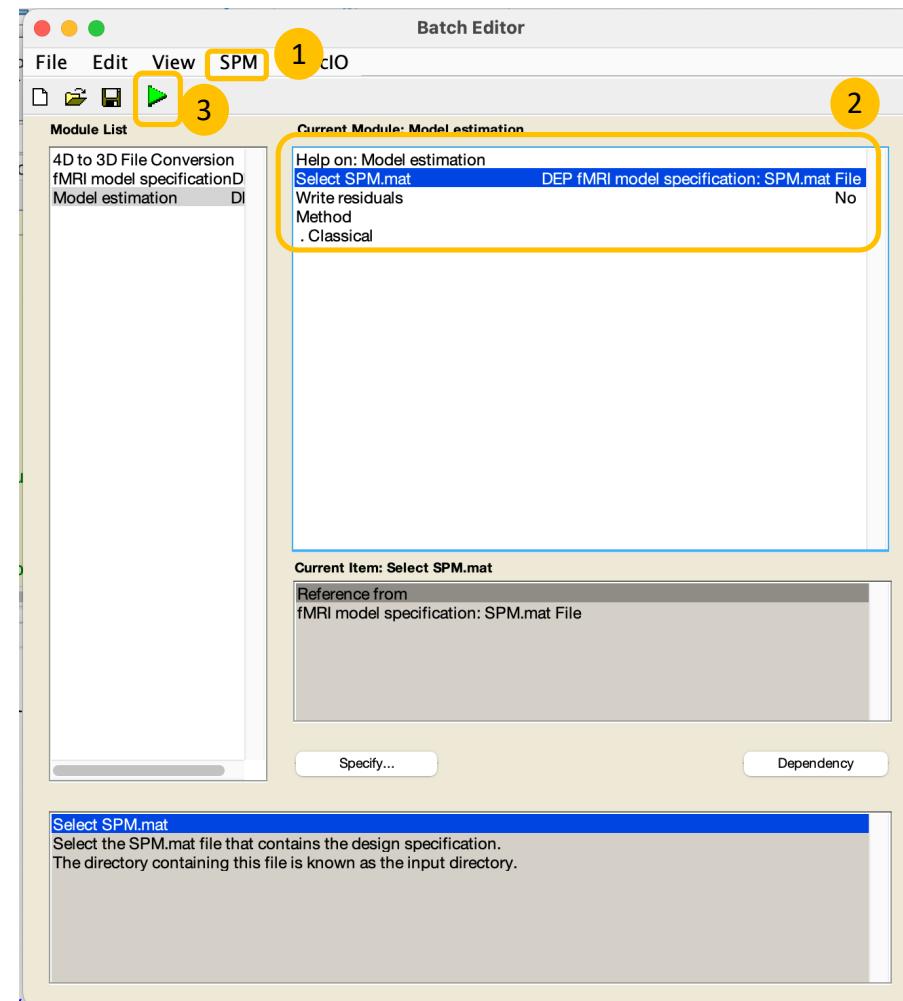
Filter
Clear
Reapply

sub-01_task-affect_run-1_events

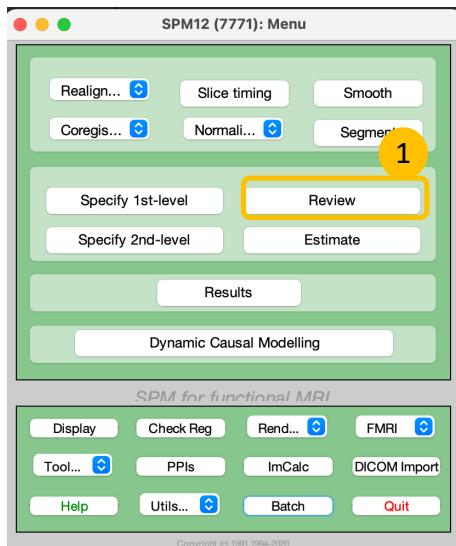
onset	duration	trial type
17.011	3	1
319.011	3	1
147.008	3	1
157.015	3	1
217.01	3	1
267.014	3	1
319.019	3	1
369.023	3	1
400.016	3	1
469.015	3	1
500.028	3	1
529.026	3	1
7.003	3	2
87.013	3	2
137	3	2
177.013	3	2
200.002	3	2
277.005	3	2
379.014	3	2
389.021	3	2
409.02	3	2
489.029	3	2
519.018	3	2
569.022	3	2

Step 3: Model estimation

1. In the top Menus: select SPM -> Stats -> Model estimation
2. Select SPM.mat (using Dependency): fMRI model specification: SPM.mat File
3. Run batch



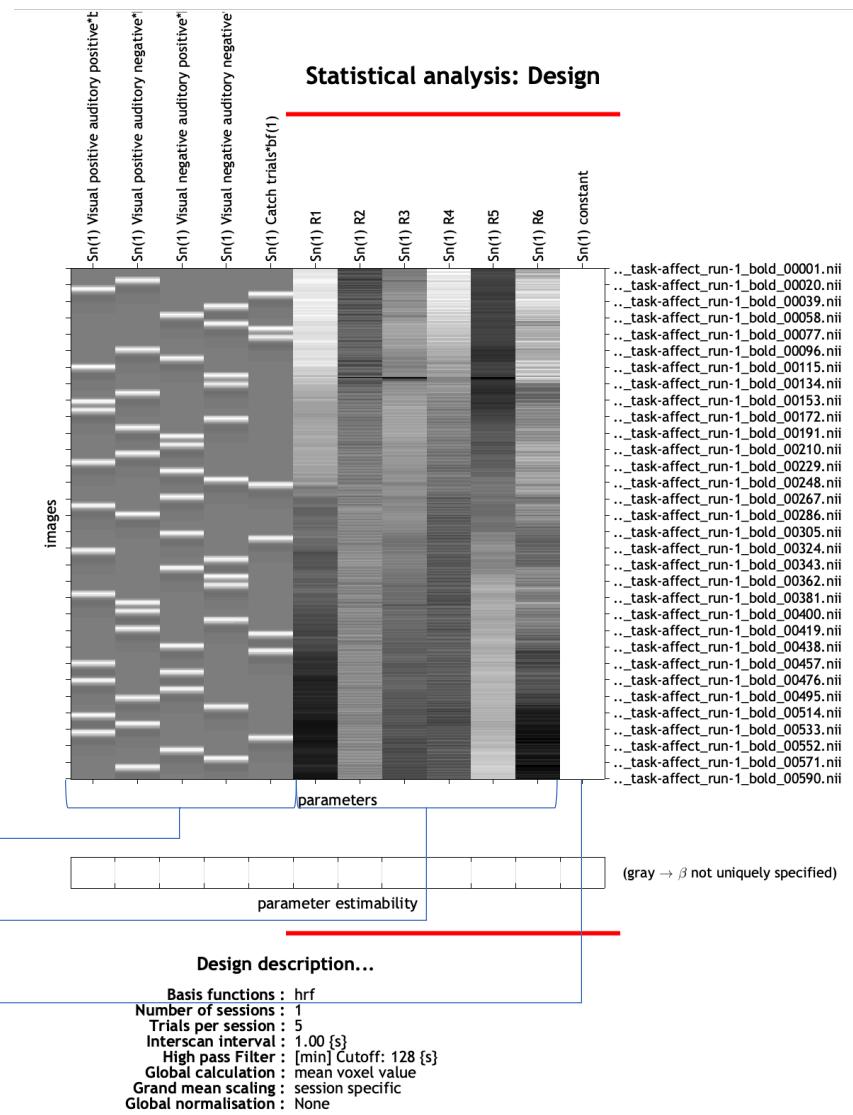
Step 3: Model review



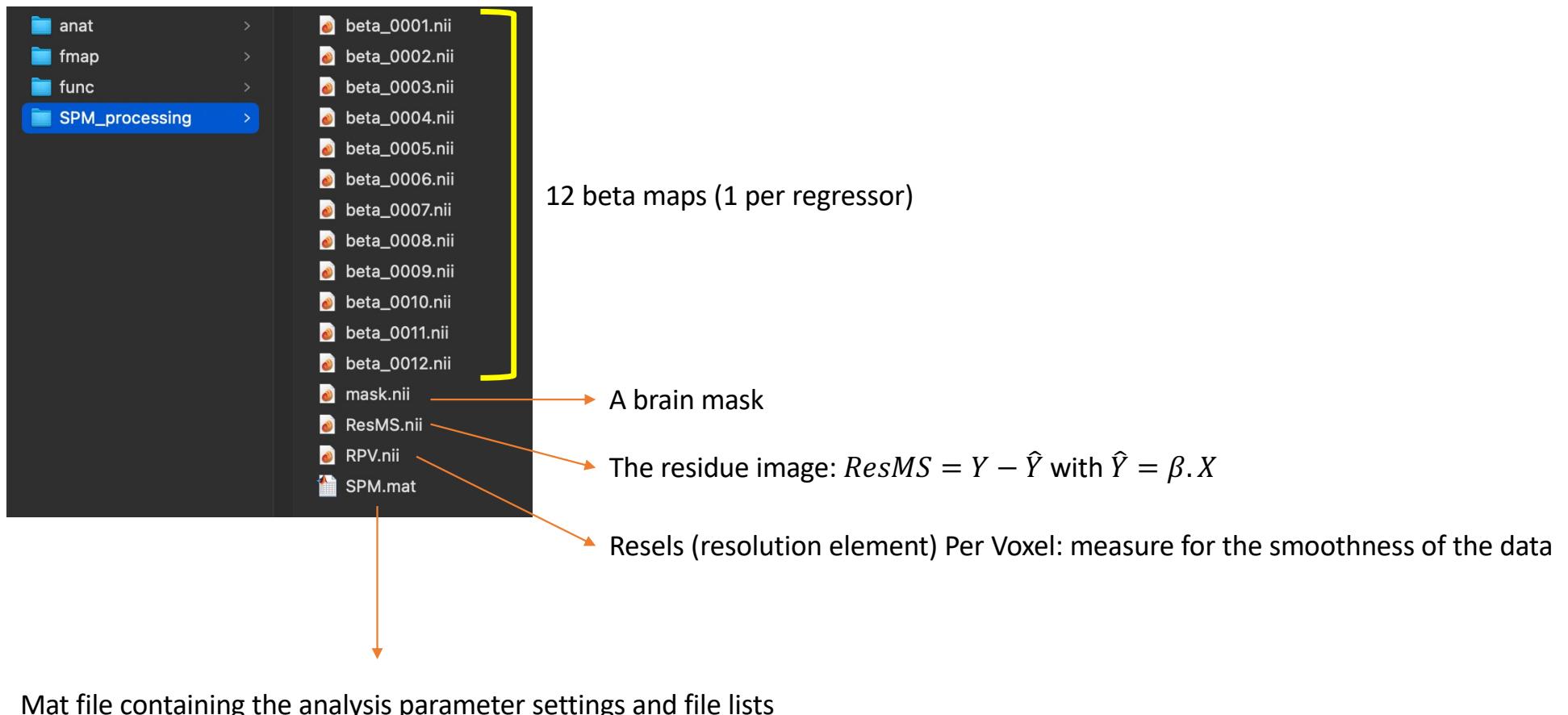
1. In the Menu window: click Review
2. Select the SPM.mat file in the output directory

12 model regressors

- 5 task regressors
- 6 motion regressors
- 1 constant



The model estimation output



The first level analysis results

The β -values

$$y_1 = x_1\beta + \varepsilon_1$$

$$y_2 = x_2\beta + \varepsilon_2$$

$$y_3 = x_3\beta + \varepsilon_3$$

 \vdots

$$y_n = x_n\beta + \varepsilon_n$$

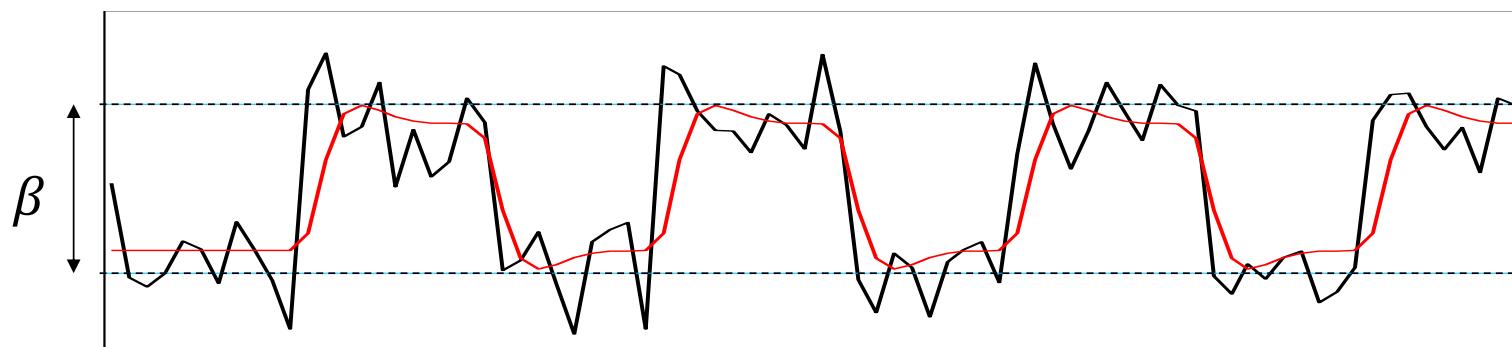


$$\begin{bmatrix} y_1 \\ y_2 \\ y_3 \\ \vdots \\ y_n \end{bmatrix} = \begin{bmatrix} x_1 \\ x_2 \\ x_3 \\ \vdots \\ x_n \end{bmatrix} [\beta] + \begin{bmatrix} \varepsilon_1 \\ \varepsilon_2 \\ \varepsilon_3 \\ \vdots \\ \varepsilon_n \end{bmatrix}$$

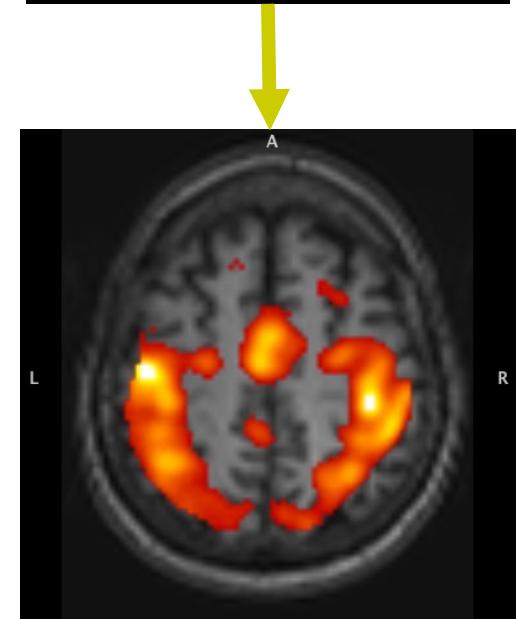
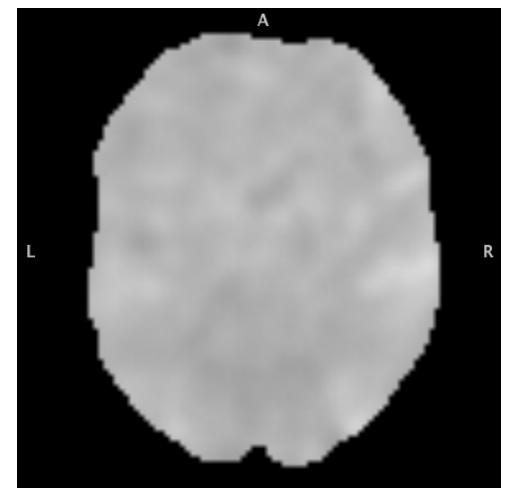


$$\mathbf{Y} = \mathbf{X} \beta + \boldsymbol{\varepsilon}$$

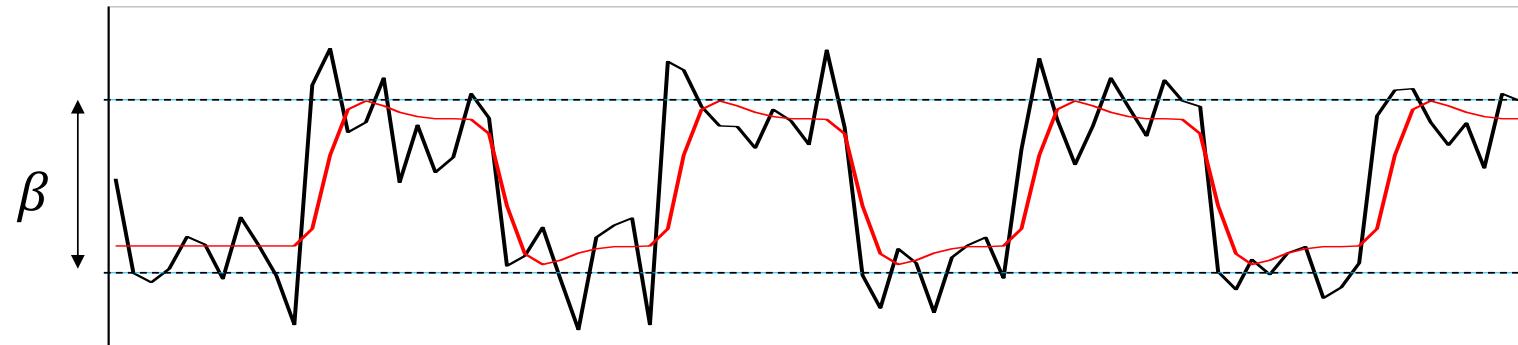
$$\hat{\beta} = (X^T X)^{-1} X^T Y$$



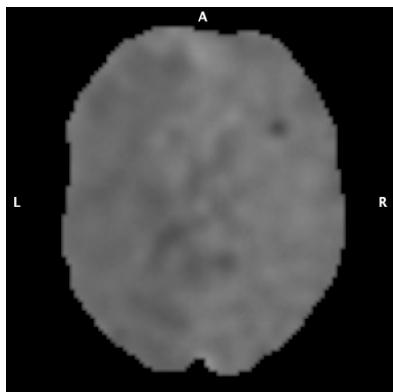
β = measure for the amount of signal variability explained by the regressor



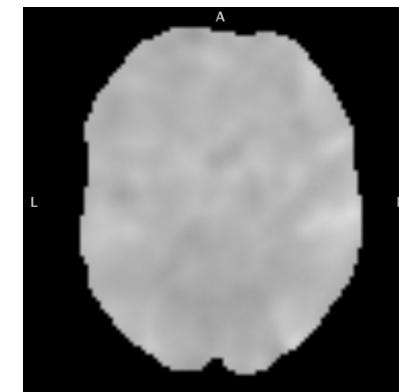
Statistical tests



$\beta = 0?$ -> T-test



$\beta_1 = \beta_2 \Rightarrow \beta_1 - \beta_2 = 0?$
↓
T or F-test

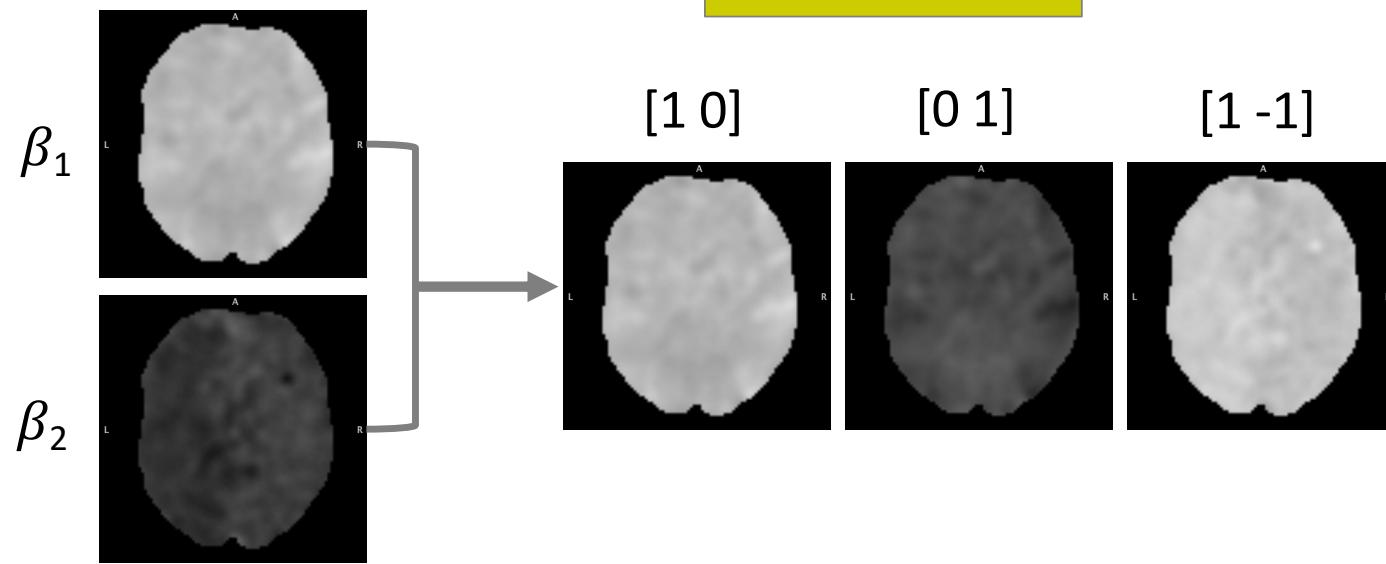


The contrast matrix

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

$$c^T \hat{\beta} = c_1 \cdot \hat{\beta}_1 + c_2 \cdot \hat{\beta}_2 + c_3 \cdot \hat{\beta}_3 \dots$$

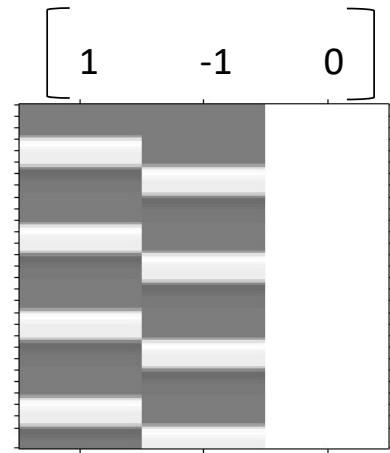
$$c^T \beta = 0$$



T-test

- Null hypothesis $H_0: c^T \hat{\beta} = 0$
- Alternative hypothesis $H_1: c^T \hat{\beta} > 0$

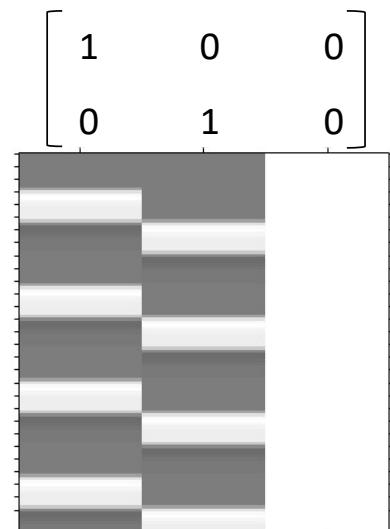
$$t_{df} = \frac{c^T \hat{\beta}}{sd(c^T \hat{\beta})}$$



F-Test

- Null hypothesis $H_0: \beta_1 = \beta_2 = \dots = 0$
- Alternative hypothesis: $H_1:$ existence of at least one $\beta \neq 0$

$$F = \frac{\text{Explained variability}}{\text{Error estimated variance}}$$



Step 4: Contrast estimation

1. In the top Menus: select SPM -> Stats -> Contrast Manager
2. Select SPM.mat (using Specify): fMRI model specification: SPM.mat File
3. Per T-contrast:
 1. Define contrast name
 2. Give the contrast weights (sum positive weights=1; sum negative weights=-1)

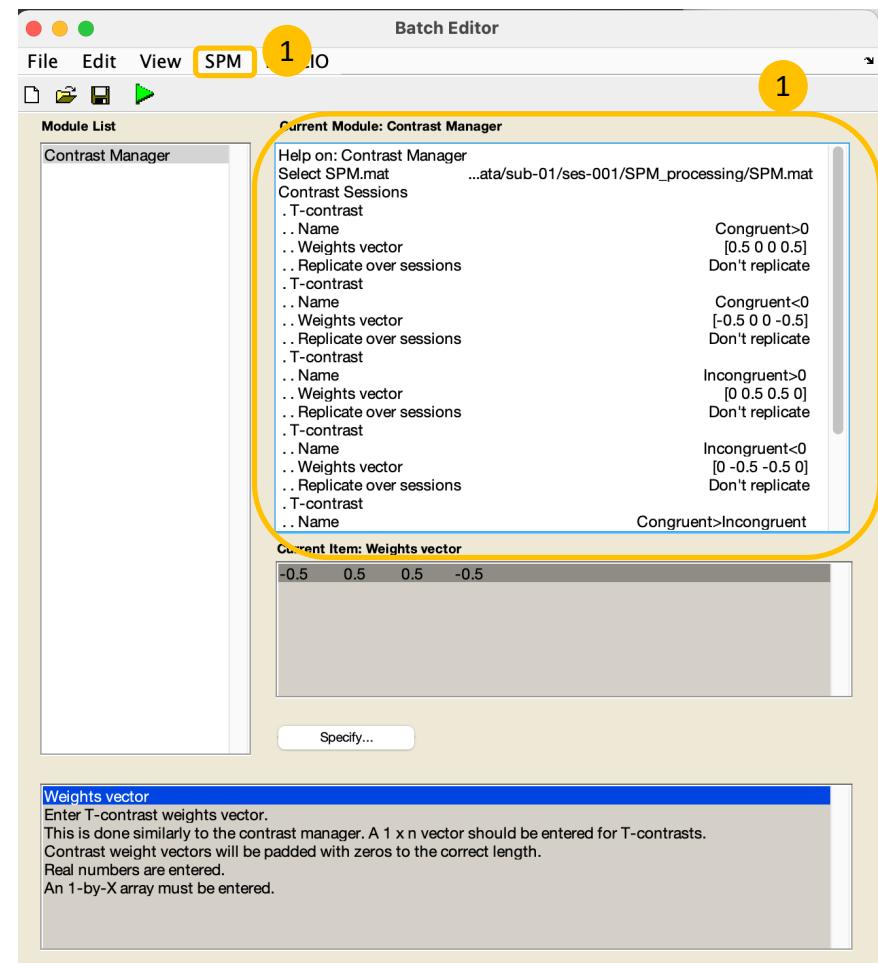
Trial conditions

1. Visual positive, Auditory positive
2. Visual positive, Auditory negative
3. Visual negative, Auditory positive
4. Visual negative, Auditory negative
5. Catch trials

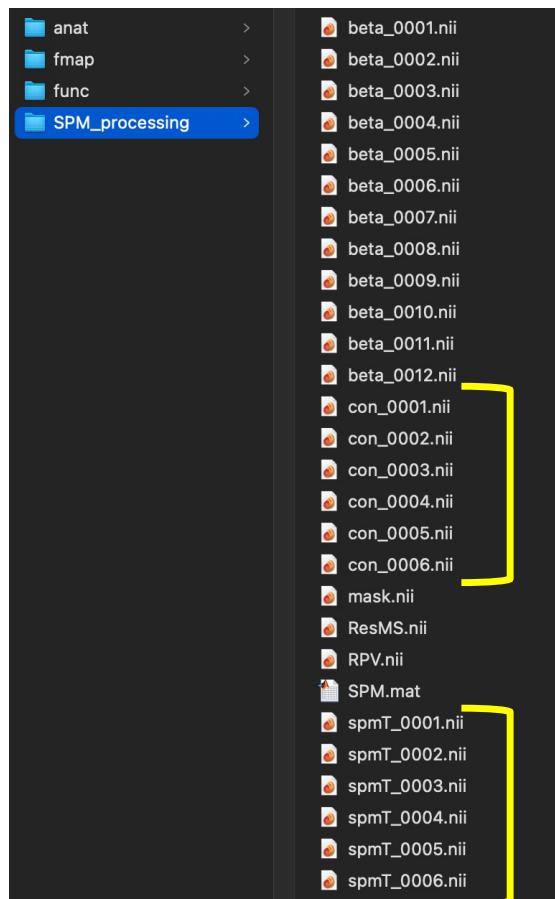
2 task conditions: Congruent and Incongruent

Contrast matrix

1. Activations for congruent (Congruent>0 = (1+4)>0): $c=[0.5 \ 0 \ 0 \ 0.5]$
2. Deactivations for congruent (Congruent<0 = (1+4)<0): $c=[-0.5 \ 0 \ 0 \ -0.5]$
3. Activations for incongruent (Incongruent>0 = (2+3)>0): $c=[0 \ 0.5 \ 0.5 \ 0]$
4. Deactivations for incongruent (Incongruent<0 = (2+3)<0): $c=[0 \ -0.5 \ -0.5 \ 0]$
5. Congruent>Incongruent ((1+4)-(2+3)>0): $c=[0.5 \ -0.5 \ -0.5 \ 0.5]$
6. Congruent<Incongruent ((2+3)-(1+4)>0): $c=[-0.5 \ 0.5 \ 0.5 \ -0.5]$



