# afni_proc.py is your *friend*

*or it will be soon*

Example scripts from

**`AFNI_data6/FT_analysis`**

Also see https://arxiv.org/abs/1709.07471

Appendix has processing scripts

# What the h--l is afni_proc.py?

- It is a Python program that
  - Takes as input a series of “options” describing processing steps to use to analyze datasets *from one subject*
  - Produces as output a Unix tcsh script file that runs all the **AFNI** programs to do the processing
- Reasons to use afni_proc.py
  - It is flexible and compact, to produce a long script
  - The output script not only does the data analysis, but also saves various diagnostic tools and files
  - All intermediate output datasets are saved to help diagnose things when results are confusing or just plain wrong
  - You can get help from us on the **AFNI** message board

# Where do afni_proc.py command lines/scripts come from?

- **Method #1**:
  - take an existing script (from yourself or a friend), and modify it to meet your needs
- **Method #2**:
  - find an approximate fit to what you want in examples, or from afni_proc.py’s help, and modify to meet your needs
- **Method #$\sqrt{-1}$**:
  - use GUI uber_subject.py
- **Method #666**:
  - beg for help on the **AFNI** message board

# Starting Simple - 1

```
afni_proc.py -subj_id FT                 \
        -dsets FT/FT_epi_r?+orig.HEAD    \
        -copy_anat FT/FT_anat+orig       \
        -tcat_remove_first_trs 2         \
        -regress_stim_times FT/AV*.txt   \
        -regress_stim_labels Vrel Arel   \
        -regress_basis 'BLOCK(20,1)'     \
        -regress_est_blur_errts          \
        -regress_opts_3dD                \
            -gltsym 'SYM: Vrel -Arel'    \
            -glt_label 1 V-A
```

Script file
s01.ap.simple

# Starting Simple - 2

```
afni_proc.py -subj_id FT                                 \
        -dsets FT/FT_epi_r?+orig.HEAD                    \
        -copy_anat FT/FT_anat+orig                       \
        -tcat_remove_first_trs 2                         \
        -regress_stim_times FT/AV*.txt                   \
        -regress_stim_labels Vrel Arel                   \
        -regress_basis 'BLOCK(20,1)'                     \
        -regress_est_blur_errts                          \
        -regress_opts_3dD                                \
            -gltsym 'SYM: Vrel -Arel'                    \
            -glt_label 1 V-A
```

ID will label output files

Script file s01.ap.simple

# Starting Simple - 3

```
afni_proc.py -subj_id FT                          \
        -dsets FT/FT_epi_r?+orig.HEAD             \
        -copy_anat FT/FT_anat+orig                \
        -tcat_remove_first_trs 2                  \
        -regress_stim_times FT/AV*.txt            \
        -regress_stim_labels Vrel Arel            \
        -regress_basis 'BLOCK(20,1)'              \
        -regress_est_blur_errts                   \
        -regress_opts_3dD                         \
            -gltsym 'SYM: Vrel -Arel'             \
            -glt_label 1 V-A
```

EPI time series datasets to analyze

Script file s01.ap.simple

# Starting Simple - 4

```
afni_proc.py -subj_id FT                        \
        -dsets FT/FT_epi_r?+orig.HEAD           \
        -copy_anat FT/FT_anat+orig              \
        -tcat_remove_first_trs 2                \
        -regress_stim_times FT/AV*.txt          \
        -regress_stim_labels Vrel Arel          \
        -regress_basis 'BLOCK(20,1)'            \
        -regress_est_blur_errts                 \
        -regress_opts_3dD                       \
            -gltsym 'SYM: Vrel -Arel'           \
            -glt_label 1 V-A
```

T1-weighted anatomical dataset for alignment to EPI datasets

Script file s01.ap.simple

# Starting Simple - 5

```
afni_proc.py -subj_id FT                            \
        -dsets FT/FT_epi_r?+orig.HEAD               \
        -copy_anat FT/FT_anat+orig                  \
        -tcat_remove_first_trs 2                    \
        -regress_stim_times FT/AV*.txt              \
        -regress_stim_labels Vrel Arel              \
        -regress_basis 'BLOCK(20,1)'                \
        -regress_est_blur_errts                     \
        -regress_opts_3dD                           \
            -gltsym 'SYM: Vrel -Arel'               \
            -glt_label 1 V-A
```

Stimulus timing files, labels, and HRF model; **Note**: timing files have ***start*** times for each task iteration

Script file s01.ap.simple

# Starting Simple - 6

```
afni_proc.py -subj_id FT                 \
        -dsets FT/FT_epi_r?+orig.HEAD    \
        -copy_anat FT/FT_anat+orig       \
        -tcat_remove_first_trs 2         \
        -regress_stim_times FT/AV*.txt   \
        -regress_stim_labels Vrel Arel   \
        -regress_basis 'BLOCK(20,1)'     \
        -regress_est_blur_errts          \
        -regress_opts_3dD                \
            -gltsym 'SYM: Vrel -Arel'    \
            -glt_label 1 V-A
```

