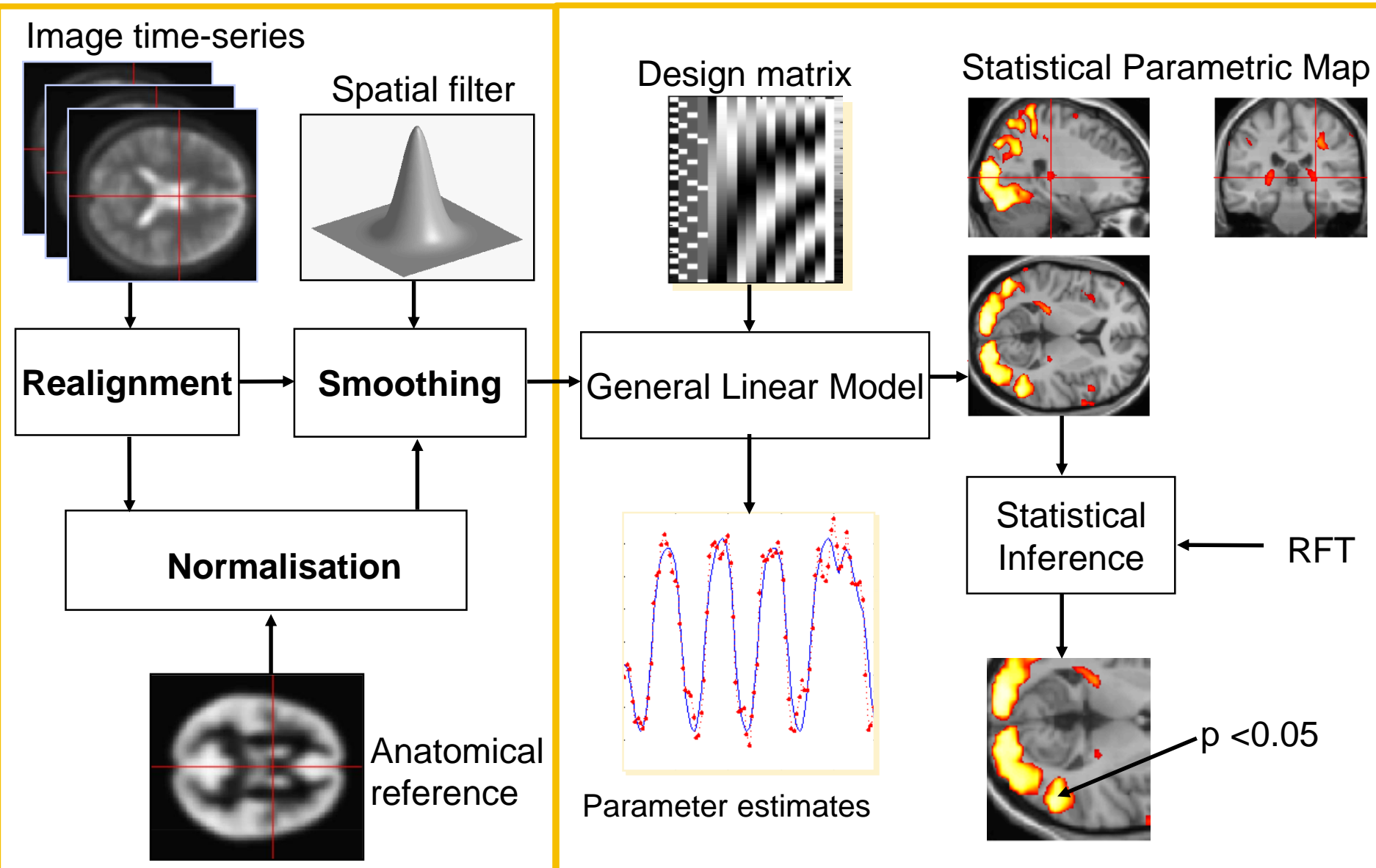


Pre-processing: Co-registration, spatial normalisation and smoothing

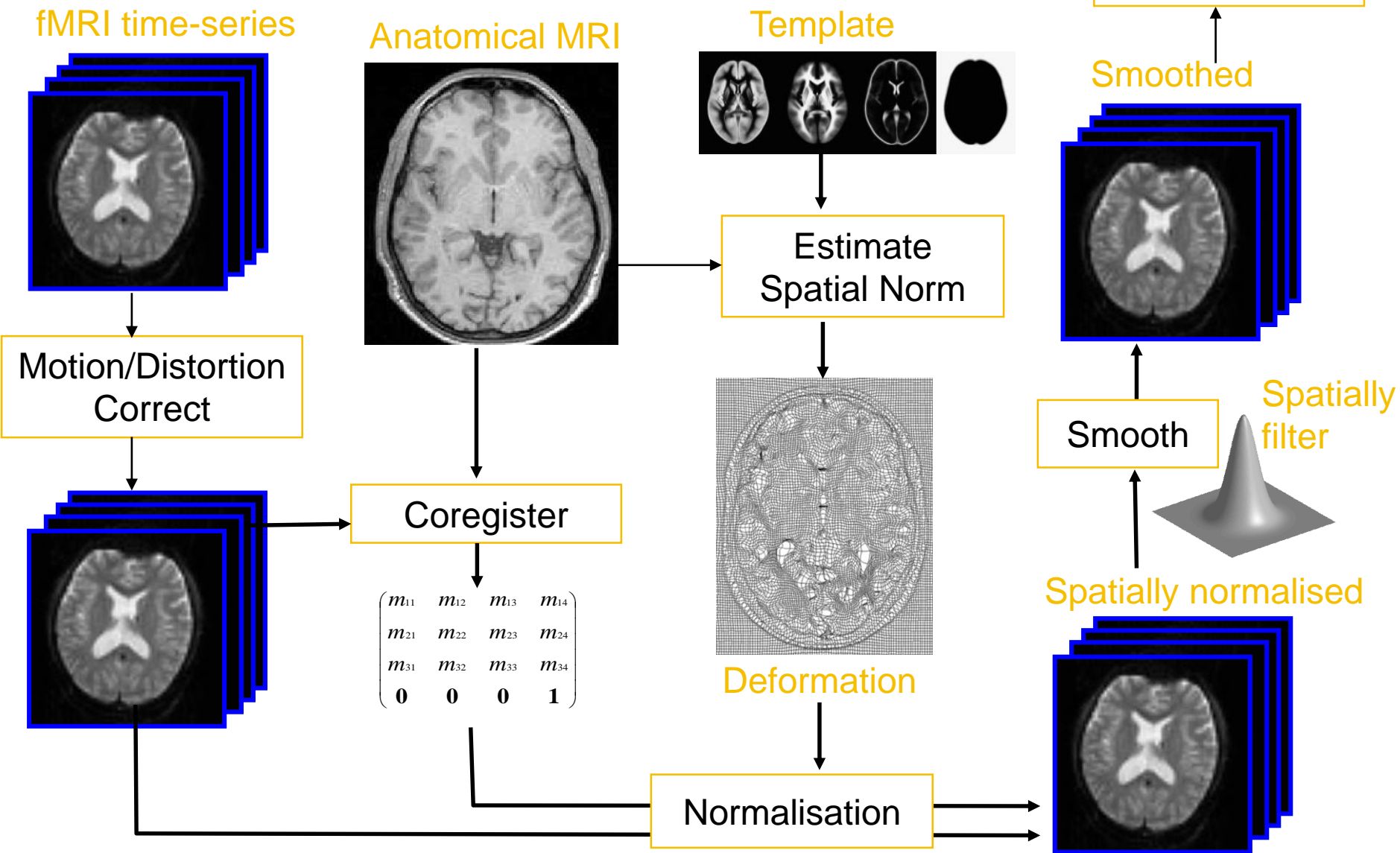
Elina Liu & Jordan Colman

With thanks to our expert **John Ashburner**

Typical SPM analysis sequence



Pre-processing sequence



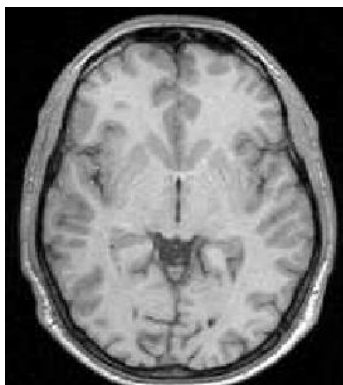
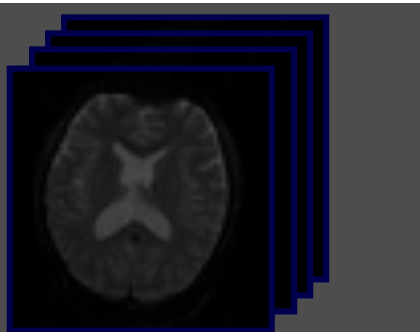
Typical pre-processing steps

1. Remove dummy scans.
2. **Realign**: correct images for movement over time.
3. **Slice timing**: correct for different slice acquisition times.
4. **Co-register**: match image modalities in individual subjects (e.g. structural and functional).
5. **Normalise**: match images to a template across subjects (sometimes preceded by segmentation of tissue types).
6. **Smooth**: apply spatial filter to adjust for residual differences and alignment errors

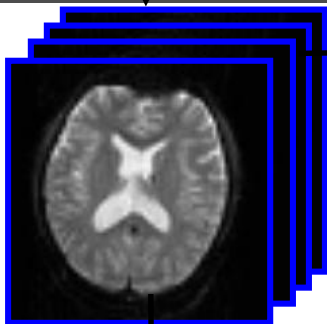
Coregistration

fMRI time-series

Anatomical MRI



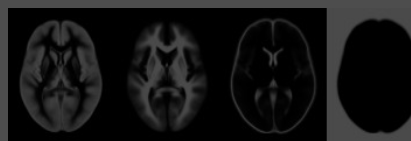
Motion/Distortion Correct



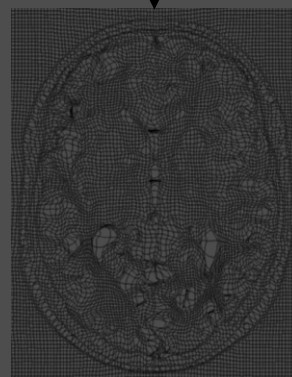
Coregister

$$\begin{pmatrix} m_{11} & m_{12} & m_{13} & m_{14} \\ m_{21} & m_{22} & m_{23} & m_{24} \\ m_{31} & m_{32} & m_{33} & m_{34} \\ \mathbf{0} & \mathbf{0} & \mathbf{0} & \mathbf{1} \end{pmatrix}$$

Template



Estimate Spatial Norm

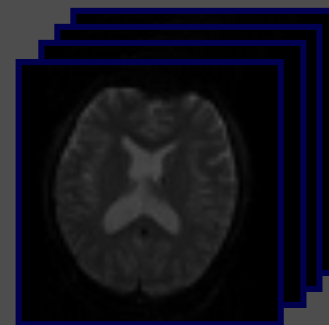


Deformation

Normalisation

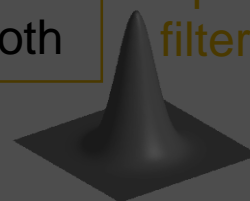
Statistics or whatever

Smoothed

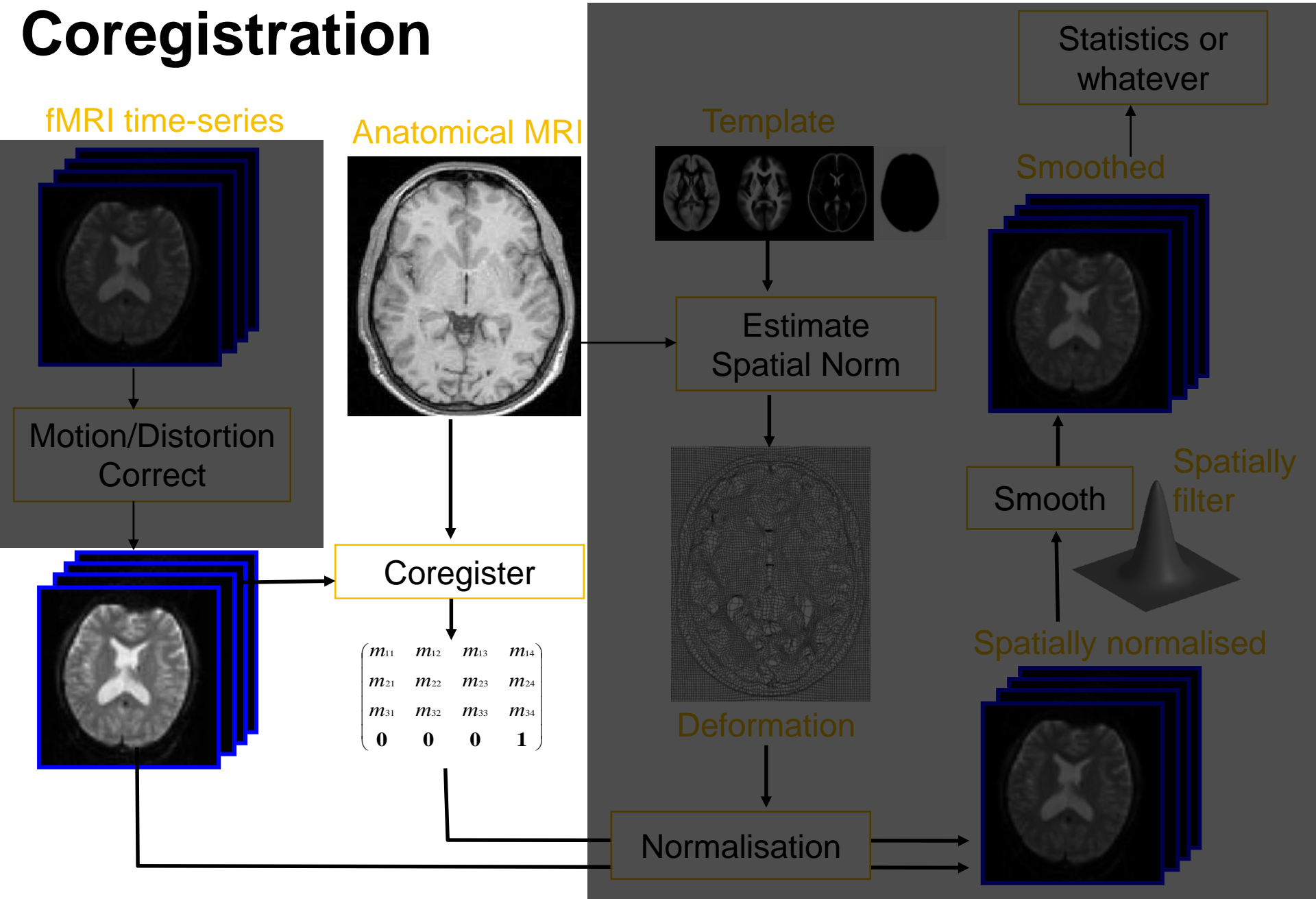
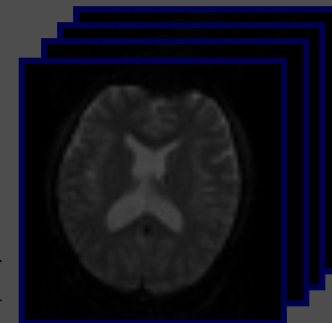


Smooth

Spatially filter

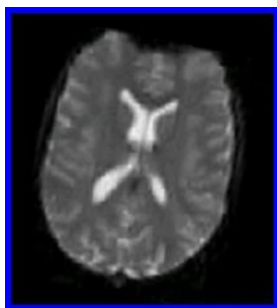


Spatially normalised



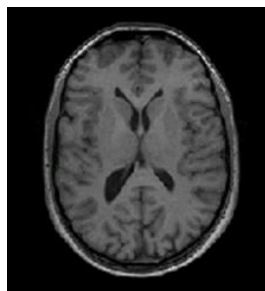
Why Coregister?