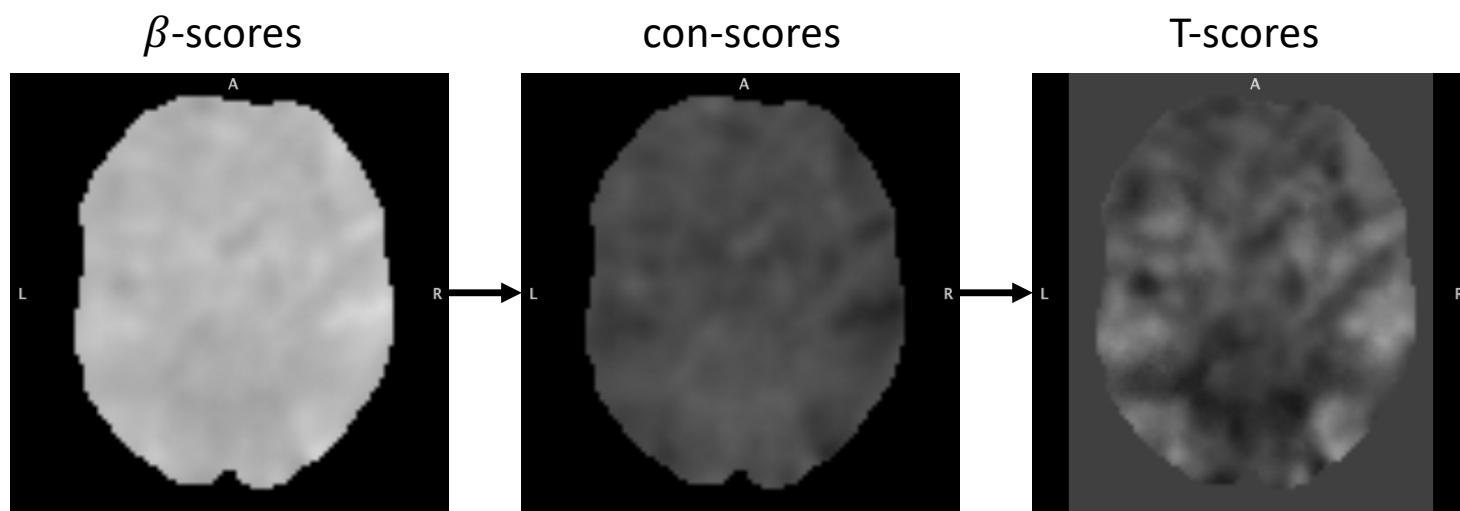
The contrast estimation output



6 contrast maps (the size of the calculated contrast)

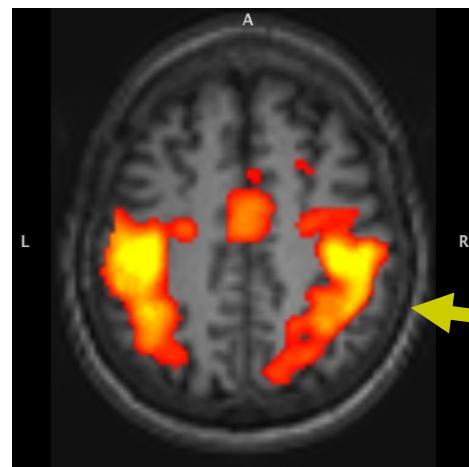
6 T-maps (the t-value of the performed T-test per contrast)

p-values

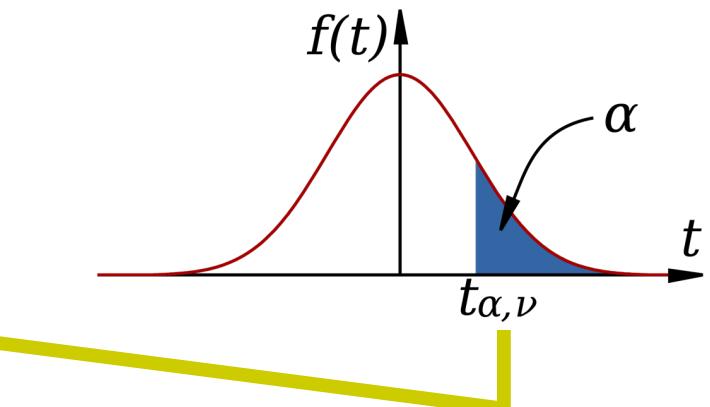


Unknown distribution

**Significant
contrast
map**



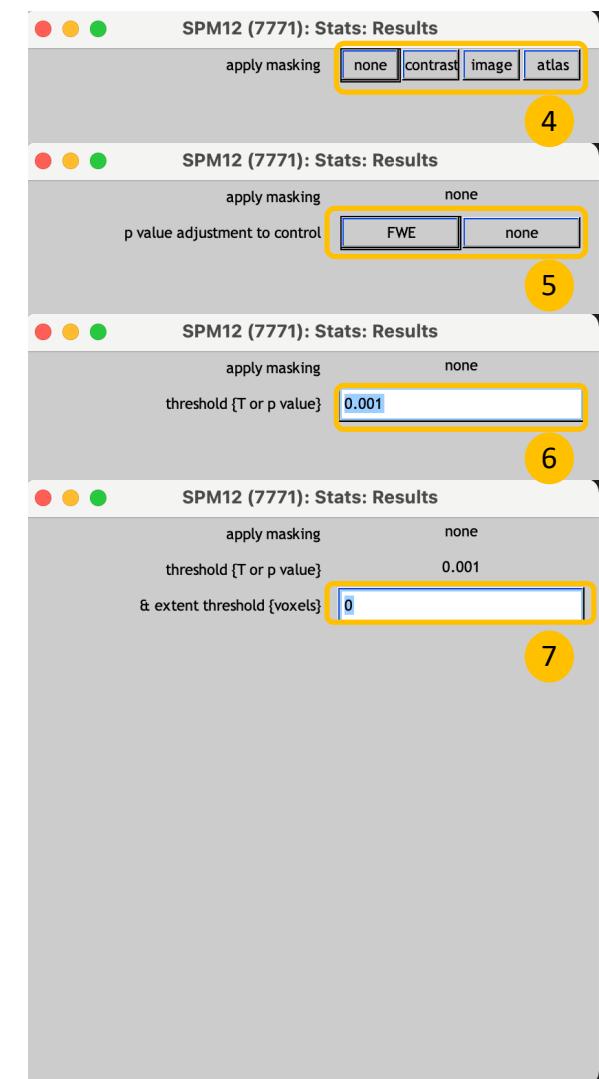
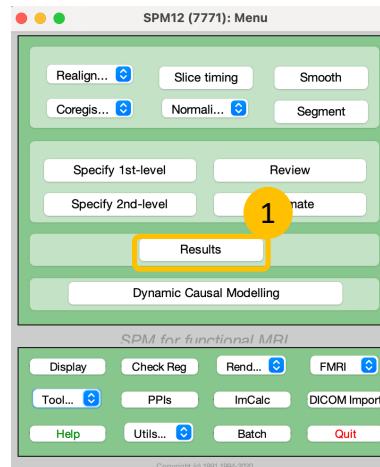
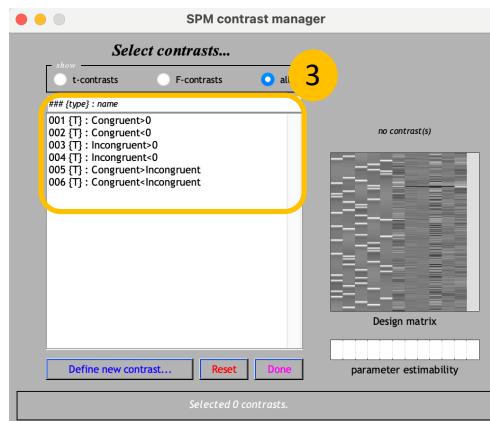
Known distribution



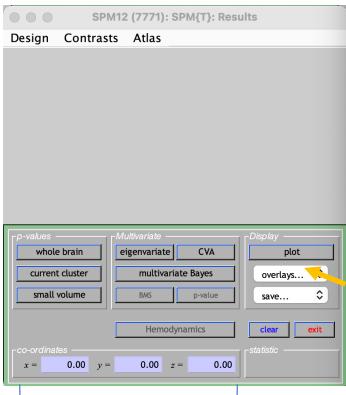
p = chance for the null-hypothesis ($H_0: c^T \beta = 0$) to be true

A look at the results

1. In the Menu window: click Results
2. Select the SPM.mat file from the analysis
3. In the contrast manager: select the contrast (e.g. 001 (T): Congruent>0)
4. Apply masking: select none
5. P value adjustment to control: select none
6. Threshold {T or p value}: 0.001
7. & extent threshold {voxels}: 0



Glass brain showing the clusters
(projection from the left, top and back)



Position of the
selected voxel
(MNI coordinates)

Select anatomical
underlay:
Overlays -> Sections
Select anat/wosub-
01_T1w.nii

Results table

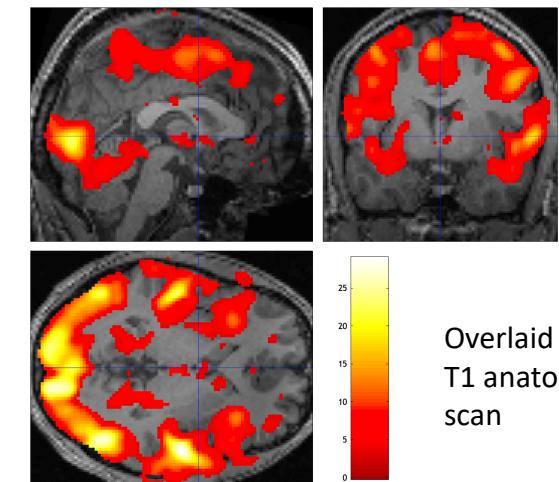
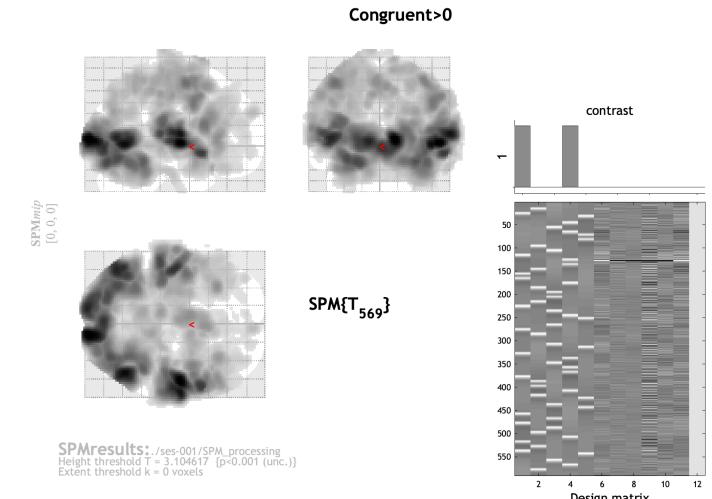
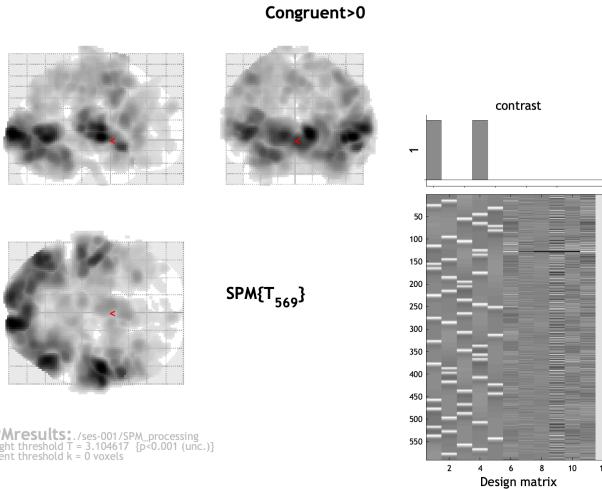
Statistics: p-values adjusted for search volume

set-level	cluster-level			peak-level			Z_{v}	P_{peak}	mm mm mm
	p	c	$P_{\text{FWE-corr}}$	$q_{\text{FDR-corr}}$	k_E	P_{uncorr}			
0.216	16	0.000	0.000	85255	0.000	0.000	0.000	28.96	inf
							0.000	0.000	12 -98 4
							0.000	0.000	28.67 inf
							0.000	0.000	inf 0.000
							0.000	0.000	48 -64 0
0.006	0.004	320	0.000	0.000	0.000	0.000	0.120	0.013	4.54 4.50 0.000
							0.120	0.013	6 58 28
							0.120	0.013	-8 52 22
0.009	0.004	289	0.001	0.000	0.000	0.000	0.142	0.013	4.50 4.45 0.000
							0.142	0.013	12 12 -4
							0.142	0.013	4 2 -2
0.210	0.059	119	0.018	0.032	0.003	0.000	0.712	0.110	3.92 3.89 0.000
0.774	0.311	47	0.117	0.072	0.008	0.000	0.712	0.110	4 6.7 0.000
							0.712	0.110	20 56 -6
							0.712	0.110	-28 -8 -44
0.166	0.057	131	0.014	0.166	0.017	0.000	0.166	0.017	4.45 4.41 0.000
							0.166	0.017	6 36 -18
							0.166	0.017	-42 18
0.950	0.527	26	0.235	0.141	0.000	0.000	0.998	0.120	3.83 3.81 0.000
0.950	0.527	23	0.203	0.097	0.000	0.000	0.998	0.120	5 36 2
0.998	0.712	9	0.490	0.929	0.220	0.000	0.998	0.120	-16 12 2
0.998	0.712	9	0.490	0.929	0.220	0.000	0.998	0.120	20 -12 -20
0.994	0.708	13	0.402	0.971	0.294	3.58	0.995	0.436	24 -36 72
1.000	0.848	2	0.370	0.995	0.436	3.44	3.42	0.000	-18 -26 6
0.946	0.709	11	0.442	0.997	0.489	3.41	3.39	0.000	18 16 10
1.000	0.812	4	0.460	1.000	0.553	3.22	3.21	0.001	-70 16 22
0.999	0.729	7	0.547	1.000	0.756	3.22	3.21	0.001	32 -2 -6
1.000	0.848	1	0.488	1.000	0.816	3.19	3.17	0.001	-12 38 -14
1.000	0.848	1	0.488	1.000	0.871	3.16	3.15	0.001	-12 38 22

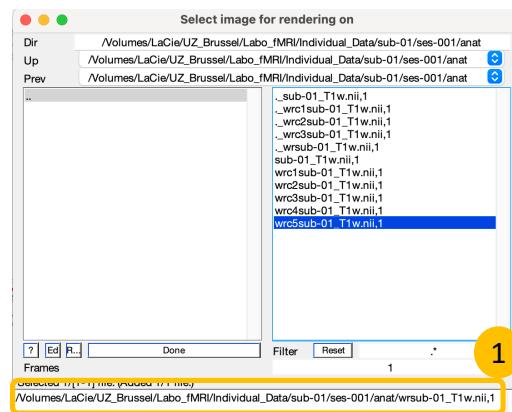
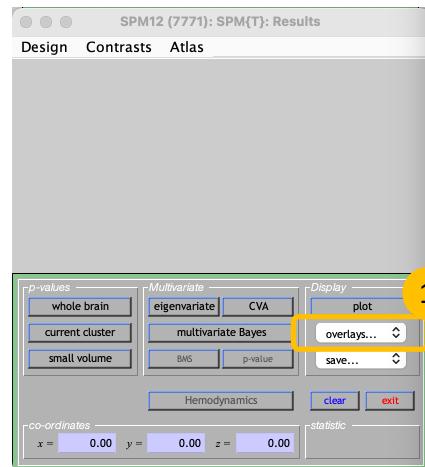
table shows 3 local maxima more than 8.0mm apart

Height threshold: T = 3.10, p = 0.001 (1.000)
Extent threshold: k = 0 voxels
Expected voxels per cluster, <k> = 19.837
Expected number of clusters, <c> = 12.76
FWEp: 4.766, FDRp: 4.168, FWEc: 289, FDRc: 289

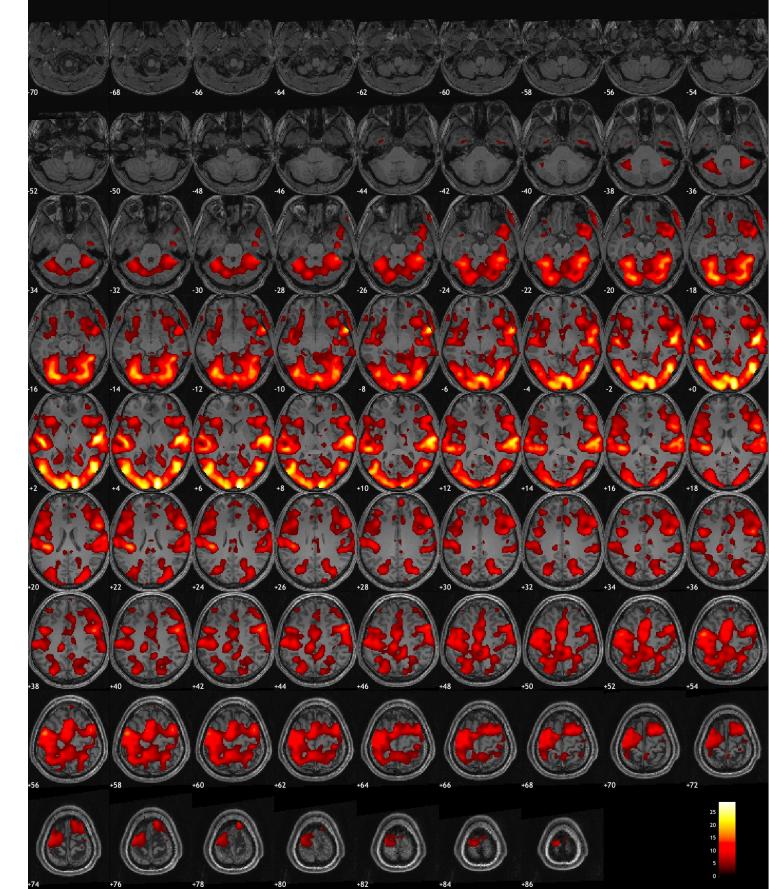
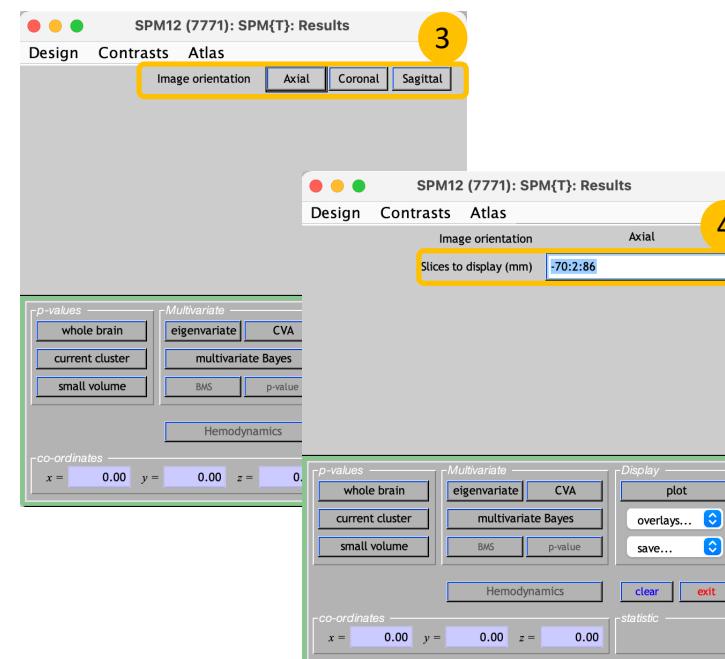
Degrees of freedom = [1, 0, 569, 0]
FWHM = 11.0 11.6 10.6 mm mm mm; 5.5 5.8 5.3 (voxels)
Volume: 1722680 - 215335 voxels = 1162.8 resels
Voxel size: 2.0 2.0 2.0 mm mm mm; (resel = 169.52 voxels)



Overlap on the
T1 anatomical
scan



1. In overlays -> Montage
2. Select the normalized anatomical scan
3. Image orientation: Axial
4. Slices to display (mm): -70:2:86



How do the results look like for the congruent contrasts?

How do the results look like for the incongruent contrasts?

How do the results look like for the congruent versus incongruent contrasts?

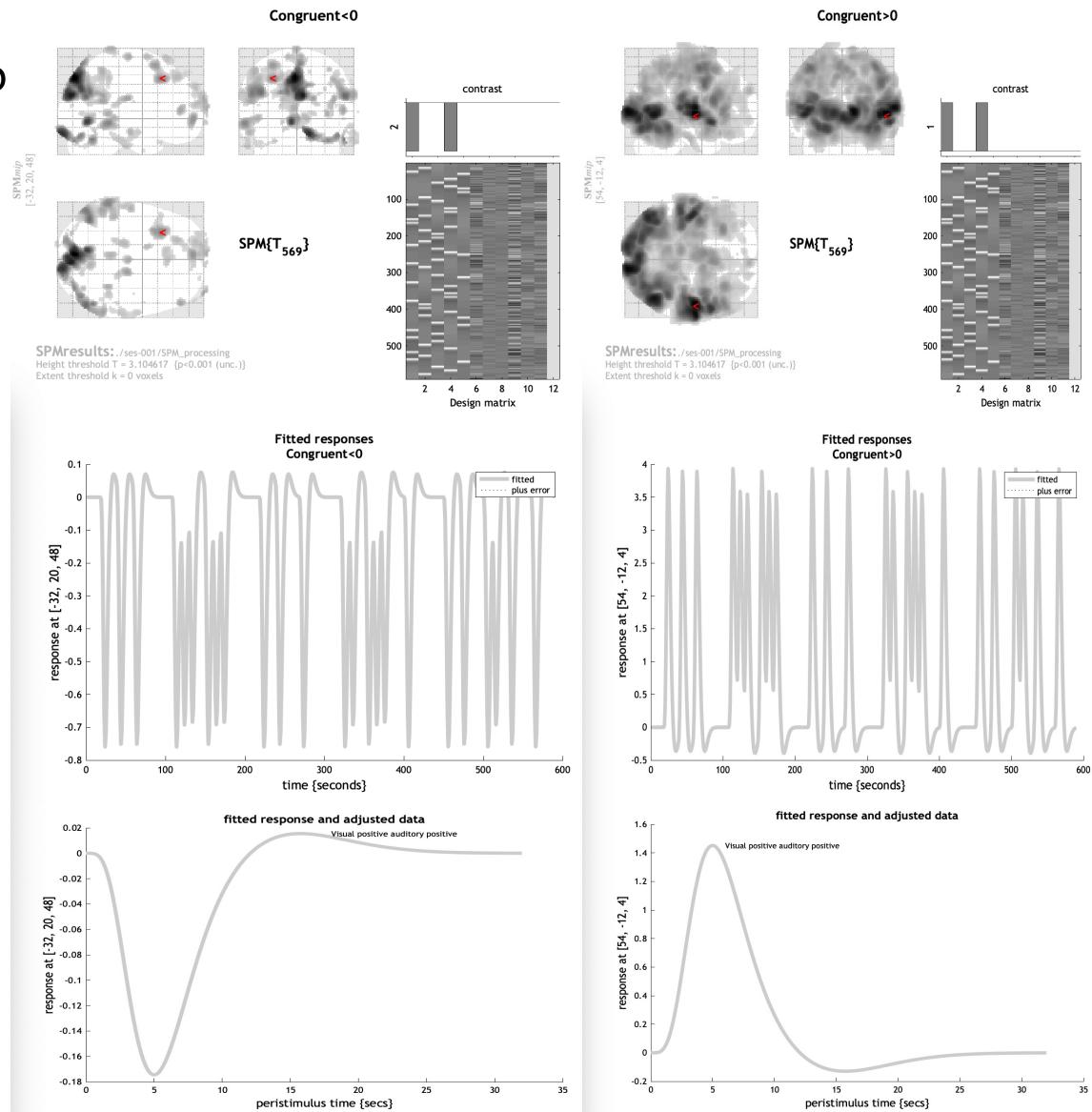
Negative contrast: deactivations?

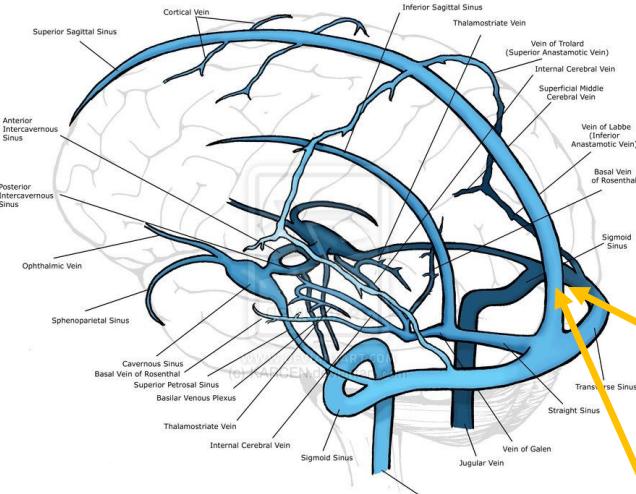
Negative beta (e.g. $\beta_1 < 0$): $\hat{y}_j - \sum_{i=2}^n \beta_{ij} \cdot x_j = \beta_1 \cdot x_j < 0$

The inverse model fits the data

Possible interpretations:

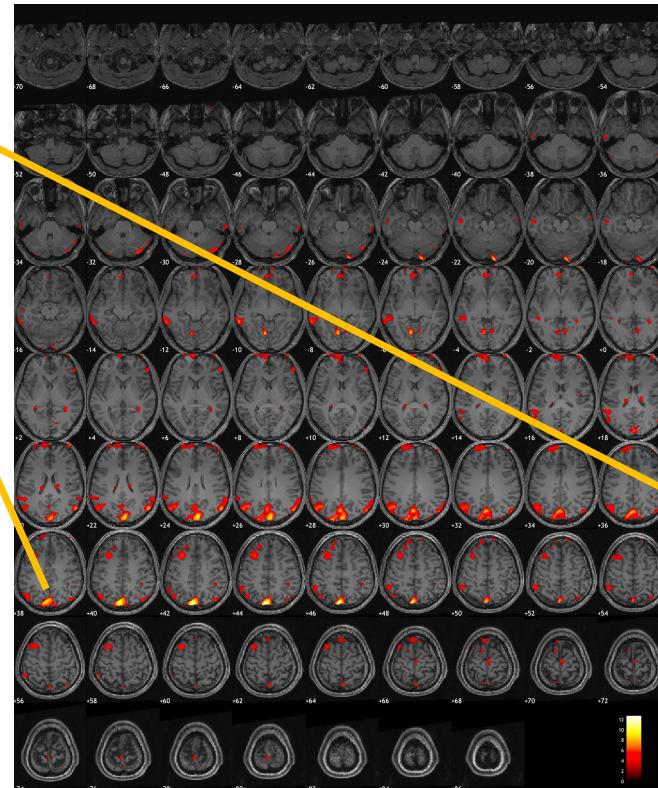
- Could be an effect of the model (e.g. the noise regressors included) -> the results are model depended!
- Deactivations (negative BOLD response)
 - Inhibited neural activity compared to the default or rest state (ongoing neural activity)
 - Decreased CBF due to an increased CBF in a nearby area
 - Increased amounts of deoxyglobine due to nearby activity (e.g. in larger veins)



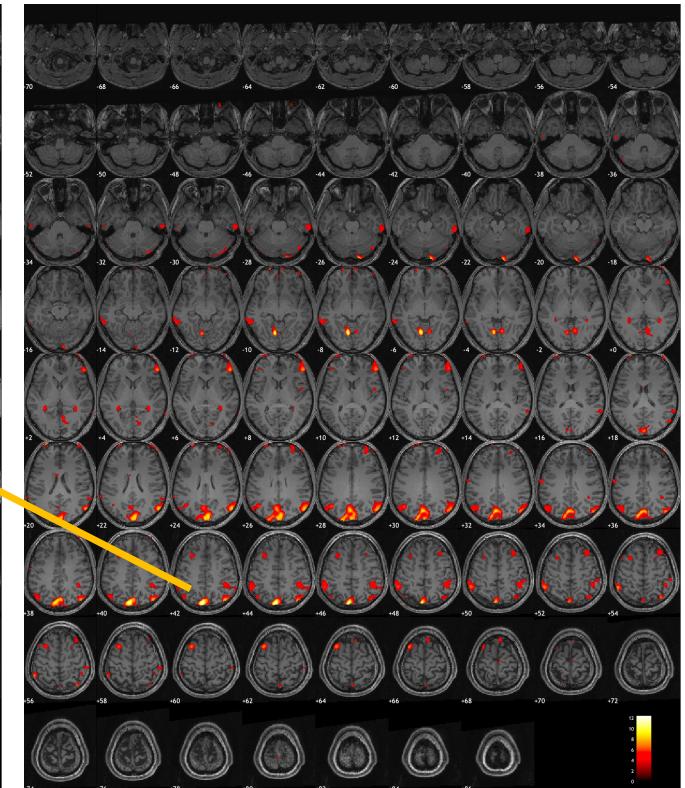


Increased amounts of deoxyglobine in
larger veins due to the increased activity?