Estimate smoothness of EPI noise for group analysis

Script file s01.ap.simple

# Starting Simple - 7

```
afni_proc.py -subj_id FT                 \
        -dsets FT/FT_epi_r?+orig.HEAD    \
        -copy_anat FT/FT_anat+orig       \
        -tcat_remove_first_trs 2         \
        -regress_stim_times FT/AV*.txt   \
        -regress_stim_labels Vrel Arel   \
        -regress_basis 'BLOCK(20,1)'     \
        -regress_est_blur_errts          \
        -regress_opts_3dD                \
            -gltsym 'SYM: Vrel -Arel'    \
            -glt_label 1 V-A
```

Set up **General** **Linear** **Test** between 2 conditions

Script file s01.ap.simple

# A Real Case - 1

Script file
s05.ap.uber

```
#!/usr/bin/env tcsh

# creation date: Thu Sep 10 14:27:59 2015

# set data directories
set top_dir    = FT
# set subject and group identifiers
set subj       = FT
set group_id   = horses
```

Not actually used here

Code subject level information
into shell variables:
Makes it easier to re-use this afni_proc.py command

# A Real Case - 2

Script file
s05.ap.uber

```
afni_proc.py -subj_id $subj                               \
 -script proc.$subj -scr_overwrite                        \
 -blocks tshift align tlrc volreg blur mask scale regress \
 -copy_anat $top_dir/FT_anat+orig                         \
-dsets                                                   \
     $top_dir/FT_epi_r1+orig.HEAD                         \
     $top_dir/FT_epi_r2+orig.HEAD                         \
     $top_dir/FT_epi_r3+orig.HEAD                         \
 -volreg_align_to MIN_OUTLIER                             \
 -volreg_align_e2a                                        \
 -volreg_tlrc_warp                                        \
 -blur_size 4.0                                           \
 -tcat_remove_first_trs 2                                 \
 -regress_stim_times                                      \
     $top_dir/AV1_vis.txt                                 \
     $top_dir/AV2_aud.txt                                 \
 -regress_stim_labels                                     \
     vis aud                                              \
 -regress_basis 'BLOCK(20,1)'                             \
 -regress_censor_motion 0.3                               \
 -regress_opts_3dD                                        \
     -jobs 2                                              \
     -gltsym 'SYM: vis -aud' -glt_label 1 V-A             \
     -gltsym 'SYM: 0.5*vis +0.5*aud' -glt_label 2 mean.VA \
 -regress_compute_fitts                                   \
 -regress_make_ideal_sum sum_ideal.1D                     \
 -regress_est_blur_epits                                  \
 -regress_est_blur_errts                                  \
 -regress_run_clustsim yes                                \
 -execute
```

The entire afni_proc.py command:
Font size will be bigger on following slides!

# A Real Case – 3a

Script file
s05.ap.uber

```
afni_proc.py -subj_id $subj                                     \
  -script proc.$subj -scr_overwrite                             \
  -blocks tshift align tlrc volreg blur mask scale regress      \
  -copy_anat $top_dir/FT_anat+orig                              \
  -dsets                                                        \
      $top_dir/FT_epi_r1+orig.HEAD                              \
      $top_dir/FT_epi_r2+orig.HEAD                              \
      $top_dir/FT_epi_r3+orig.HEAD                              \
  -volreg_align_to MIN_OUTLIER                                  \
  -volreg_align_e2a                                             \
  -volreg_tlrc_warp                                             \
  -blur_size 4.0                                                \
```

Set up which
processing
”blocks”
will be run

Script file
s05.ap.uber

# A Real Case – 3b

```
afni_proc.py -subj_id $subj                               \
 -script proc.$subj -scr_overwrite                        \
 -blocks tshift align tlrc volreg blur mask scale regress \
 -copy_anat $top_dir/FT_anat+orig                         \
 -dsets                                                   \
     $top_dir/FT_epi_r1+orig.HEAD                         \
     $top_dir/FT_epi_r2+orig.HEAD                         \
     $top_dir/FT_epi_r3+orig.HEAD                         \
 -volreg_align_to MIN_OUTLIER                             \
 -volreg_align_e2a                                        \
 -volreg_tlrc_warp                                        \
 -blur_size 4.0                                           \
```

Select input datasets (anat and EPI)

Script file
s05.ap.uber

# A Real Case – 3c

```
afni_proc.py -subj_id $subj                               \
 -script proc.$subj -scr_overwrite                        \
 -blocks tshift align tlrc volreg blur mask scale regress \
 -copy_anat $top_dir/FT_anat+orig                         \
 -dsets                                                   \
     $top_dir/FT_epi_r1+orig.HEAD                         \
     $top_dir/FT_epi_r2+orig.HEAD                         \
     $top_dir/FT_epi_r3+orig.HEAD                         \
 -volreg_align_to MIN_OUTLIER                             \
 -volreg_align_e2a                                        \
 -volreg_tlrc_warp                                        \
 -blur_size 4.0                                           \
```

Specify how
“volreg” step
will operate

# A Real Case – 3d

Script file
s05.ap.uber

```
afni_proc.py -subj_id $subj                                   \
 -script proc.$subj -scr_overwrite                            \
 -blocks tshift align tlrc volreg blur mask scale regress \
 -copy_anat $top_dir/FT_anat+orig                             \
 -dsets                                                        \
     $top_dir/FT_epi_r1+orig.HEAD                             \
     $top_dir/FT_epi_r2+orig.HEAD                             \
     $top_dir/FT_epi_r3+orig.HEAD                             \
 -volreg_align_to MIN_OUTLIER                                 \
 -volreg_align_e2a                                            \
 -volreg_tlrc_warp                                            \
 -blur_size 4.0                                               \
```