- Intermodal
- Intrasubject
- Aim – match images of same subject but different modalities
 - Link functional images (T2* weighted) with a structural image (T1)
 - Superior anatomical localisation
 - More precise spatial normalisation



FUNCTIONAL – T2*

Note the poor
structural definition



STRUCTURAL – T1



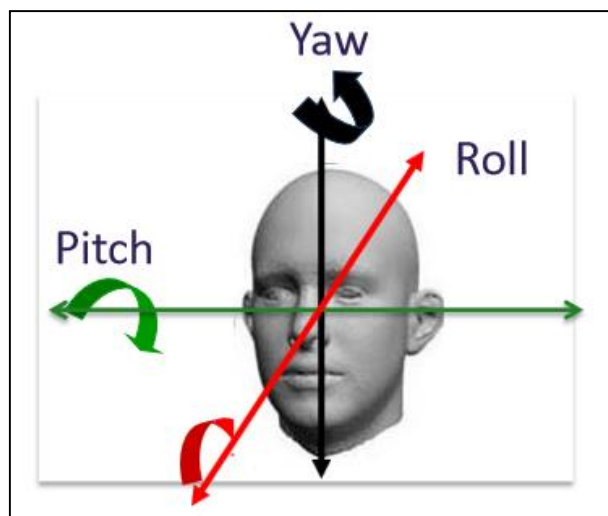
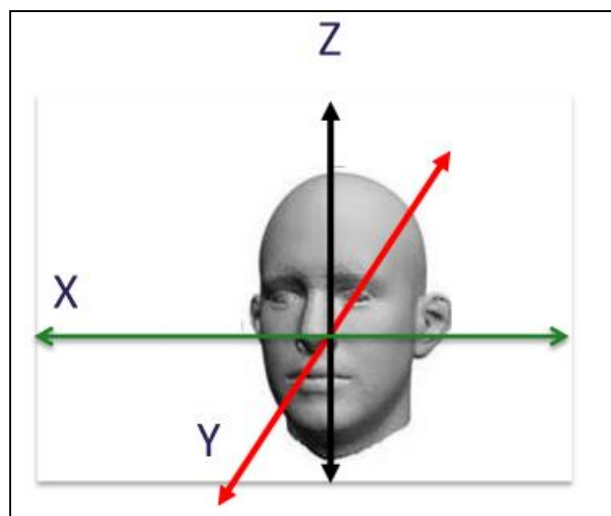
CO-REGISTRATION

How Coregister? – Procedure

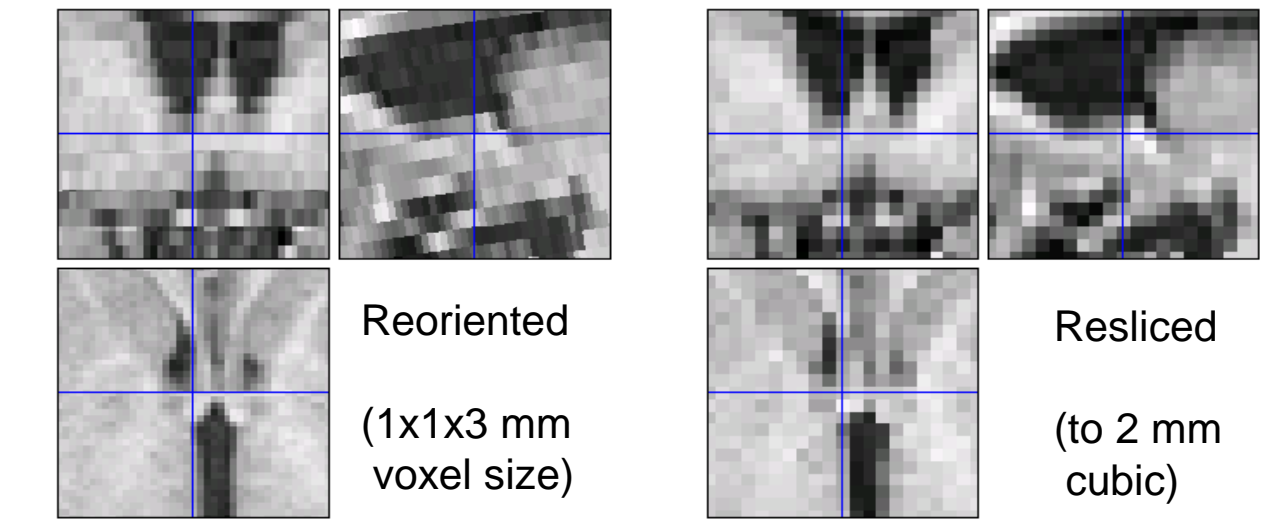
- It's not simple...
 - Cannot rely on simply using intensity difference
 - Identifying individual landmarks too labour intensive
- Need a measure of similarity...
 - Seek to measure shared information
 - Registration intuitively relies on the concept of aligning images to increase their similarity
- Two-step process
 - i. Registration
 - ii. Reslicing (optional)

How Coregister? – i. Registration

- Similar to realignment
- Affine transformation
 - Estimate parameters
 - Apply these transformation
- Fit source image to reference image (static)
 - → Intensity nonuniformity (INU) in T1



How Coregister? – ii. Reslicing (optional)



- Resliced to the same dimensions, voxel sizes, orientation etc.
 - E.g., Match 3mm fMRI with 1mm T1 → produce 1mm fMRI that fill up the disk

Coregistration – advise ESTIMATE only

- [Coregister (Estimate)]
- The algorithm only changes the headers of the images in order to reflect their new orientation/position
 - The changed headers are accounted for by the spatial normalisation routines

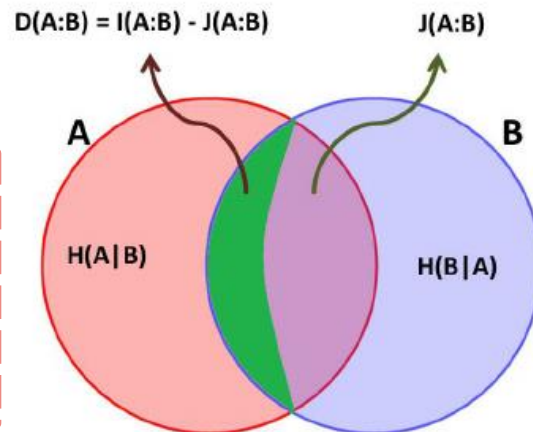
Measure of similarity

- SPM mathematically measures the similarity of the two images
 - Images are too different to use mean squared difference
 - → Mutual Information (Information Theory)
 - More flexible measures of image similarity
 - Derived from joint histograms

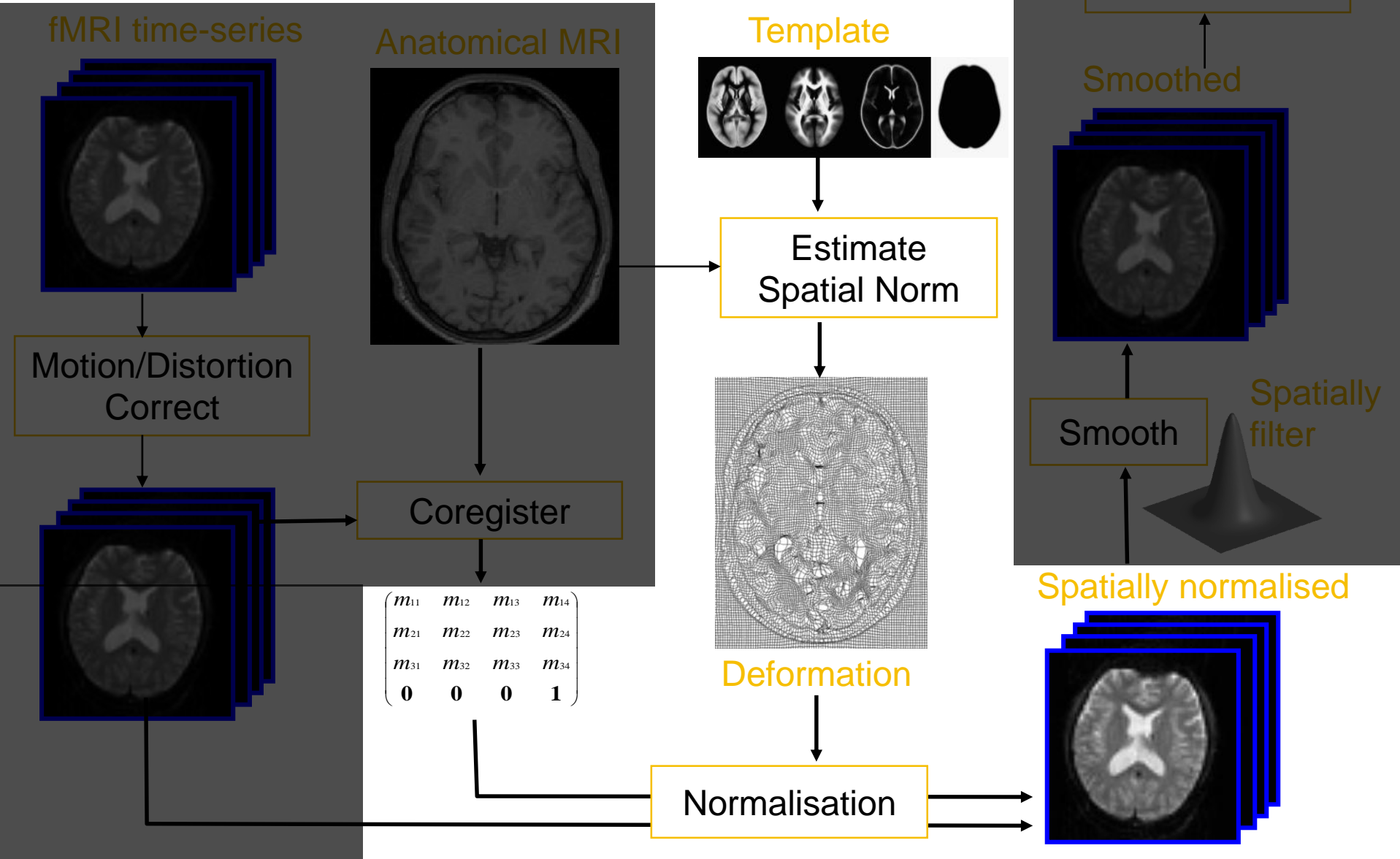
$$MI = \int_{ab} P(a,b) \log_2 [P(a,b) / (P(a) P(b))]$$

* Related to entropy: $MI = -H(a,b) + H(a) + H(b)$

* Where $H(a) = -\int_a P(a) \log_2 P(a)$ and $H(a,b) = -\int_{a,b} P(a,b) \log_2 P(a,b)$

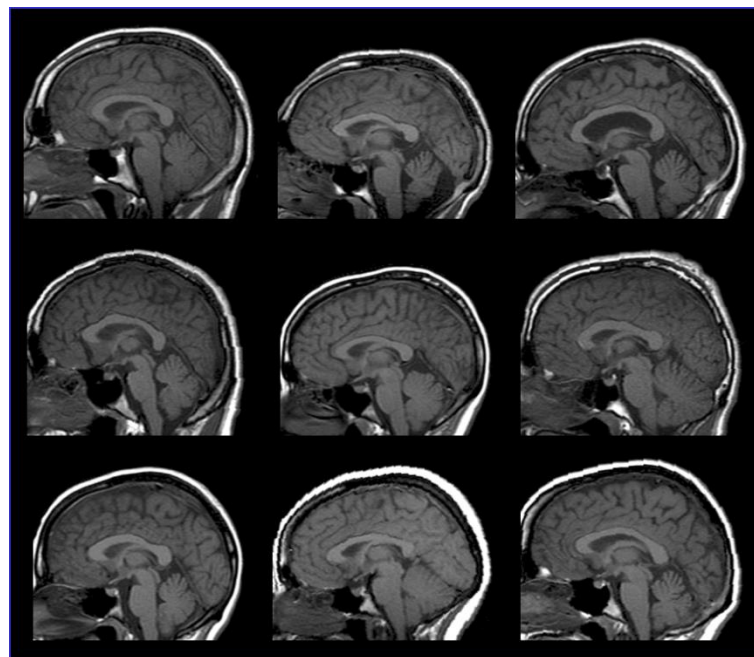


Spatial Normalisation



Spatial Normalisation

- Inter-subject
- Aim – voxel to voxel correspondence between the subjects
 - Motivation – a shared space to compare subjects



Different individuals' structural scans

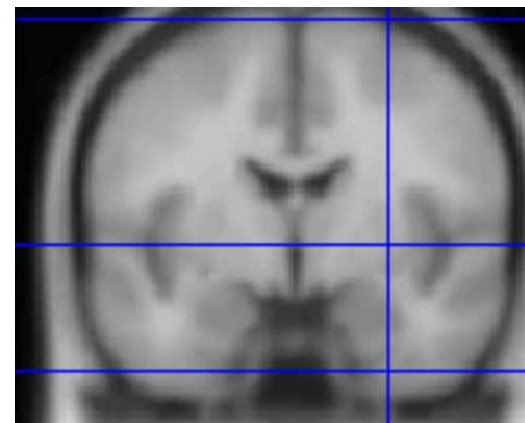
Why Normalise?

- Increases statistical power
- Generalise to population level
- Allows group-level analysis
- Allows cross-study comparisons

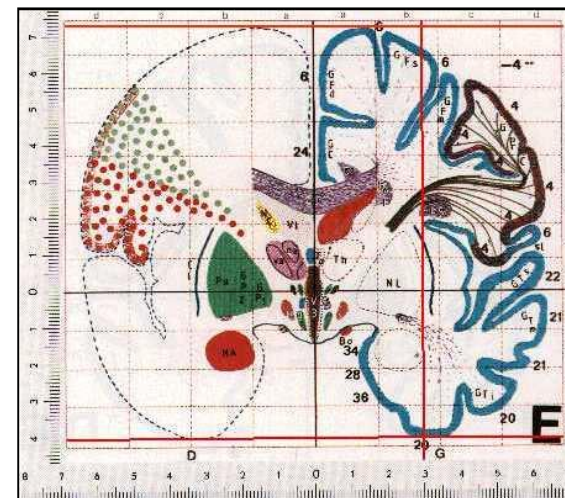


How Normalise? – Templates

- Native Space → Standard Space
- Montreal Neurological Institute (MNI)
 - Different templates (e.g., Colin27, MNI305)
- Talairach and Tournoux (1988) atlas
 - Based on left hemisphere of 60-year-old woman
 - Smaller than MNI templates (→ `mni2tal.m` can convert)
- Individual/ averaged structural scans from own study
- SPM uses MNI ICBM152!



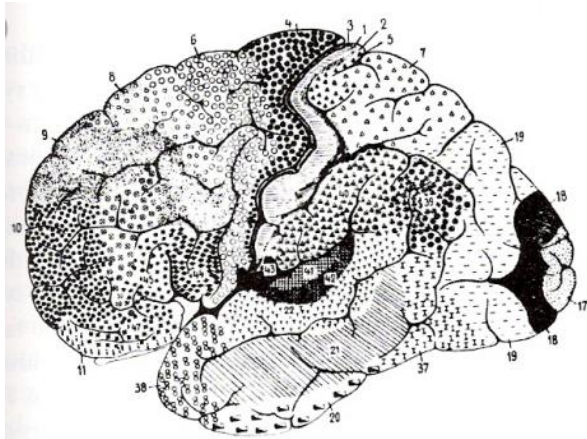
MNI Space



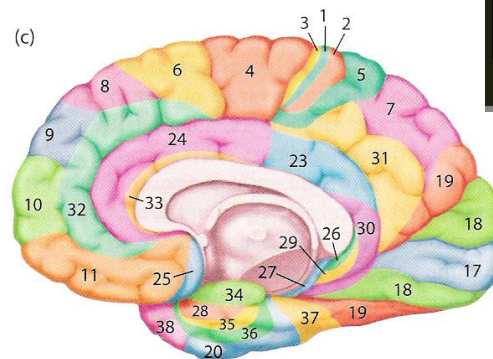
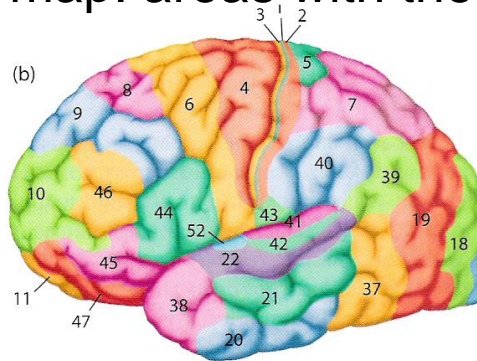
Talairach

Normalise outcome – Localisation

- Standard coordinates [x y z] in mm
 - [0 0 0] = middle of brain
- Brodmann's area
 - Cytoarchitectonic map: areas with the same physiology



Original Brodmann's



MNI<->Talairach with Brodmann Areas (1.09) BioImage Suite

MNI-Y=-19 MNI-X=10

R L R L

MNI-Z=6

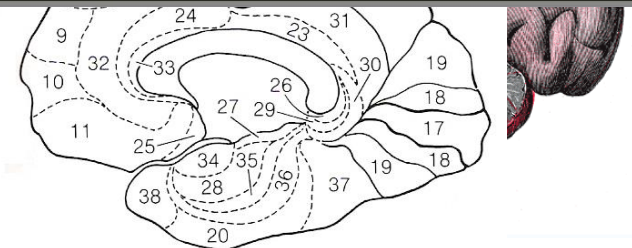
Show Brodmann areas overlay

MNI: 10 -19 6

TAL: 9 -20 8

Area: Right-Thalamus (50)

[About this application](#)

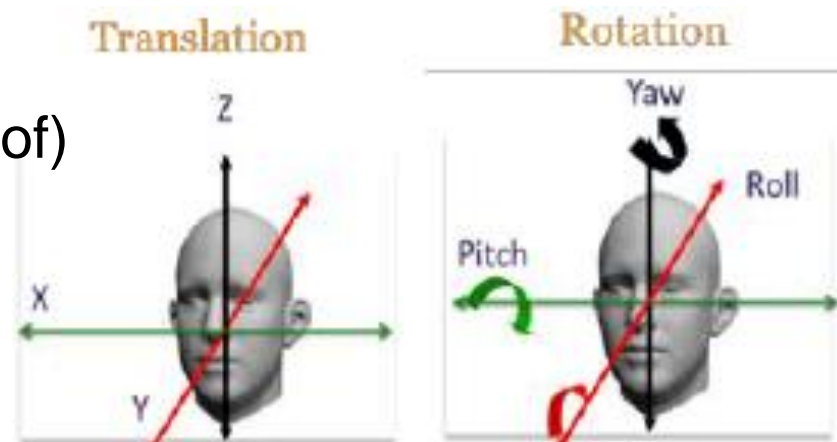


+Y
(front)

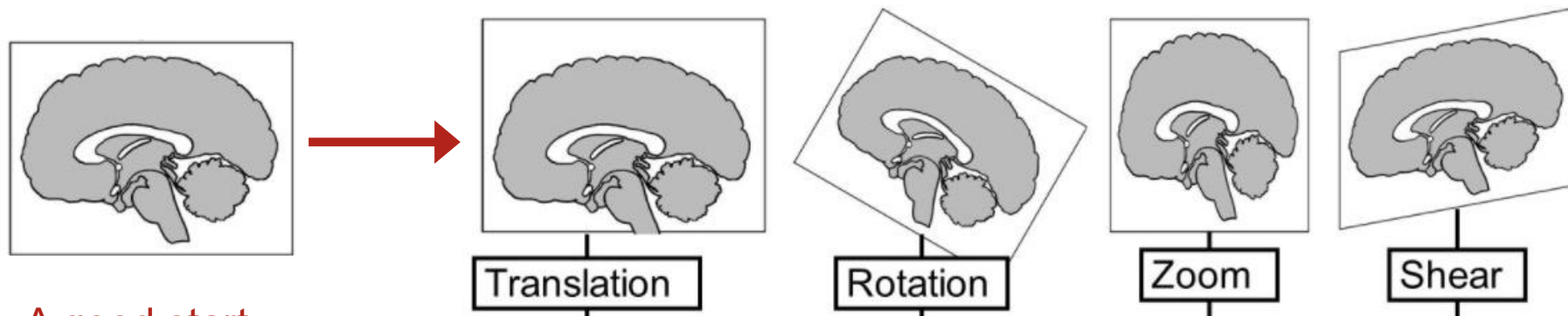
How Normalise? – Procedure 1

i. Affine Linear transformation

- optimum 12-parameter
- Translations across axes (3 dof)
- Rotations around axes (3 dof)
- Scaling or Zooming (3 dof)
- Shearing or Skewing (3 dof)