Incongruent<0

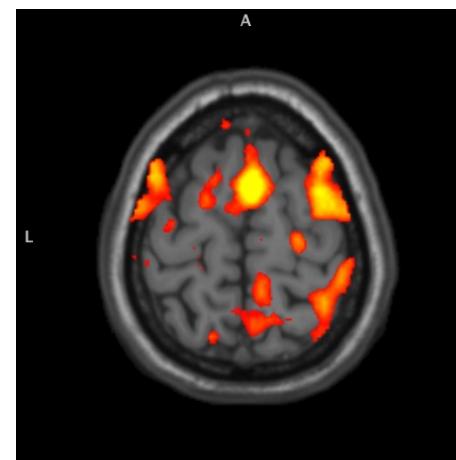
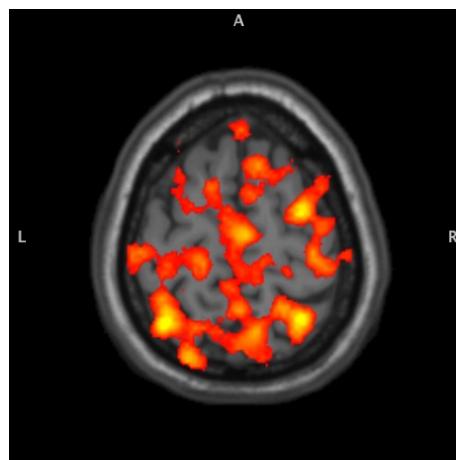
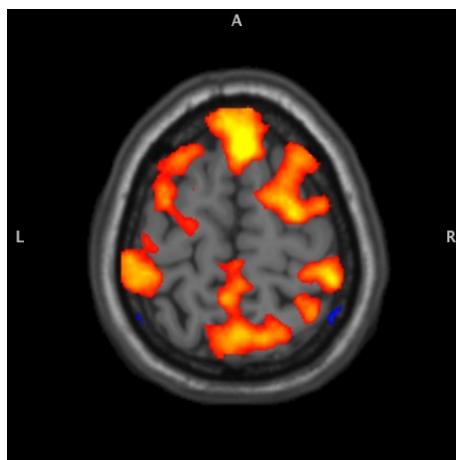
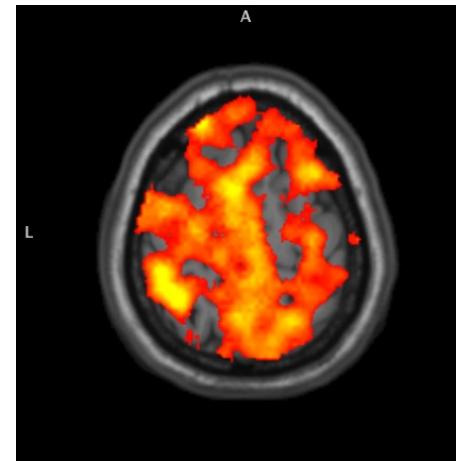
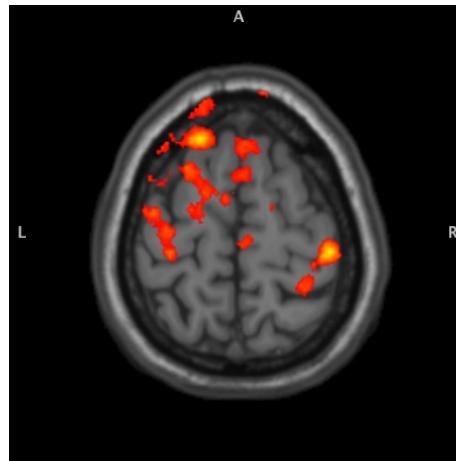
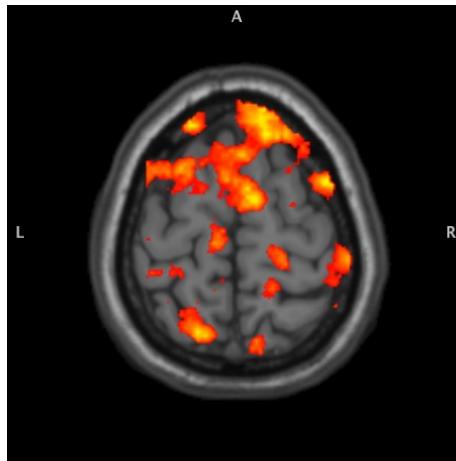


Congruent<0



Second level analysis

Individual results



Variable

- Between subjects
- In time

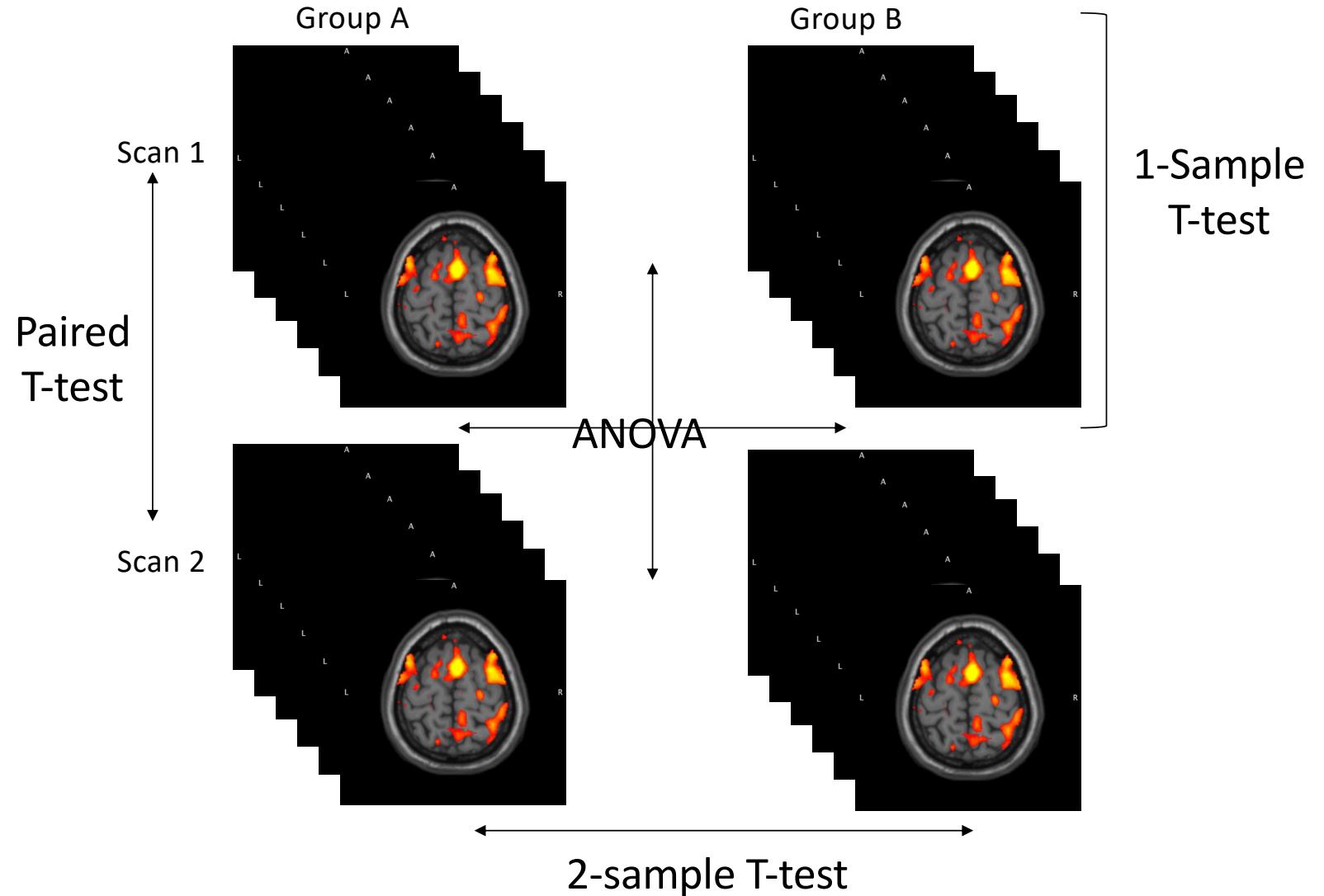
Depending on

- Scanner
- Analysis settings
- Accidental activations
- Uncontrollable factors
(fatigue, caffeine,...)



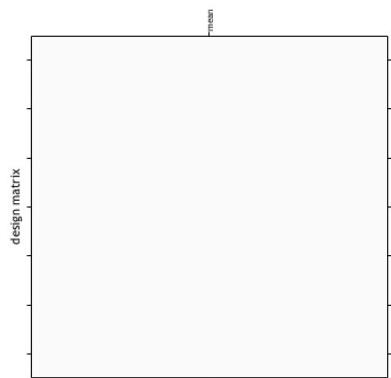
Low repeatability

Study setup and statistical tests

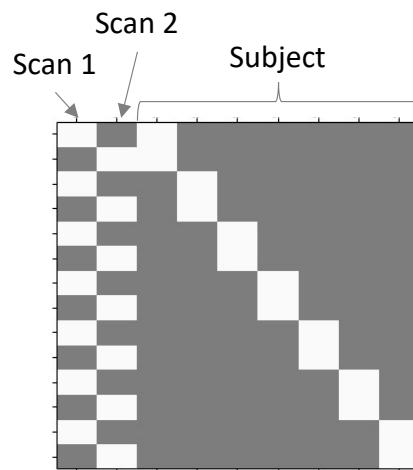


Statistical tests in GLM

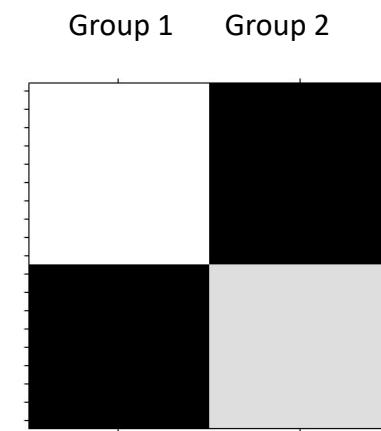
1-sample T-test



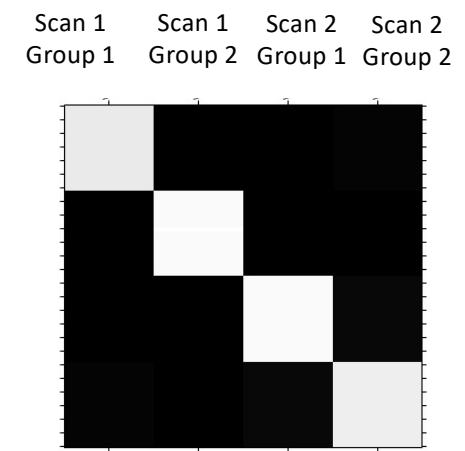
Paired T-test



2-sample T-test



ANOVA



Significant within group
Contrast matrix: [1]

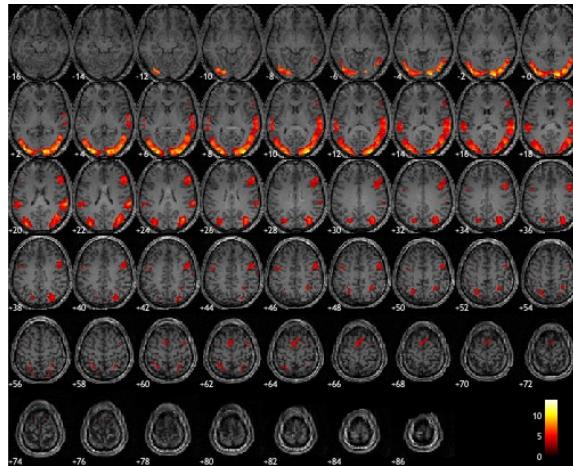
Significant differences
between scans
Contrast matrix:
Scan 1>Scan 2: [1 -1]
Scan 1<Scan 2: [-1 1]

Significant differences
between groups
Contrast matrix:
Group 1>Group 2: [1 -1]
Group 1<Group 2: [-1 1]

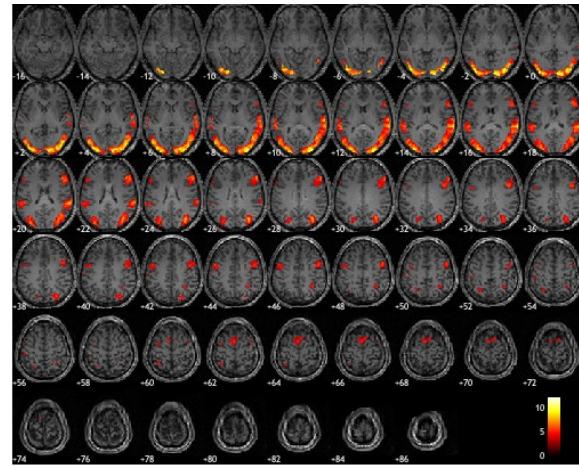
Main effect of task: [1 1 1 1]
Main effect of scan: [1 1 -1 -1]
Main effect of group: [1 -1 1 -1]
Interaction effect group x scan:
[1 -1 -1 1]

1-sample T-test

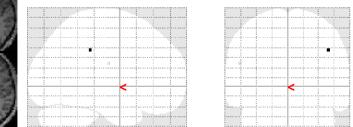
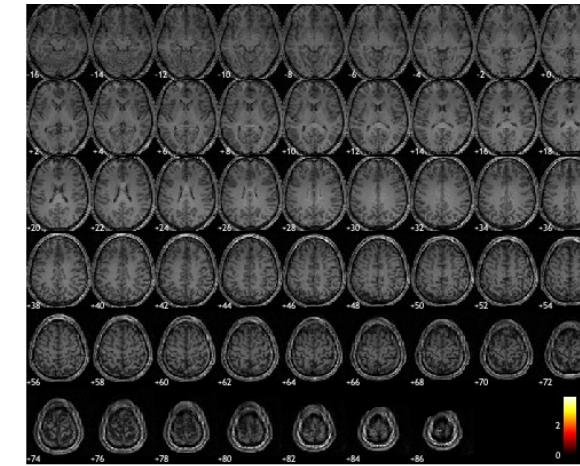
Congruent>0



Incongruent>0

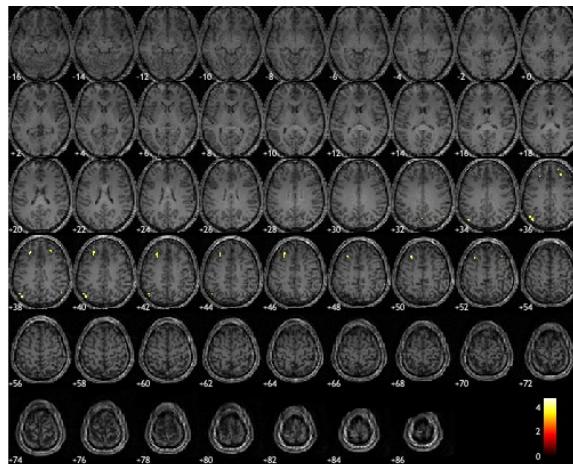


Congruent>Incongruent

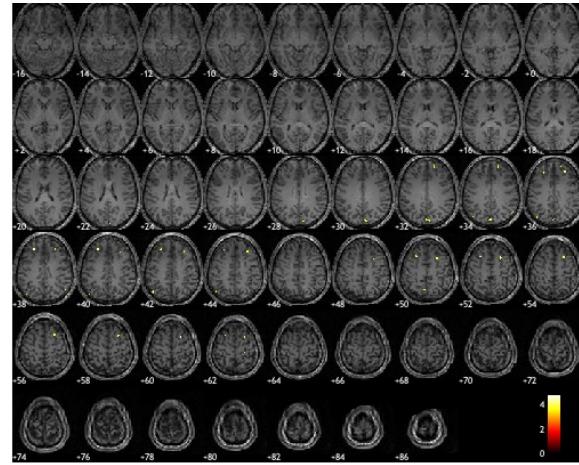


SPM{T₁₉}

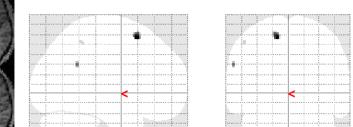
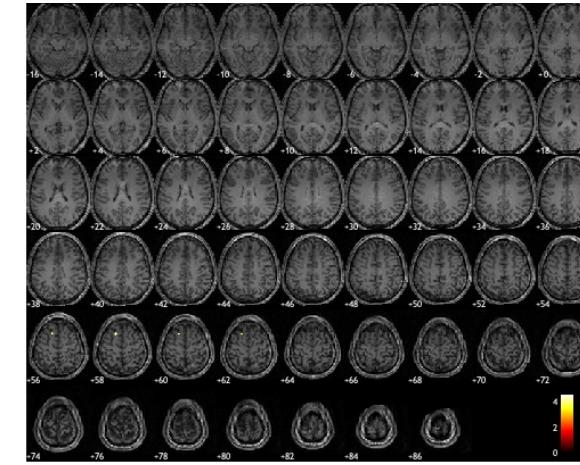
Congruent<0



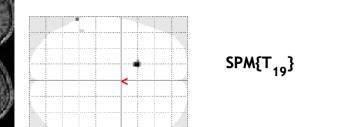
Incongruent<0



Congruent<Incongruent

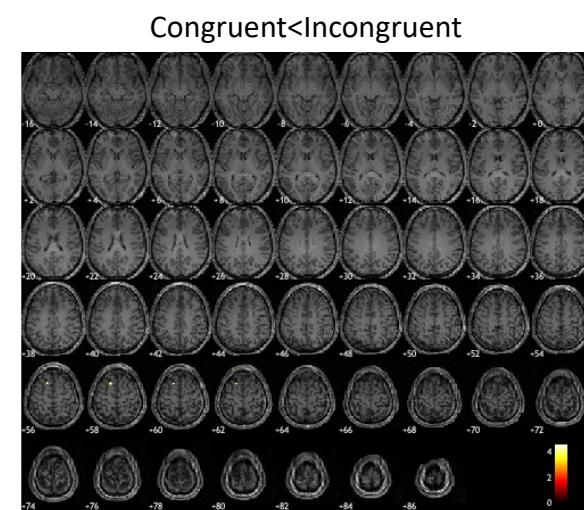
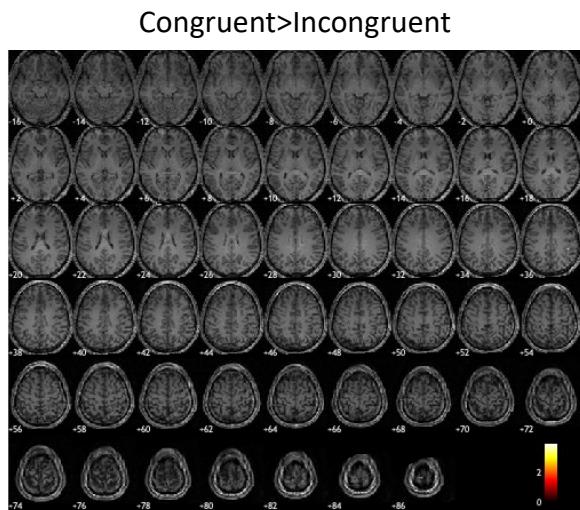


SPM{T₁₉}



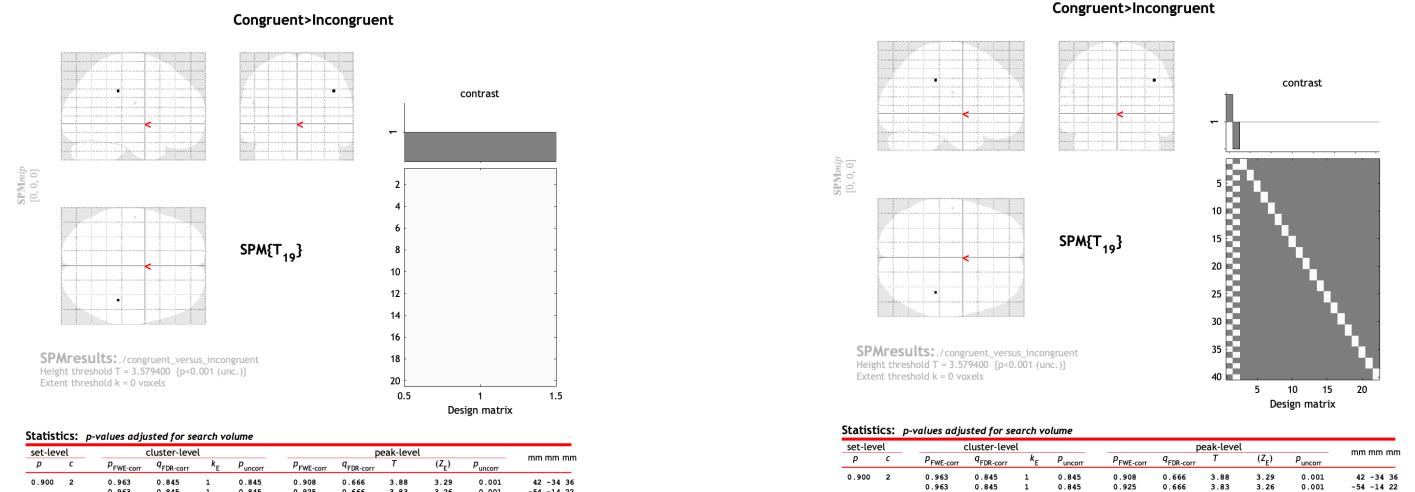
p(uncorrected)<0.001, k>0

Paired T-test: Congruent (con_0001) versus Incongruent (con_0003)



p(uncorrected)<0.001, k>0

Why are the result for the 1 sample T-test for the contrast “Congruent>Incongruent” the same as the results of the paired T-test “Congruent>Incongruent”



1 sample T-test “Congruent>Incongruent”

paired T-test “Congruent>Incongruent”

table shows 3 local maxima more than 8.0mm apart

Height threshold: T = 3.88, p = 0.001 (0.979)
 Degrees of freedom: 1, 1.0, 19.0
 Extent threshold: k = 0 voxels
 Expected voxels per cluster, <k> = 19.330
 Expected number of clusters, <c> = 3.89
 FWeP: 6.136, FDRP: inf, FWEC: inf, FDRC: inf

Degrees of freedom: 1, 1.0, 19.0
 PWeH: 13.1 12.0 12.4 mm mm mm; 6.6 6.0 6.2 (voxels)
 Volume: 392632 - 49079 voxels = 151.5 resels
 Voxel size: 2.0 2.0 2.0 mm mm mm; (resel = 243.47 voxels)

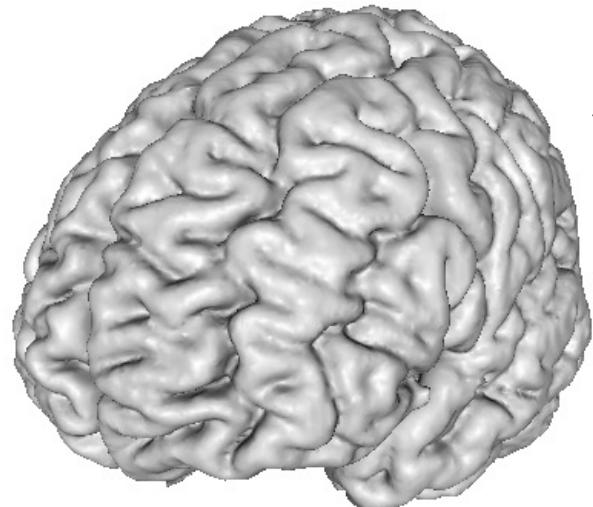
table shows 3 local maxima more than 8.0mm apart

Height threshold: T = 3.88, p = 0.001 (0.979)
 Degrees of freedom: 1, 1.0, 19.0
 Extent threshold: k = 0 voxels
 Expected voxels per cluster, <k> = 19.330
 Volume: 392632 - 49079 voxels = 151.5 resels
 Expected number of clusters, <c> = 3.89
 Voxel size: 2.0 2.0 2.0 mm mm mm; (resel = 243.47 voxels)

Degrees of freedom: 1, 1.0, 19.0
 PWeH: 13.1 12.0 12.4 mm mm mm; 6.6 6.0 6.2 (voxels)
 Volume: 392632 - 49079 voxels = 151.5 resels
 Voxel size: 2.0 2.0 2.0 mm mm mm; (resel = 243.47 voxels)

Statistical Thresholds

Multiple comparisons problem

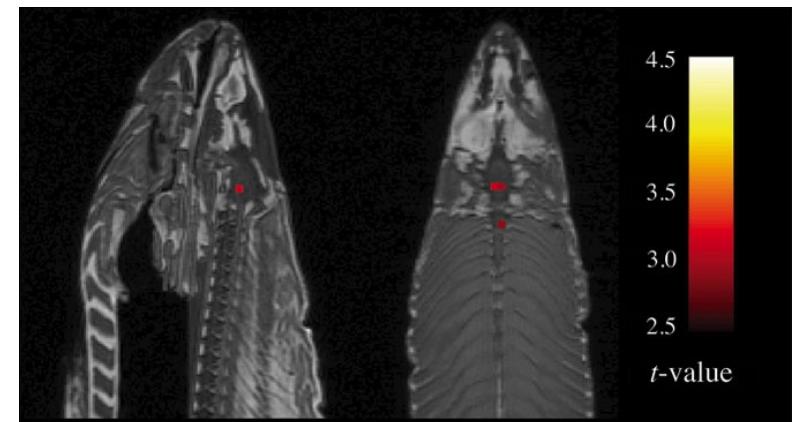


→ >200 000 voxels

↓
>200 000 statistical tests

High chance for false positive results

($200000 \times 0.001 = 200$ expected false positive activations)



Family-Wise Error (FWE) correction

Family-wise null-hypothesis: no activation in any voxel

Family-wise error: a false positive activation somewhere in the image

Family-wise error rate (α): p-value corrected for the chance to get a false positive result in at least 1 voxel

Adjusting the rejecting threshold for multiple comparisons: $p = \frac{\alpha}{N}$

If $\alpha=0.05$ and $N=200\ 000$ the corrected $p=2.5 \cdot 10^{-7}$



TO CONSERVATIVE

Random field theory

FWE correction assumes N independent voxels <- not true, contrast maps are smooth



Spatial correlation between neighboring voxels



Number of independent observations $n_i < N$



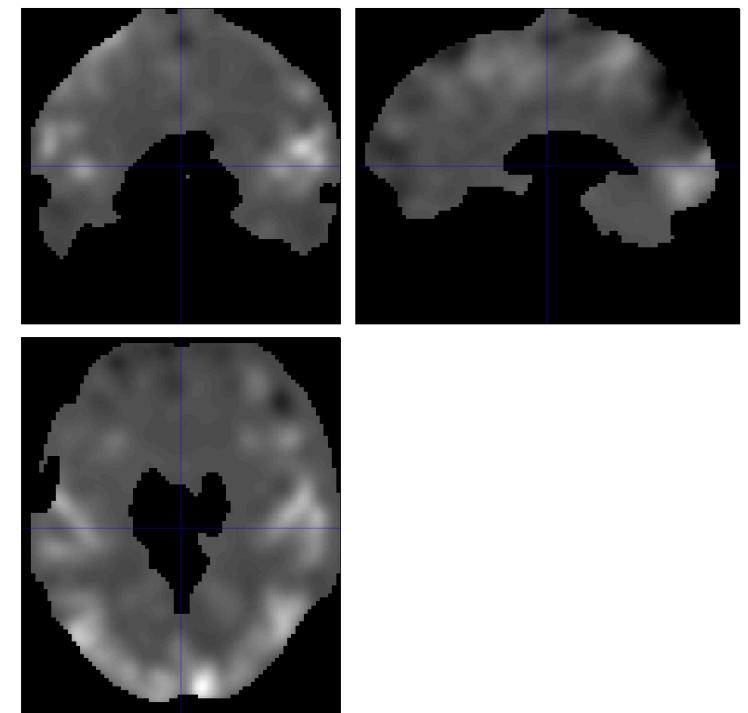
$$\text{FWE corrected } p = \frac{\alpha}{n_i}$$



But ... $n_i = ?$

Resels (resolution element) = number of independent observations in the image

$$R = \frac{\text{Image Volume}}{FWHM_x \cdot FWHM_y \cdot FWHM_z}$$



Random field theory

FWE correction: $\alpha = E[EC]$

$E[EC]$ = expected EC = chance to have at least 1 significant activation voxels above threshold

(null hypothesis: no significant activation voxel above the threshold)

Statistics: p-values adjusted for search volume					Significant at $\alpha < 0.05$								
set-level		cluster-level			peak-level				mm mm mm mm				
p	c	P _{FWE-corr}	q _{FDR-corr}	k _E	P _{uncorr}	P _{FWE-corr}	q _{FDR-corr}	T	(Z _E)	P _{uncorr}	mm	mm	mm
0.000	7	0.000	0.000	7612	0.000	0.000	0.000	14.02	6.72	0.000	22	-90	4
						0.000	0.000	12.32	6.39	0.000	-36	-88	0
						0.000	0.000	11.84	6.29	0.000	-26	-92	4
						0.013	0.008	7.11	4.91	0.000	54	28	26
						0.082	0.036	5.97	4.43	0.000	42	6	34
						0.150	0.061	5.60	4.25	0.000	44	-2	40
						0.014	0.008	7.06	4.89	0.000	-52	-34	12
						0.020	0.011	6.84	4.80	0.000	-48	-32	20
						0.079	0.035	6.00	4.44	0.000	-54	-16	8
						0.002	0.002	246	4.02	0.000	-6	0	68
						0.299	0.112	5.14	4.02	0.000	4	72	0
						0.503	0.196	4.74	3.81	0.000	4	4	72
						0.516	0.201	4.72	3.79	0.000	-10	6	76
						0.338	0.127	5.05	3.97	0.000	-46	0	34
						0.340	0.127	5.05	3.97	0.000	-40	-2	46
						0.543	0.215	4.68	3.77	0.000	-54	-2	42
						0.368	0.137	4.99	3.94	0.000	-26	-62	52
						0.617	0.260	4.56	3.70	0.000	-26	-54	62
						0.866	0.483	4.12	3.44	0.000	-34	-46	60
						0.416	0.158	4.90	3.89	0.000	32	-52	48
						0.783	0.379	4.29	3.54	0.000	28	-56	60