Specify how much spatial blurring will be used (FWHM mm)

# A Real Case – 4a

Script file
s05.ap.uber

```
-tcat_remove_first_trs 2                                 \
-regress_stim_times                                      \
    $top_dir/AV1_vis.txt                                 \
    $top_dir/AV2_aud.txt                                 \
-regress_stim_labels                                     \
    vis aud                                              \
-regress_basis 'BLOCK(20,1)'                             \
-regress_censor_motion 0.3                               \
-regress_opts_3dD                                        \
    -jobs 2                                              \
    -gltsym 'SYM: vis -aud' -glt_label 1 V-A             \
    -gltsym 'SYM: 0.5*vis +0.5*aud' -glt_label 2 mean.VA \
-regress_compute_fitts                                   \
-regress_make_ideal_sum sum_ideal.1D                     \
-regress_est_blur_epits                                  \
-regress_est_blur_errts                                  \
-regress_run_clustsim yes                                \
-execute
```

Specify task timing, labels, and response model;
**Note**: task timing files contain ***start*** times for each task iteration

# A Real Case – 4b

Script file
s05.ap.uber

```
-tcat_remove_first_trs 2                                   \
-regress_stim_times                                        \
    $top_dir/AV1_vis.txt                                   \
    $top_dir/AV2_aud.txt                                   \
-regress_stim_labels                                       \
    vis aud                                                \
-regress_basis 'BLOCK(20,1)'                               \
-regress_censor_motion 0.3                                 \
-regress_opts_3dD                                          \
    -jobs 2                                                \
    -gltsym 'SYM: vis -aud' -glt_label 1 V-A               \
    -gltsym 'SYM: 0.5*vis +0.5*aud' -glt_label 2 mean.VA \
-regress_compute_fitts                                     \
-regress_make_ideal_sum sum_ideal.1D                       \
-regress_est_blur_epits                                    \
-regress_est_blur_errts                                    \
-regress_run_clustsim yes                                  \
-execute
```

Maximum motion (in mm) to accept between successive TRs

# A Real Case – 4c

Script file
s05.ap.uber

```
-tcat_remove_first_trs 2                                 \
-regress_stim_times                                      \
    $top_dir/AV1_vis.txt                                 \
    $top_dir/AV2_aud.txt                                 \
-regress_stim_labels                                     \
    vis aud                                              \
-regress_basis 'BLOCK(20,1)'                             \
-regress_censor_motion 0.3                               \
-regress_opts_3dD                                        \
    -jobs 2                                              \
    -gltsym 'SYM: vis -aud' -glt_label 1 V-A             \
    -gltsym 'SYM: 0.5*vis +0.5*aud' -glt_label 2 mean.VA \
-regress_compute_fitts                                   \
-regress_make_ideal_sum sum_ideal.1D                     \
-regress_est_blur_epits                                  \
-regress_est_blur_errts                                  \
-regress_run_clustsim yes                                \
-execute
```

Other regression options:
Use 2 CPUs
Set up GLTs

# A Real Case – 4d

Script file
s05.ap.uber

```
-tcat_remove_first_trs 2                                 \
-regress_stim_times                                      \
    $top_dir/AV1_vis.txt                                 \
    $top_dir/AV2_aud.txt                                 \
-regress_stim_labels                                     \
    vis aud                                              \
-regress_basis 'BLOCK(20,1)'                             \
-regress_censor_motion 0.3                               \
-regress_opts_3dD                                        \
    -jobs 2                                              \
    -gltsym 'SYM: vis -aud' -glt_label 1 V-A             \
    -gltsym 'SYM: 0.5*vis +0.5*aud' -glt_label 2 mean.VA \
-regress_compute_fitts                                   \
-regress_make_ideal_sum sum_ideal.1D                     \
-regress_est_blur_epits                                  \
-regress_est_blur_errts                                  \
-regress_run_clustsim yes                                \
-execute
```

Other regression options:
Compute fitted model
(best fit to data);
Create sum of task ideal
response time series
(for display purposes)

# A Real Case – 4e

Script file
s05.ap.uber

```
-tcat_remove_first_trs 2                                 \
-regress_stim_times                                      \
    $top_dir/AV1_vis.txt                                 \
    $top_dir/AV2_aud.txt                                 \
-regress_stim_labels                                     \
    vis aud                                              \
-regress_basis 'BLOCK(20,1)'                             \
-regress_censor_motion 0.3                               \
-regress_opts_3dD                                        \
    -jobs 2                                              \
    -gltsym 'SYM: vis -aud' -glt_label 1 V-A             \
    -gltsym 'SYM: 0.5*vis +0.5*aud' -glt_label 2 mean.VA \
-regress_compute_fitts                                   \
-regress_make_ideal_sum sum_ideal.1D                     \
-regress_est_blur_epits                                  \
-regress_est_blur_errts                                  \
-regress_run_clustsim yes                                \
-execute
```

Estimate smoothness of ***noise*** in the data: From the dataset itself, From the *residuals* (=data-model fit).

