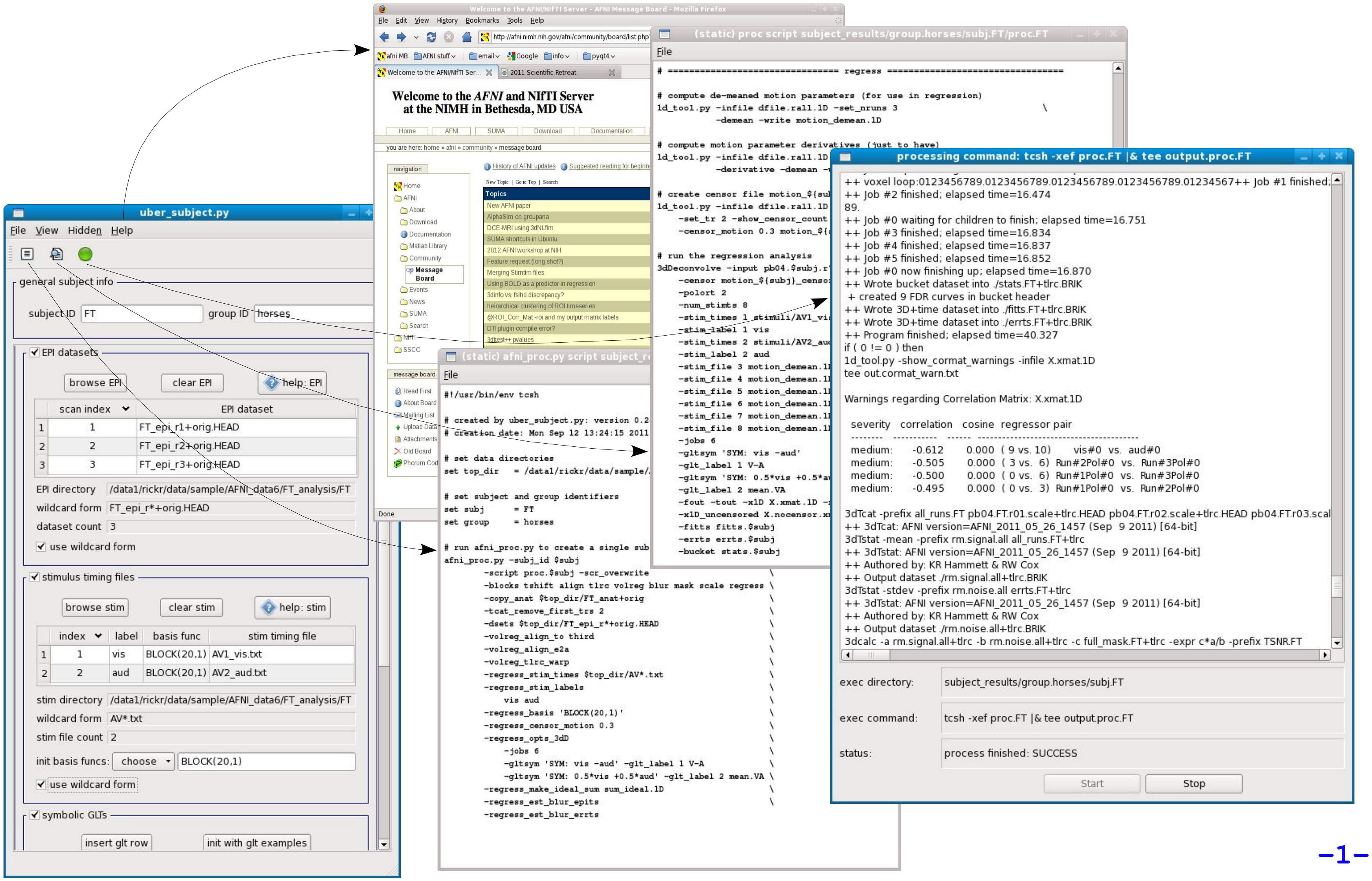
AFNI Start to Finish:
How to Analyze Data with AFNI



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@ROI_Corr_Mat -roi and my output matrix labels
DTI plugin compile error?
3dttest++ pvalues
Done
uber_subject.py
File View Hidden Help
browse EPI
clear EPI
help: EPI
browse stim
clear stim
help: stim
choose
insert glt row
init with glt examples
Start
Stop

uber_subject.py

general subject info

subject ID: FT group ID: horses

☑ EPI datasets

	scan index	EPI dataset
1	1	FT_epi_r1+orig.HEAD
2	2	FT_epi_r2+orig.HEAD
3	3	FT_epi_r3+orig.HEAD

EPI directory: /data1/rickr/data/sample/AFNI_data6/FT_analysis/FT
wildcard form: FT_epi_r*+orig.HEAD
dataset count: 3
☑ use wildcard form