Euler Characteristic (EC) = number of voxel found after thresholding

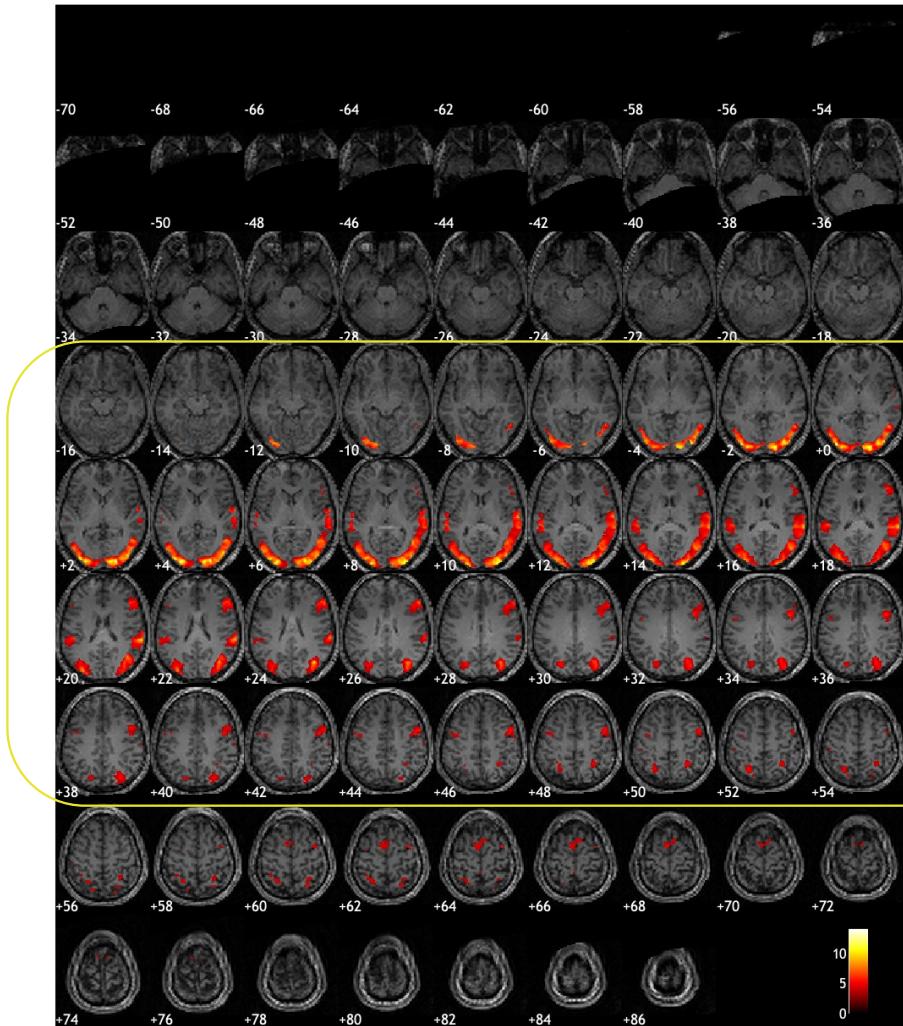
table shows 3 local maxima more than 8.0mm apart

Height threshold: T = 3.58, p = 0.001 (0.992)
Extent threshold: k = 126 voxels, p = 0.006 (0.030)
Expected voxels per cluster, <k> = 14.679
Expected number of clusters, <n> = 0.03
FWEp: 6.281, FDRp: 5.858, FWEc: 126, FDRc: 126

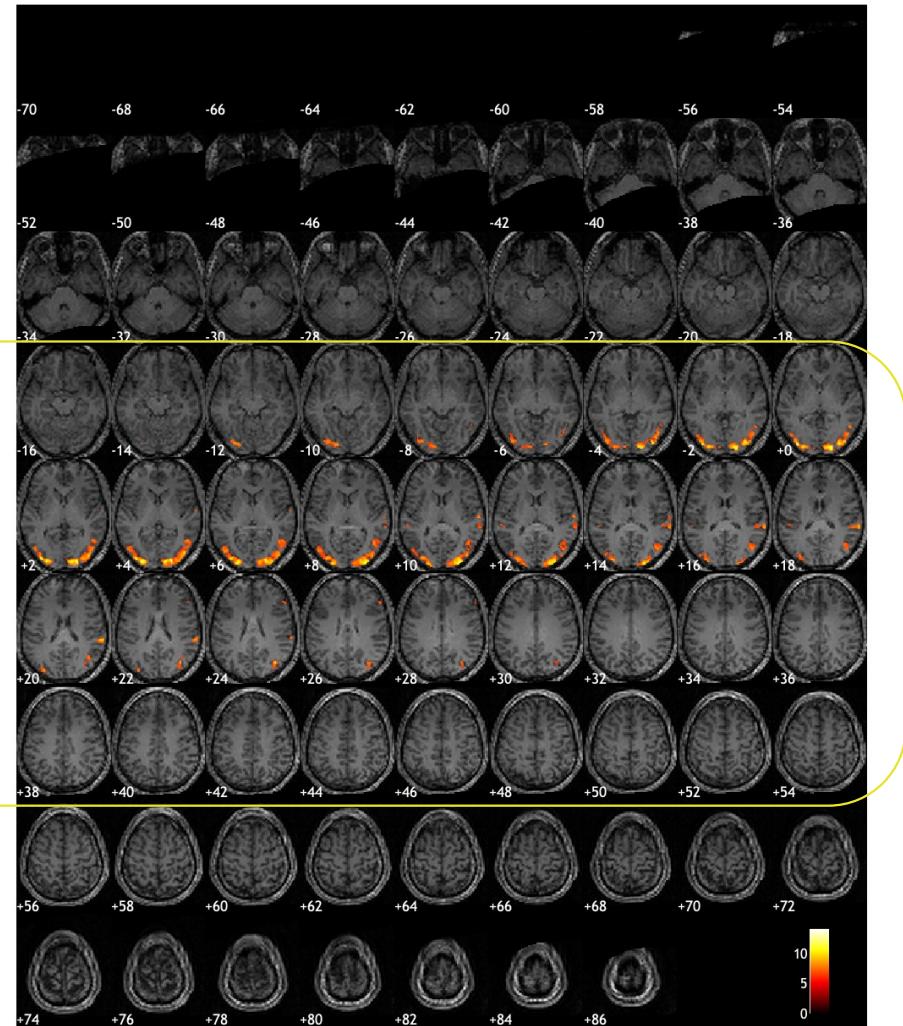
Degrees of freedom = [1.0, 19.0]
FWHM = 11.9 10.5 11.8 mm mm mm; 5.9 5.3 5.9 {voxels}
Volume: 392632 = 49079 voxels = 199.5 resels
Voxel size: 2 0 2 0 2 0 mm mm mm; (resel = 184 90 voxels)

Result for a 1-sample T-test at group level

Congruent>0 ($p(\text{uncorrected})<0.001$)



Congruent>0 ($p(\text{FWE})<0.05$)



Result for a 1-sample T-test at group level

In our single subject analysis, for the results of congruent>0 with the threshold set at p(FWE)<0.05

The number of resulting clusters is =

The smallest cluster size =

The largest cluster size =

Significance threshold at cluster level

Significant at $\alpha < 0.05$

Statistics: p-values adjusted for search volume

set-level	c	cluster-level			peak-level					mm mm mm			
		$p_{\text{FWE-corr}}$	$q_{\text{FDR-corr}}$	k_E	p_{uncorr}	$p_{\text{FWE-corr}}$	$q_{\text{FDR-corr}}$	T	(Z_E)	p_{uncorr}	mm	mm	mm
0.000	7	0.000	0.000	7612	0.000	0.000	0.000	14.02	6.72	0.000	22	-90	4
						0.000	0.000	12.32	6.39	0.000	-36	-88	0
						0.000	0.000	11.84	6.29	0.000	-26	-92	4
						0.013	0.008	7.11	4.91	0.000	54	28	26
						0.082	0.036	5.97	4.43	0.000	42	6	34
						0.150	0.061	5.60	4.25	0.000	44	-2	40
						0.014	0.008	7.06	4.89	0.000	-52	-34	12
						0.020	0.011	6.84	4.80	0.000	-48	-32	20
						0.079	0.035	6.00	4.44	0.000	-54	-16	8
						0.299	0.112	5.14	4.02	0.000	-6	0	68
						0.503	0.196	4.74	3.81	0.000	4	4	72
						0.516	0.201	4.72	3.79	0.000	-10	6	76
						0.338	0.127	5.05	3.97	0.000	-46	0	34
						0.340	0.127	5.05	3.97	0.000	-40	-2	46
						0.543	0.215	4.68	3.77	0.000	-54	-2	42
						0.368	0.137	4.99	3.94	0.000	-26	-62	52
						0.617	0.260	4.56	3.70	0.000	-26	-54	62
						0.866	0.483	4.12	3.44	0.000	-34	-46	60
						0.416	0.158	4.90	3.89	0.000	32	-52	48
						0.783	0.379	4.29	3.54	0.000	28	-56	60

Chance to find the selected cluster by chance

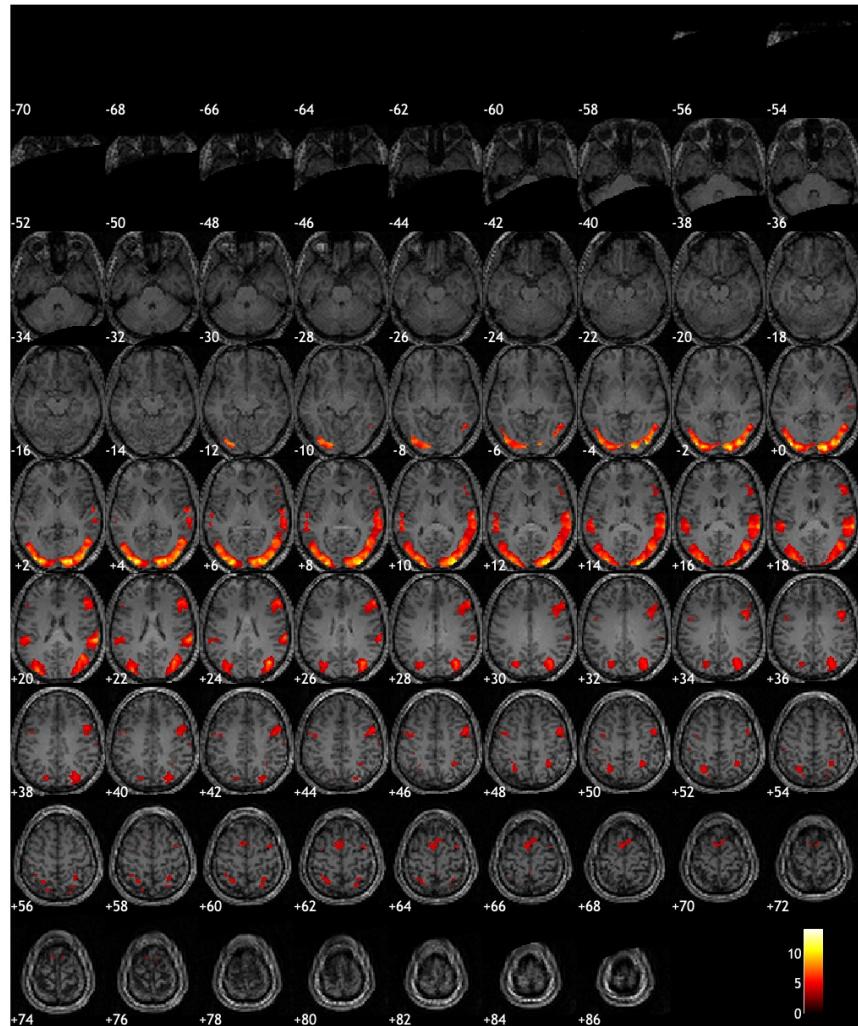
table shows 3 local maxima more than 8.0mm apart

Height threshold: $T = 3.58$, $p = 0.001$ (0.992)
 Extent threshold: $k = 126$ voxels, $p = 0.006$ (0.030)
 Expected voxels per cluster, $\langle k \rangle = 14.679$
 Expected number of clusters, $\langle c \rangle = 0.03$
 FWEp: 6.281, FDRp: 5.858, FWEc: 126, FDRc: 126

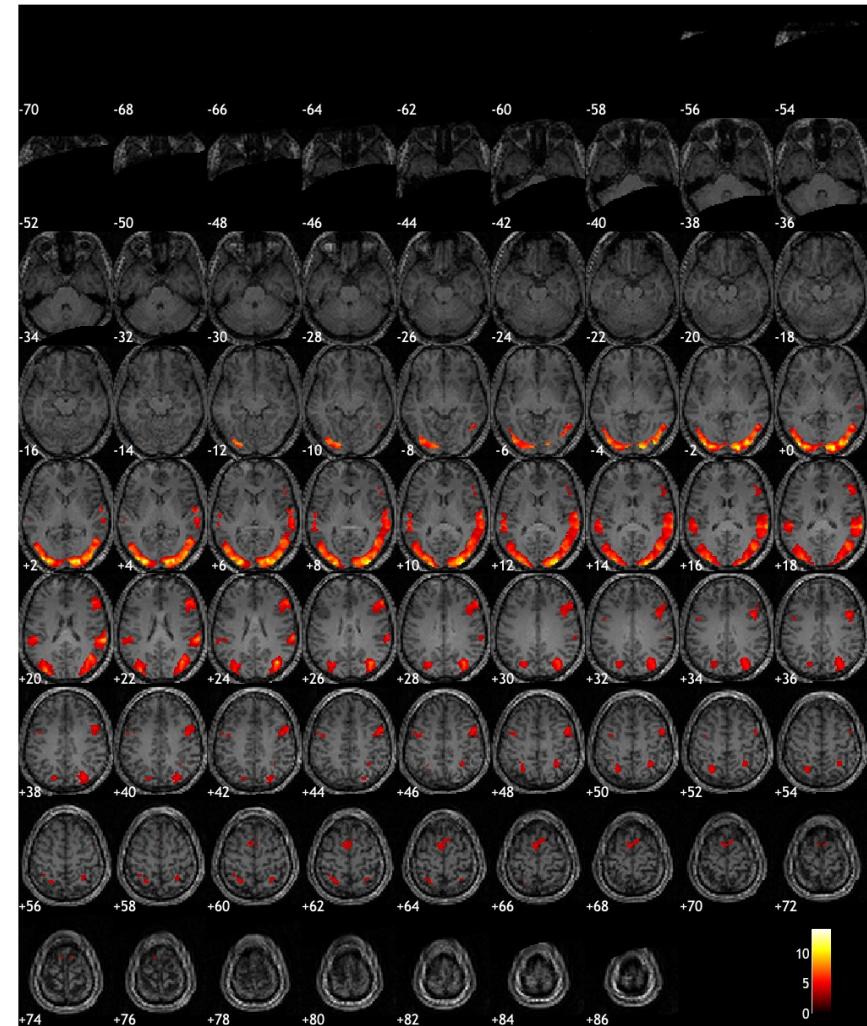
Degrees of freedom = [1.0, 19.0]
 FWHM = 11.9 10.5 11.8 mm mm mm; 5.9 5.3 5.9 {voxels}
 Volume: 392632 = 49079 voxels = 199.5 resels
 Voxel size: 2.0 2.0 2.0 mm mm mm; (resel = 184.90 voxels)

Result for a 1-sample T-test at group level

Congruent>0 ($p(\text{uncorrected})<0.00$, $k=0$)



Congruent>0 ($p(\text{uncorrected})<0.001$, $k>126$)



Result for a 1-sample T-test at group level

In our single subject analysis, for the results of congruent>0 with the voxel significance threshold set at p(uncorrected)<0.001 and the cluster significance set at p(FWE)<0.05

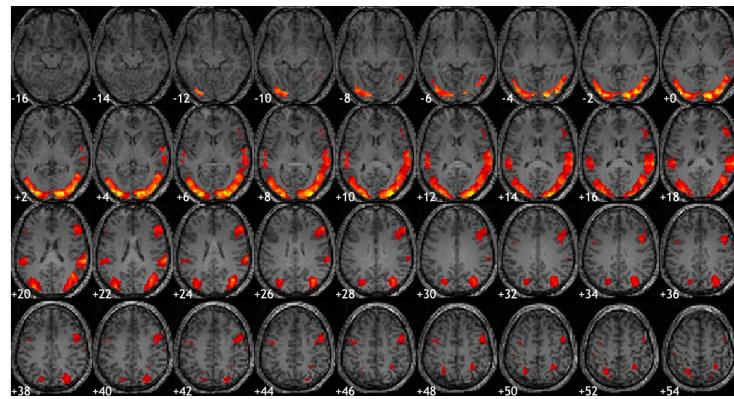
The minimum cluster size is =

The number of resulting clusters is =

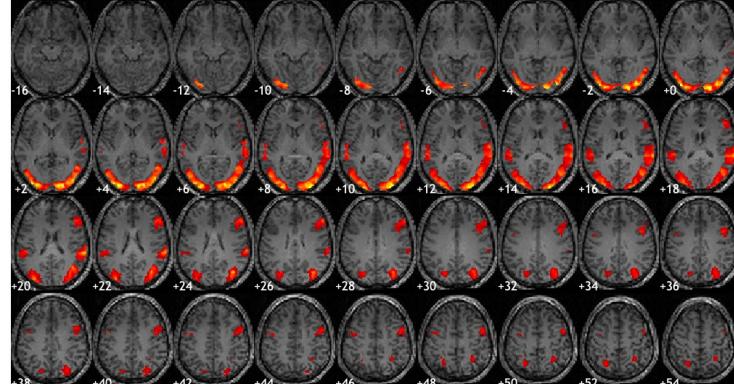
The largest cluster size =

Uncorrected:
 $p(\text{uncorrected}) < 0.001, k > 0$

Highest chance Type I errors

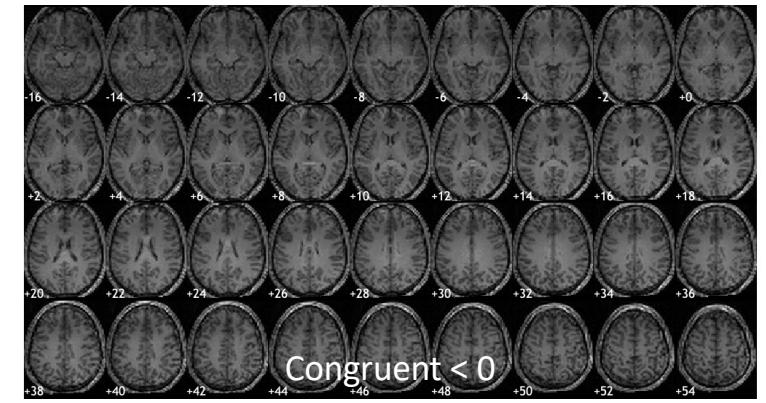
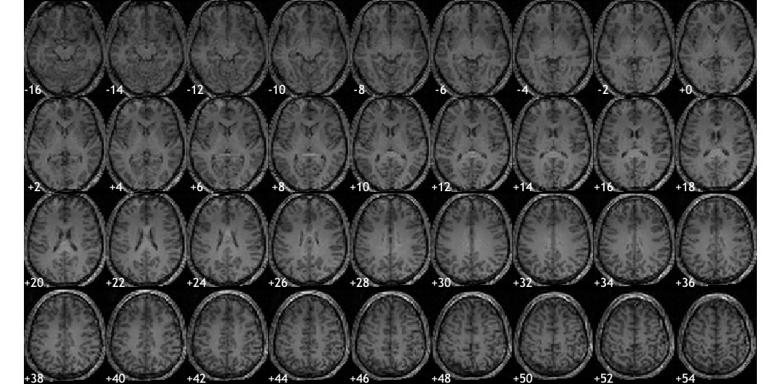
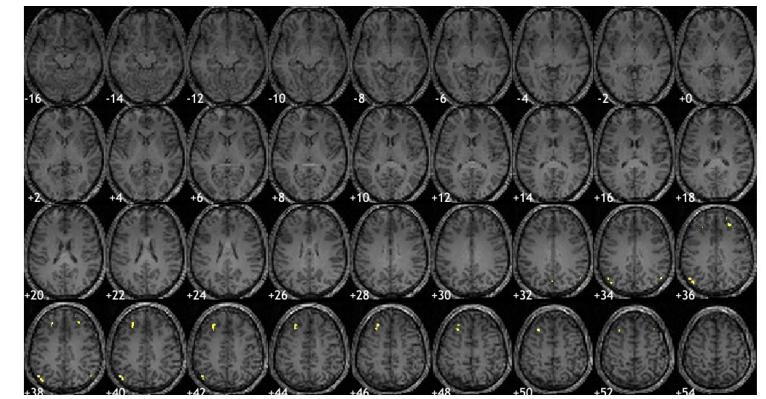
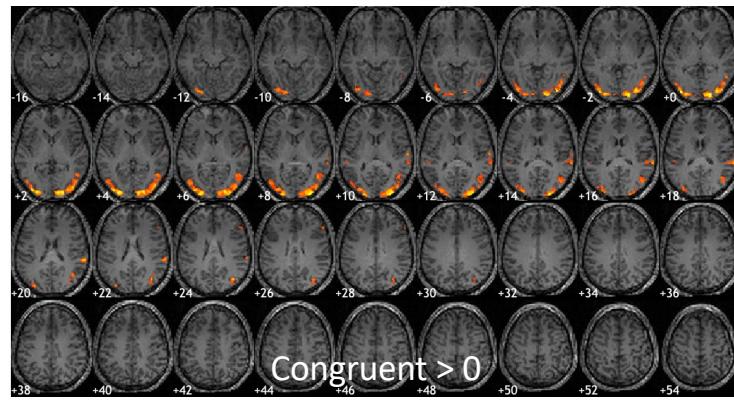


FWE correction at cluster level:
 $p(\text{uncorrected}) < 0.001, k > 126$



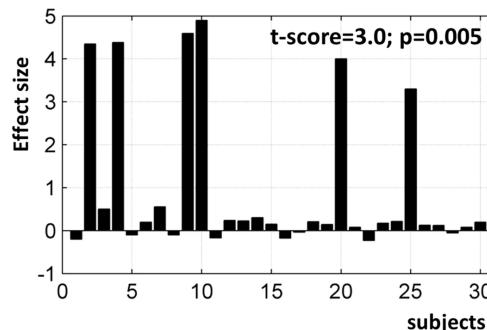
FWE correction at cluster level:
 $p(\text{FWE}) < 0.05, k > 0$

Highest chance Type II errors



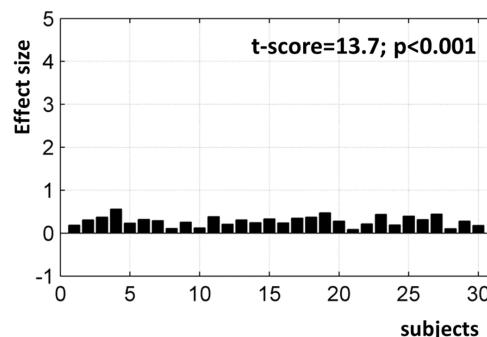
Remarque: Significant \neq Large or important

Significant:
 $mean(effect) \neq 0$



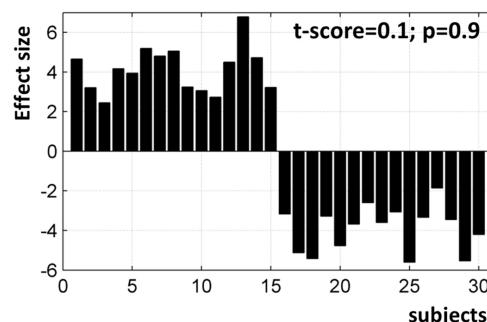
Significant effect

But ... only a large effect in a few subjects



Significant effect

But ... it's a small effect in all subjects



No significant effect

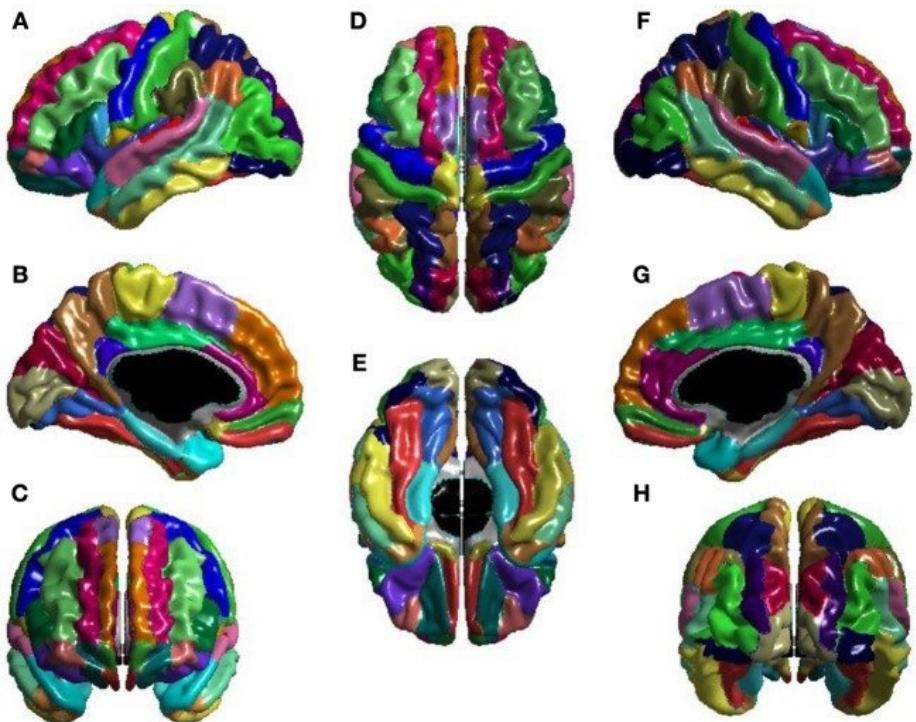
But ... a large positive effect in one halve of the subjects
and a large negative effect in the other halve

Anatomical labeling

AAL: Automated Anatomical Labeling atlas

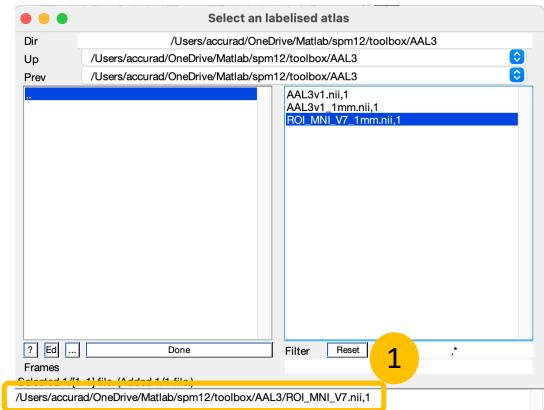
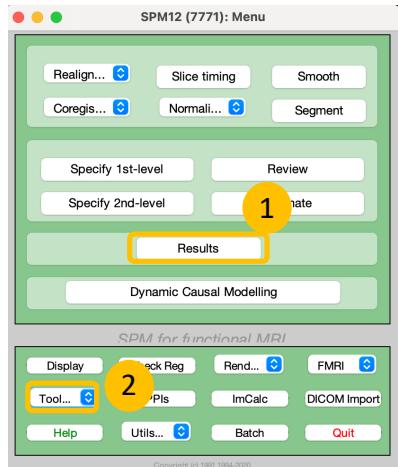
170 anatomical brain areas