And estimate cluster-size thresholds from smoothness estimates

# A Real Case – 4f

Script file
s05.ap.uber

```
-tcat_remove_first_trs 2                                        \
-regress_stim_times                                             \
    $top_dir/AV1_vis.txt                                        \
    $top_dir/AV2_aud.txt                                        \
-regress_stim_labels                                            \
    vis aud                                                     \
-regress_basis 'BLOCK(20,1)'                                    \
-regress_censor_motion 0.3                                      \
-regress_opts_3dD                                               \
    -jobs 2                                                     \
    -gltsym 'SYM: vis -aud' -glt_label 1 V-A                    \
    -gltsym 'SYM: 0.5*vis +0.5*aud' -glt_label 2 mean.VA \
-regress_compute_fitts                                          \
-regress_make_ideal_sum sum_ideal.1D                            \
-regress_est_blur_epits                                         \
-regress_est_blur_errts                                         \
-regress_run_clustsim yes                                       \
-execute
```

Run script after
it is created

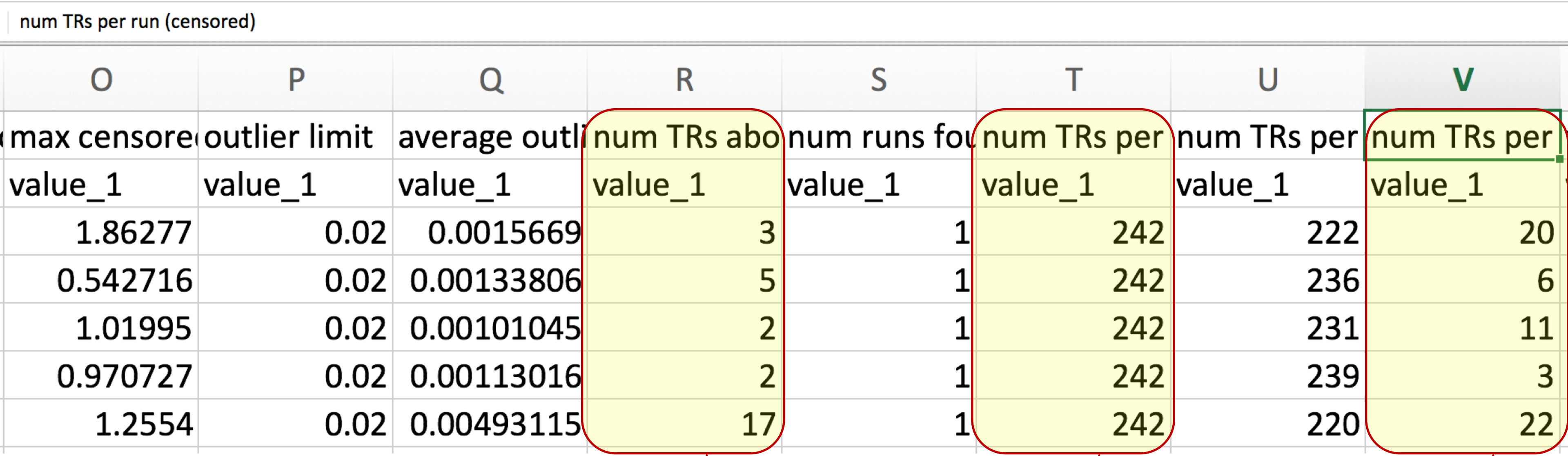
# Summarizing Results

- Each afni_proc.py **`results`** directory has a file with a name like **`out.ss_review.SUBJECT-ID.txt`**
  - Each line give some information about the data and the processing results, such as number of time points censored
- A command like this will generate a table with all these summary results from all subjects:

```
gen_ss_review_table.py
 -infiles data_orig/sub*/*.results/out.ss_review.*.txt
 -tablefile UCLA.xls
```

- Next slide: what part of this output looks like in Excel
  - On Linux, you can use LibreOffice **http://www.libreoffice.org/**

# Excel Snapshot Image of UCLA.xls



num TRs per run (censored)

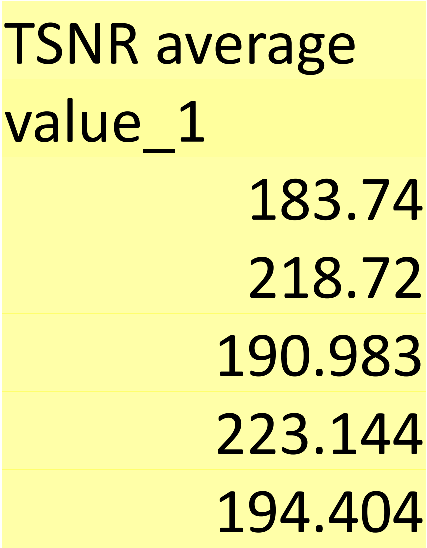
| O | P | Q | R | S | T | U | V |
|---|---|---|---|---|---|---|---|
| max censore | outlier limit | average outl | num TRs abo | num runs fou | num TRs per | num TRs per | num TRs per |
| value_1 | value_1 | value_1 | value_1 | value_1 | value_1 | value_1 | value_1 |
| 1.86277 | 0.02 | 0.0015669 | 3 | 1 | 242 | 222 | 20 |
| 0.542716 | 0.02 | 0.00133806 | 5 | 1 | 242 | 236 | 6 |
| 1.01995 | 0.02 | 0.00101045 | 2 | 1 | 242 | 231 | 11 |
| 0.970727 | 0.02 | 0.00113016 | 2 | 1 | 242 | 239 | 3 |
| 1.2554 | 0.02 | 0.00493115 | 17 | 1 | 242 | 220 | 22 |