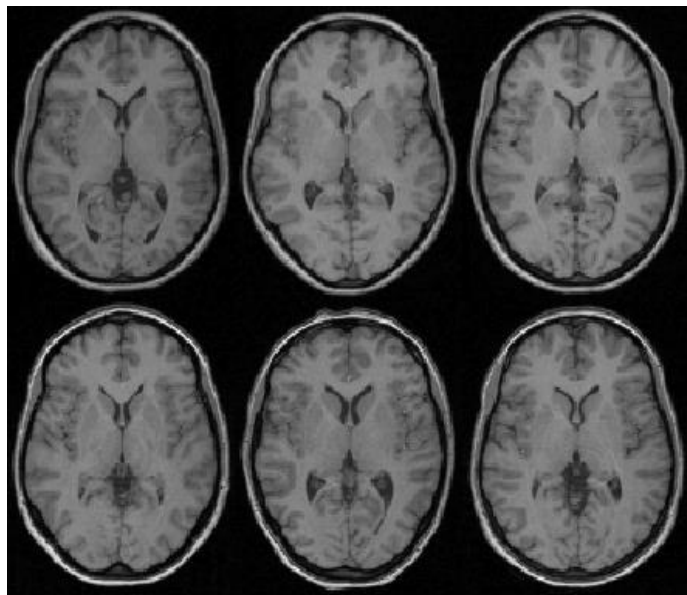
- Major differences in size, shape and position



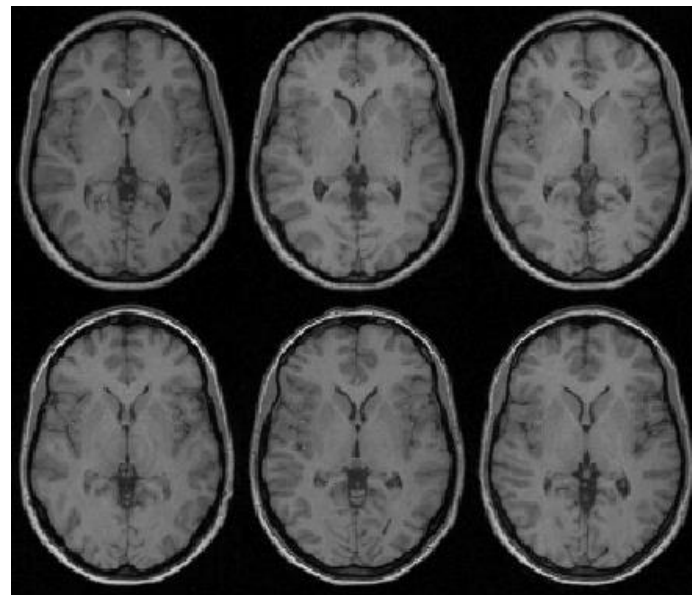
A good start...

How Normalise? – Procedure 2

- ii. Non-linear transformation – Warping
 - Constructs **non-linear deformations** to move voxels from original location to template location
- Small-scale differences
 - Very flexible (1000s dof's)



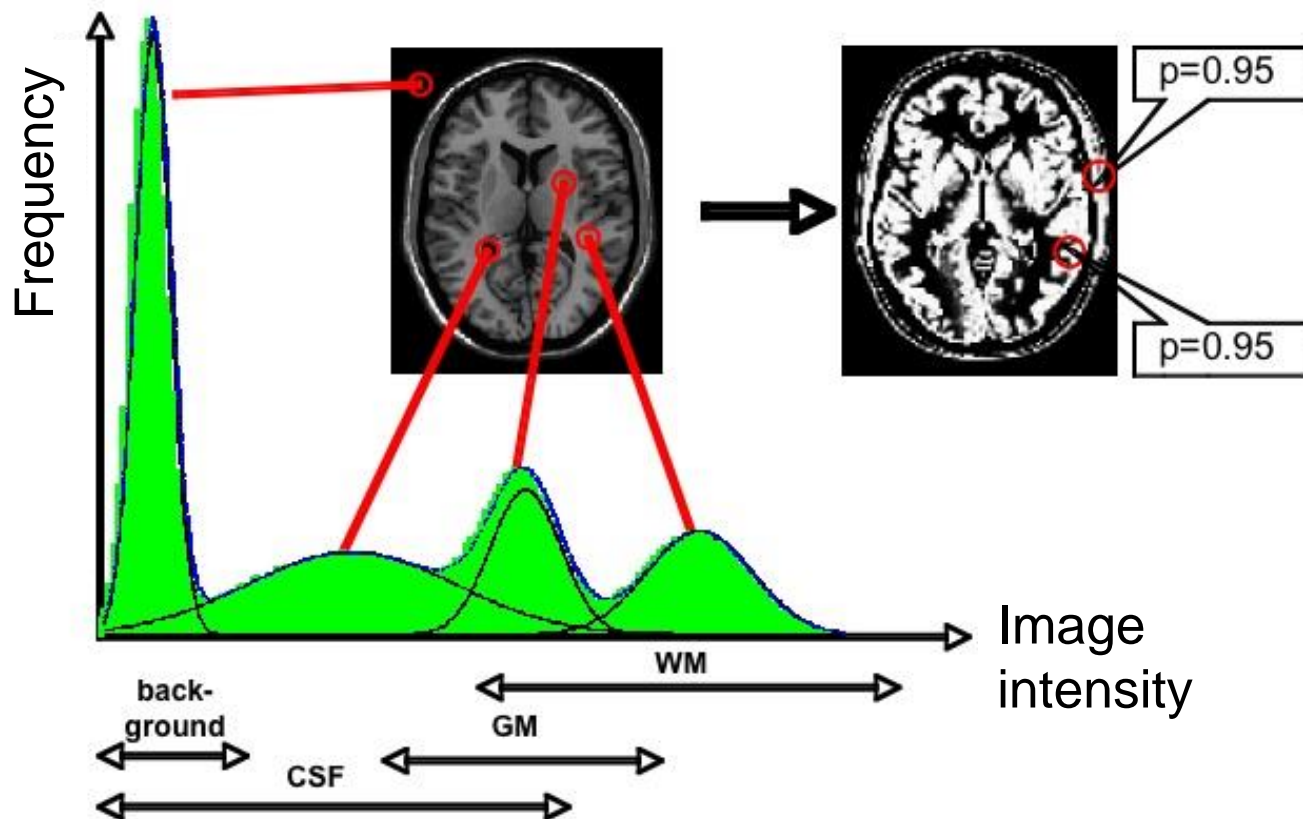
Affine Registration



Non-linear Registration

2. Warping: how does it work?

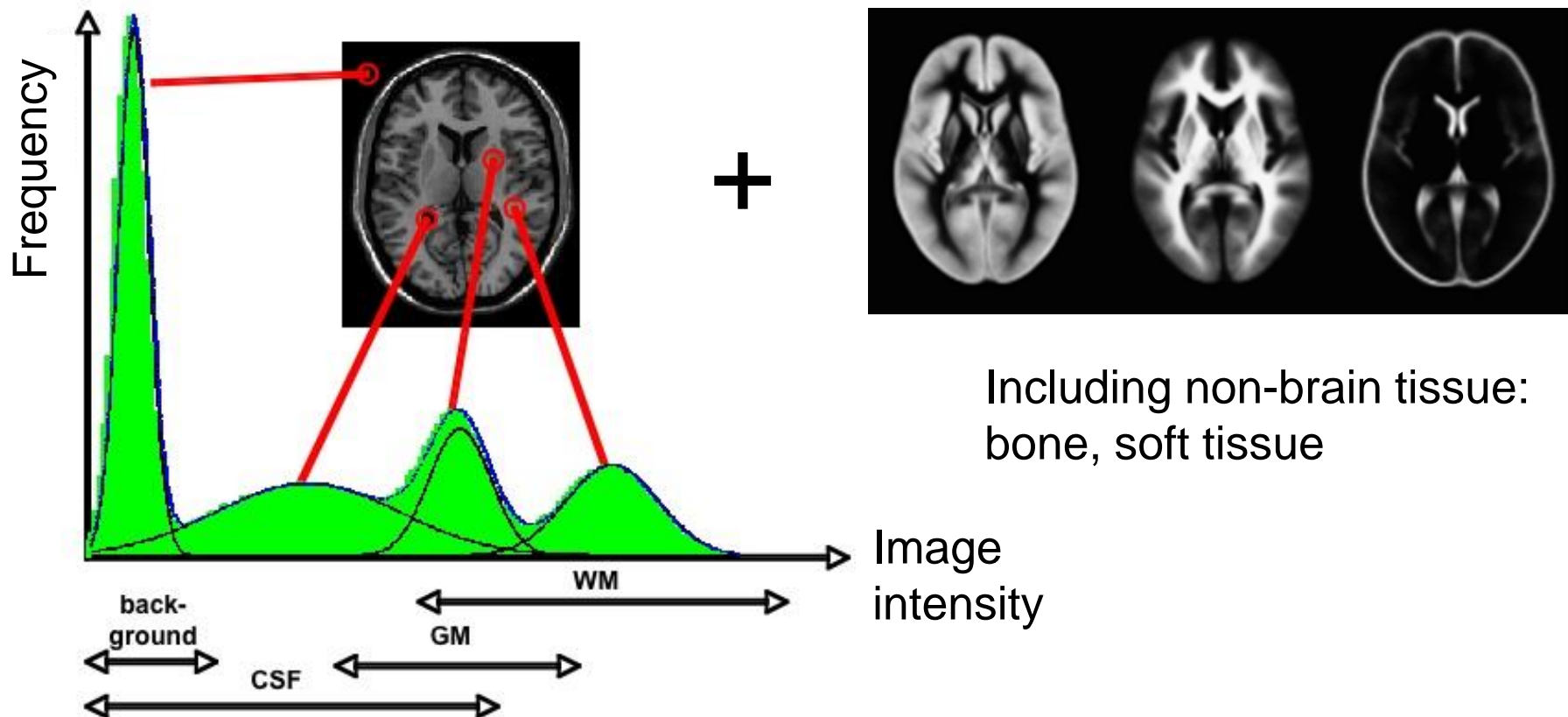
Warping in SPM12: segment the source image into tissue types, using a **Gaussian Mixture Model** of signal intensities.



grey matter,
white matter,
etc.

2. Warping: how does it work?

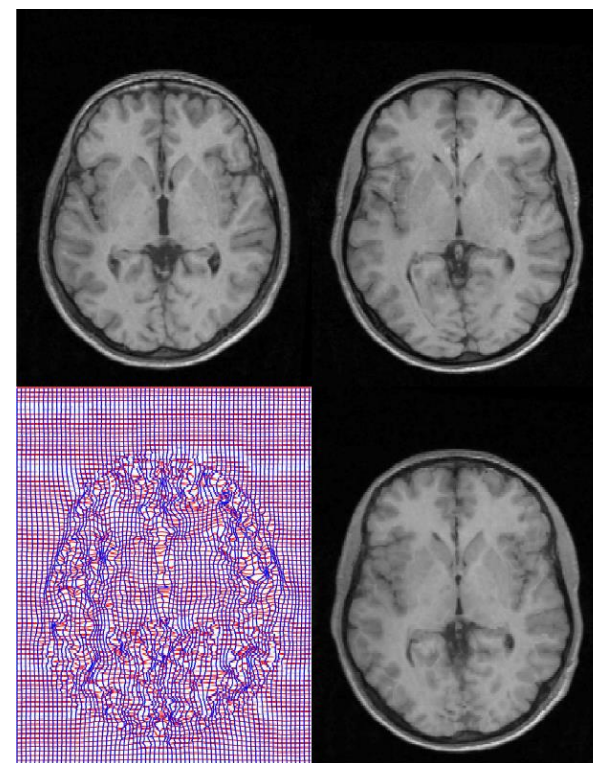
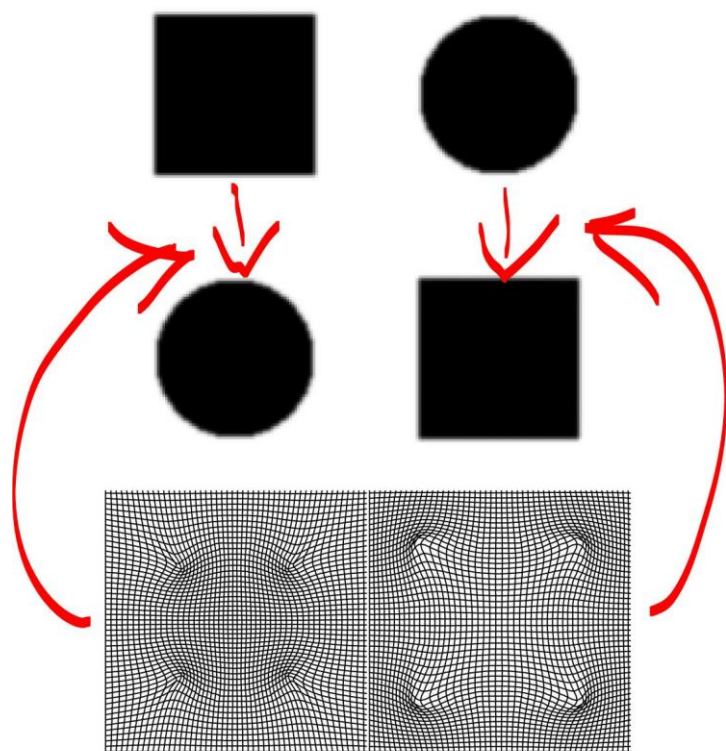
Alignment with the standard **Tissue Probability Maps**



2. Warping in more detail

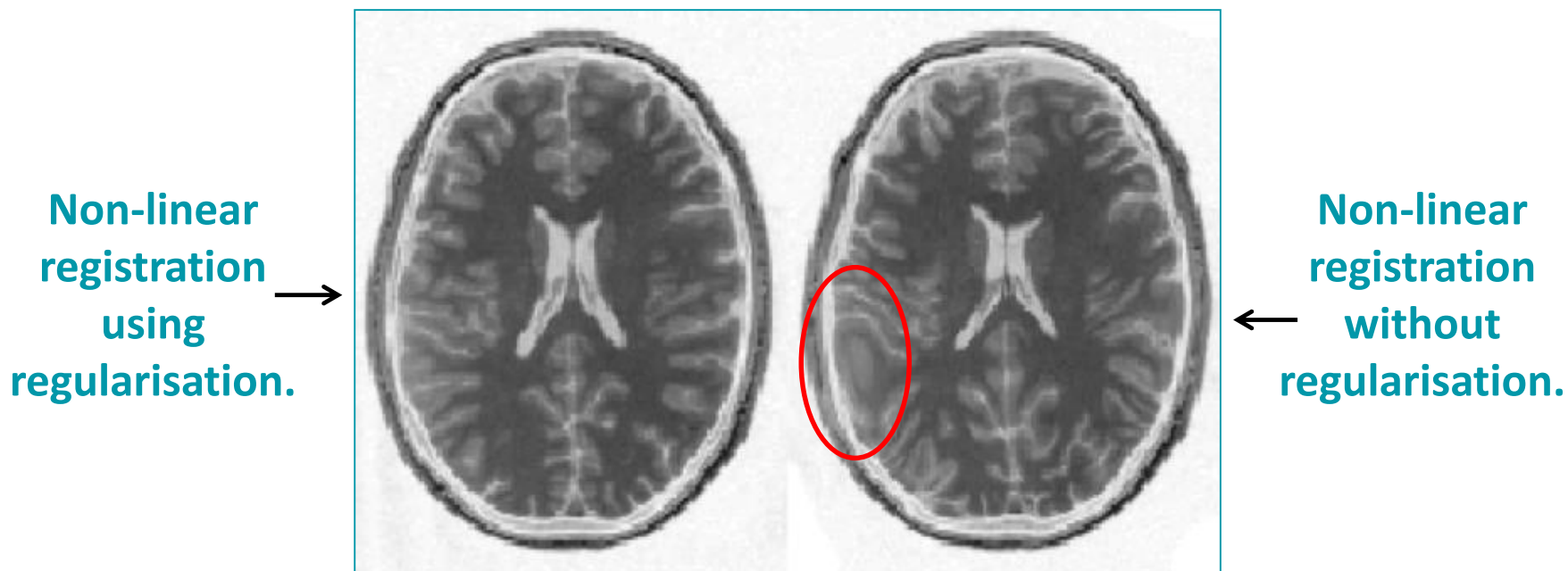
Deformations: modelled with a linear combination of basis functions