Locations defined in MNI template space

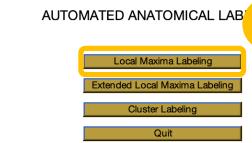


Label	Anatomy	Label	Anatomy	Label	Anatomy	Label	Anatomy
1	Precentral L	51	Lingual L	101	Cerebellum 4 5 L	151	ACC sub L
2	Precentral R	52	Lingual R	102	Cerebellum 4 5 R	152	ACC sub R
3	Frontal Superior 2 L	53	Occipital Superior L	103	Cerebellum 6 L	153	ACC pre L
4	Frontal Superior 2 R	54	Occipital Superior R	104	Cerebellum 6 R	154	ACC pre R
5	Frontal Middle 2 L	55	Occipital Middle L	105	Cerebellum 7b L	155	ACC Superior L
6	Frontal Middle 2 R	56	Occipital Middle R	106	Cerebellum 7b R	156	ACC Superior R
7	Frontal Inferior Opercular L	57	Occipital Inferior L	107	Cerebellum 8 L	157	N Acc L
8	Frontal Inferior Opercular R	58	Occipital Inferior R	108	Cerebellum 8 R	158	N Acc R
9	Frontal Inferior Triangular L	59	Fusiform L	109	Cerebellum 9 L	159	VTA L
10	Frontal Inferior Triangular R	60	Fusiform R	110	Cerebellum 9 R	160	VTA R
11	Frontal Inferior Orbital 2 L	61	Posteriorcentral L	111	Cerebellum 10 L	161	SN pc L
12	Frontal Inferior Orbital 2 R	62	Posteriorcentral R	112	Cerebellum 10 R	162	SN pc R
13	Rolandic Opercular L	63	Parietal Superior L	113	Vermis 1 2	163	SN pr L
14	Rolandic Opercular R	64	Parietal Superior R	114	Vermis 3	164	SN pr R
15	Superiorp Motor Area L	65	Parietal Inferior L	115	Vermis 4 5	165	Red N L
16	Superiorp Motor Area R	66	Parietal Inferior R	116	Vermis 6	166	Red N R
17	Olfactory L	67	SuperiorMarginal L	117	Vermis 7	167	LC L
18	Olfactory R	68	SuperiorMarginal R	118	Vermis 8	168	LC R
19	Frontal Superior Medial L	69	Angular L	119	Vermis 9	169	Raphe D
20	Frontal Superior Medial R	70	Angular R	120	Vermis 10	170	Raphe M
21	Frontal Medial Orbital L	71	Precuneus L	121	Thalamus AV L		
22	Frontal Medial Orbital R	72	Precuneus R	122	Thalamus AV R		
23	Rectus L	73	Paracentral Lobule L	123	Thalamus LP L		
24	Rectus R	74	Paracentral Lobule R	124	Thalamus LP R		
25	OFCMedial L	75	Caudate L	125	Thalamus VA L		
26	OFCMedial R	76	Caudate R	126	Thalamus VA R		
27	OFCAnterior L	77	Putamen L	127	Thalamus VL L		
28	OFCAnterior R	78	Putamen R	128	Thalamus VL R		
29	OFCPosterior L	79	Pallidum L	129	Thalamus VPL L		
30	OFCPosterior R	80	Pallidum R	130	Thalamus VPL R		
31	OFClat L	81	Thalamusamus L	131	Thalamus IL L		
32	OFClat R	82	Thalamusamus R	132	Thalamus IL R		
33	Insula L	83	Heschl L	133	Thalamus Re L		
34	Insula R	84	Heschl R	134	Thalamus Re R		
35	Cingulate Anterior L	85	Temporal Superior L	135	Thalamus MDm L		
36	Cingulate Anterior R	86	Temporal Superior R	136	Thalamus MDm R		
37	Cingulate Middle L	87	Temporal Pole Superior L	137	Thalamus MDI L		
38	Cingulate Middle R	88	Temporal Pole Superior R	138	Thalamus MDI R		
39	Cingulate Posterior L	89	Temporal Middle L	139	Thalamus LGN L		
40	Cingulate Posterior R	90	Temporal Middle R	140	Thalamus LGN R		
41	Hippocampus L	91	Temporal Pole Middle L	141	Thalamus MGN L		
42	Hippocampus R	92	Temporal Pole Middle R	142	Thalamus MGN R		
43	ParaHippocampal L	93	Temporal Inferior L	143	Thalamus PuL L		
44	ParaHippocampal R	94	Temporal Inferior R	144	Thalamus PuL R		
45	Amygdala L	95	Cerebellum Crus1 L	145	Thalamus PuM L		
46	Amygdala R	96	Cerebellum Crus1 R	146	Thalamus PuM R		
47	Calcarine L	97	Cerebellum Crus2 L	147	Thalamus PuA L		
48	Calcarine R	98	Cerebellum Crus2 R	148	Thalamus PuA R		
49	Cuneus L	99	Cerebellum 3 L	149	Thalamus PuL L		
50	Cuneus R	100	Cerebellum 3 R	150	Thalamus PuL R		

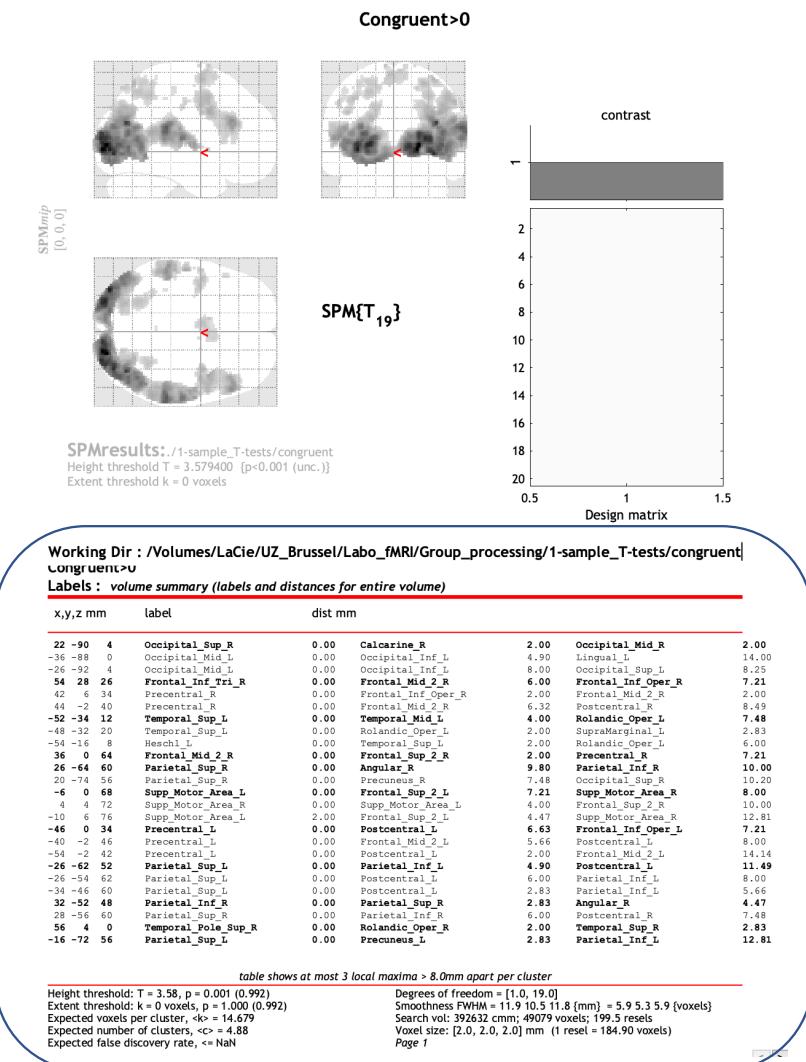
AAL: Automated Anatomical Labeling atlas



1. Open the results first
2. In the Menus Window: Toolbox: -> AAL3
3. In the AAL window -> Local Maxima Labeling
4. Select an labeling atlas: ROI_MNI_V7.nii file



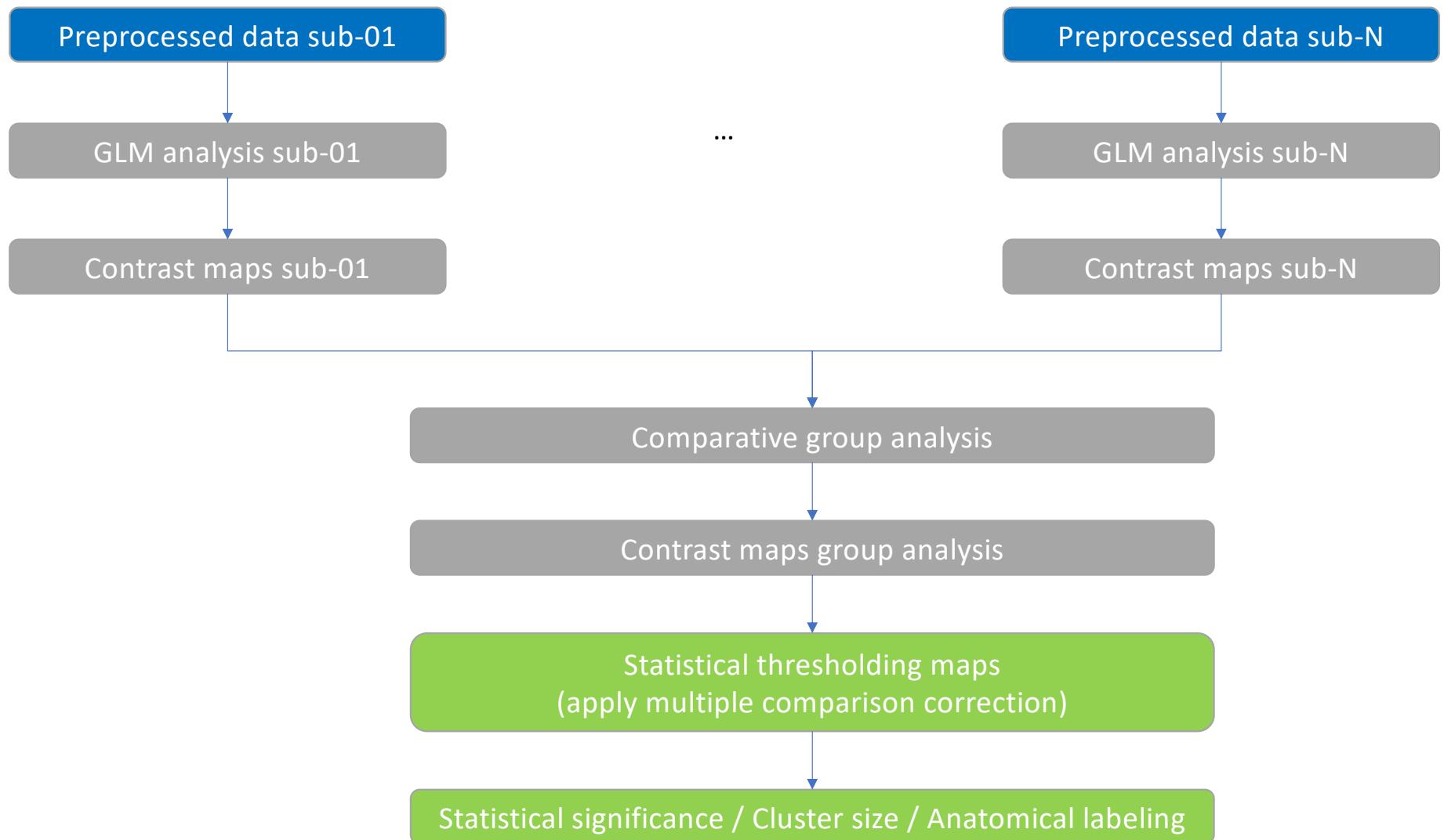
Result for a 1-sample T-test at group level



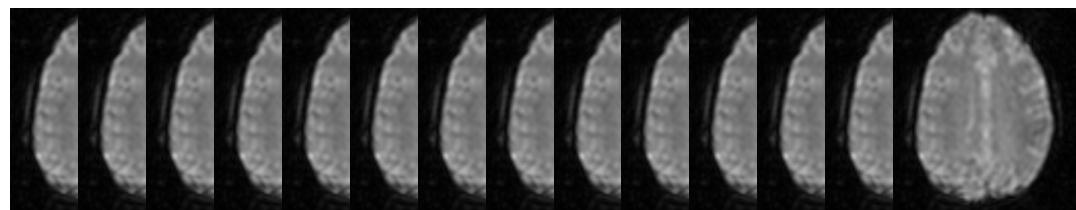
In our single subject analysis, for the results of congruent>0 with the voxel significance threshold set at p(uncorrected)<0.001.

What is the AAL label for the cluster at

1. [12, -98, 4]:
2. [-28, -8, -44]:
3. [20, -12, -20]:
4. [6, 58, 28]:



Resting state fMRI



Time: 5~10 minutes



Table 1.

Presents a brief comparison between task based fMRI and Resting state fMRI.

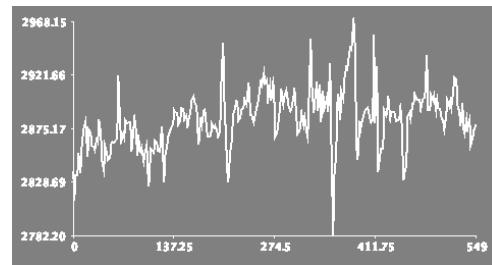
SI no.	Task-based fMRI	Rs-fMRI
I	Analyses of the spontaneous modulations in the BOLD signal in the presence of a particular activity (e.g. finger-tapping, eye-blinking, naming, memorizing, etc.)	Analyses of the spontaneous BOLD signal in the absence of any explicit task or an input
II	Task-related increase in neuronal metabolism are less than 5%	60–80% of brain's energy is consumed during resting state
III	During task-based activity the focus is only on a very small fraction of the brain's overall activity	In terms of overall brain function, the resting state brain activity is far more significant than task-related activity
IV	The signal during a task-related activity is very small compared to the noise, i.e. 80% of the BOLD modulation is discarded as noise	The signals which are discarded as noise in task fMRI is taken as signals in rs-fMRI as they are the low frequency spontaneous fluctuations in the BOLD signal
V	Due to discarding of signal as noise, task fMRI has a low SNR	Have improved SNR since it takes the overall spontaneous low frequency fluctuations
VI	For the interpretation of results, a large number of trials are required in task fMRI	No need of more trials like task fMRI
VII	If one wants to analyse the motor function and language function, a separate task may be required to analyse each function in task-based fMRI	In rs-fMRI, the acquired may be used to analyse one or more functions
IX	Patient cooperation is essential to do task fMRI	Paediatric patients, patients with low IQ and even patients in the vegetative and coma state are able to do rs-fMRI
X	Repeated sessions of task-based activity to assess the disease prognosis, treatment effect etc. will result in familiarity with the task which will affect the output adversely	In rs-fMRI even we are taking different sessions, due to the absence of task, we are able to avoid the task-related confusions and uncertainties faced by task fMRI

fMRI: functional magnetic resonance imaging; rs-fMRI: resting state functional magnetic resonance imaging; BOLD: blood oxygenation level-dependent; SNR: signal to noise ratio.

From Smith et al. Resting state fMRI: A review on methods in resting state connectivity analysis and resting state networks. Neuroradiol J. 2017 30(4): 305–317

The brain is never at rest

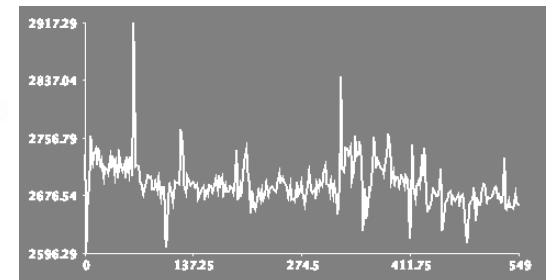
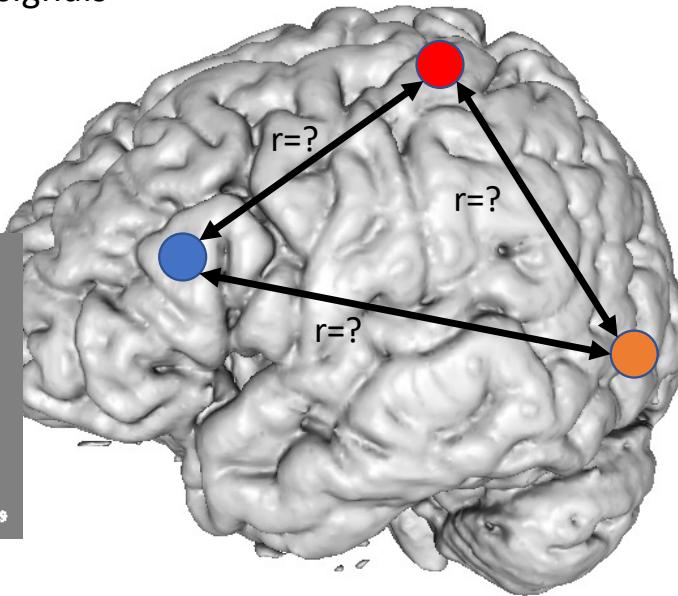
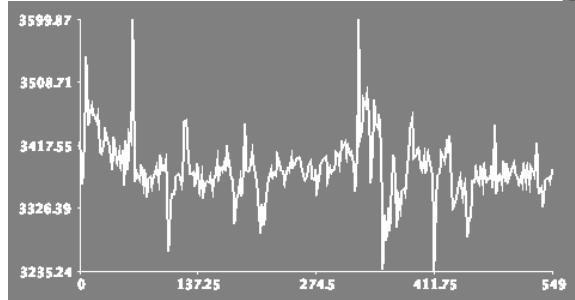
Spontaneous ongoing neural activity -> BOLD signals (BS)



Interactions between brain areas -> correlated signals



Neural networks

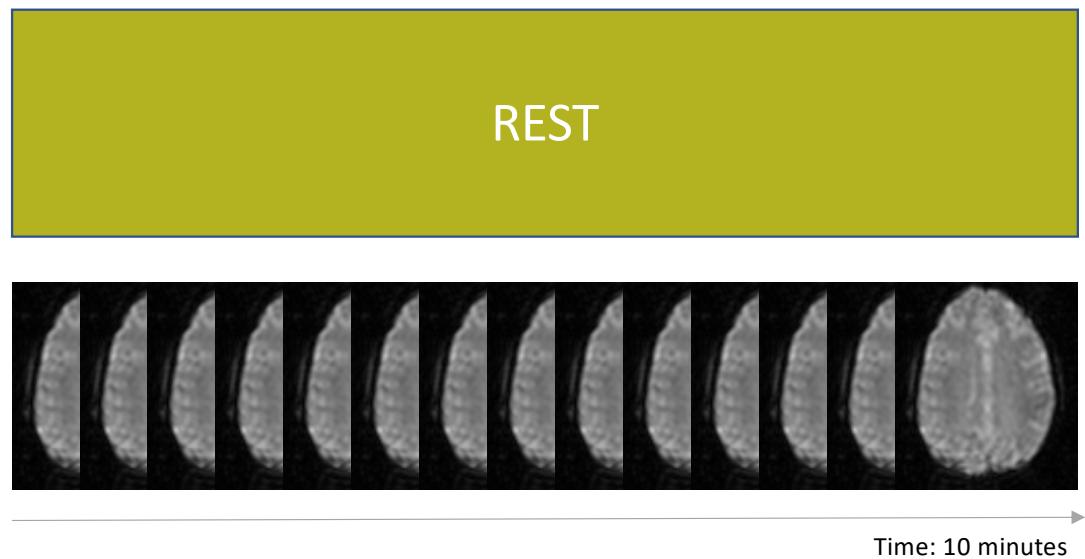


Resting-state functional connectivity measures **temporal correlation** of spontaneous BOLD signal among spatially distributed brain regions, with the assumption that regions with correlated activity form functional networks.

Why didn't we look at connectivity (correlations between the neural activity in different brain areas) in our task-fMRI experiment?

1. There is no connectivity between brain areas during processing a task
2. The neural activity in all brain areas correlate with the task
3. The correlations between the neural activity and the task lead to correlations between brain areas
4. All areas involved in processing the task are connected to each other

Resting state fMRI paradigm

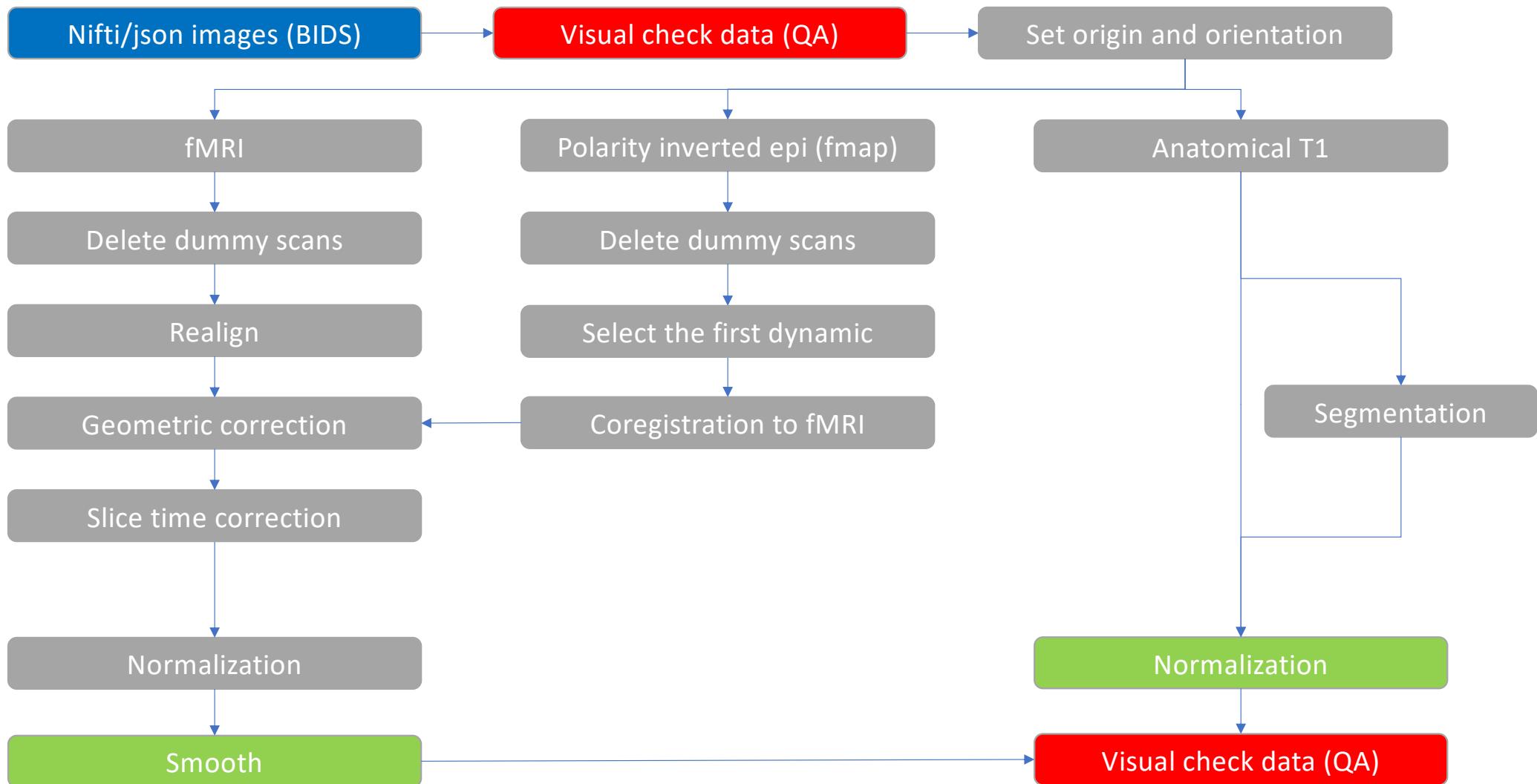


GE-EPI at temporal resolution of 2s

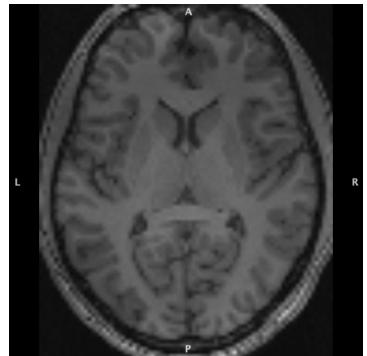
Instructions:

- Relax but don't fall asleep
- Try not to move
- Try not to think of anything
- Keep eyes open

20 healthy subjects



The preprocessing results



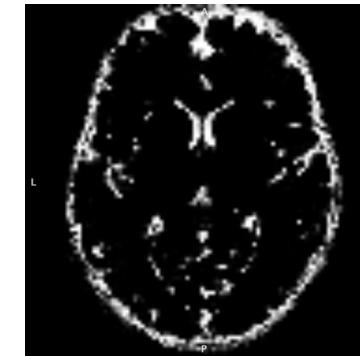
wosub-01_T1w



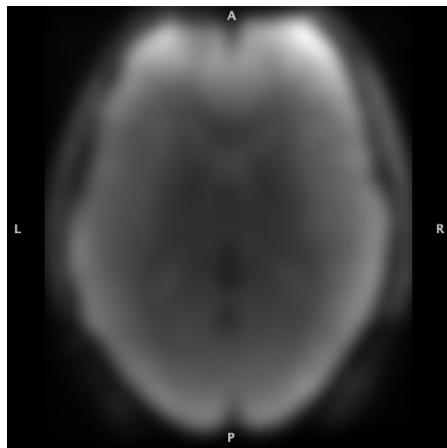
wc1osub-01_T1w



wc2osub-01_T1w



wc3osub-01_T1w



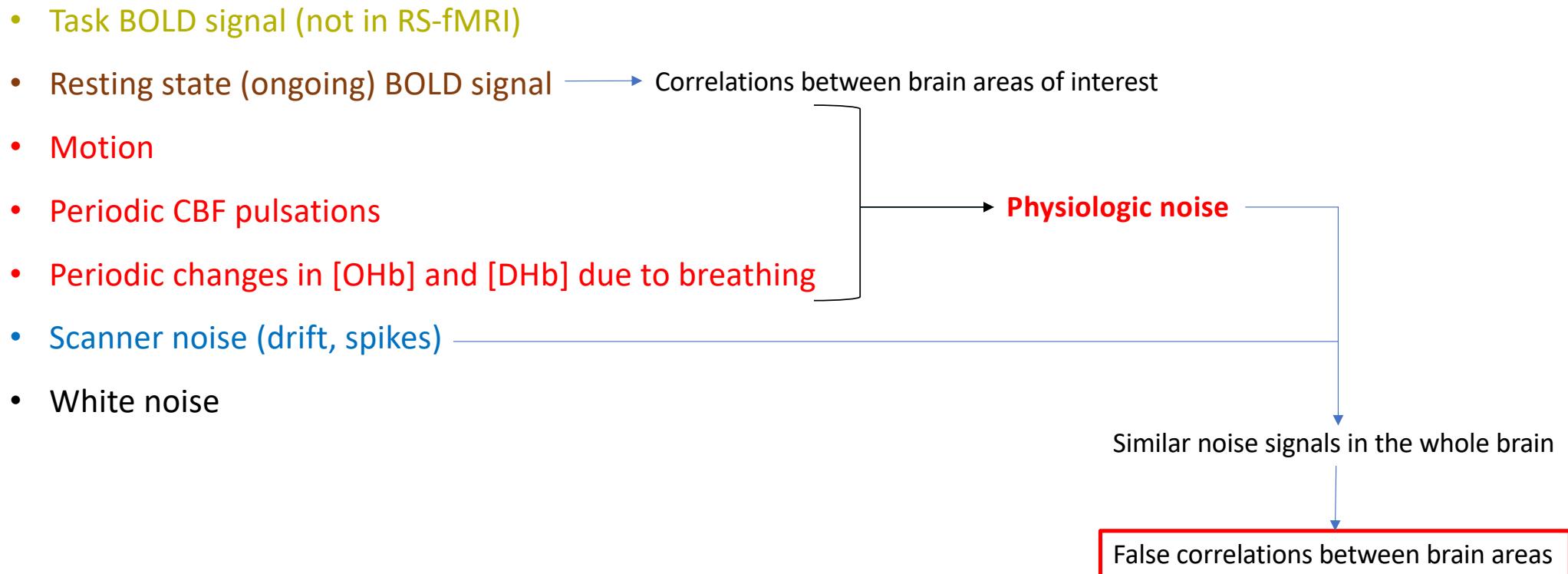
swuraosub-01_..._bold

	Trans x	Trans y	Trans z	Rot x	Rot y	Rot z
rp_aosub-01_task-affect_run-1_bold_00001.txt ~	0.000000e+00	-4.7340067e-16	0.000000e+00	0.000000e+00	0.000000e+00	1.2588970e-17
	1.7513722e-02	-6.7856195e-03	6.0396937e-03	4.4362332e-04	-1.0208815e-04	3.4466021e-04
	4.8461260e-02	-3.7386199e-02	6.2362634e-02	1.4502508e-03	-4.0640802e-04	9.5685461e-04
	6.9180189e-02	-3.9871672e-02	6.4310601e-02	1.3979599e-03	-3.5460454e-04	1.4147925e-03
	5.20911583e-02	3.2066219e-02	7.7718133e-02	4.3498257e-04	-9.9844301e-05	8.9267421e-04
	5.7427248e-02	1.6258788e-02	7.6479148e-02	7.0890674e-04	1.6059046e-04	7.9906206e-04
	8.6317280e-02	-3.5120548e-02	7.2283943e-02	1.3903804e-03	-2.0507305e-05	1.3339979e-03
	1.1731055e-01	-3.0335500e-02	6.3979068e-02	1.7311331e-03	-1.9270127e-04	1.7468015e-03
	8.0937031e-02	5.0152872e-02	9.1561197e-02	6.9045654e-04	2.2436062e-04	1.1304025e-03
	6.3606665e-02	3.5731932e-02	7.7646949e-02	5.5901698e-04	1.8457663e-04	8.6840523e-04
	5.2421874e-02	-1.0953733e-02	1.0717907e-01	9.7029852e-04	-6.1821191e-05	1.0495726e-03
	7.4589833e-02	-4.0247471e-04	1.2329963e-01	7.9186069e-04	1.8275389e-05	1.1582123e-03
	1.0715947e-01	-5.0387869e-02	1.0422207e-01	7.68870931e-04	-7.4170782e-05	1.4709182e-03
	1.4121567e-01	-7.7419048e-02	1.30771721e-01	1.4501688e-03	-3.1955366e-04	1.8303812e-03
	1.7744815e-01	-6.8792732e-02	5.1087038e-03	8.2285381e-04	-7.5366066e-05	2.0172927e-03
	1.2917189e-01	1.7510493e-02	5.8319505e-02	-2.8877083e-06	-1.3831180e-05	1.5282397e-03
	1.3561558e-01	3.8867613e-03	8.8903376e-02	6.1257401e-04	-2.0483346e-04	1.6858282e-03
	1.5266451e-01	1.3644321e-02	1.3092410e-02	1.41526926e-04	3.26510861e-03	