Number of TRs with too many outlier values for each subject

Number of TRs found for each subject

Number of TRs censored for each subject

# Another Valuable Summary: TSNR



| TSNR average value_1 |
|---:|
| 183.74 |
| 218.72 |
| 190.983 |
| 223.144 |
| 194.404 |

- Measures magnitude of EPI signal strength divided by standard deviation of noise
- For 3 Tesla data, TSNR values near 180-200 are usual – with ”standard” scanning parameters (TR 2-3s, voxel size 2-3mm)
- If some subject’s TSNR is much lower than others, examine the data to find the problem!

# Masking for Group Analysis

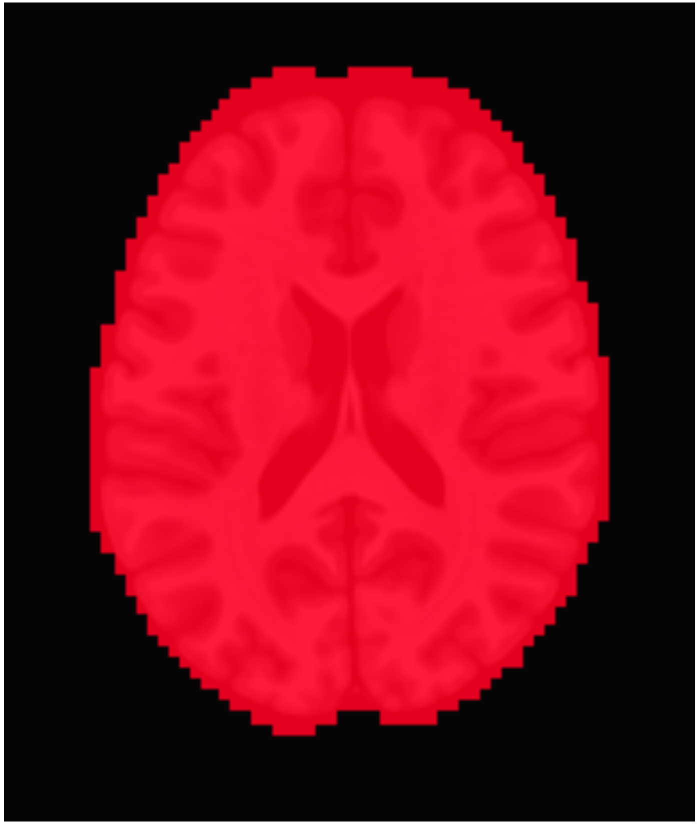
- In each `results` directory, the output dataset `mask_epi_anat.SUBJECT-ID+tlrc.HEAD` is the 0-1 brain mask of the EPI dataset in the template space
- Combine all these masks into one mask dataset:

```
3dmask_tool -input
 data_orig/sub-*/*.results/mask_epi_anat.*.HEAD
 -prefix mask_all.nii -frac 0.8
```

- Another way: use a gray matter plus CSF mask from MNI template (***if*** you have used nonlinear alignment to that template):

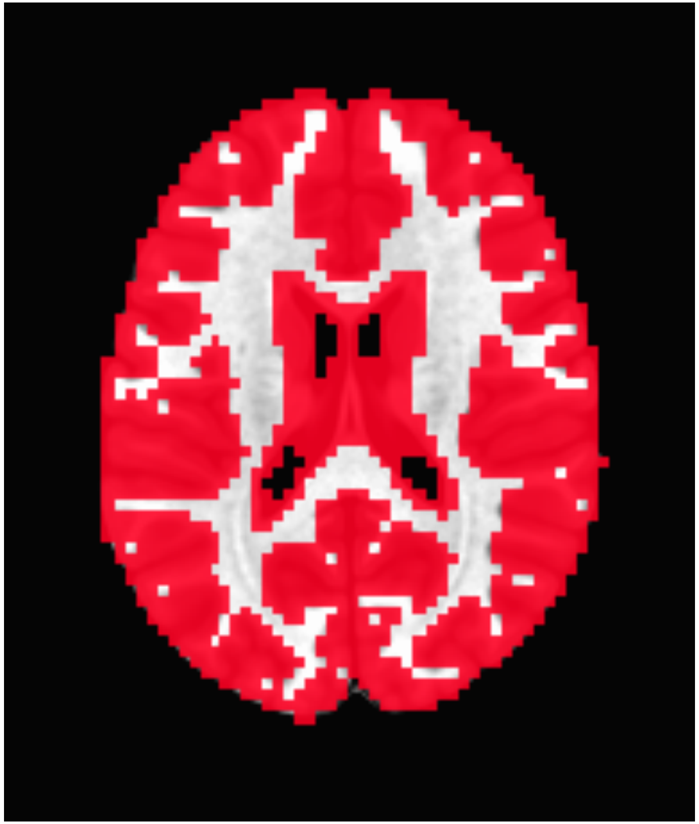
```
3dresample -master mask_all.nii -prefix mask_GC.nii
 -rmode NN
 -input ~/abin/MNI152_2009_template.nii.gz'[4]'
```