→ Using another updated algorithm

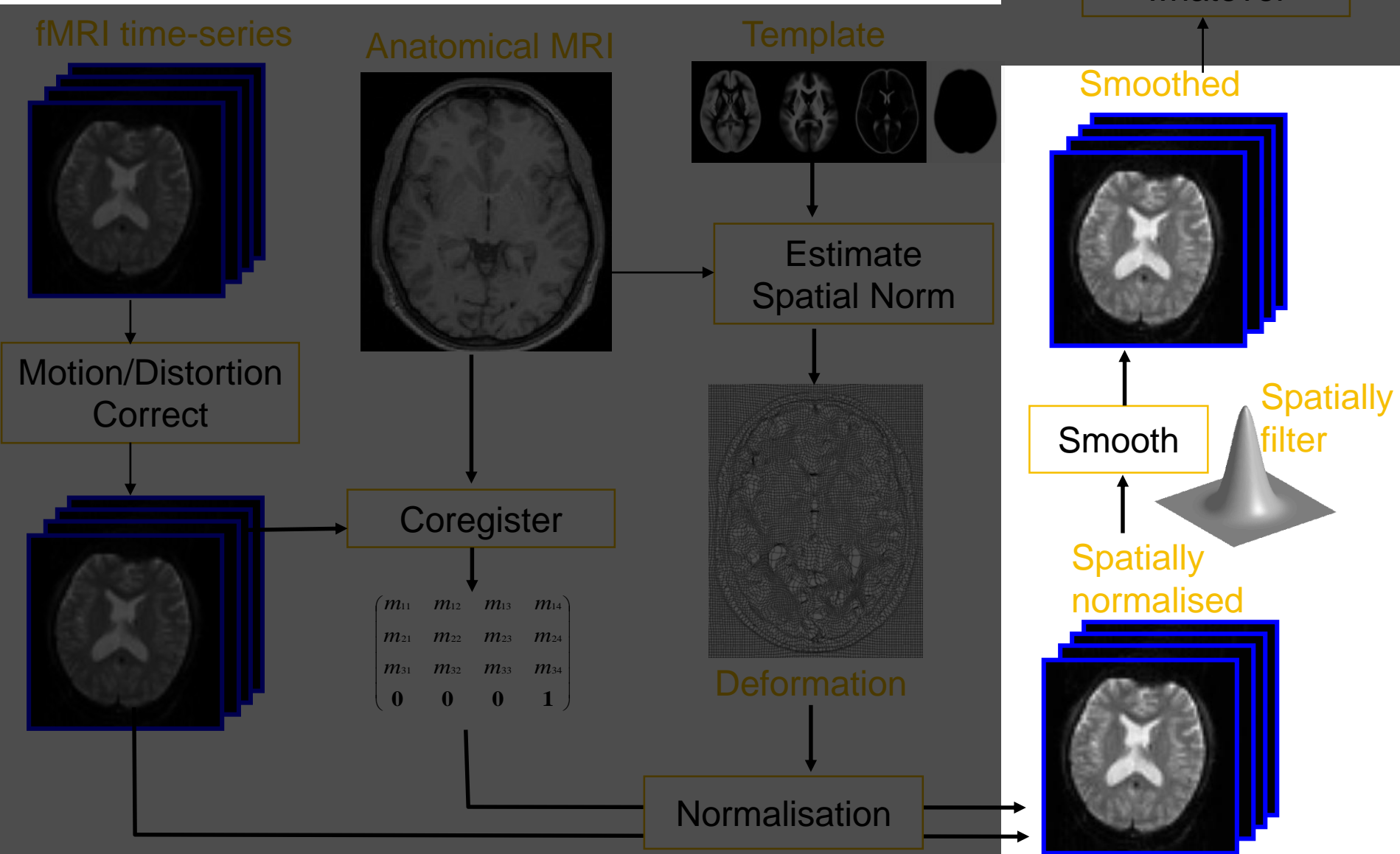


Bad Normalise? – problems of overfitting

- Flexibility of warping can lead to unrealistic results
- → Regularisation – constraints to protect against overfitting

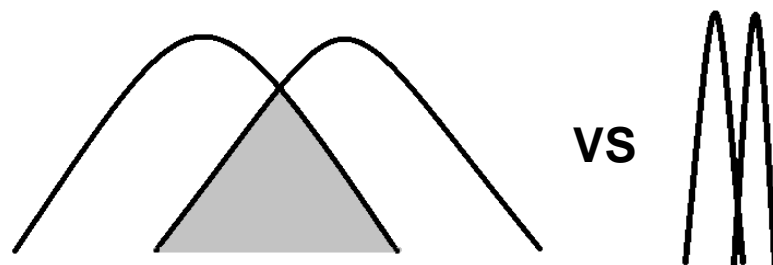
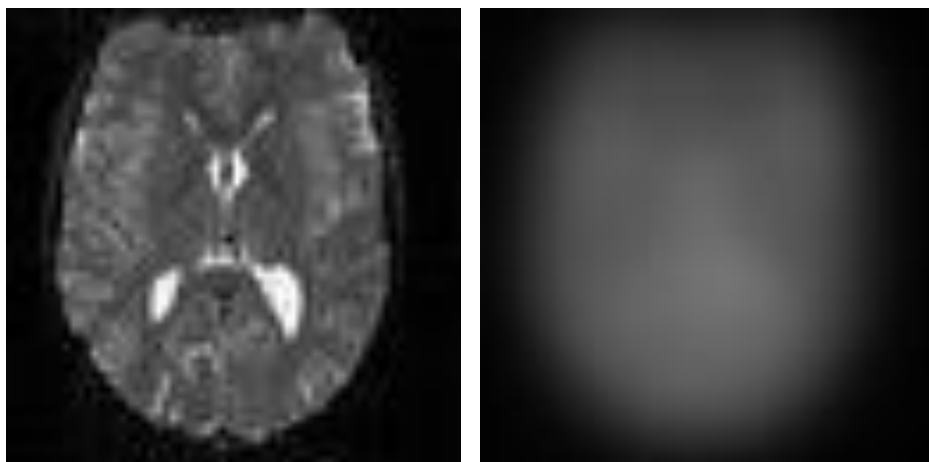


Smoothing



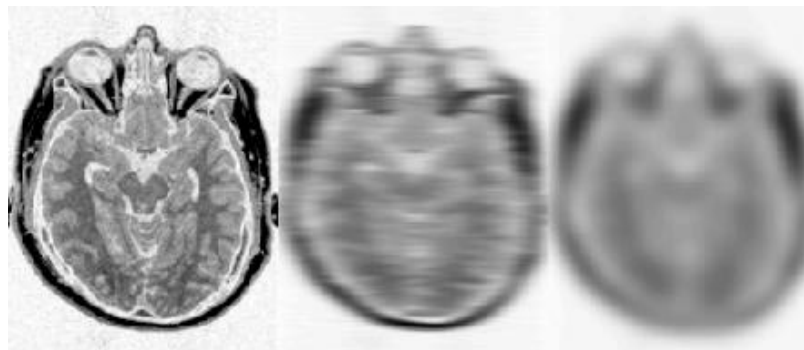
Smoothing

- Blurs over residual anatomical differences and registration errors
- Evens out signal across space.
- Removes high spatial frequencies (signal that changes rapidly across voxels).



Why do Smoothing?

- Suppresses noise
 - signal reinforced and noise averaged out
- Superior spatial overlap
- Data becomes more normally distributed
- Increases sensitivity to effects of similar scale to kernel



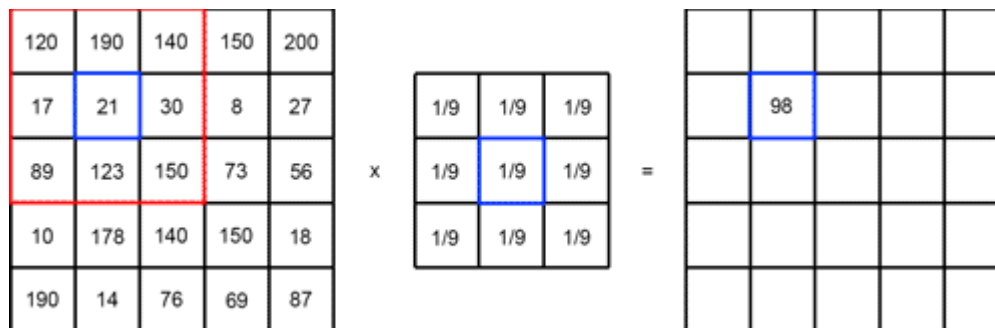
No smoothing

Smoothing in
X direction

Smoothing in
X&Y direction

How to Smooth? - applying a kernel / filter

- A square section of the image is multiplied in an element wise fashion by the filter and summed
- The central voxel is reassigned to this new value
- A square filter is run across the whole image

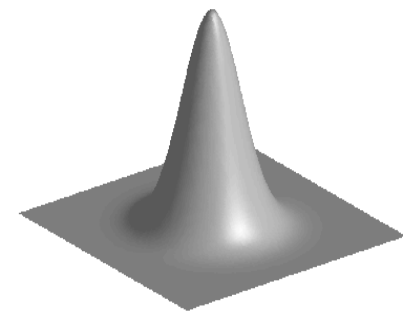
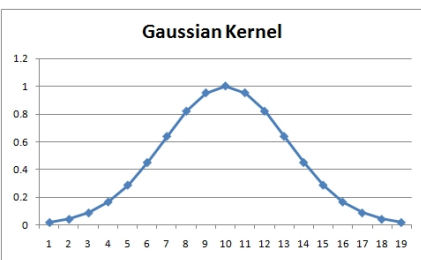


Here is an example of an average filter being applied to an image

Outlier pixels with lower values than the rest of the image are smoothed out

How to Smooth? - The Gaussian filter

- Rather than an average Kernel the central information can be more heavily weighted
- A gaussian does this and can be turned into a discrete kernel with varying widths
- This can then be applied to an image as in the previous slide



1/16

1	2	1
2	4	2
1	2	1

1/273

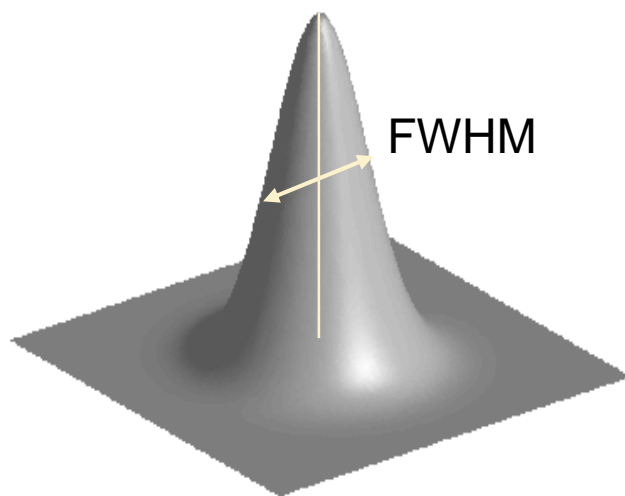
1	4	7	4	1
4	16	26	16	4
7	26	41	26	7
4	16	26	16	4
1	4	7	4	1

1/1003

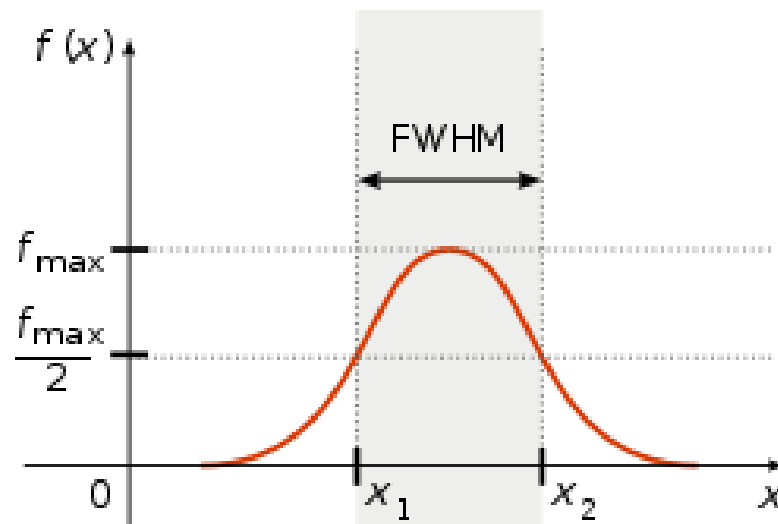
0	0	1	2	1	0	0
0	3	13	22	13	3	0
1	13	59	97	59	13	1
2	22	97	159	97	22	2
1	13	59	97	59	13	1
0	3	13	22	13	3	0
0	0	1	2	1	0	0

Smoothing: FWHM

- Defined by full-width at half maximum (FWHM)
- Width of smoothing kernel at half its maximum amplitude

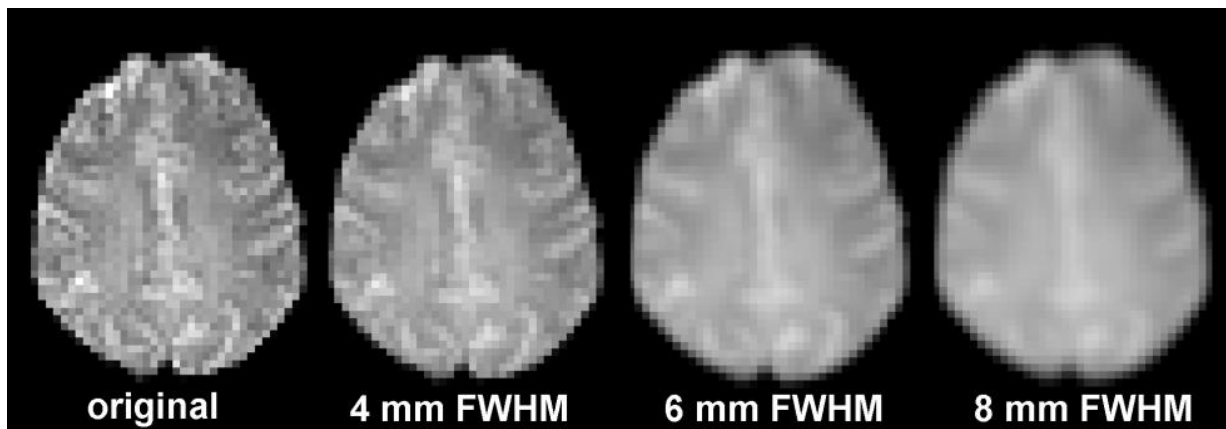


Gaussian smoothing kernel



Smoothing: FWHM value

- Ideally, size of smoothing kernel should match the expected signal (“matched filter” theorem)
- However, signal rarely known and varies across regions
- Typically, 5-8 mm FWHM
- The larger the FWHM, the more sensitive the statistical analyses, but less spatial resolution



Smoothing: disadvantages?

- Reduction of spatial resolution of the data
- Edge Artifacts
- Merging
- Extinction
- Misslocalisation of activation peaks

Summary

- Coregistration links functional data with a structural image
→ **Intrasubject**
- Normalisation links images from one subject to standardised space
→ **Intersubject**
- Smoothing blurs over any residual errors and anatomical differences

Keep in mind

- Multiple data adjustments are made during fMRI analysis, and these alter the raw data considerably

References

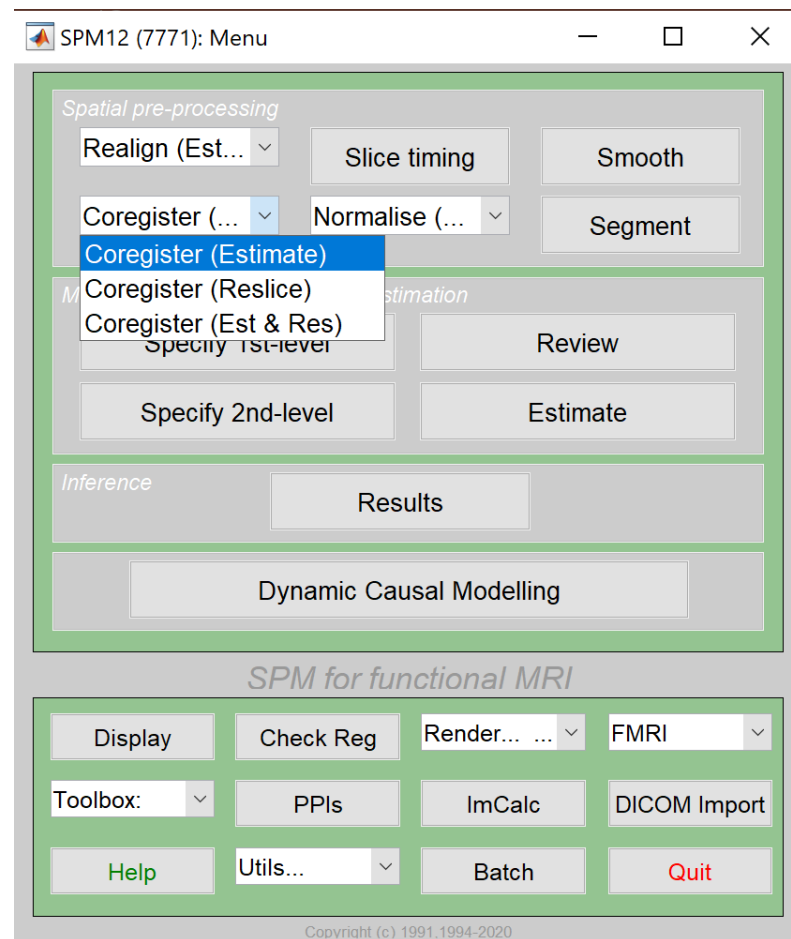
- John's Lecture
- Ged Ridgway's preprocessing lecture
- SPM Homepage
- Mfd Resources 2018

Tutorial – Pre-processing #2

- We will use the Unwarped auditory data from last week
- Find the data @ <https://www.fil.ion.ucl.ac.uk/spm/data/auditory/>
& Select ‘Auditory - single subject’
- Steps:
 - 1) Co-register fMRI data to structural image
 - 2) Spatially normalise structural images and apply to fMRI data
 - 3) Smooth the fMRI data
- Additional info on using ‘segment’ tool