Realignment parameters

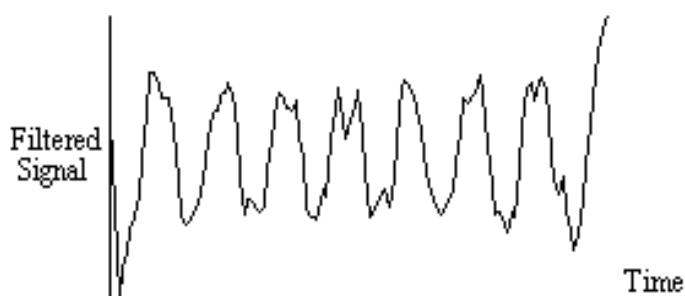
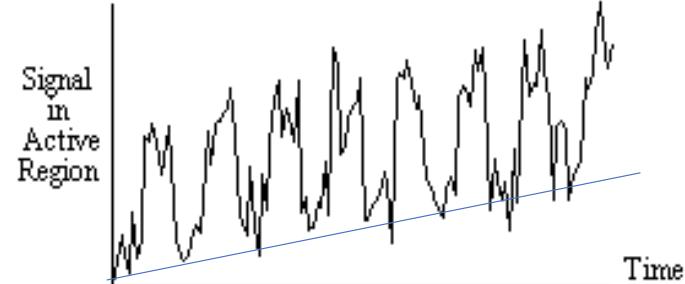
But ... the fMRI signal

$$Y = \beta \cdot X + \gamma \cdot RS + \delta \cdot N + \epsilon \cdot SN + \varepsilon$$



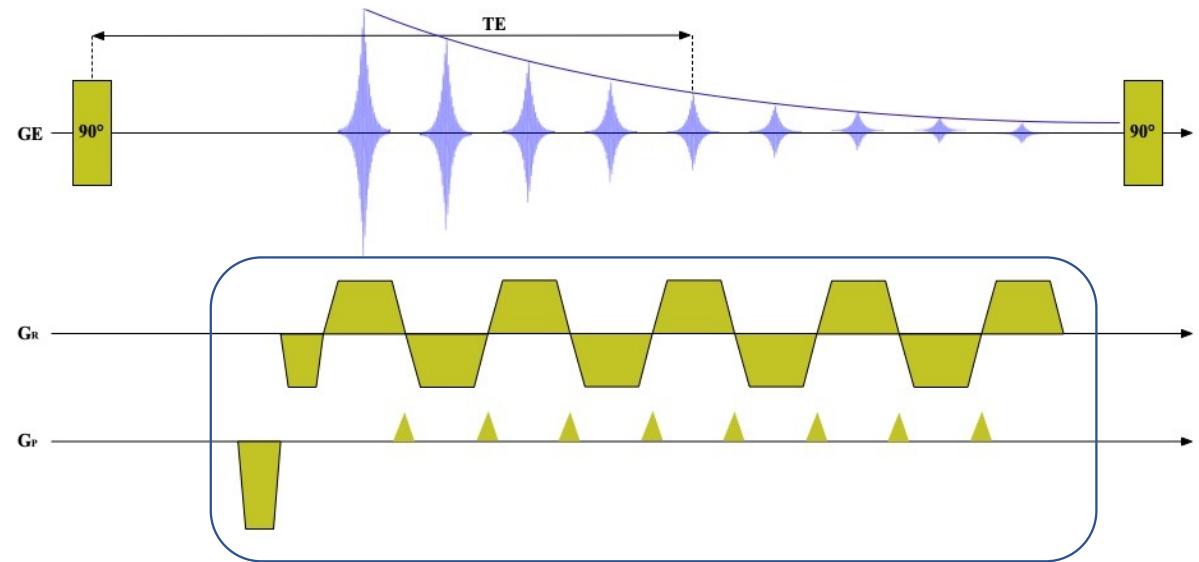
Denoising

fMRI signal drift

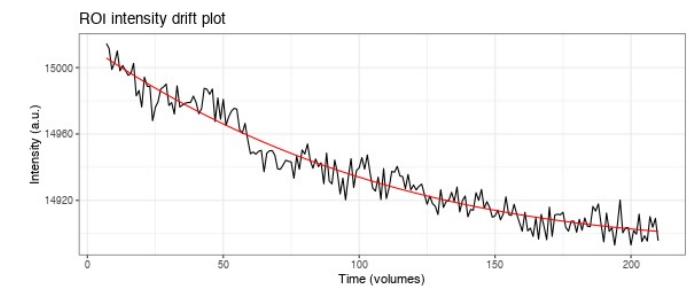
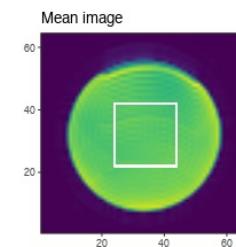


Subtracting a trendline

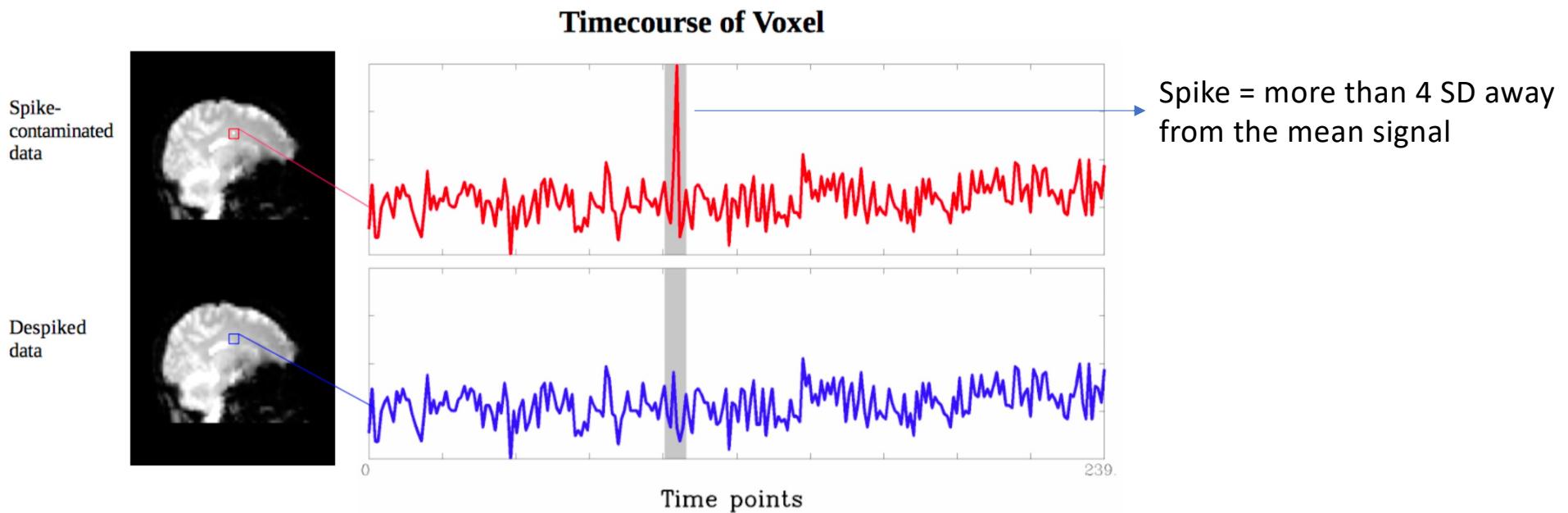
or by doing a high pass filter (done in SPM)



Repeated fast gradient switching -> heating of the gradient coil



fMRI signal spikes: abnormal high or low signals due to hardware errors or severe head motion



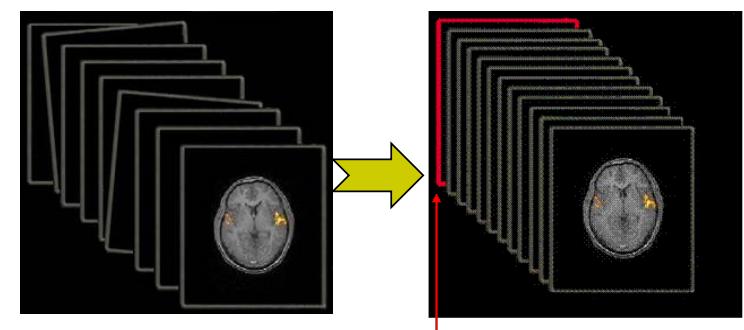
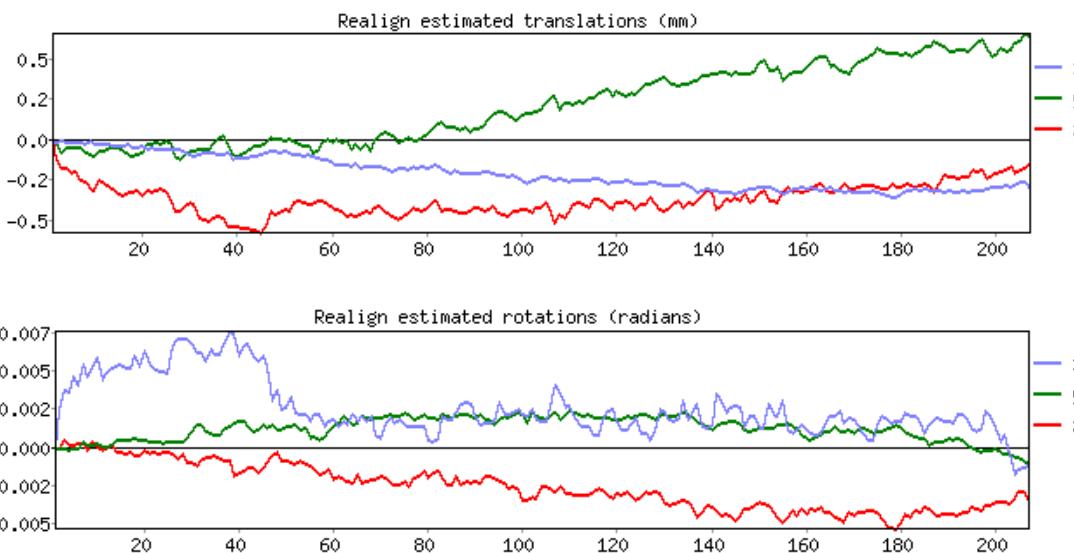
Solutions:

Scrubbing: nulling (removing) bad volumes

Despiking: (voxelwise) truncation of fMRI time series

Affects the temporal correlation of the fMRI signal

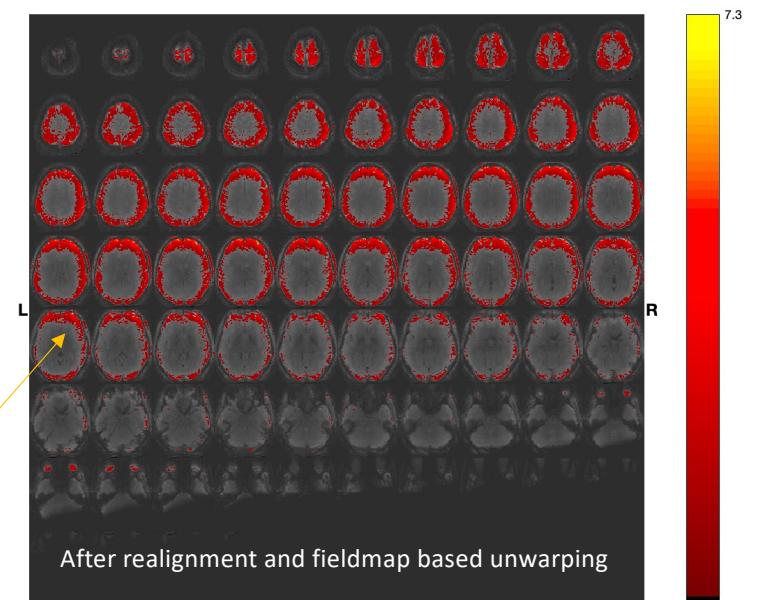
Head motion



realignment

Motion effects on the signal:

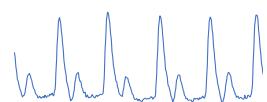
- The signal in a voxel is coming from a collection of neighboring voxels (most visible at brain edges)
- Since the susceptibility artifact at the sinuses depends on the position and orientation of the head in the magnetic field B_0 , motion affect the susceptibility artifact
- Spins that move from one slice to the other will get excited at irregular intervals -> effects on their signal saturation



Respiration



Pulsatile flow (cardiac)



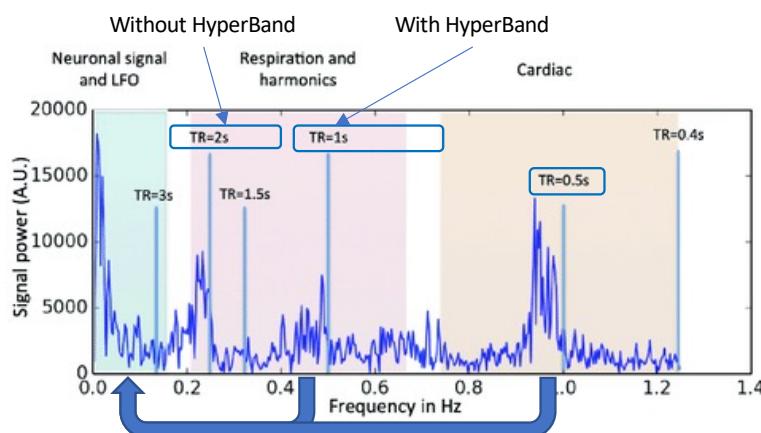
Non-BOLD effects: motion

BOLD effects: dynamic changes in OHb/dHB and CBF

Noise BOLD signals @ $f \in [0.8, 1.2]$ Hz for cardiac and $f \in [0.2, 0.7]$ Hz for respiration

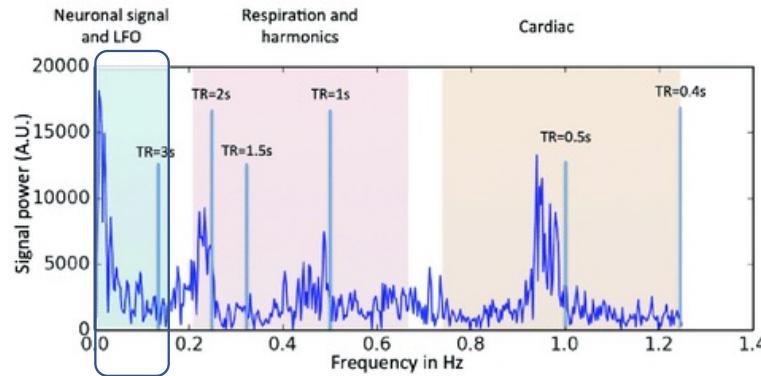
But if TR>0.5s -> aliasing into $f < 0.2$ Hz

Overlap with task and resting state BOLD frequencies



Denoising

Bandpass filtering



High-pass: $f > 0.01$ (drift)

Low-pass: $f < 0.1$ (respiratory and cardiac noise)

Noise regression

Prior to the analysis

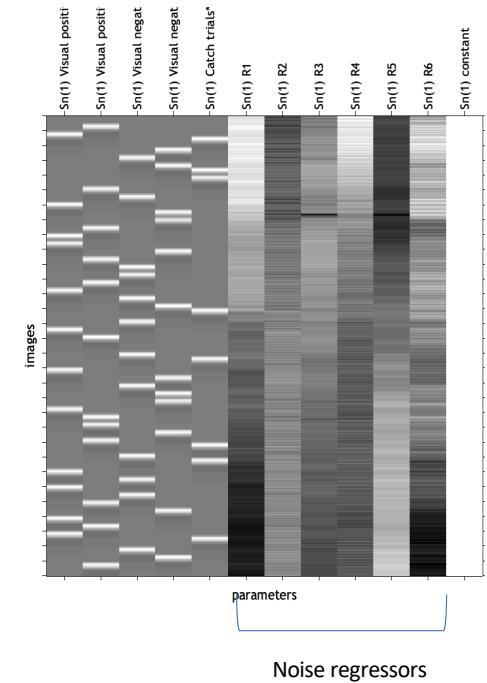
$$Y_c = Y - \gamma \cdot N$$

Resting state analysis

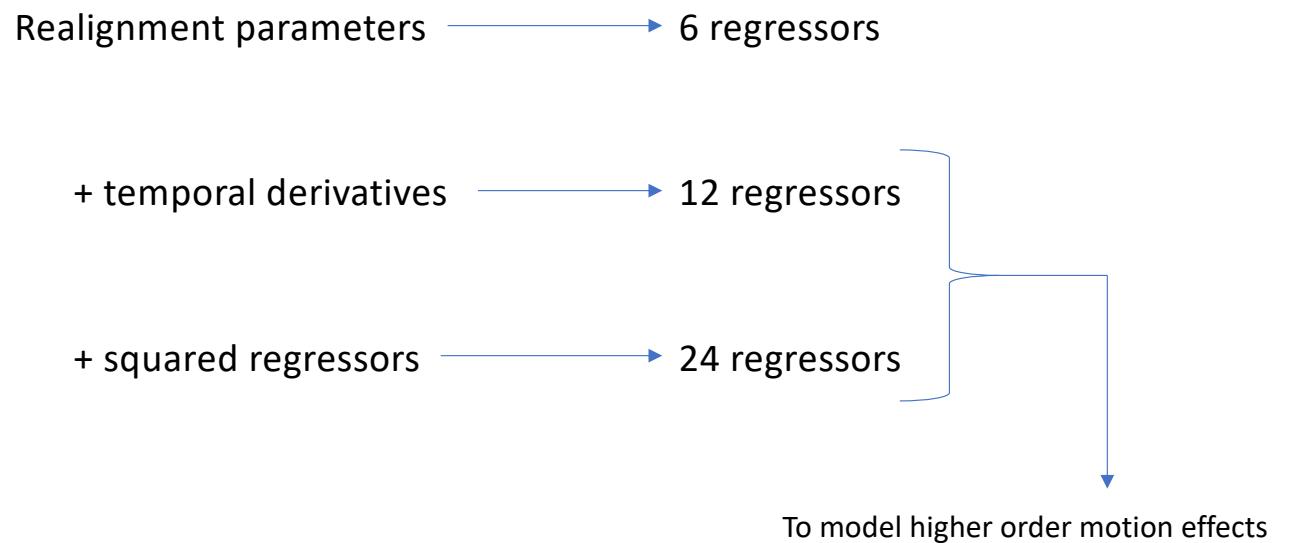
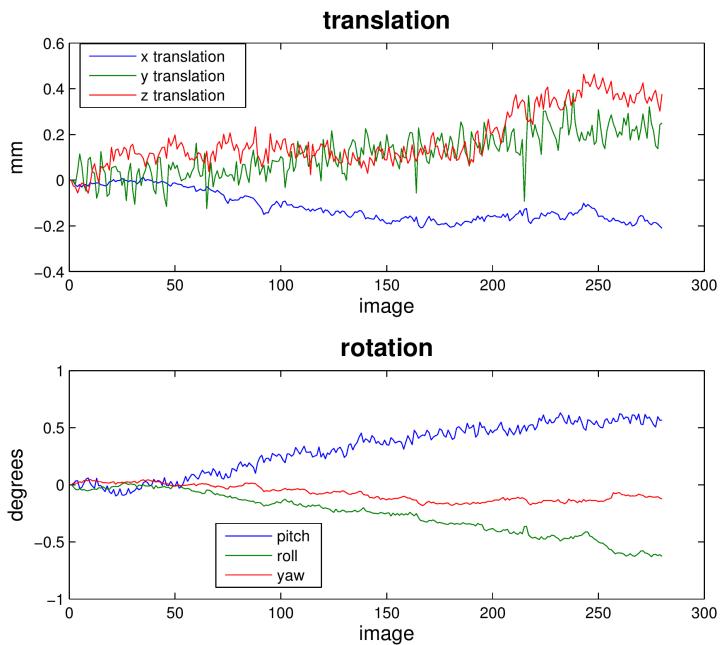
GLM regressors

$$Y = \beta \cdot X$$

Which noise regressors?

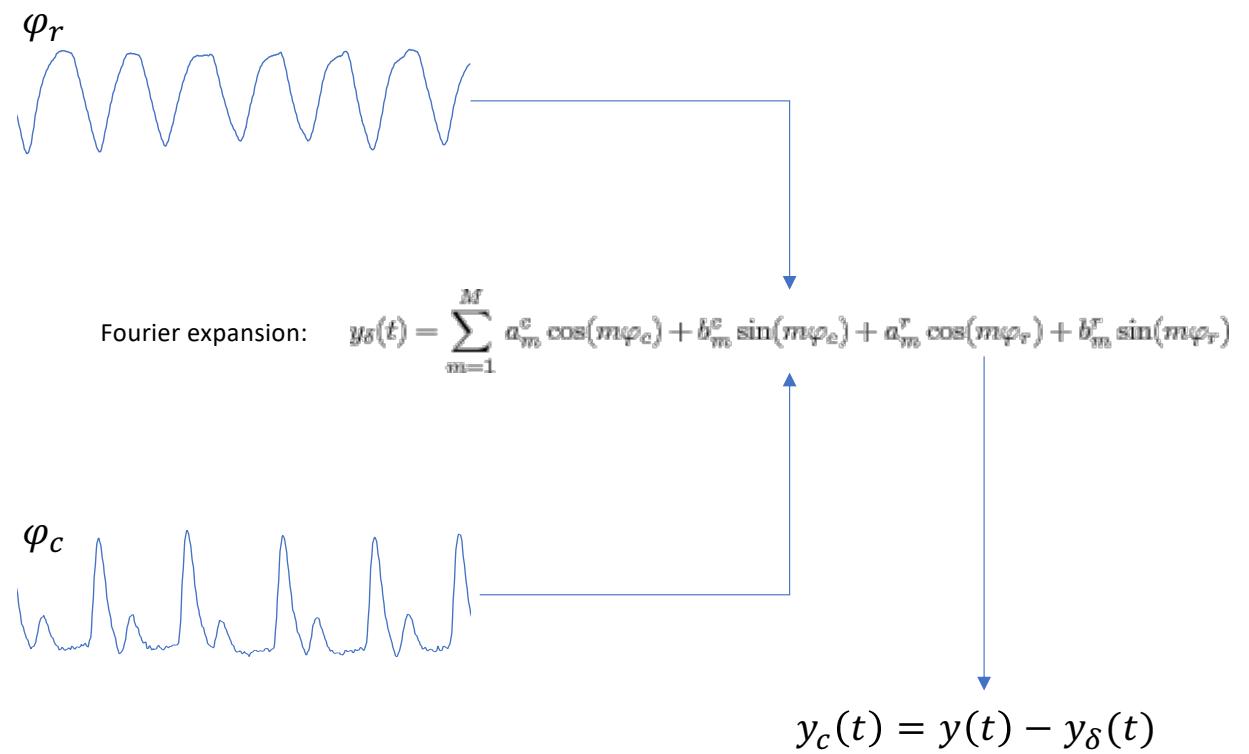
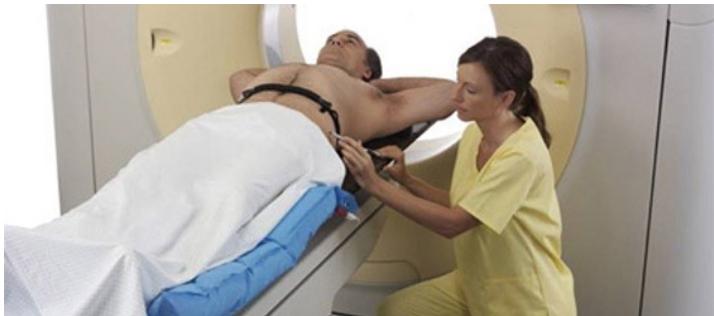


Motion regressors



Caution: the number of regressors affects the degrees of freedom of the statistical analysis!

RETROICOR

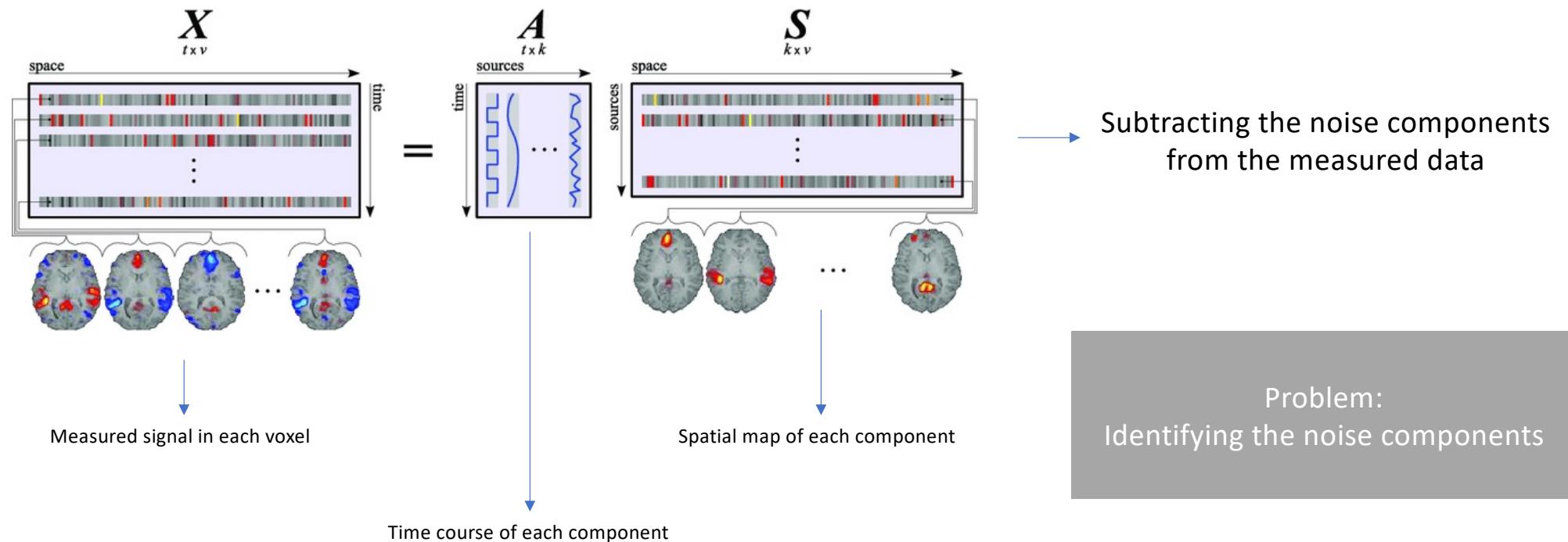


In our task fMRI experiment, we only added the 6 motion regressors as noise regressors because

1. We can not determine the physiologic noise signals in task fMRI
2. Physiologic noise does not correlate with the task
3. Adding more noise regressors to the design matrix can reduce the study outcome
4. The effect of adding more noise regressors to the design matrix is negligible

Independent component analysis (ICA) based denoising

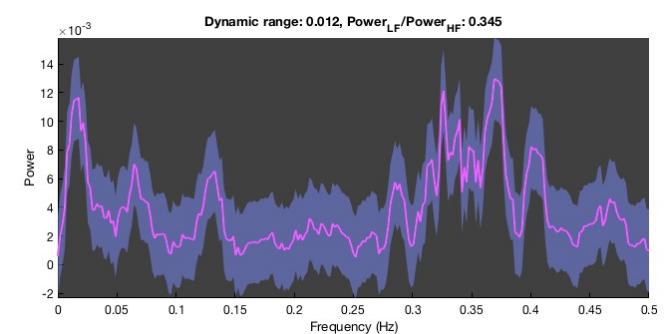
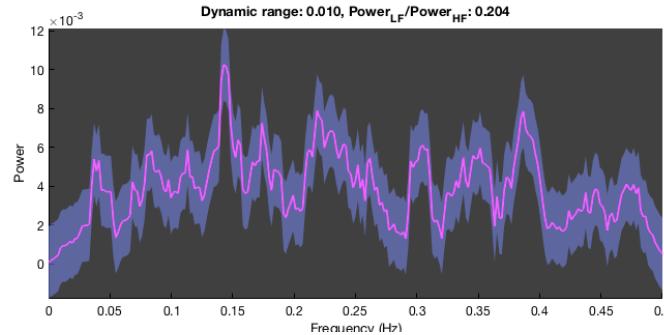
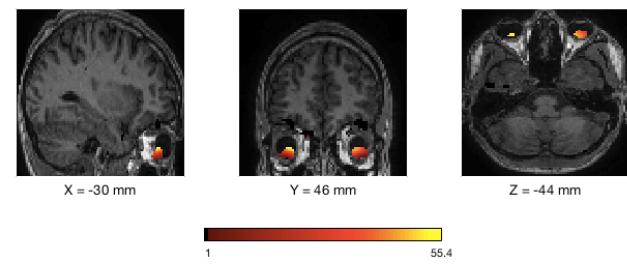
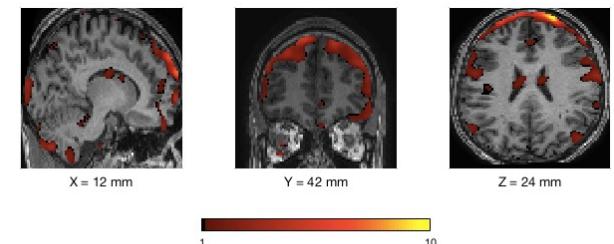
Assumption: The signal in each voxel is the weighted sum of a limited number of independent components (task, resting state and noise components)