# Whole Brain and GM+CSF Masks



73517 voxels

3 mm$^3$ voxels

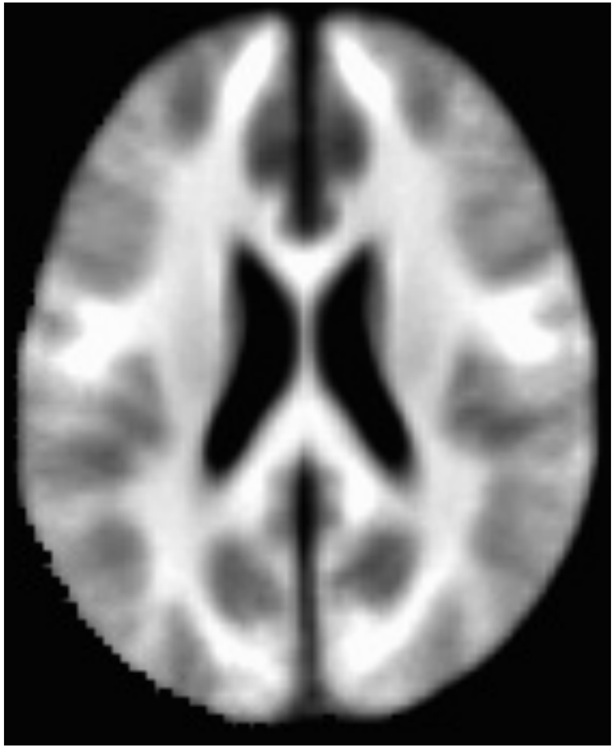


53104 voxels

# Nonlinear Warping to MNI Template

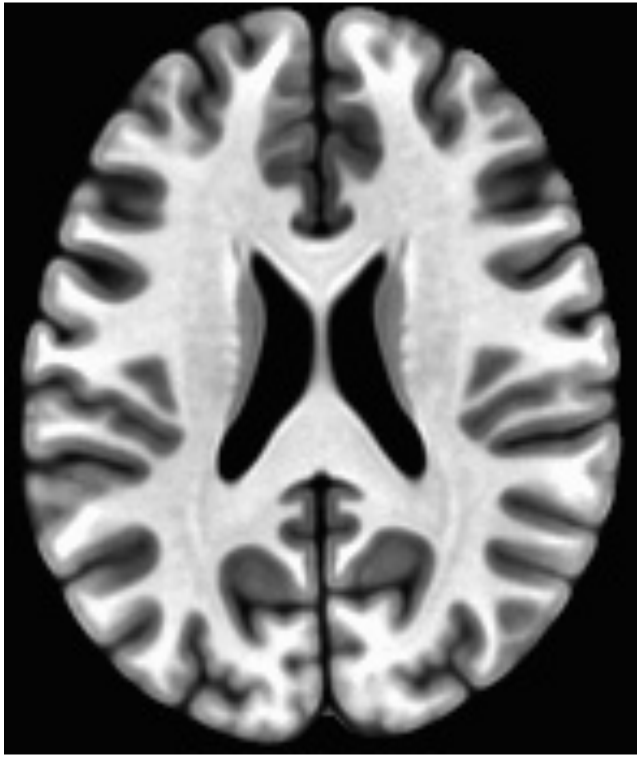
- afni_proc.py *can* do the nonlinear warping for you
  - But, nonlinear warping is slow (in fact, slowly slow)
  - If you need to re-rerun subject analysis, nonlinear warping will slow the re-run script down *a lot*
- Solution: do the nonlinear warping *before* using afni_proc.py, then supply the warping results so that afni_proc.py will skip doing the warping itself
- Mechanism: the **@SSwarper** script (tcsh)
  - Does Skull Stripping ("SS") and nonlinear warping
  - Base dataset is **MNI152_2009_template_SSW.nii.gz**
    - Nonlinearly warped, not too blurry