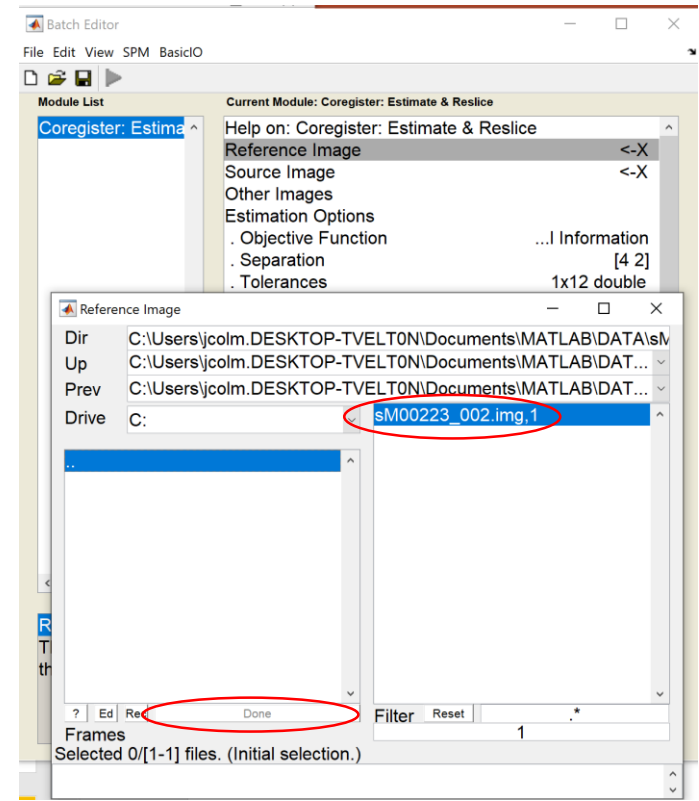
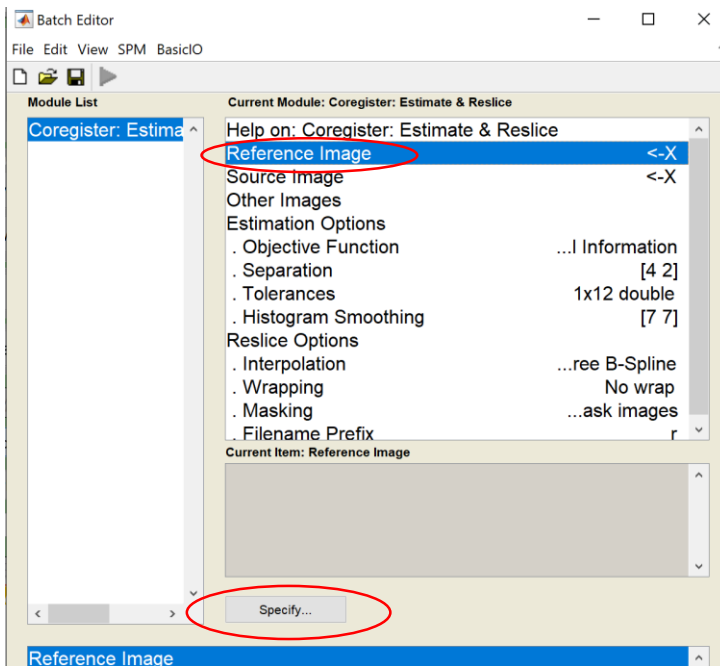
Step 1) Co-register to Structural image

- Open SPM by typing: 'spm fmri' into Matlab
- Select Coregister (Estimate)
- Coregister (Est & Res) will reslice the fMRI data to the same resolution as the structural data



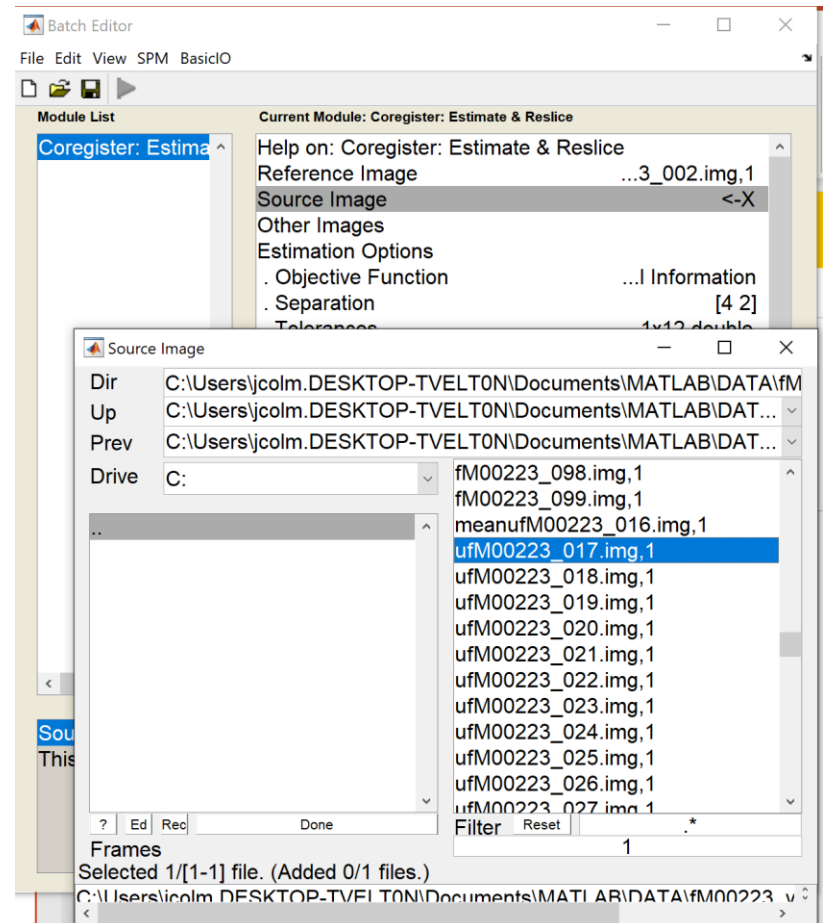
Step 1) Select Reference image

- Click new subject
- Then click 'Reference image'
- Go to structural image folder and select it
- Then press 'Done' button



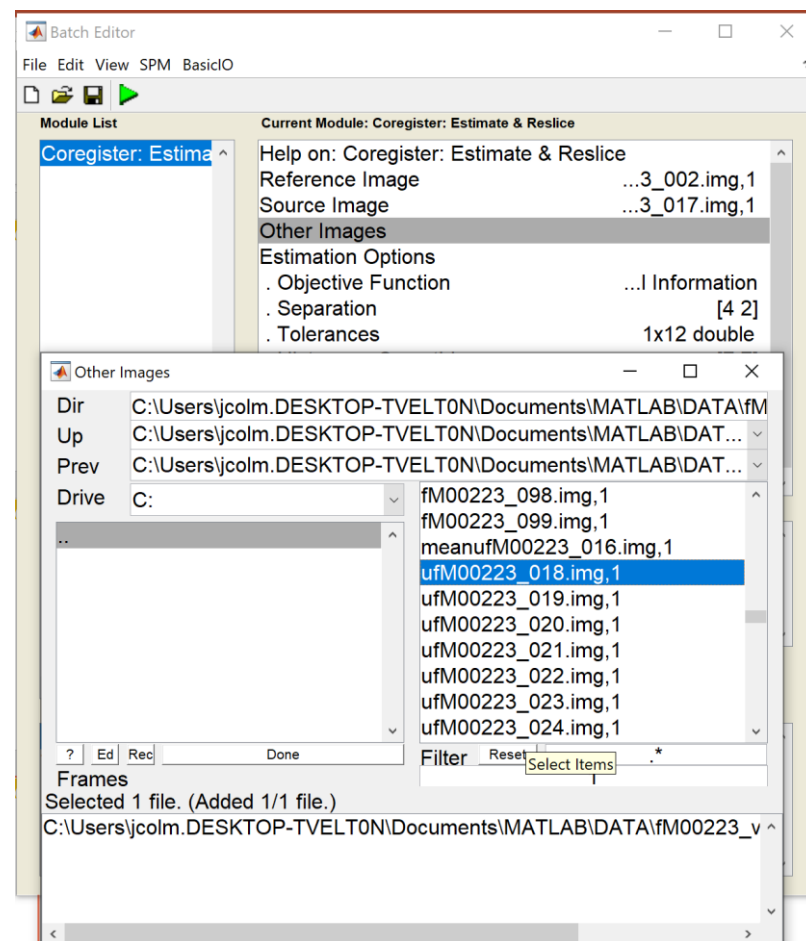
Step 1) Select image to Co-register

- Select the first ‘Unwarped’ image from last week beginning with a u
- ‘ufM00223_016.img’



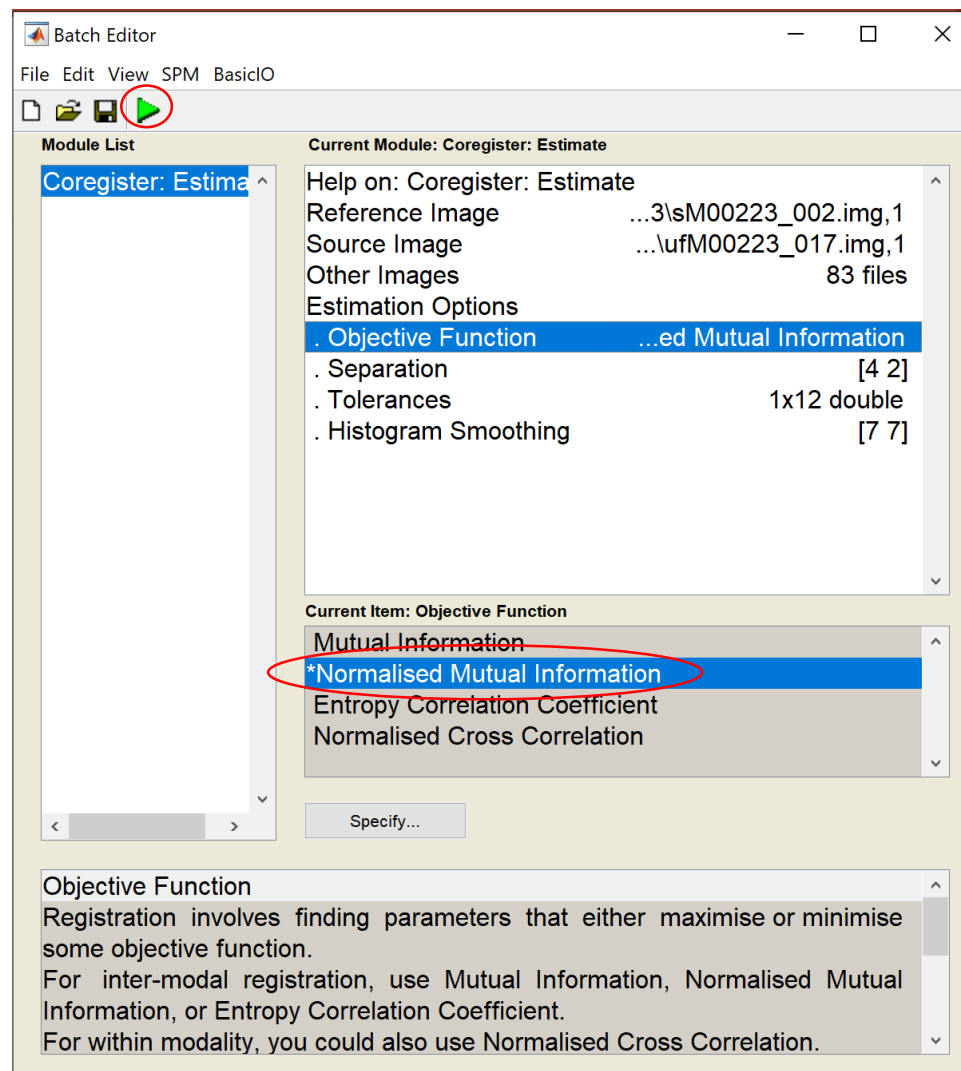
Step 1) Select other images already in alignment to also co-register

- Select all fMRIs already matched to the 016 image to also co-register



Step 1) Objective function

- As the images are different sequences ensure one of the 'Mutual information' options is selected
- Then press 'Run' to process



Step 1) SPM- Coregistration Output

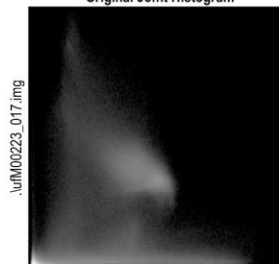
Normalised Mutual Information Coregistration

$$X1 = 3.000*X + 0.024*Y + 0.019*Z + 28.444$$

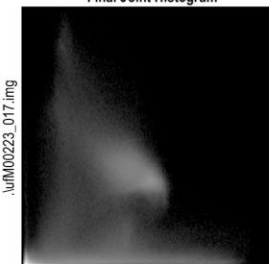
$$Y1 = -0.024*X + 3.000*Y + 0.045*Z + 33.018$$

$$Z1 = -0.006*X - 0.015*Y + 1.000*Z - 8.394$$

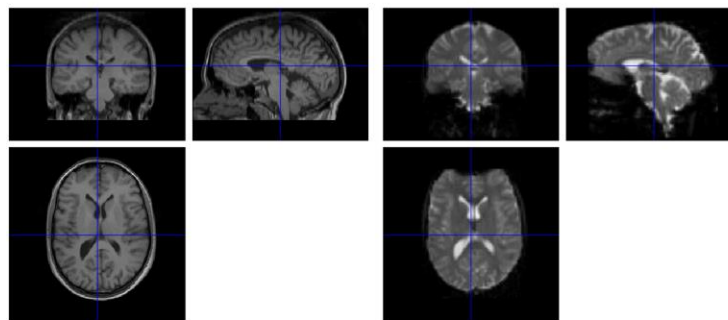
Original Joint Histogram



Final Joint Histogram

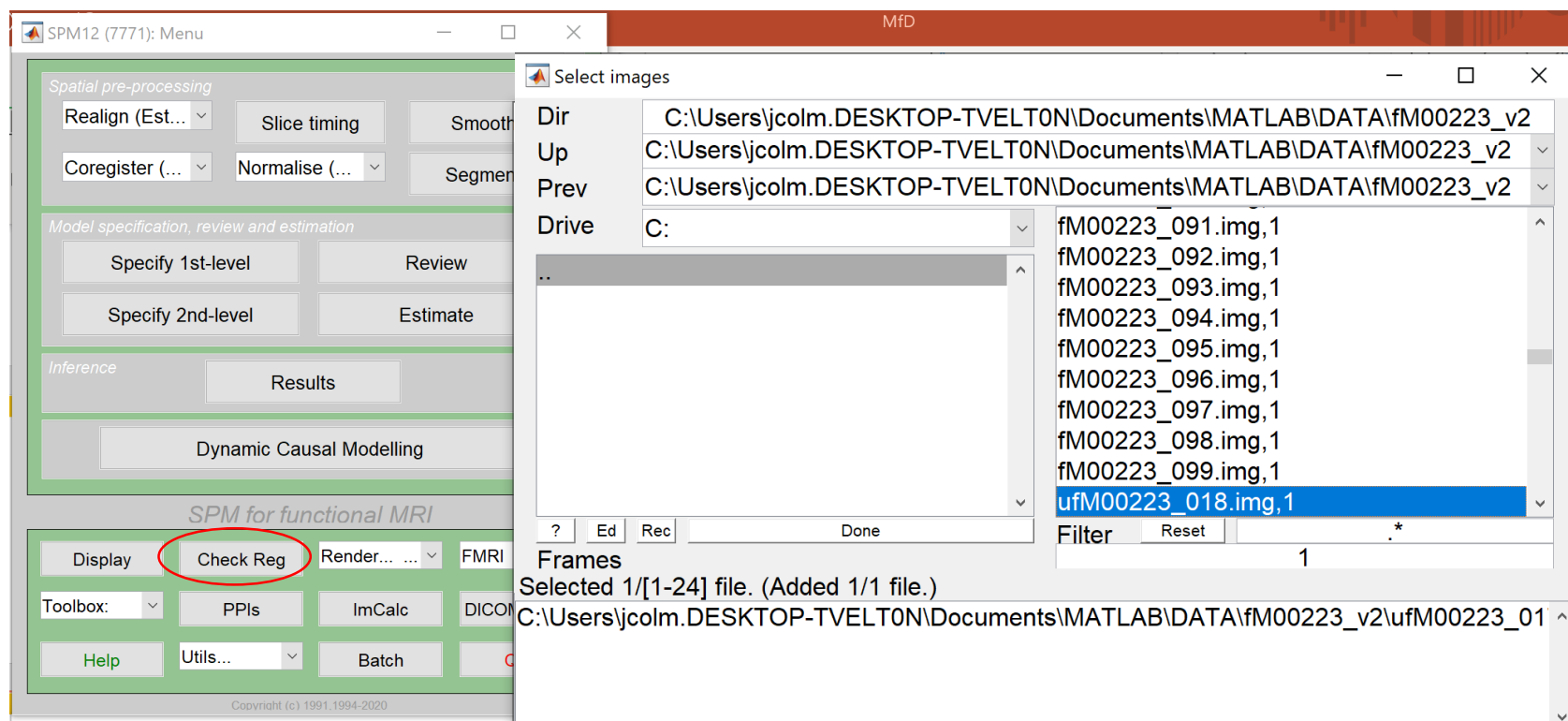


Very important to check your images- it automatically loads these up for you!



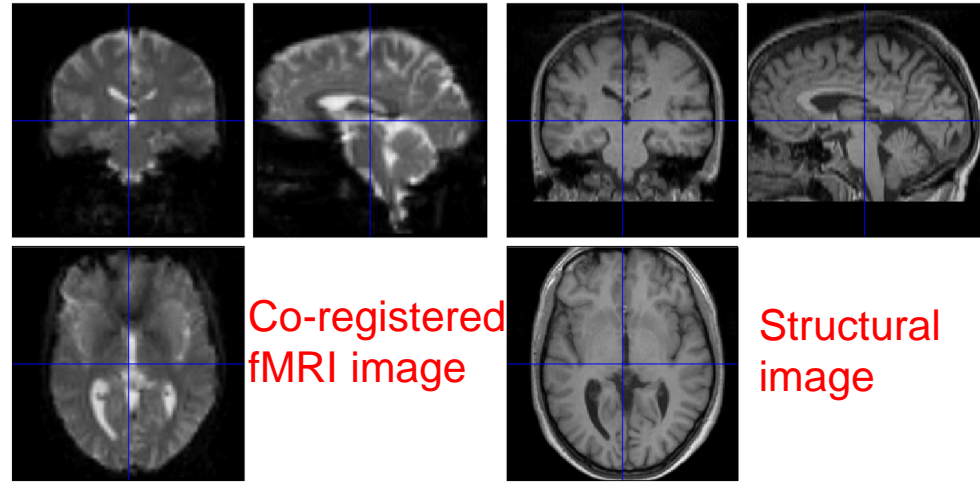
Step 1) Check Co-registration with 'Check Reg'

- Use can also use the 'Check Reg' function to ensure alignment worked
- Select structural image, original fMRI image and Co-registered fMRI image (still has a u prefix as doesn't make new images)

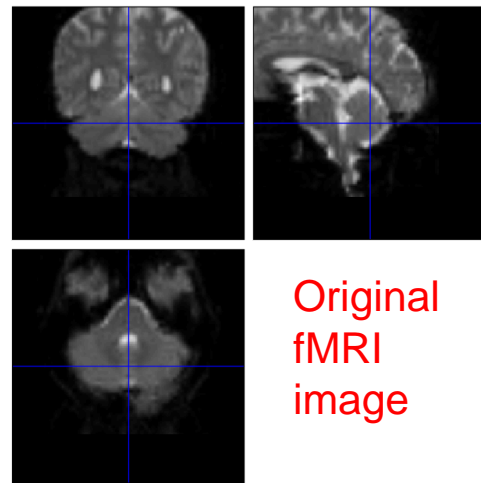


Step 1) Check Reg

- You Can see good alignment with the T1 structural image of the same subject!