ICA based denoising: noise component identification

Based on the spatial location: brain edges, CSF, non-brain areas

Based on high (> 0.1 Hz) frequency content

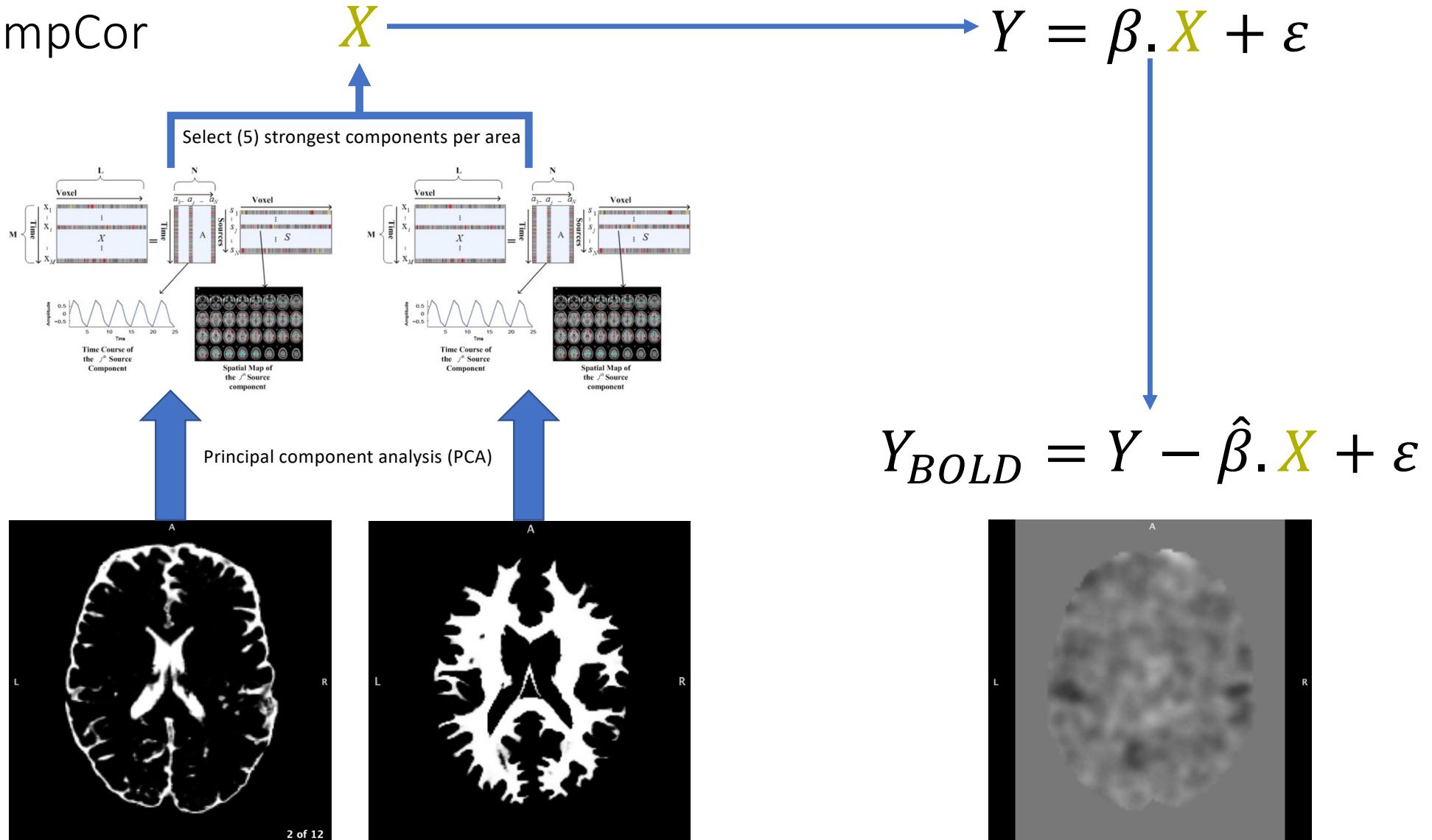


Manual → time consuming and rater dependent

ICA-AROMA → automatic motion artifact removal

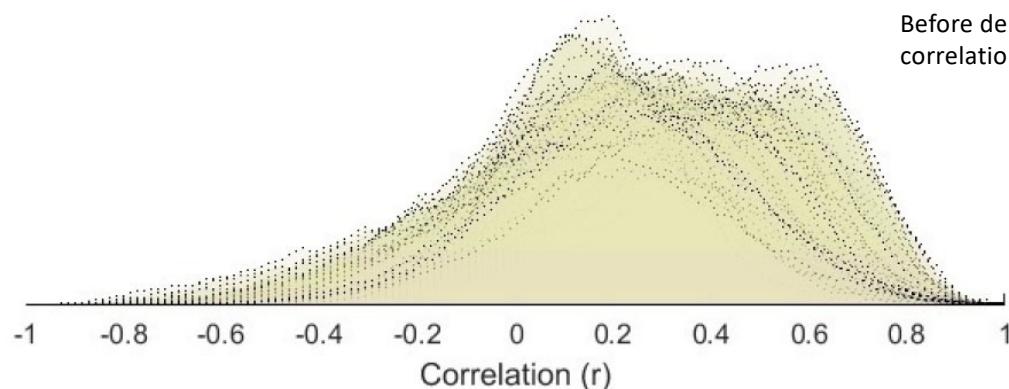
Deep learning (DL) or AI based (e.g. Melodic) → training of the algorithm

(a)CompCor



Effect of denoising

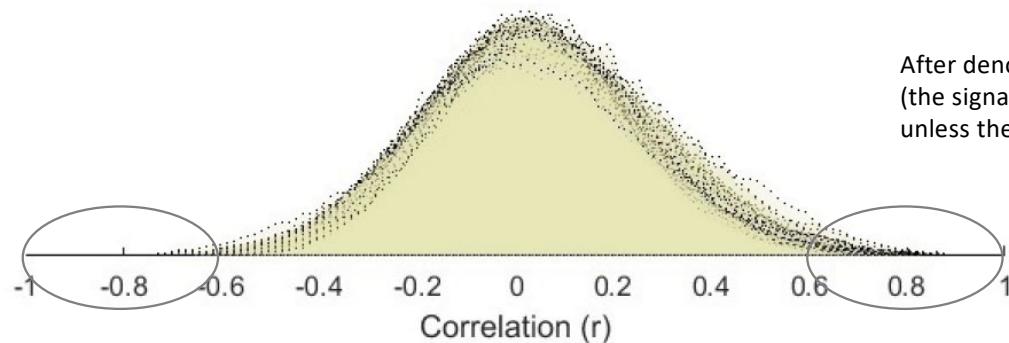
Connectivity histogram before denoising



Before denoising: skewed distribution due to correlations induced by noise regressors

Evaluation of the denoising step:
Connectivity values between
random selected voxel pairs

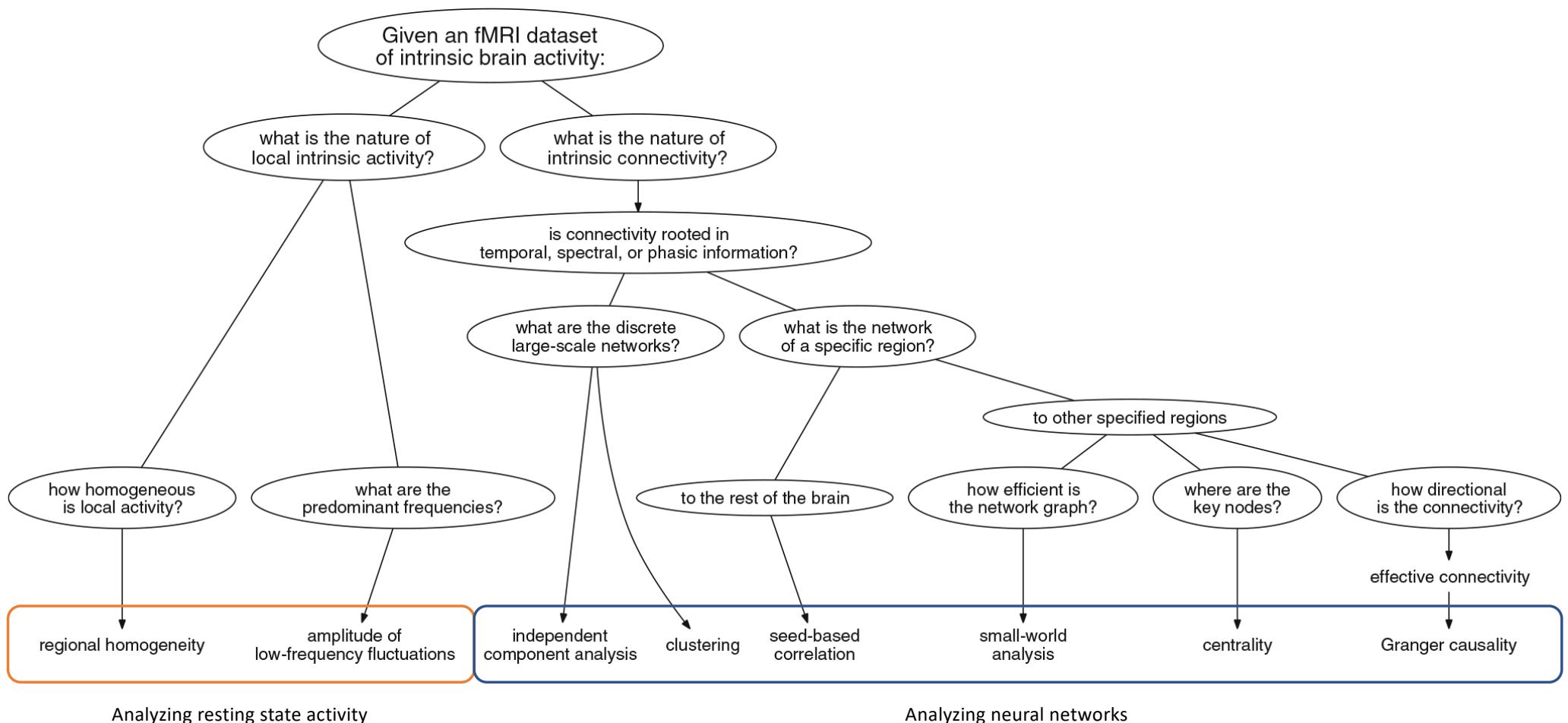
Connectivity histogram after denoising



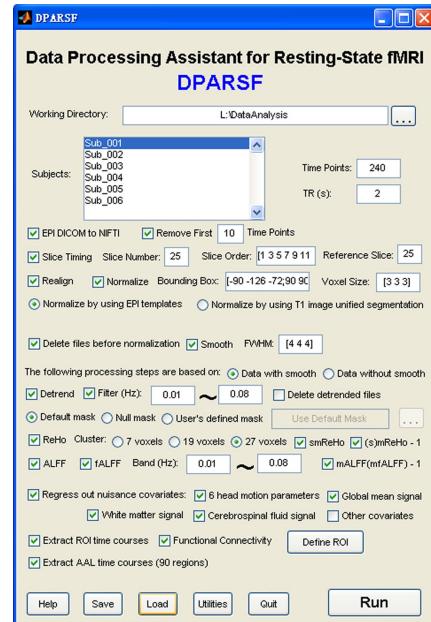
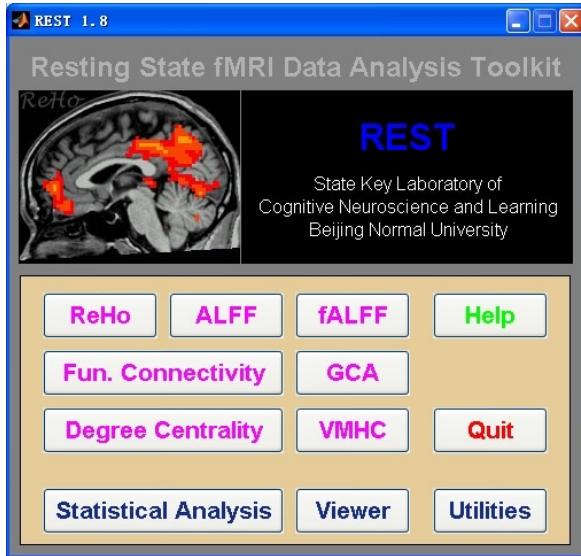
After denoising: normally distributed correlations
(the signals of 2 randomly selected voxels normally do not correlate unless they are functionally connected)

Analysis of RS-fMRI

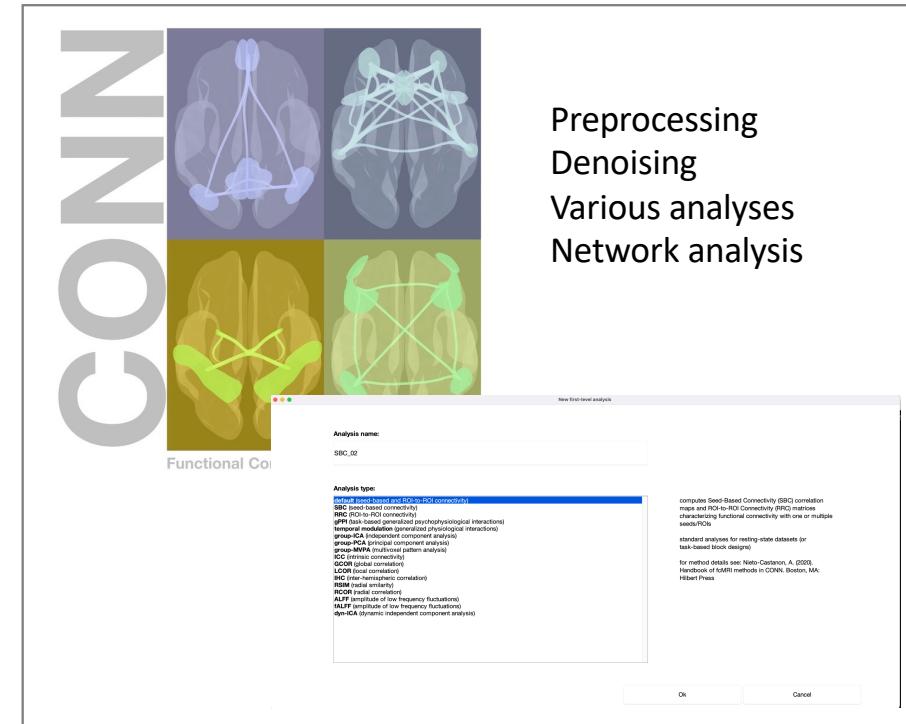
RS-fMRI analysis



RS-fMRI toolboxes

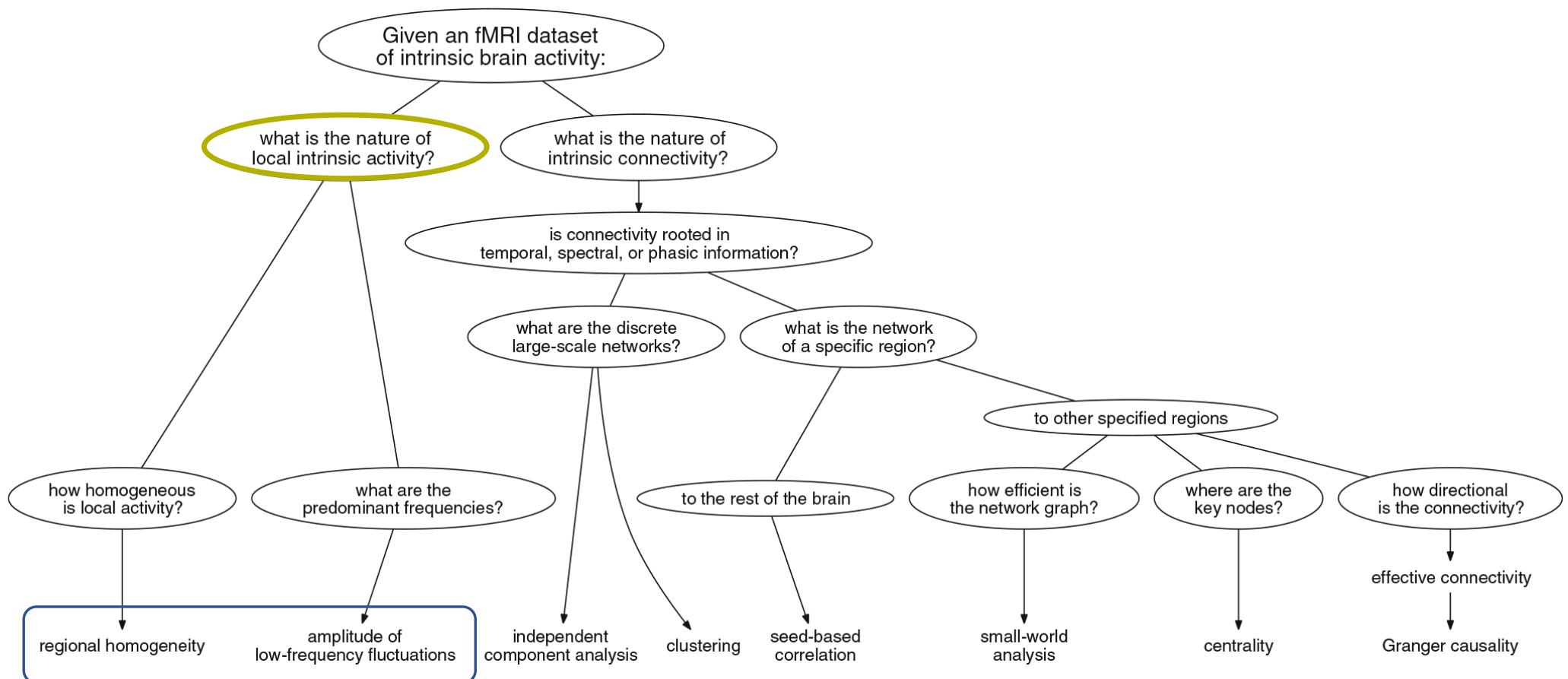


Preprocessing
Basic denoising (detrend+filter)
Connectivity analyses



Only ICA

RS-fMRI analysis



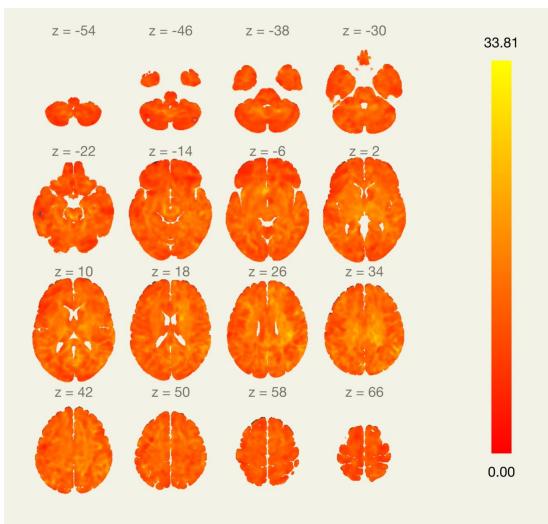
(Fractional) Amplitude of Low-Frequency Fluctuations ((f)ALFF)

(f)ALFF is an R-fMRI indicator that is used to detect **the regional intensity of spontaneous fluctuations in the BOLD signal**, which pinpoints the spontaneous neural activity of specific regions and physiological states of the brain.

ALFF maps represent a measure of BOLD signal power within the frequency band of interest (e.g. 0.01 - 0.10 Hz).
ALFF is defined as the root mean square of BOLD signal at each individual voxel after low- or band- pass filtering:

fALFF maps represent a relative measure of BOLD signal power within the frequency band of interest (e.g. 0.01 - 0.10 Hz) compared to that over the entire frequency spectrum.

fALFF is defined as the ratio of root mean square of BOLD signal at each individual voxel after vs. before low- or band- pass filtering:



$$ALFF(x) = \sqrt{\frac{1}{N} \cdot \sum_t (h(t) * S(x, t))^2}$$

$$fALFF(x) = \sqrt{\frac{\sum_t (h(t) * S(x, t))^2}{\sum_t S(x, t)^2}}$$

S is original BOLD timeseries before band- or low- pass filtering
h is a low- or band-pass filter
N is the number of timepoints

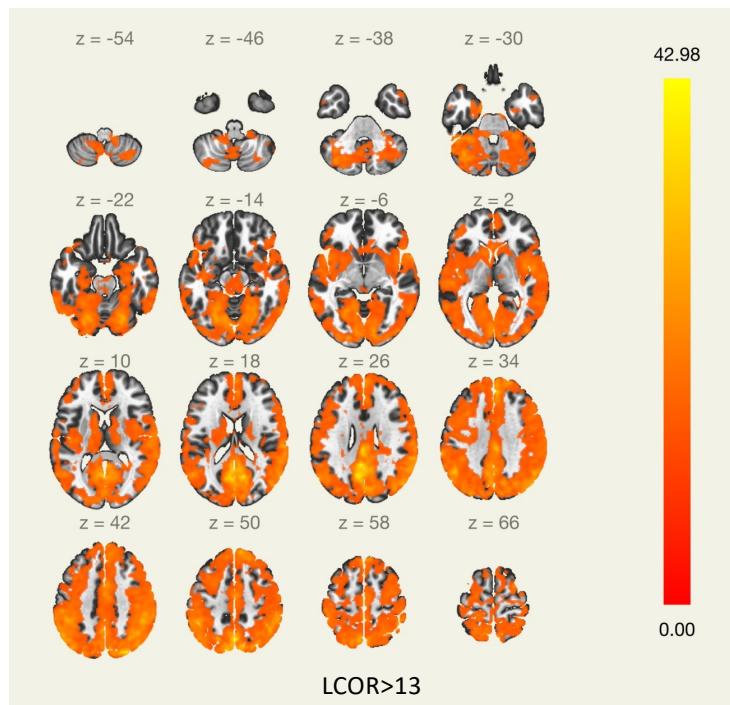
Different sensitivity and reliability for changes due to aging, disease states and treatment effects

↓
Best to look at both

Local correlation (LCOR) = Regional Homogeneity (ReHo)

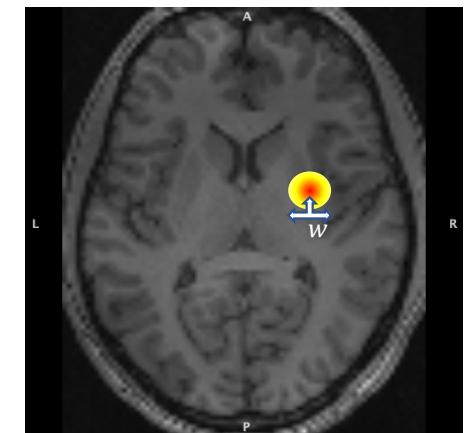
LCOR maps represent a measure of local coherence at each voxel, characterized by the strength and sign of connectivity between a given voxel and its neighboring areas.

LCOR is defined as the average of correlation coefficients between each individual voxel and a region of neighboring voxels

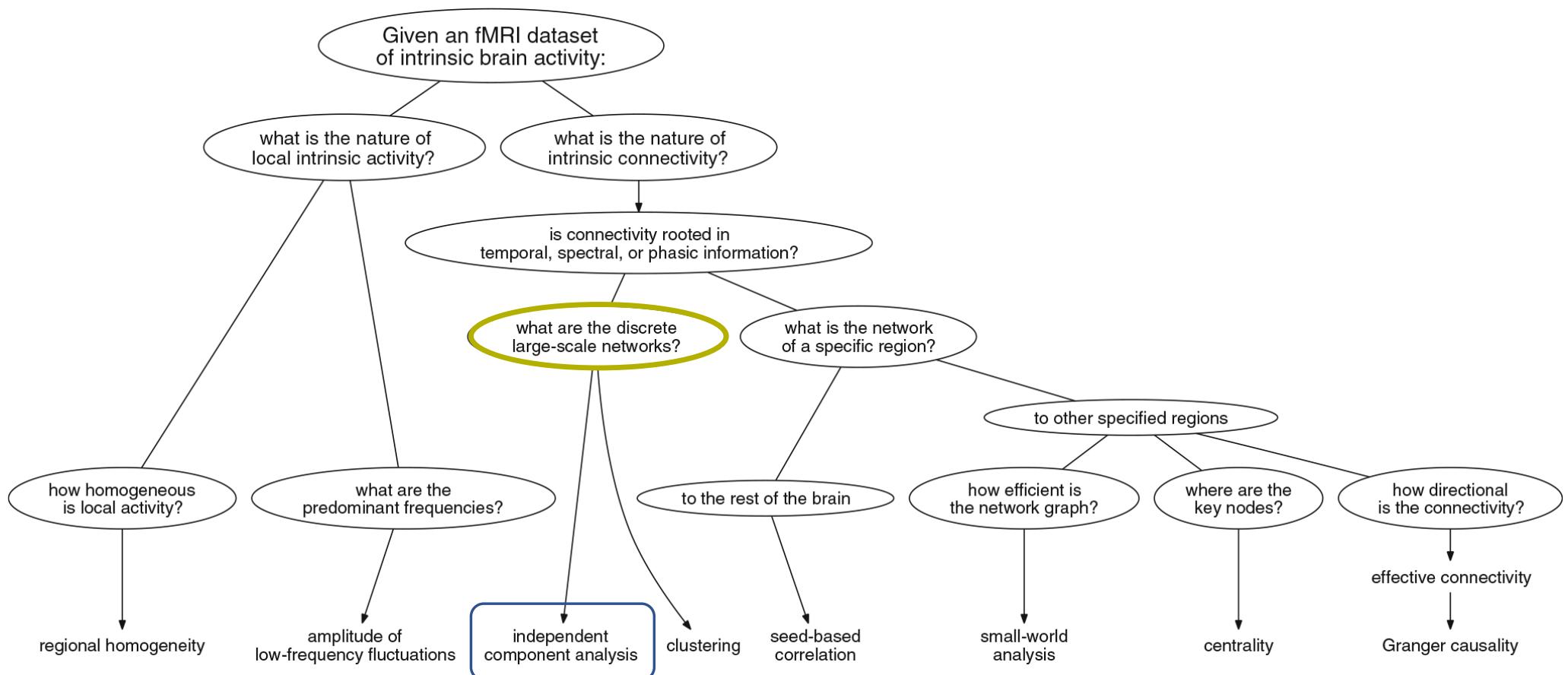


$$LCOR(x) = \frac{\int w(x - y)r(x, y)dy}{\int w(x - y)dy}$$

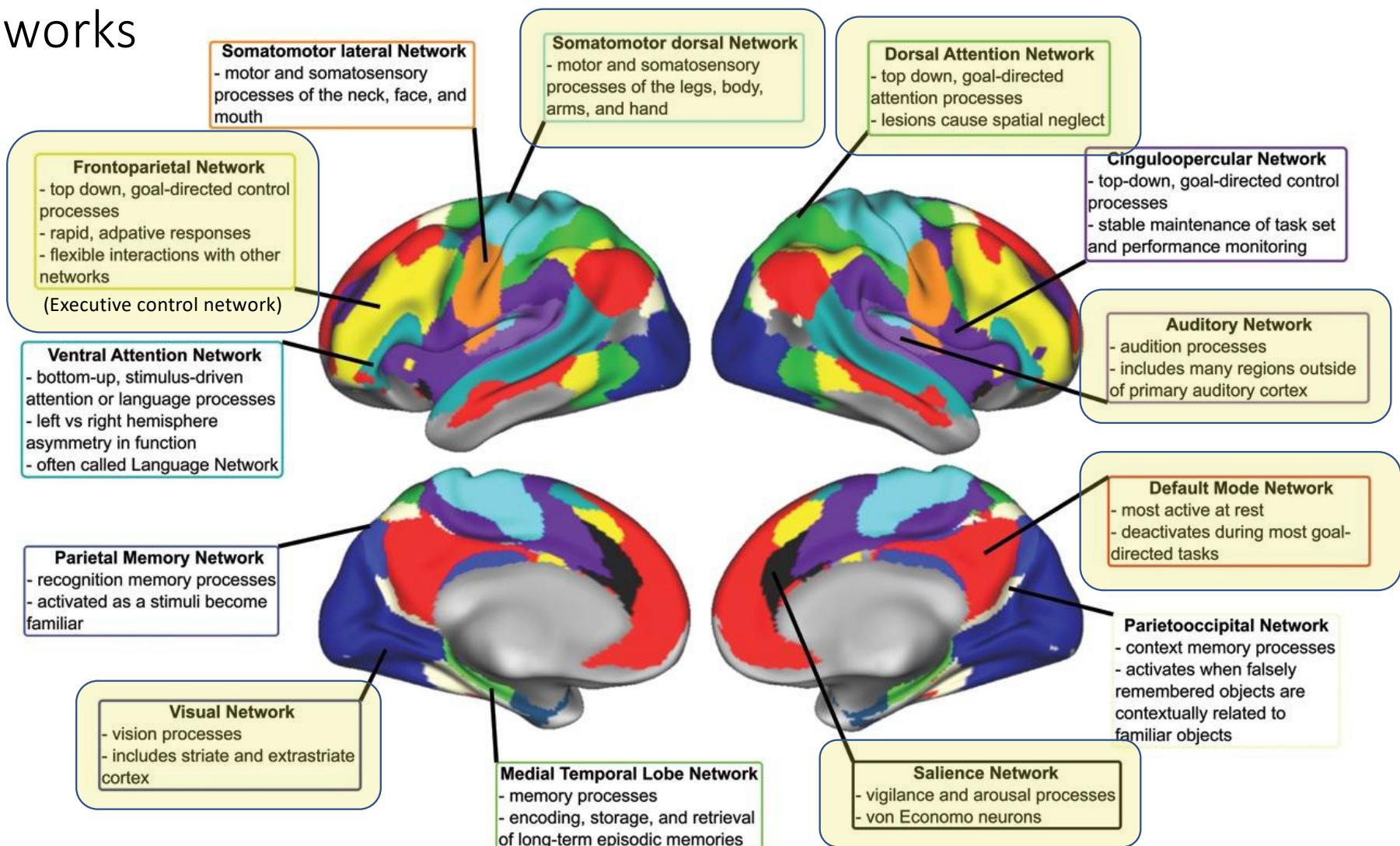
w is an isotropic Gaussian weighting function with size σ characterizing the size of the local neighborhood ($w(z) = e^{-\frac{|z|^2}{2\sigma^2}}$)