# Two MNI Templates



**MNI152_1mm_uni+tlrc**

Affine alignments



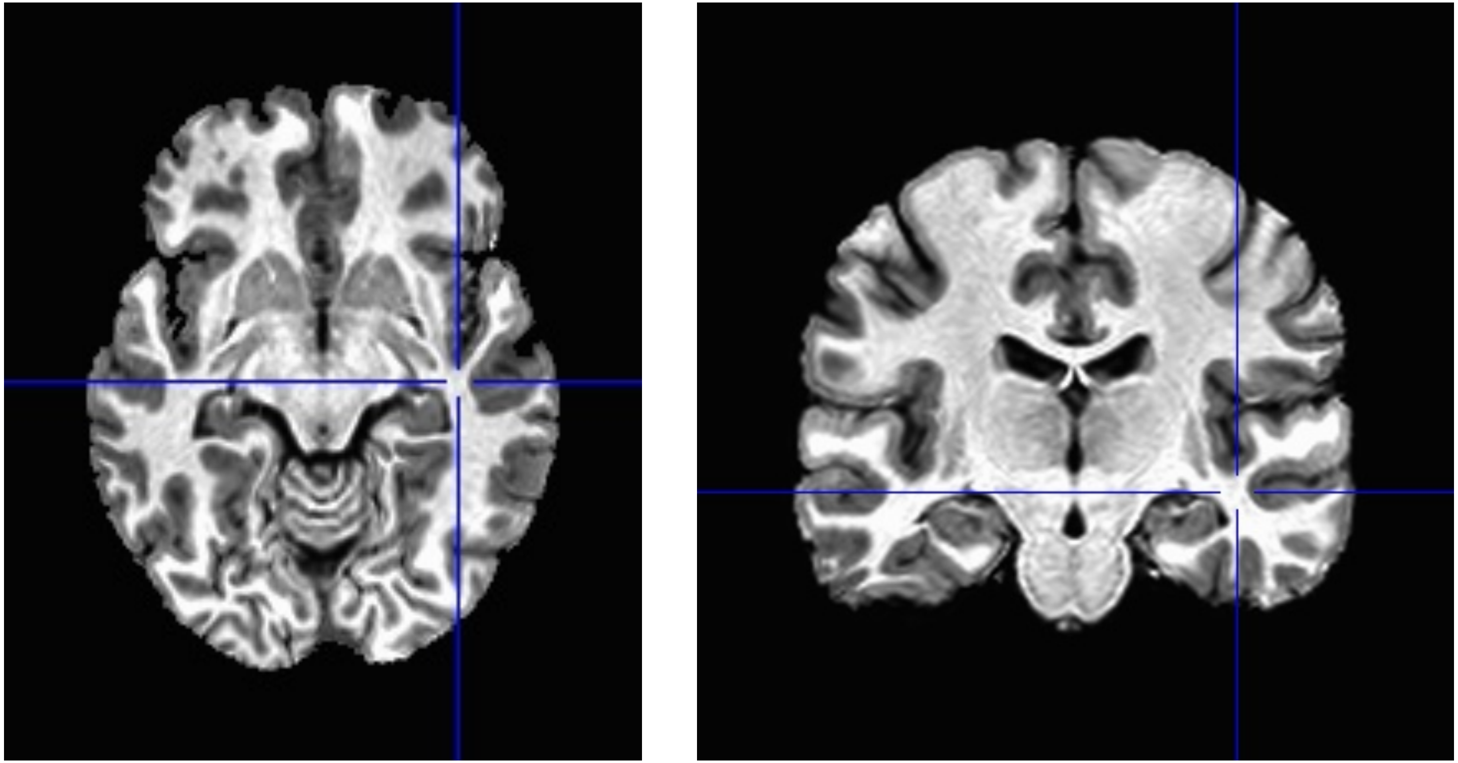
**MNI152_2009_template_SSW.nii.gz**

Nonlinear alignments

# What @SSwarper Reads and Writes

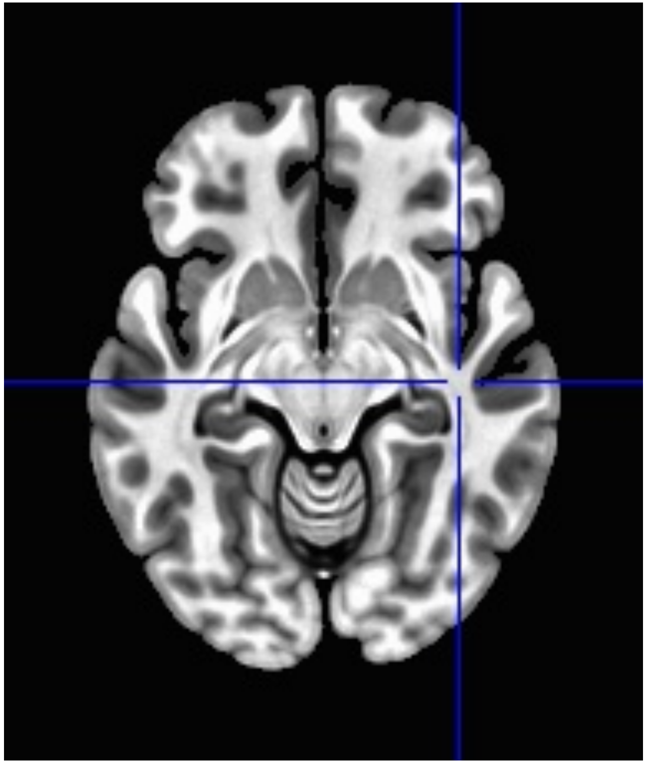
- Inputs:
  - T1-weighted anatomical image of subject (skull-on)
  - Subject ID code, for names of output files
- Outputs (subject ID = **sub007**):
  - **anatSS.sub007.nii**
    - skull-stripped dataset in original coordinates
  - **anatQQ.sub007.nii**
    - skull-stripped dataset, nonlinearly warped to MNI template
  - **anatQQ.sub007.aff12.1D**
    - affine matrix to transform original dataset to MNI template
  - **anatQQ.sub007_WARP.nii**
    - incremental warp from affine transformation to nonlinearly aligned dataset
- These files are needed for later use in afni_proc.py