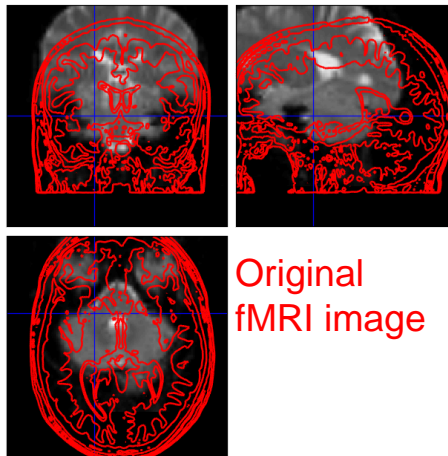
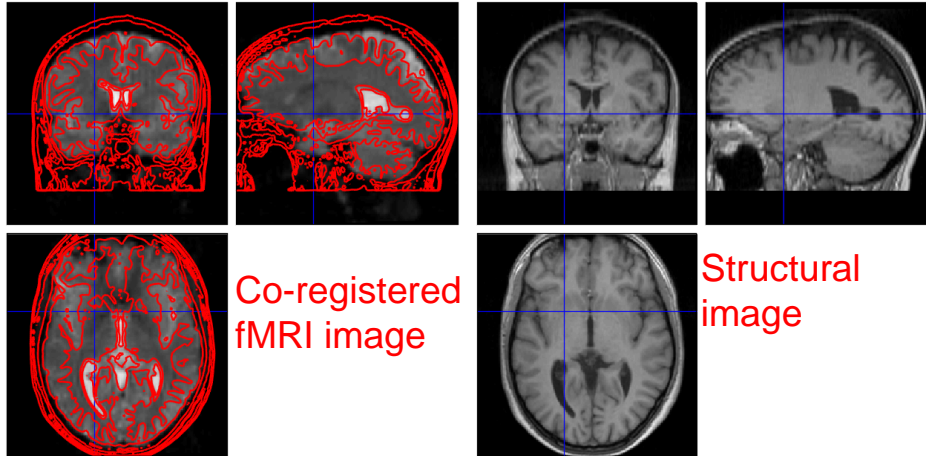
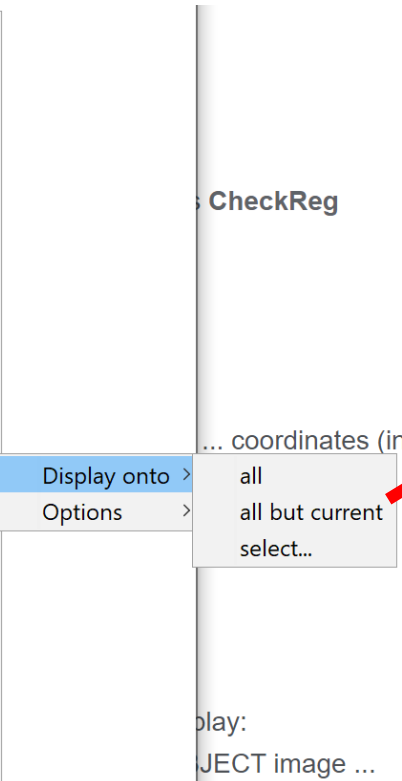
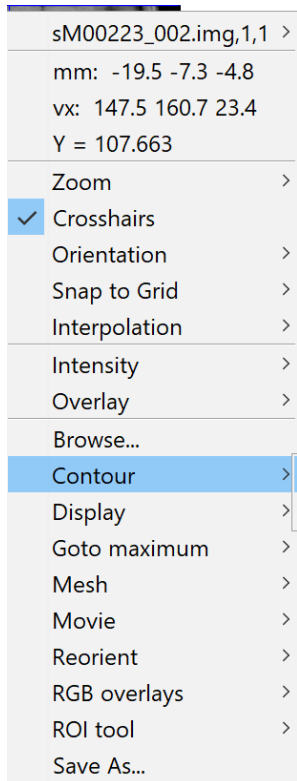


sM00223_002.img,1,1 >		
mm: -19.5 -7.3 -4.8		
vx: 147.5 160.7 23.4		
Y = 107.663		
Zoom >		
<input checked="" type="checkbox"/>	Crosshairs	
	Orientation >	
	Snap to Grid >	
	Interpolation >	
	Intensity >	
	Overlay >	
	Browse...	
	Contour >	
	Display onto >	... coordinates (ir
	Options >	all
		all but current
		select...
	Goto maximum >	
	Mesh >	
	Movie >	
	Reorient >	
	RGB overlays >	
	ROI tool >	
	Save As...	



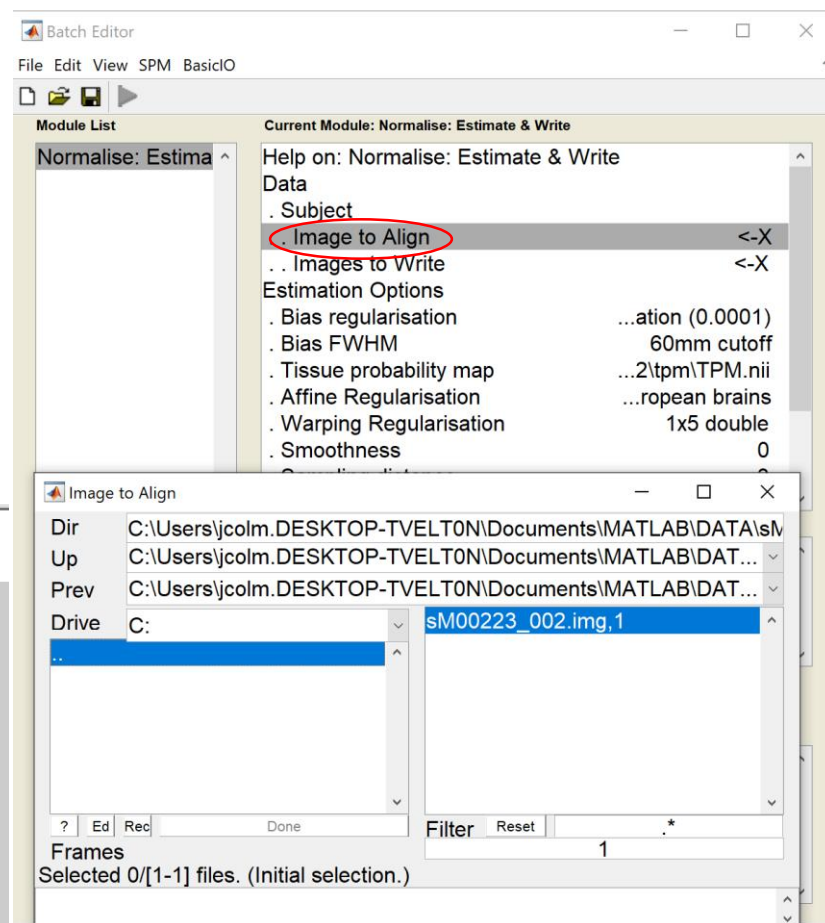
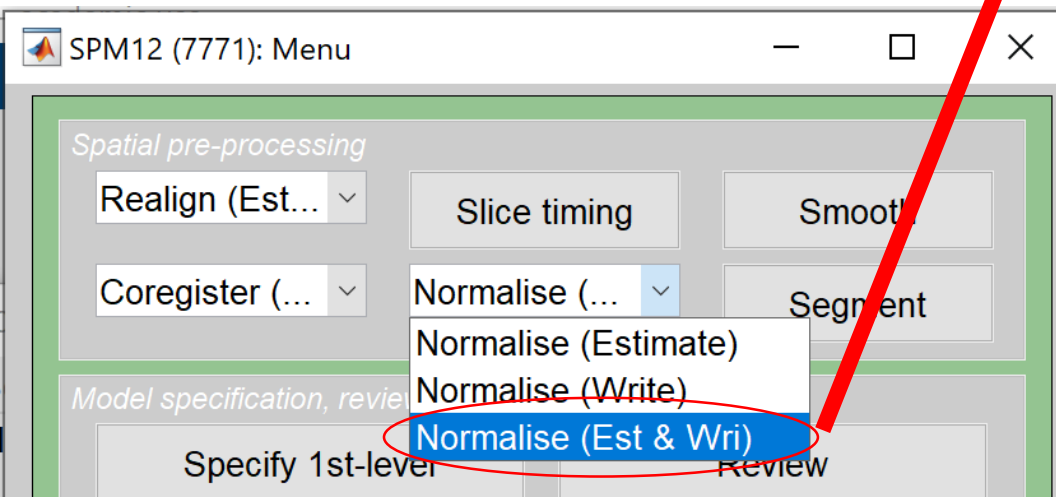
Step 1) Check Reg – contour option

- Right click the structural image and go to the contour option



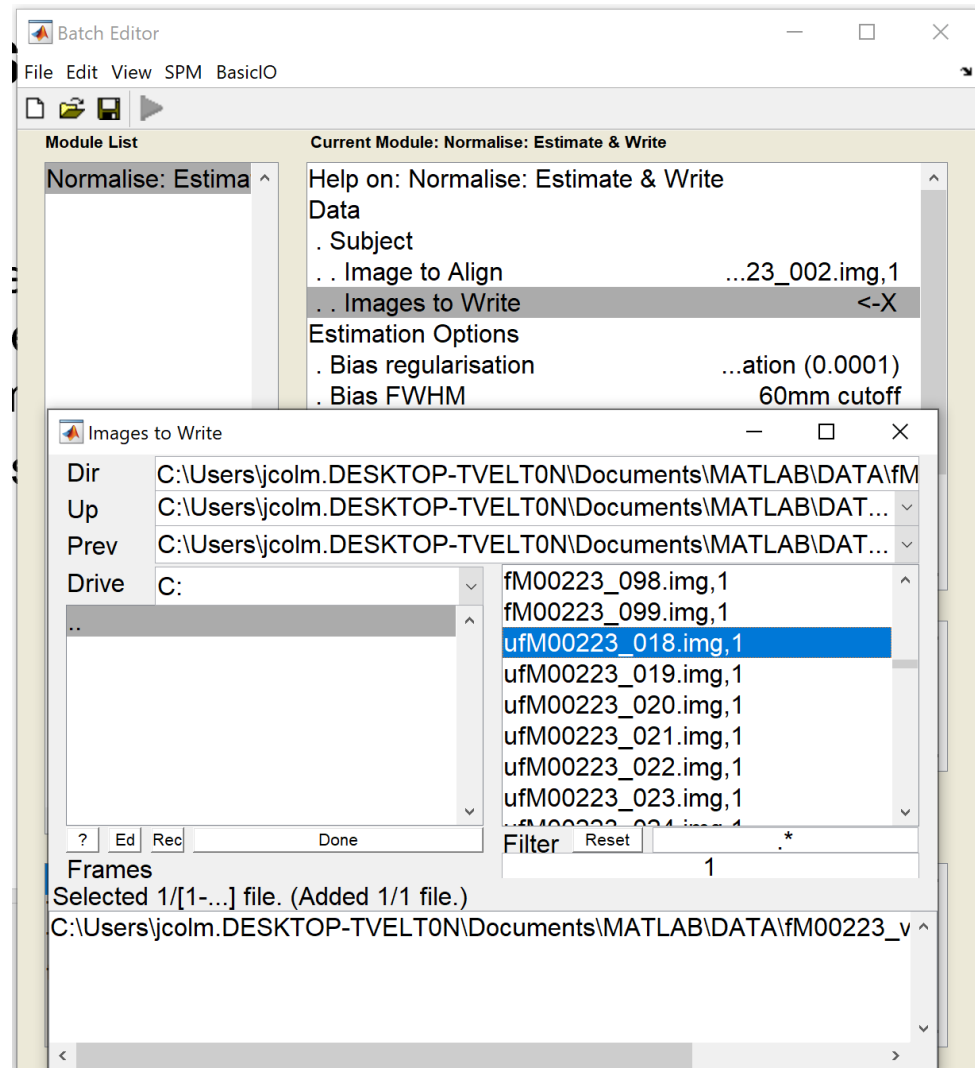
Step 2) Spatial normalisation

- Use the Normalise (Est & Wri) tool which normalises to 'MNI space'
- Select the structural image to align



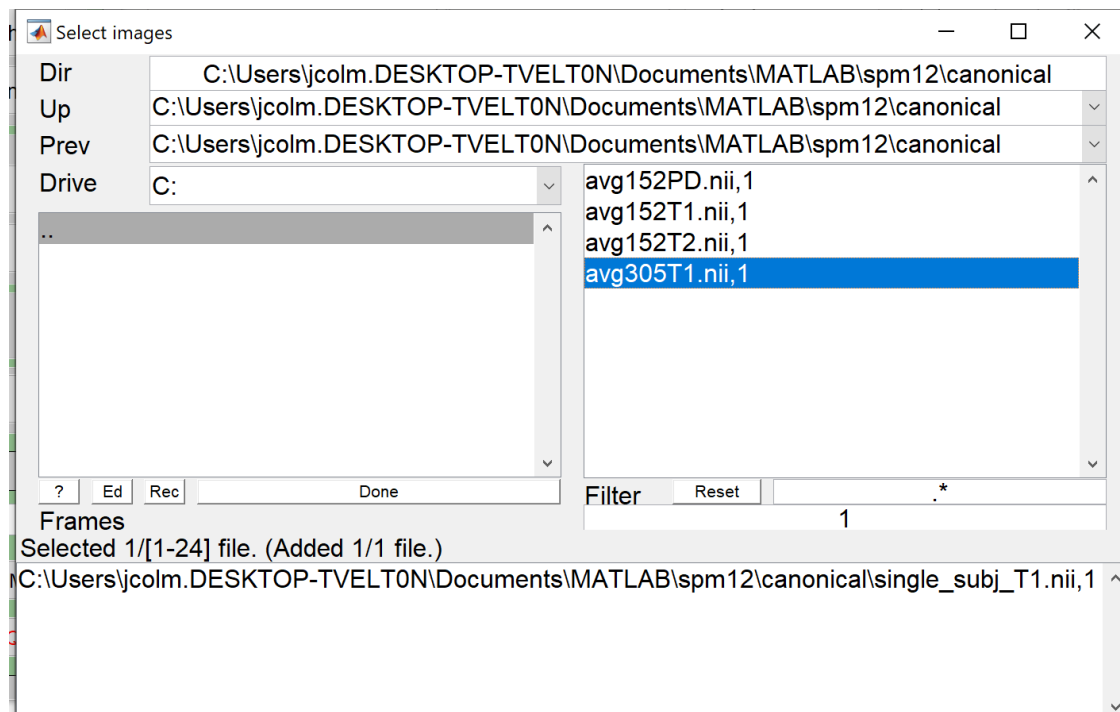
Step 2) Select images to write

- Select all the co-registered fMRI images to spatially normalised
- Also select the structural images to check the normalisation if you want
- Press run and have a cup of tea!