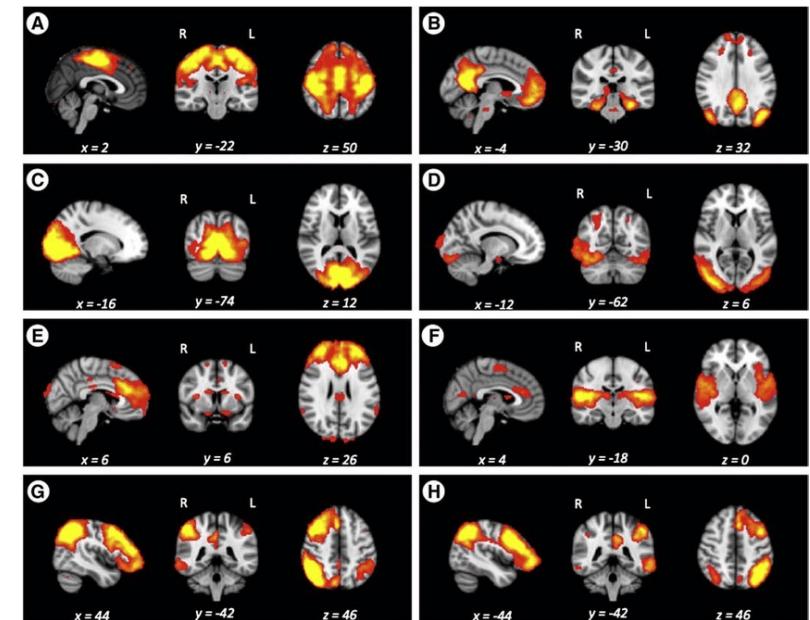
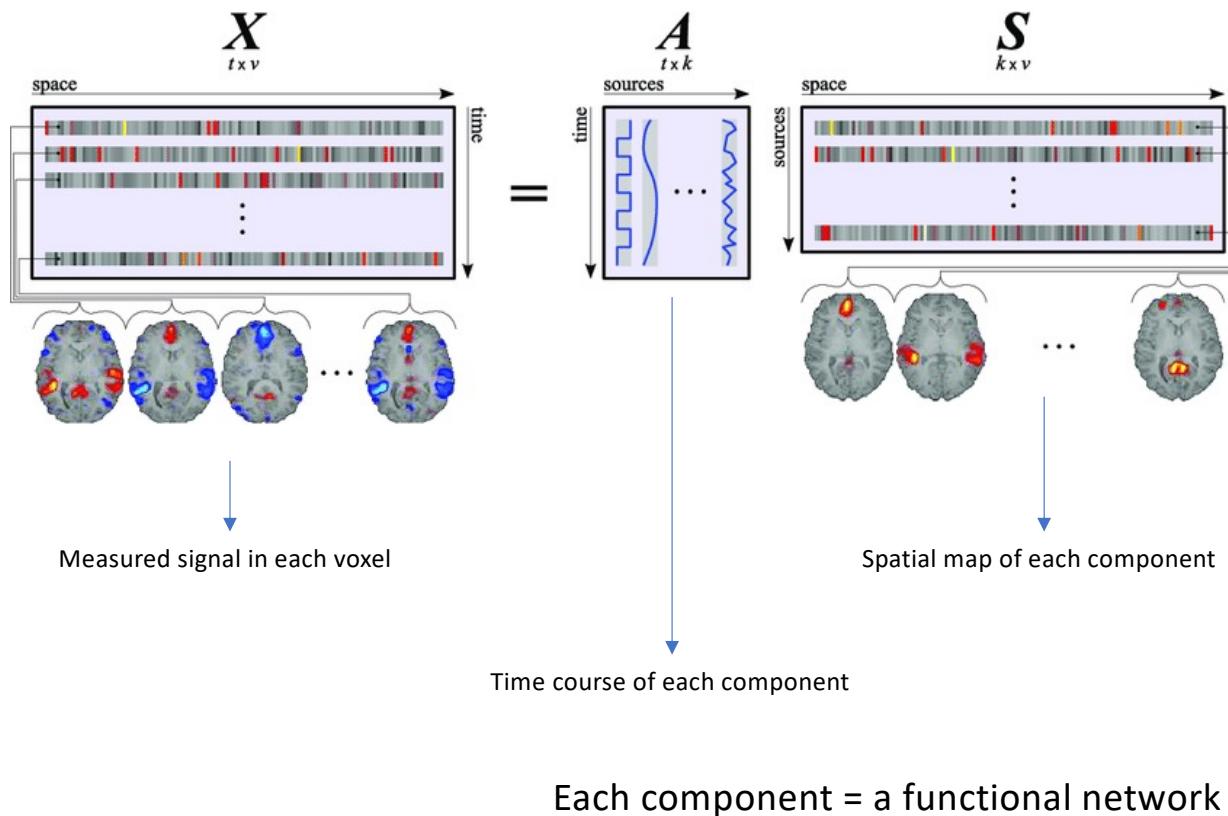
RS-fMRI analysis



Brain networks

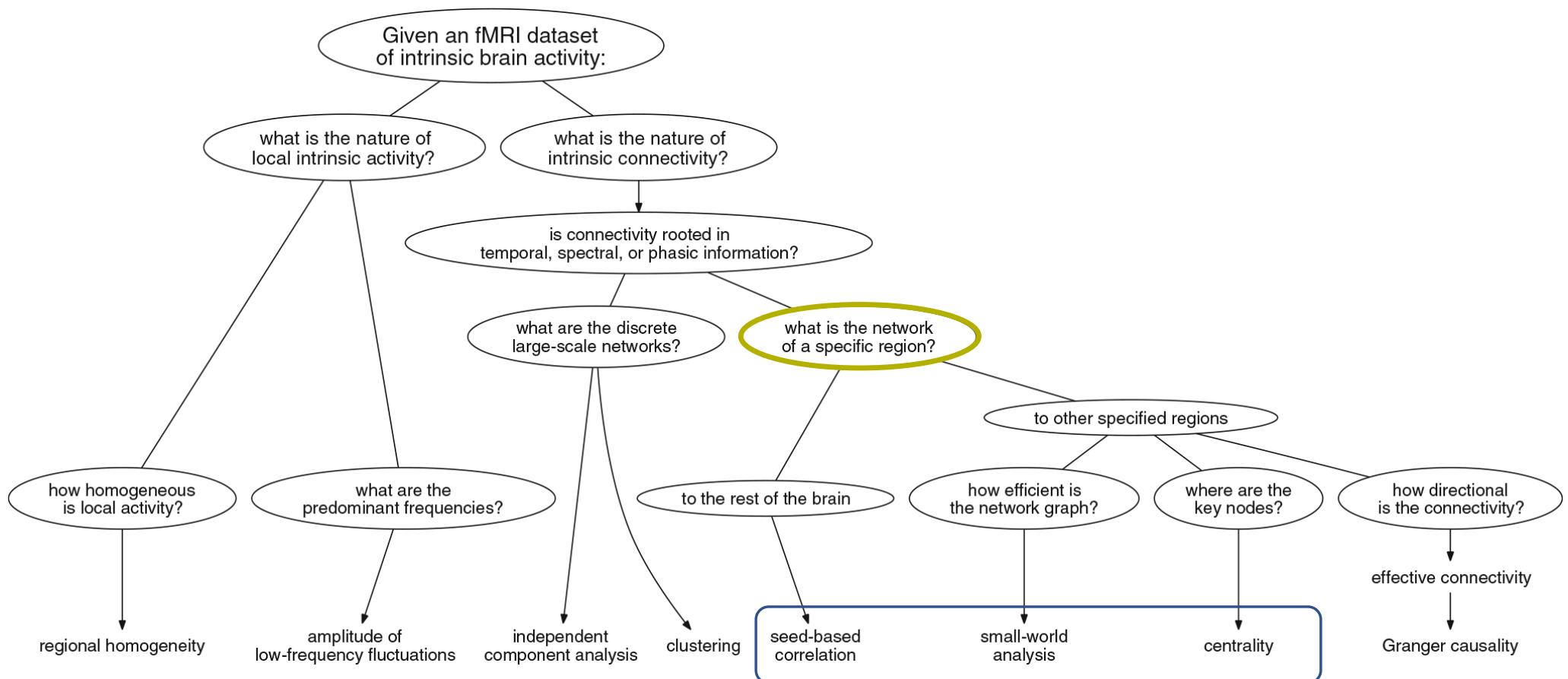


Splitting the fMRI signals in independent networks = ICA



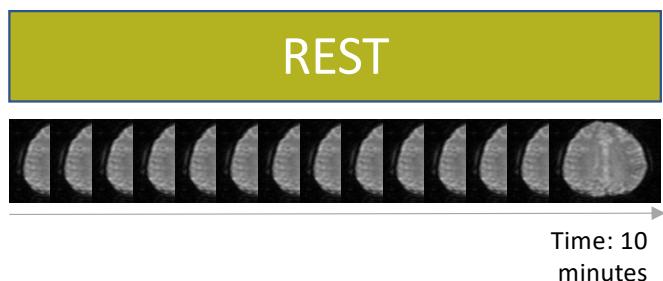
The resting state networks. Group mean (A) motor, (B) default mode, (C) medial visual, (D) lateral visual, (E) executive, (F) auditory, (G) right dorsal visual stream, and (H) left dorsal visual stream networks.

RS-fMRI analysis



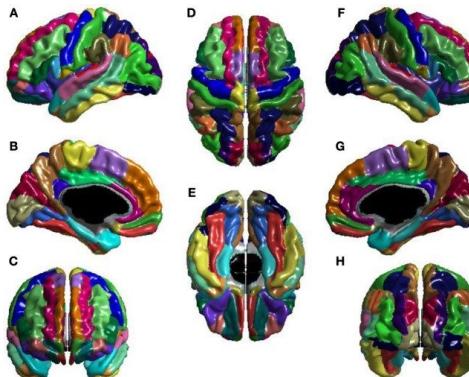
Seed-based connectivity (SBC) / seed-to-voxel connectivity

A

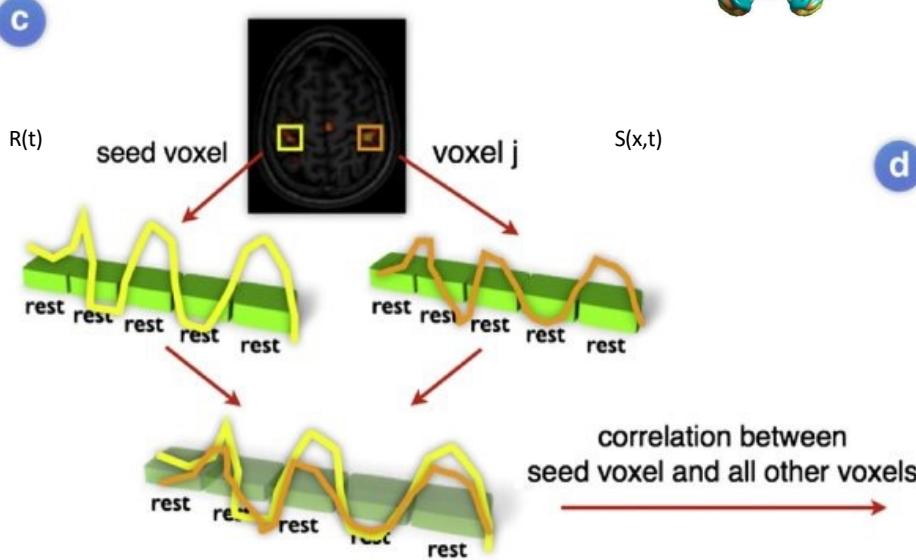


B

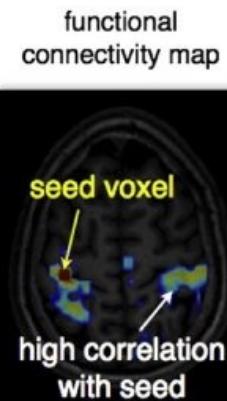
Choose 1 area from the 170 anatomical regions from the MNI atlas as seed area



C



d



SBC calculates in each voxel the correlation between its temporal signal and the temporal signal of a chosen seed region.

$$r(x) = \frac{\int S(x, t)R(t)dt}{\sqrt{\int R^2(t)dt \int S^2(x, t)dt}}$$

S is the BOLD timeseries at each voxel
 R is the average BOLD timeseries within a ROI
 r is the spatial map of Pearson correlation coefficients

Seed-based connectivity (SBC): Default mode network (DMN)

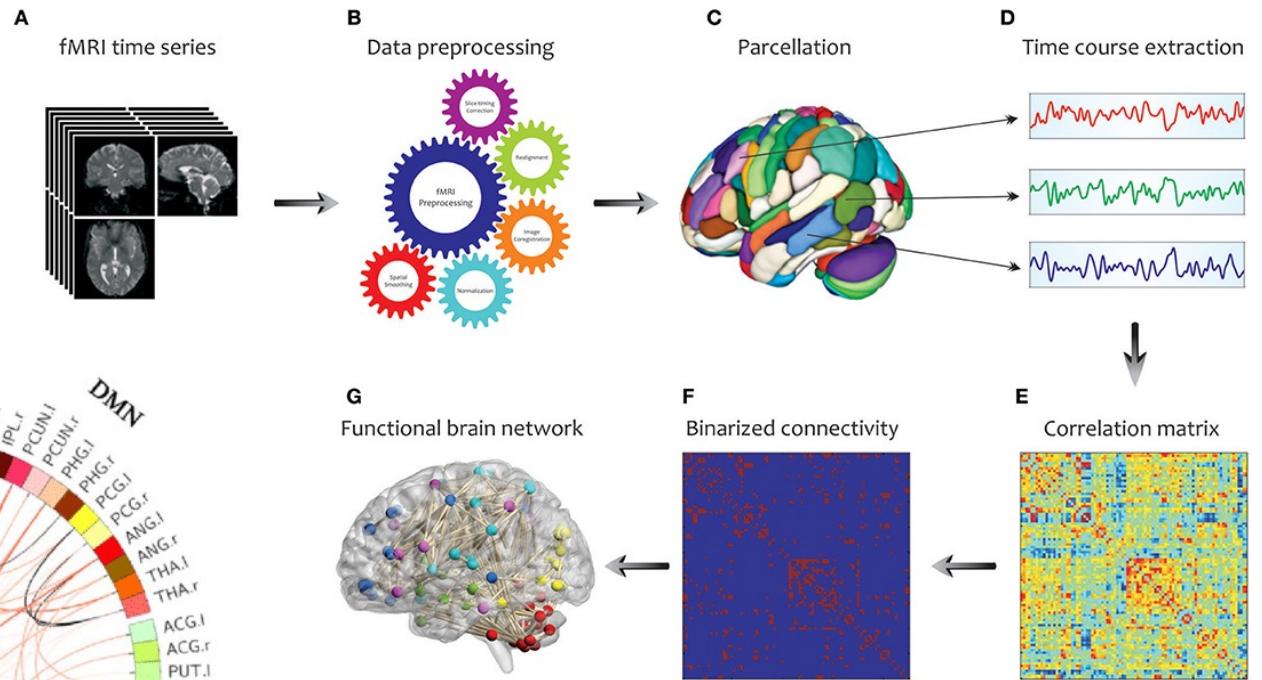
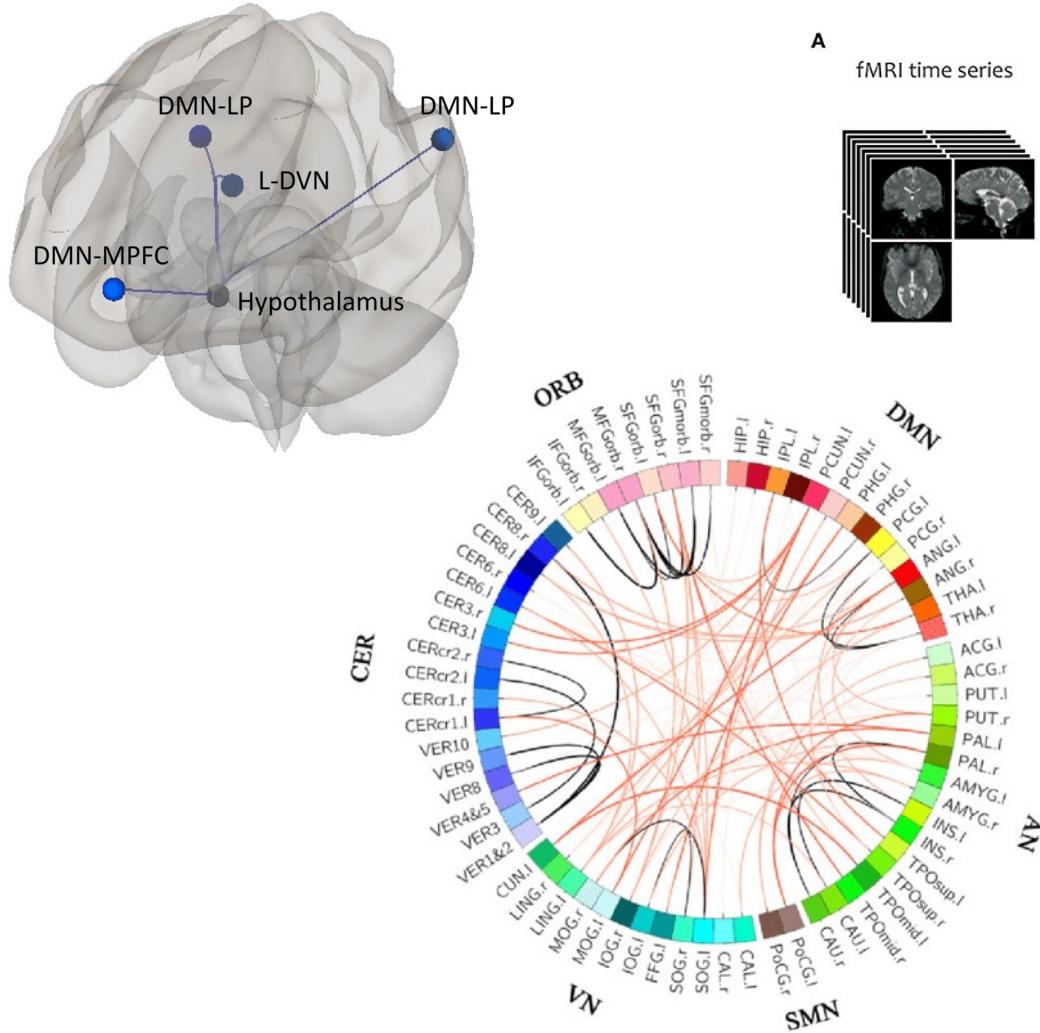


Seed: MPFC



Seed: PCC

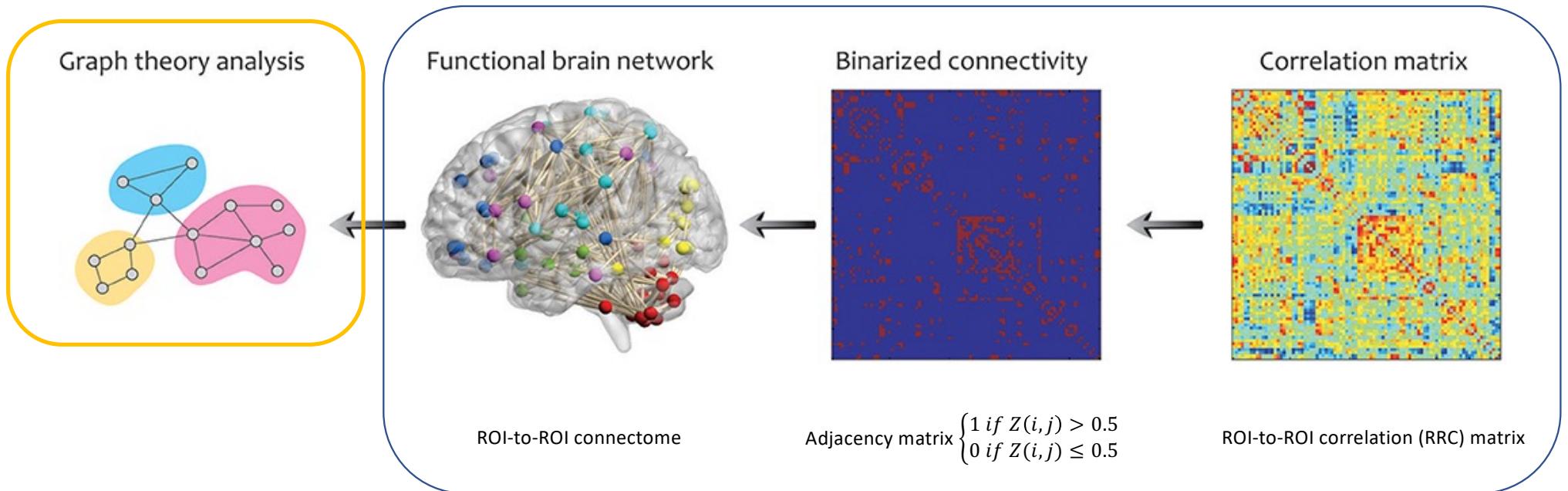
ROI-to-ROI connectivity (RRC)



$$r(i,j) = \frac{\int R_i(t)R_j(t)dt}{\sqrt{\int R_i^2(t)dt \int R_j^2(t)dt}}$$

R is the BOLD timeseries within each ROI
 r is a matrix of correlation coefficients

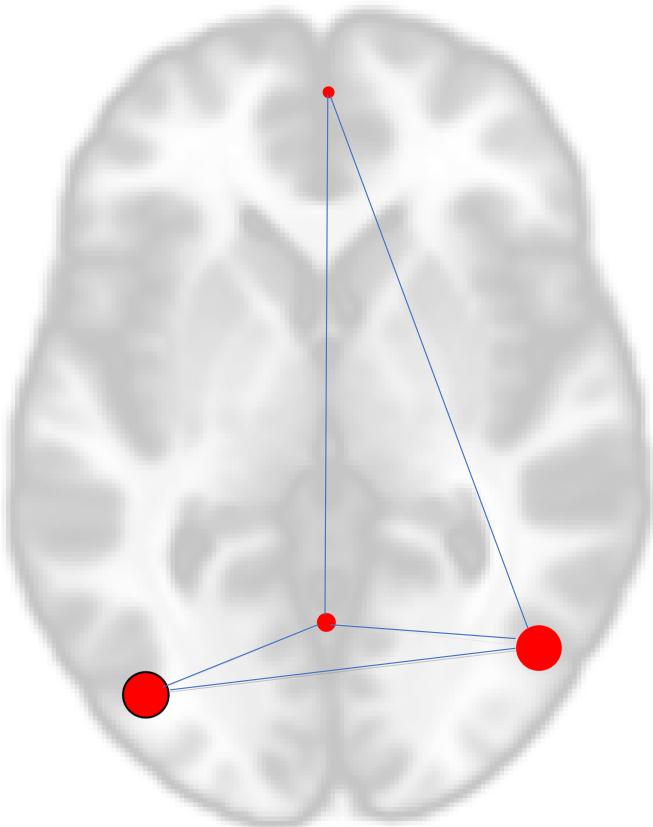
ROI-to-ROI connectivity -> functional networks



Graph theory based network measures

Nodes = ROIs in the graph

Edges = connections between the nodes



Degree and Cost at each node/ROI represent **measures of network centrality**, characterizing **the degree of local connectedness** of each ROI within a graph

Degree = number of edges from/to each node

$$\text{At ROI level: } d_i = \sum_j A_{i,j}$$

$$\text{At network level: } d = \frac{\sum_i d_i}{N}$$

A is the adjacency matrix

N is the total number of nodes

Cost = proportion of edges from/to each node

$$\text{At ROI level: } c_i = \frac{\sum_j A_{i,j}}{N-1}$$

$$\text{At network level: } c = \frac{\sum_i c_i}{N}$$

Average path distance and Global efficiency at a node represents a measure of **this node centrality within the network**, characterizing **the degree of global connectedness** of each ROI.

Average path distance = average number of edges traversed in an optimal path between one node and an other node

$$\text{At ROI level: } L_i = \frac{\sum_{j \in \Omega_i} D_{i,j}}{N-1}$$

D is the shortest-path distance matrix

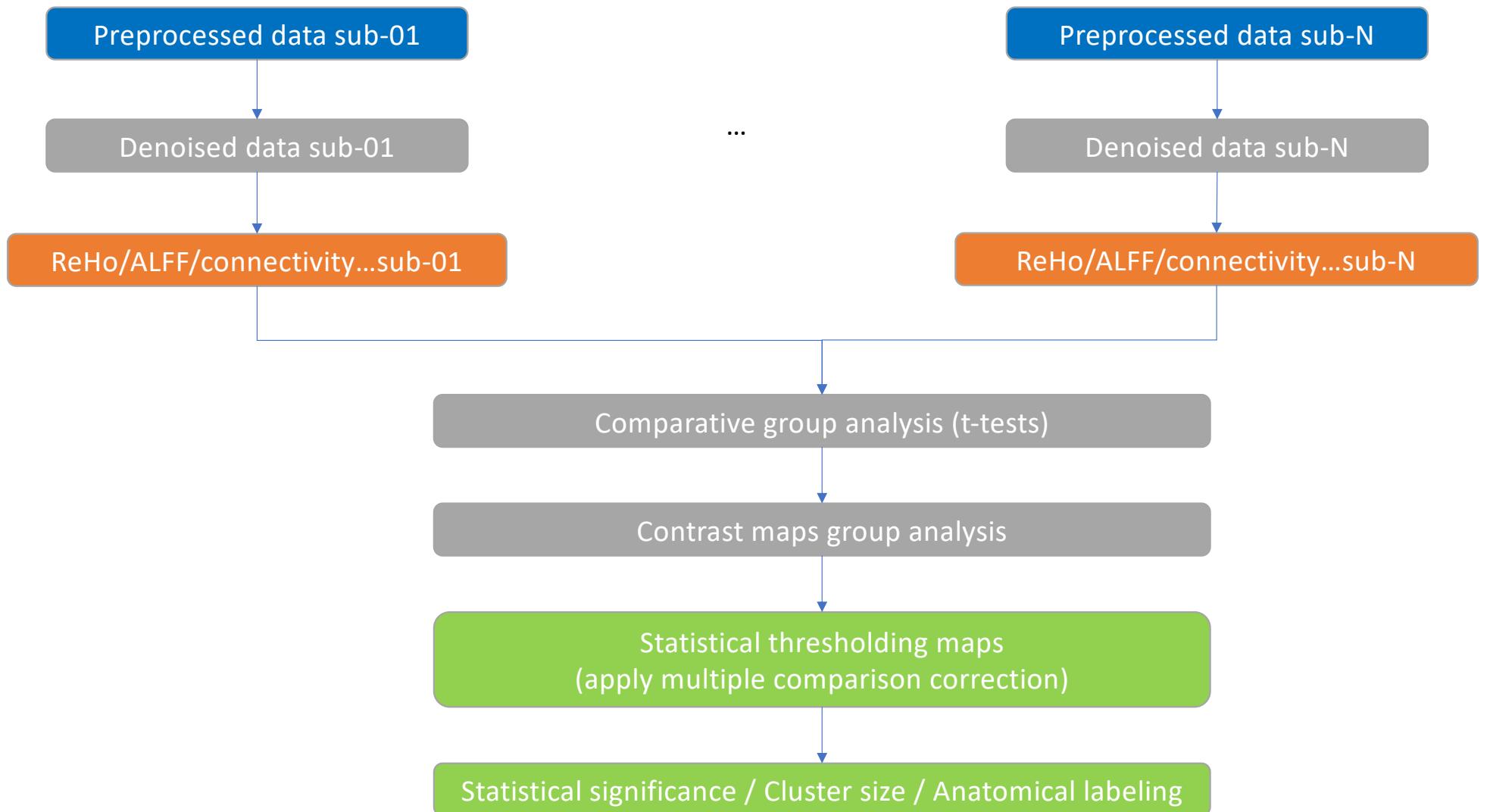
N is the total number of nodes

$$\text{At network level: } L = \frac{\sum_i L_i}{N}$$

Global efficiency = average inverse distance between a node and all other nodes

$$\text{At ROI level: } GE_i = \frac{\sum_{j \neq i} 1/D_{i,j}}{N-1}$$

$$\text{At network level: } GE = \frac{\sum_i GE_i}{N}$$



DANK U - THANK YOU