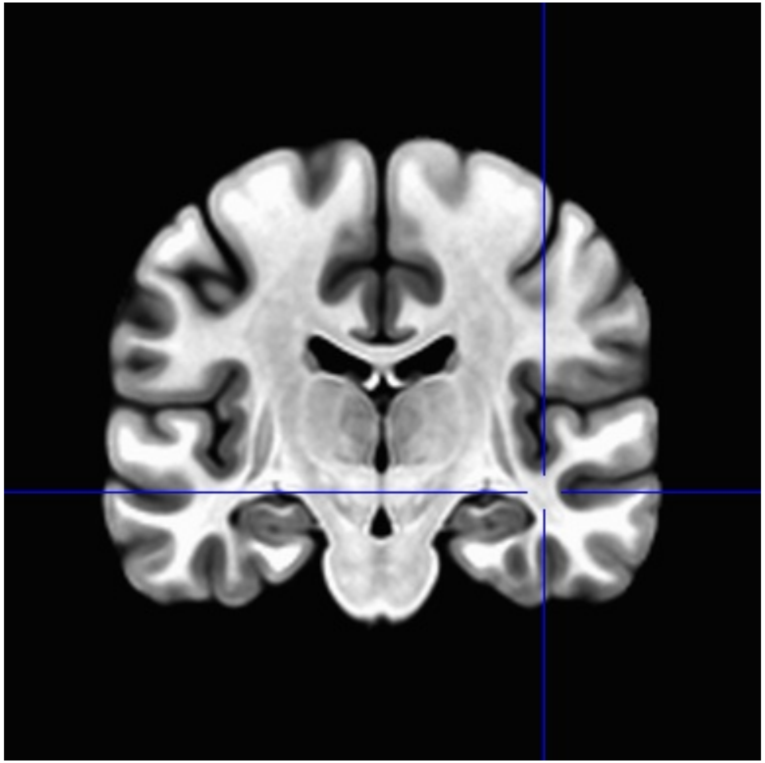
# @SSwarper Results



sub00440 from Beijing-Zang
in the FCON-1000 collection

# MNI Template Slices





For comparison

# Nonlinear Registration Script

- What follows is a script for doing nonlinear warping (registration) of *one* anatomical dataset to an MNI template
- In a real study, this script is run once for each subject
- Takes a long time, so the script should be submitted to a multi-node cluster

# Nonlinear Registration - 1

```
#!/bin/tcsh
### This script nonlinear warps one anatomical dataset,
### taken from the anat_orig directory, to the MNI 2009
### nonlinear template (supplied with AFNI binaries), and
### puts the resulting files into anat_warped directory.
### The only command line argument is the subject ID

set subj = $argv[1]
set tempdir = .

# don't log AFNI programs in ~/.afni.log
# don't try any version checks
# don't auto-compress output files
setenv AFNI_DONT_LOGFILE  YES
setenv AFNI_VERSION_CHECK NO
setenv AFNI_COMPRESSOR    NONE
```

# Nonlinear Registration - 2

```
### go to data directory
# topdir = directory above this Scripts directory
set topdir  = `dirname $cwd`
cd $topdir/anat_orig

### create final output directories
mkdir -p $topdir/anat_warped
mkdir -p $topdir/anat_warped/snapshots

### create temp directory to hold work, and copy anat there
mkdir -p temp_$subj
cp anat_$subj.nii.gz temp_$subj
cd temp_$subj
```

# Nonlinear Registration - 3

```
### process the anat dataset, using the AFNI script
### that does the warping and skull-stripping
@SSwarper –input anat_$subj.nii.gz –subid $subj \
          –base MNI152_2009_template_SSW.nii.gz
# compress the output datasets
gzip -1v *.nii
### move the results to where they belong
# skull-stripped original, Q-warped dataset, and the warps
\mv -f anatSS.${subj}.nii.gz   anatQQ.${subj}.nii.gz      \
       anatQQ.${subj}.aff12.1D anatQQ.${subj}_WARP.nii.gz \
       $topdir/anat_warped
# snapshots for visual inspection
\mv -f *.jpg $topdir/anat_warped/snapshots
# delete the temporary directory
cd ..
\rm -rf temp_$subj
exit 0
```

# Nonlinear Registration - 4

*Add these lines above **afni_proc.py** command:*

```
set basedset = MNI152_2009_template_SSW.nii.gz
set tpath = `@FindAfniDsetPath $basedset`
if( "$tpath" == '' ) then
  echo "***** @SSwarper -- Failed to find $basedset :(“
  exit 1
endif
set basedset = $tpath/$basedset
```

*Add these options to **afni_proc.py** command:*

```
-copy_anat anat_warped/anatSS.${subj}.nii          \
-tlrc_base $basedset                               \
-tlrc_NL_warp                                      \
-tlrc_NL_warped_dsets                              \
$warpdir/anatQQ.${subj}.nii.gz                     \
$warpdir/anatQQ.${subj}.aff12.1D                   \
$warpdir/anatQQ.${subj}_WARP.nii.gz                \
```