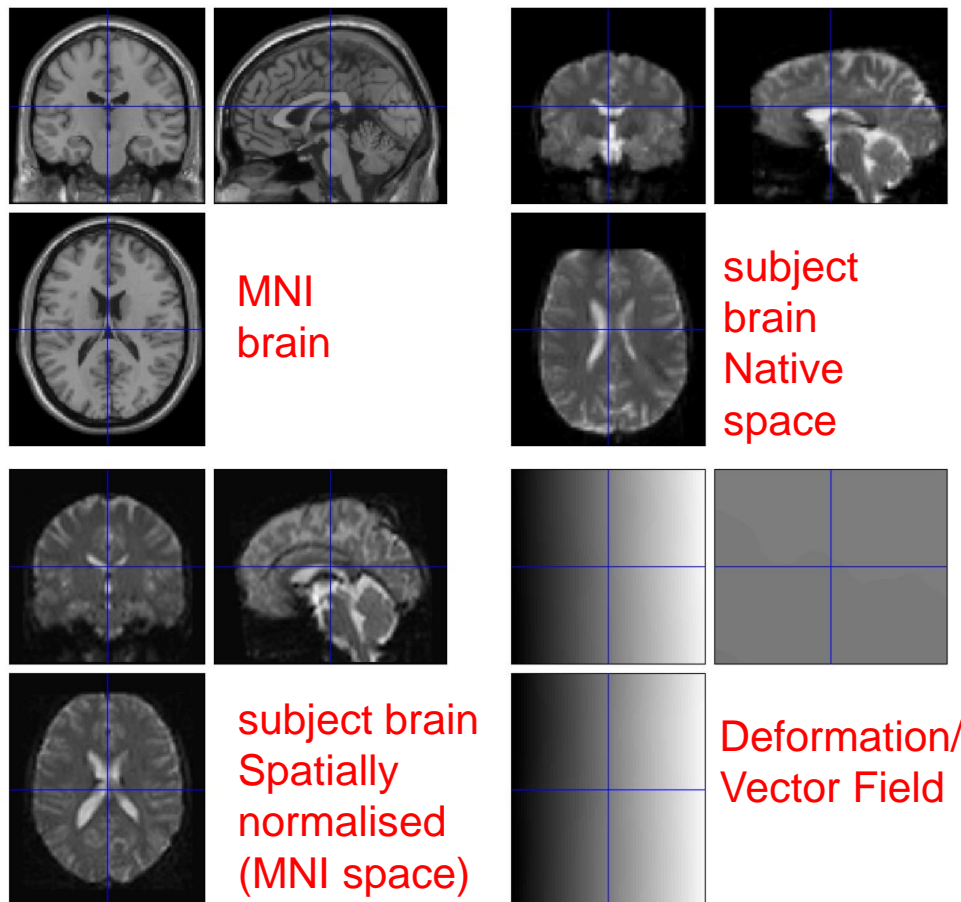
Step 2) Check against MNI space

- Go to the 'canonical' folder in the spm12 directory to find the MNI MRI images



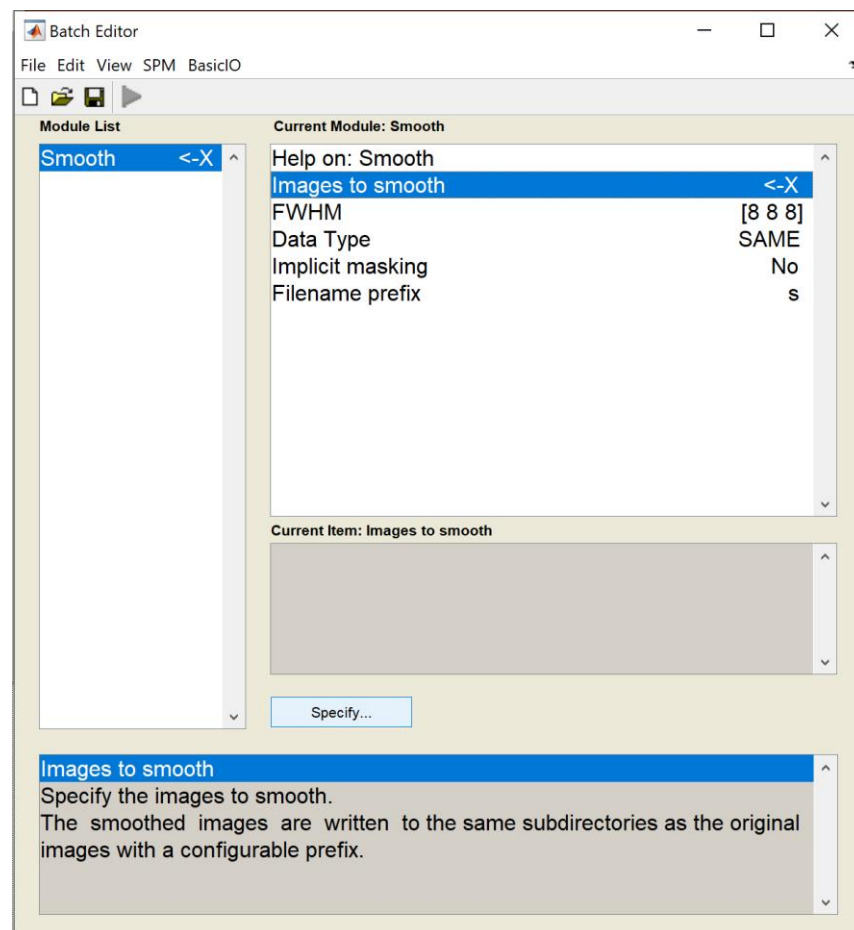
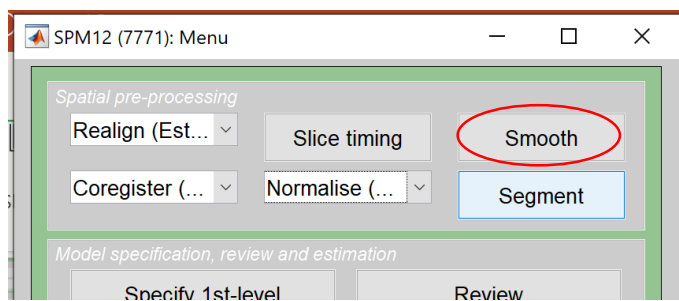
Step 2) Results

- You can use the 'Check Reg' function to check the results



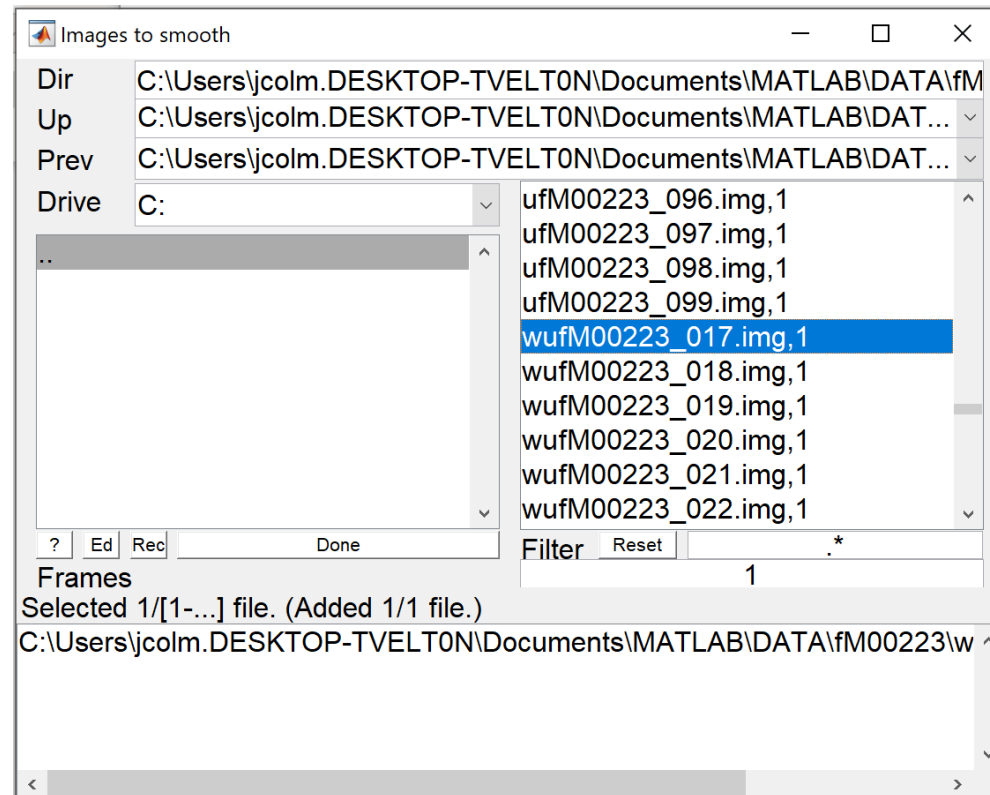
Step 3) Smoothing

- Select 'Smooth' function
- You can alter the width of the smooth by changing on the FWHM section
- 8mm is selected here
- The images will be prefixed with an s



Step 3) Smoothing

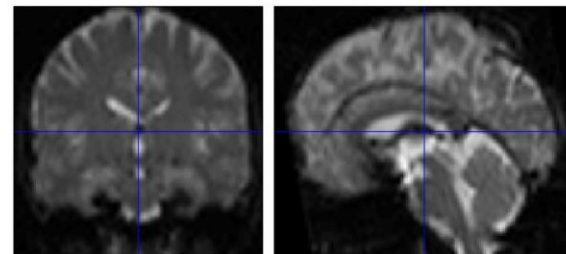
- Select all images which were spatially normalised
- Prefixed here with w for warped



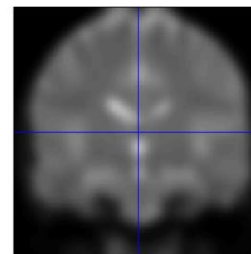
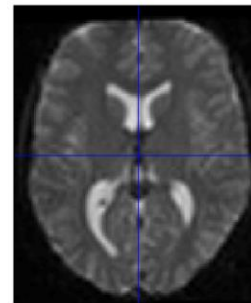
Step 3) Check results

- Use 'Check Reg' to compare smoothed and unsmoothed images
- The smoothed images are prefixed with s
- And you're done!

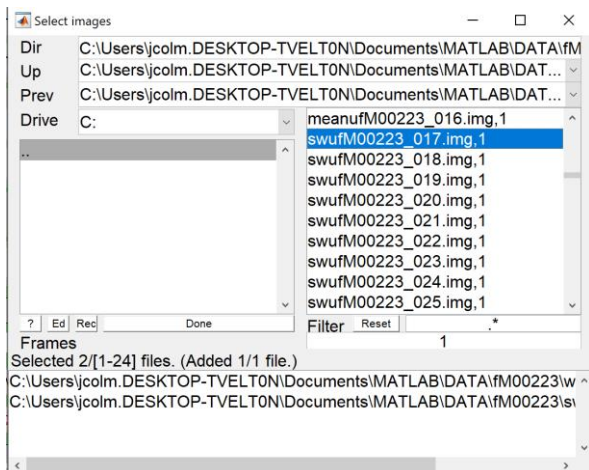
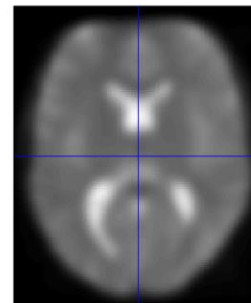
SPM12 (7771): Graphics
File Edit View Insert Tools Desktop Window SPM Figure Help



Unsmoothed



Smoothed

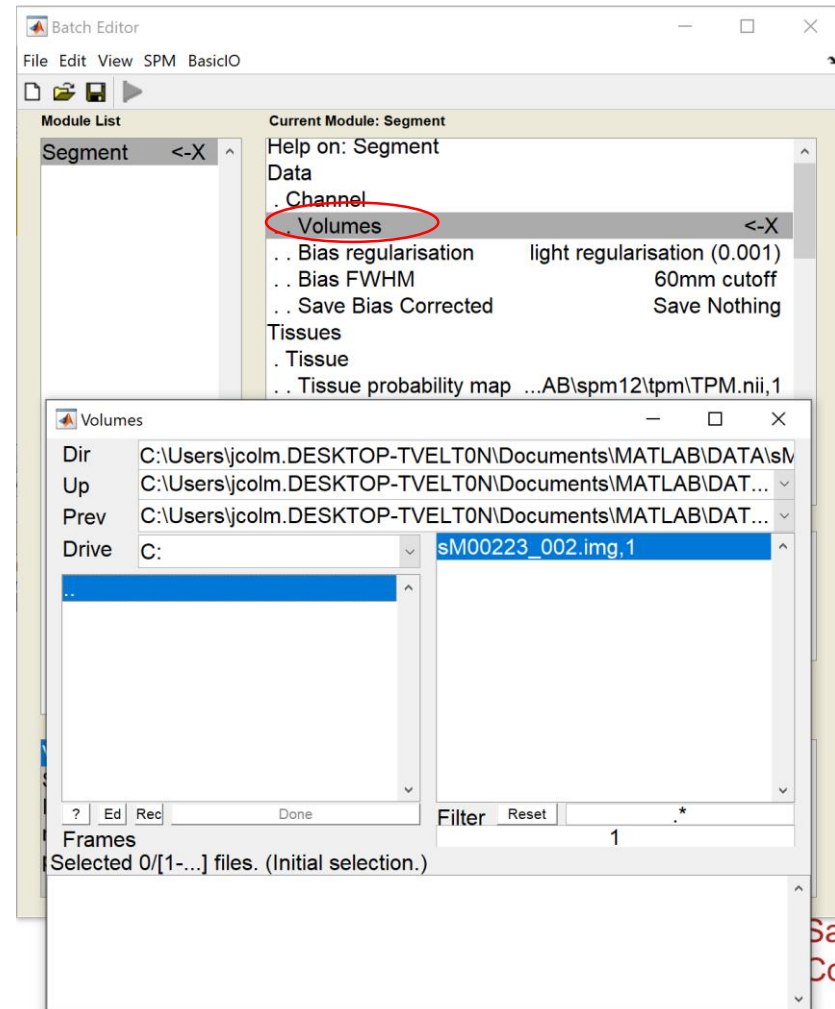
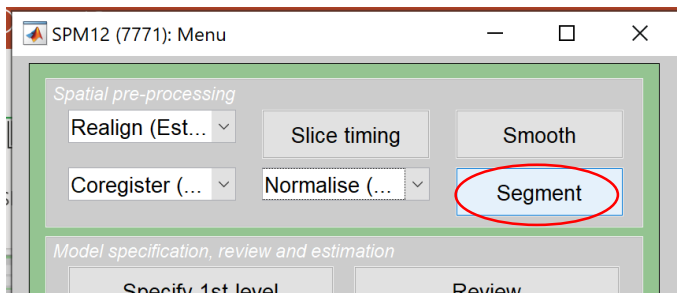


Alternate Step 2) Segment tool

- The segment tool can be used instead of the normalisation tool
- This is just here for extra info
- We include the following as an alternative to Step 2)

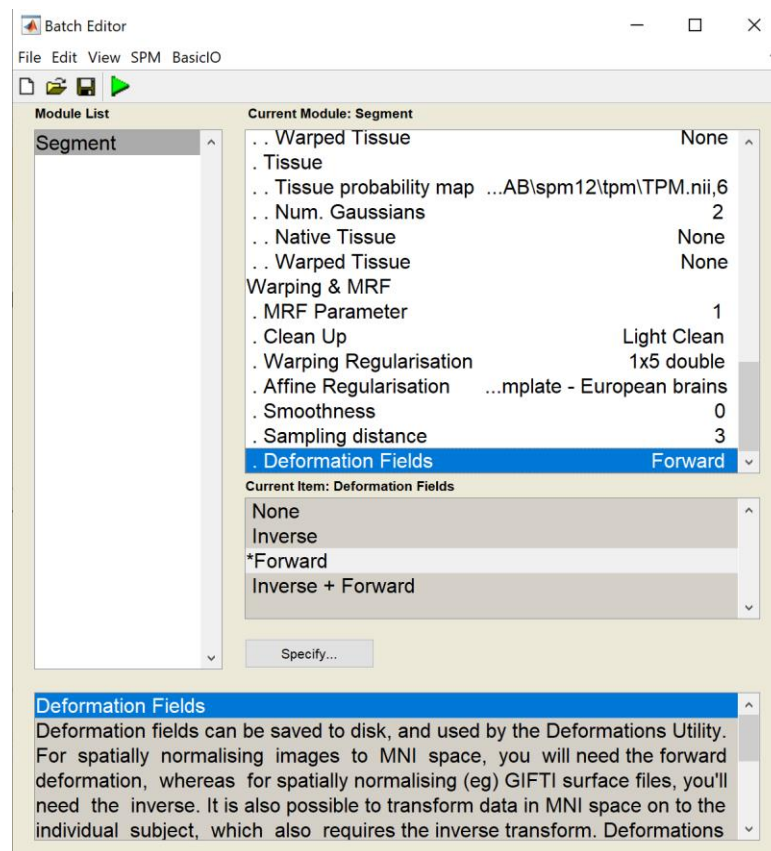
Spatial normalisation using 'Segment'

- Use the Segment tool which normalises to MNI
- It also produces a tissue segmentation and extracts the brain
- Select the structural image



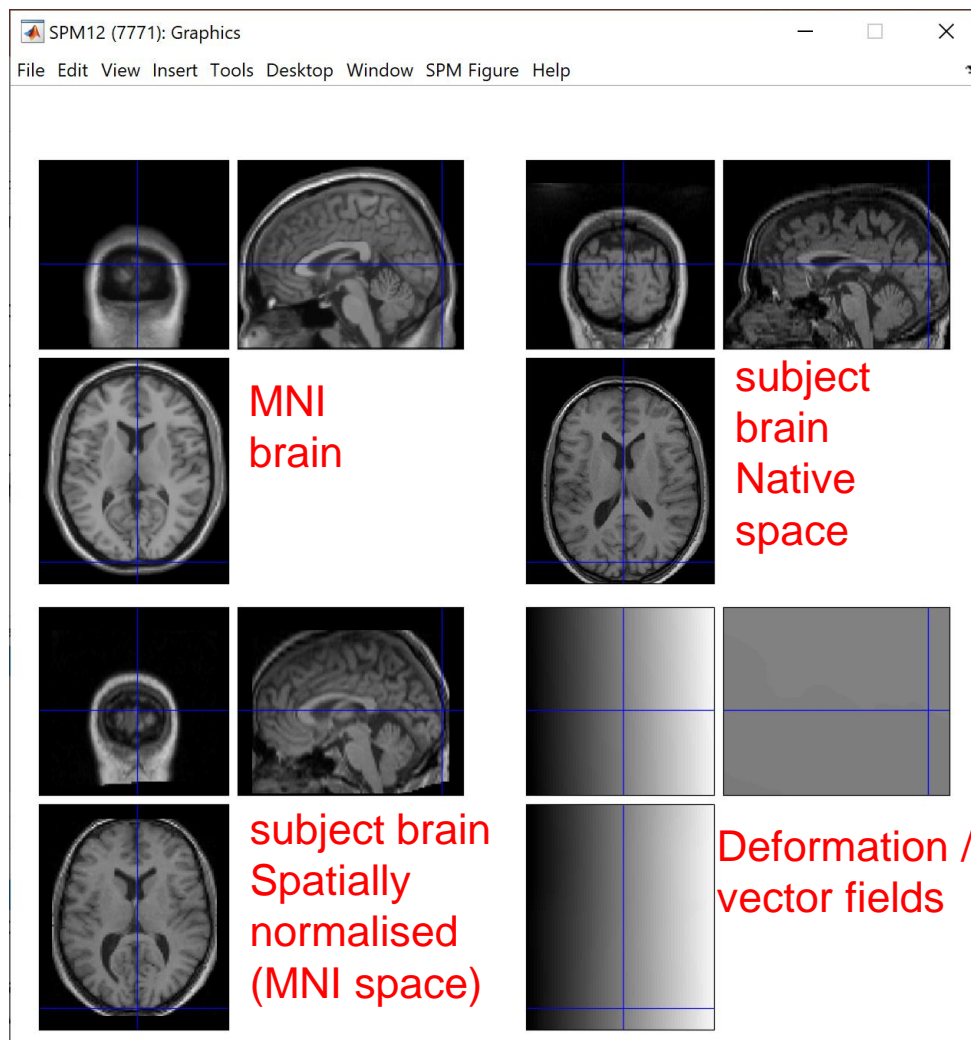
Segment tool: 'Deformation Fields'

- Ensure the 'Forward' option is selected for the Deformation Fields
- Then press run



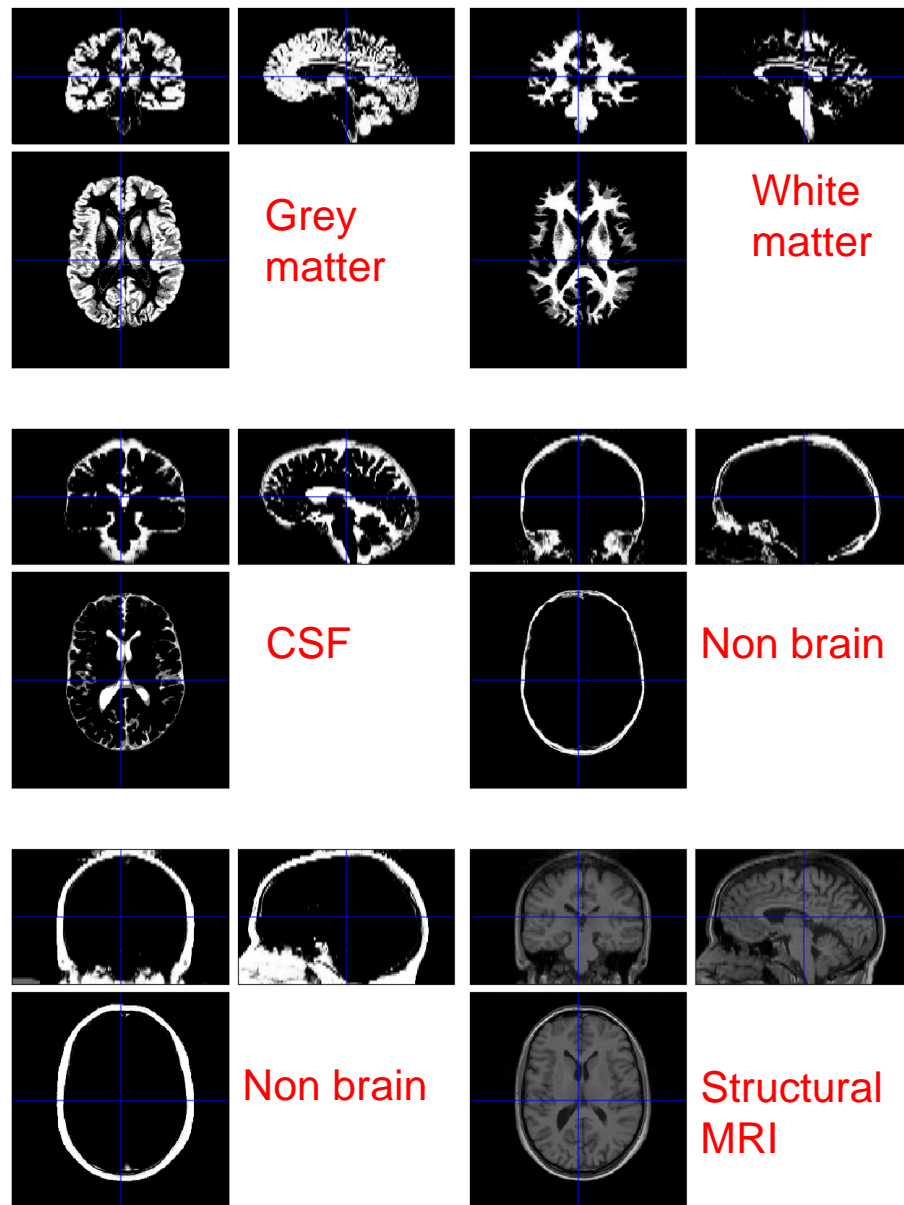
Structural image Results

- You can use the 'Check Reg' function to check the results



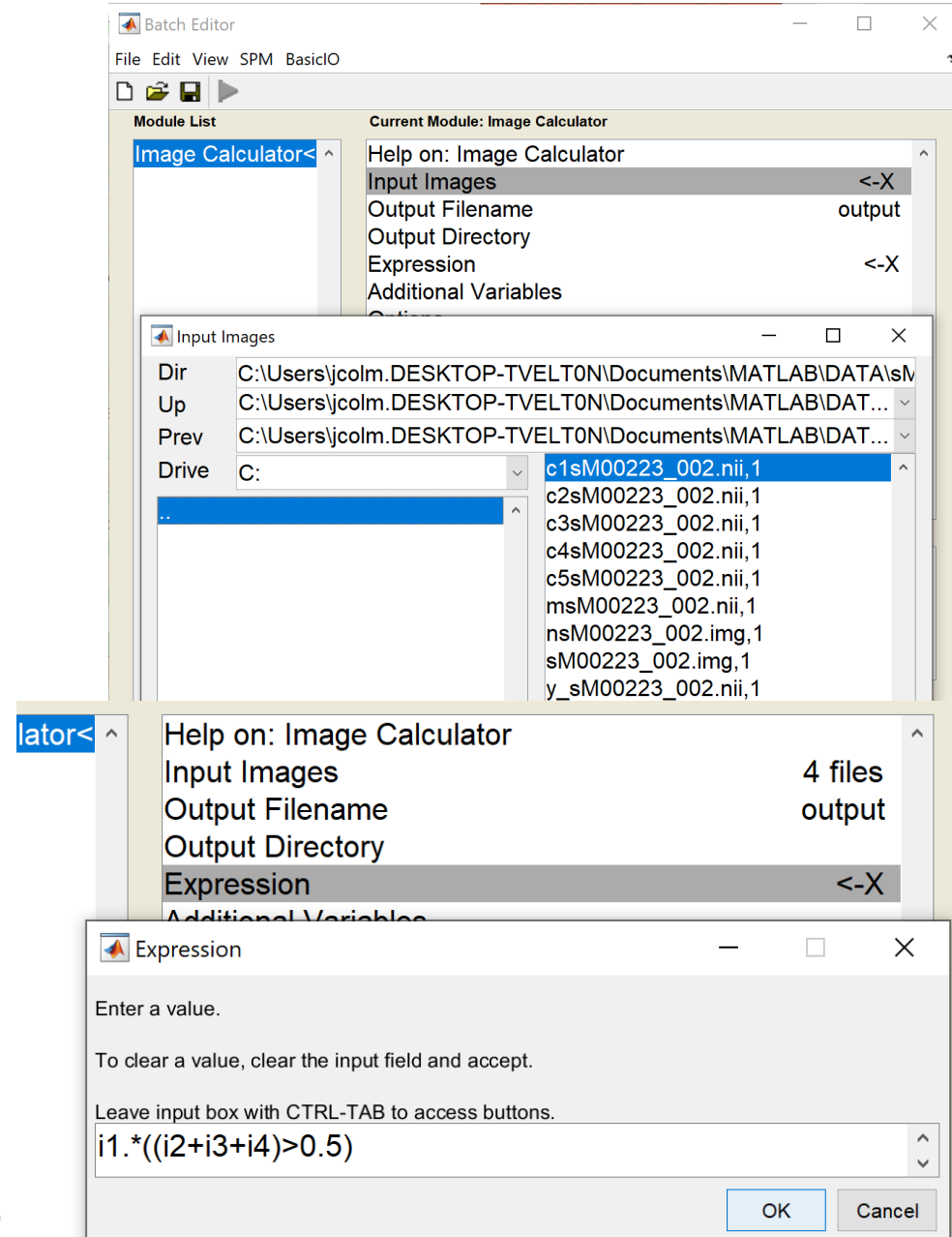
Segmentation Results

- A set of tissue segments are also produced



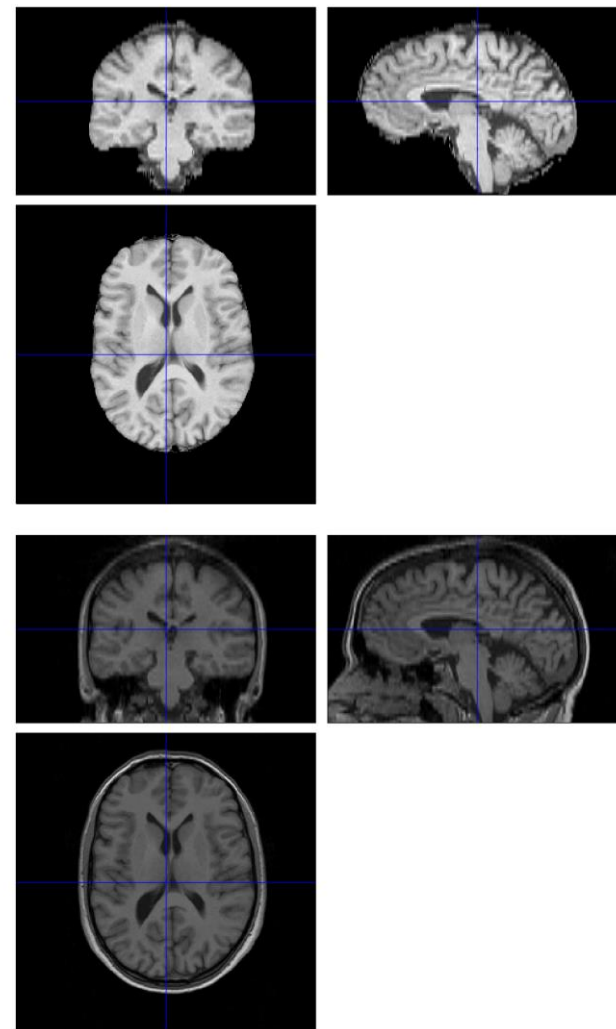
Skull stripping using segment output

- ImCalc can be used to extract the brain see expression
- the 'm' prefixed image is intensity nonuniformity corrected
- Tissue segmentations will be prefixed by 'c'
- Load 'm' image first then then c1 ,(grey matter) c2 (white matter) and c3 (CSF)



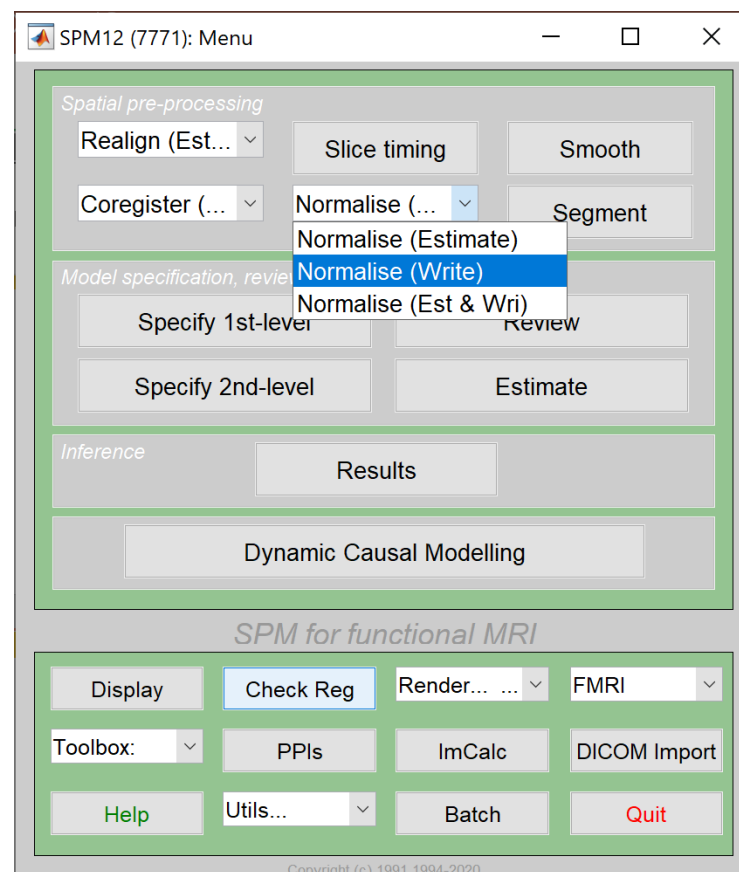
Skull stripping result

- See final INU and brain extracted result
- This new image can produce better co-registration results in step 1
- This help especially with severely effected INU images e.g. 7T MRI



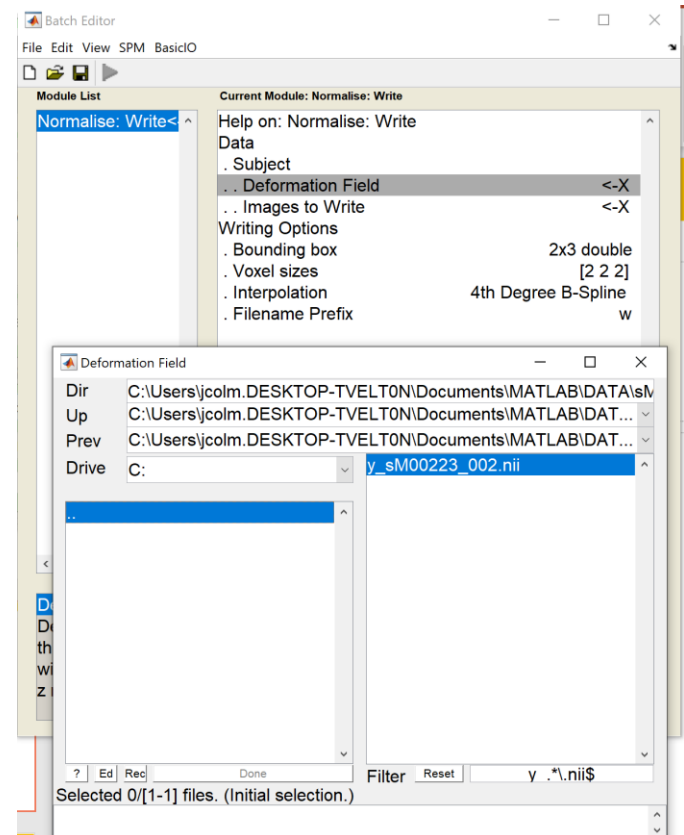
Now Apply spatial normalisation to fMRI data

- Now you can use the ‘Deformation / vector field’ to warp the fMRI images
- Select the ‘Normalise (Write)’ tool



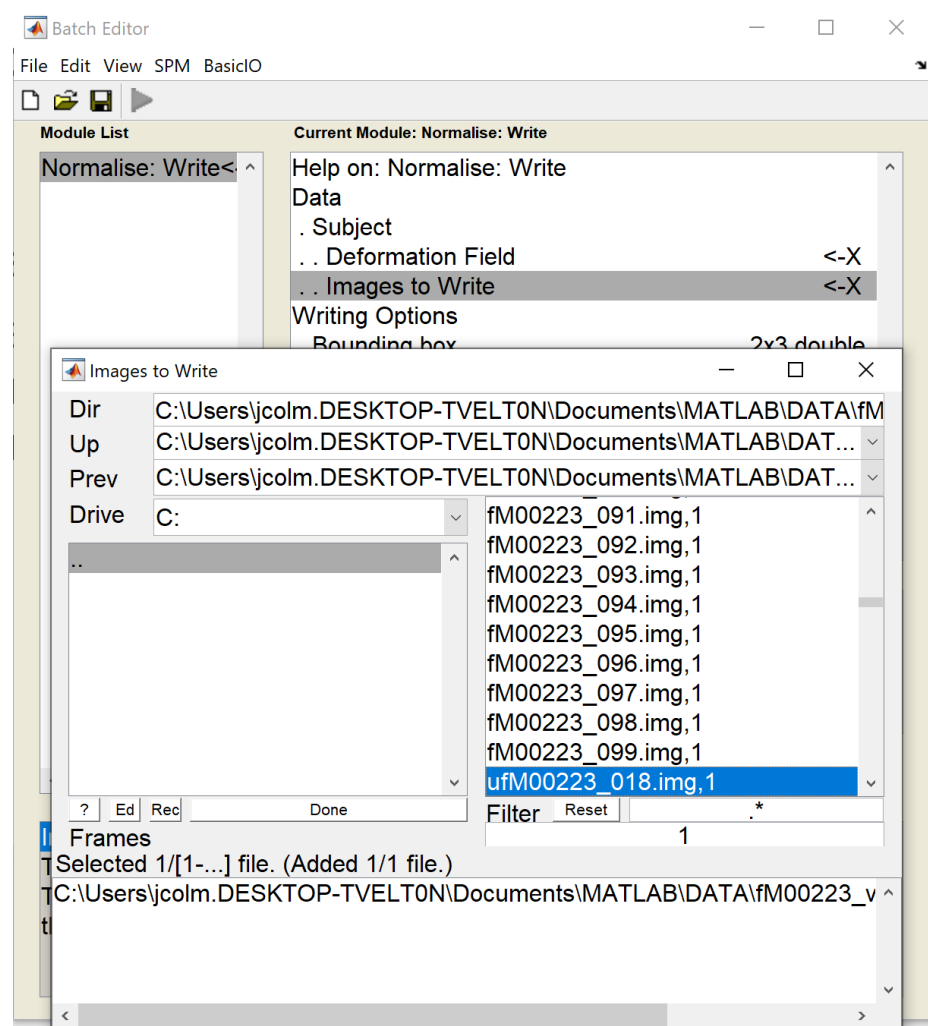
Select Deformation Field made by the segment tool

- Go to the structural image folder
- The deformation field will have a y prefix



Select fMRI data

- Press 'Images to Write'
- Select all fMRI images
Co-registered to the
Structural image
- Press run
- You can now proceed
with step 3 / smoothing



Q & A