☑ stimulus timing files

	index	label	basis func	stim timing file
1	1	vis	BLOCK(20,1)	AV1_vis.txt
2	2	aud	BLOCK(20,1)	AV2_aud.txt

stim directory: /data1/rickr/data/sample/AFNI_data6/FT_analysis/FT
wildcard form: AV*.txt
stim file count: 2
init basis funcs: choose BLOCK(20,1)
☑ use wildcard form

☑ symbolic GLTs

(static) afni_proc.py script subject_r

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

# created by uber_subject.py: version 0.2
# creation date: Mon Sep 12 13:24:15 2011

# set data directories
set top_dir   = /data1/rickr/data/sample/

# set subject and group identifiers
set subj      = FT
set group     = horses

# run afni_proc.py to create a single sub
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                         \
        -tcat_remove_first_trs 2                                 \
        -dsets $top_dir/FT_epi_r*+orig.HEAD                      \
        -volreg_align_to third                                   \
        -volreg_align_e2a                                        \
        -volreg_tlrc_warp                                        \
        -regress_stim_times $top_dir/AV*.txt                     \
        -regress_stim_labels                                     \
            vis aud                                              \
        -regress_basis 'BLOCK(20,1)'                             \
        -regress_censor_motion 0.3                               \
        -regress_opts_3dD                                        \
            -jobs 6                                              \
            -gltsym 'SYM: vis -aud' -glt_label 1 V-A             \
            -gltsym 'SYM: 0.5*vis +0.5*aud' -glt_label 2 mean.VA \
        -regress_make_ideal_sum sum_ideal.1D                     \
        -regress_est_blur_epits                                  \
        -regress_est_blur_errts
```

(static) proc script subject_results/group.horses/subj.FT/proc.FT

```
# ================================ regress =================================

# compute de-meaned motion parameters (for use in regression)
1d_tool.py -infile dfile.rall.1D -set_nruns 3                  \
           -demean -write motion_demean.1D

# compute motion parameter derivatives (just to have)
1d_tool.py -infile dfile.rall.1D
           -derivative -demean -

# create censor file motion_${su
1d_tool.py -infile dfile.rall.1D
    -set_tr 2 -show_censor_count
    -censor_motion 0.3 motion_${

# run the regression analysis
3dDeconvolve -input pb04.$subj.r
    -censor motion_${subj}_censo
    -polort 2
    -num_stimts 8
    -stim_times 1 stimuli/AV1_vi
    -stim_label 1 vis
    -stim_times 2 stimuli/AV2_au
    -stim_label 2 aud
    -stim_file 3 motion_demean.1
    -stim_file 4 motion_demean.1
    -stim_file 5 motion_demean.1
    -stim_file 6 motion_demean.1
    -stim_file 7 motion_demean.1
    -stim_file 8 motion_demean.1
    -jobs 6
    -gltsym 'SYM: vis -aud'
    -glt_label 1 V-A
    -gltsym 'SYM: 0.5*vis +0.5*a
    -glt_label 2 mean.VA
    -fout -tout -x1D X.xmat.1D -
    -x1D_uncensored X.nocensor.x
    -fitts fitts.$subj
    -errts errts.$subj
    -bucket stats.$subj
```

processing command: tcsh -xef proc.FT |& tee output.proc.FT

```
++ voxel loop:0123456789.0123456789.0123456789.0123456789.01234567++ Job #1 finished;
++ Job #2 finished; elapsed time=16.474
89.
++ Job #0 waiting for children to finish; elapsed time=16.751
++ Job #3 finished; elapsed time=16.834
++ Job #4 finished; elapsed time=16.837
++ Job #5 finished; elapsed time=16.852
++ Job #0 now finishing up; elapsed time=16.870
++ Wrote bucket dataset into ./stats.FT+tlrc.BRIK
 + created 9 FDR curves in bucket header
++ Wrote 3D+time dataset into ./fitts.FT+tlrc.BRIK
++ Wrote 3D+time dataset into ./errts.FT+tlrc.BRIK
++ Program finished; elapsed time=40.327
if ( 0 != 0 ) then
1d_tool.py -show_cormat_warnings -infile X.xmat.1D
tee out.cormat_warn.txt

Warnings regarding Correlation Matrix: X.xmat.1D

  severity   correlation   cosine  regressor pair
  --------   -----------   ------  ----------------------------------------
  medium:     -0.612       0.000  ( 9 vs. 10)       vis#0  vs.  aud#0
  medium:     -0.505       0.000  ( 3 vs.  6)  Run#2Pol#0  vs.  Run#3Pol#0
  medium:     -0.500       0.000  ( 0 vs.  6)  Run#1Pol#0  vs.  Run#3Pol#0
  medium:     -0.495       0.000  ( 0 vs.  3)  Run#1Pol#0  vs.  Run#2Pol#0

3dTcat -prefix all_runs.FT pb04.FT.r01.scale+tlrc.HEAD pb04.FT.r02.scale+tlrc.HEAD pb04.FT.r03.scal
++ 3dTcat: AFNI version=AFNI_2011_05_26_1457 (Sep  9 2011) [64-bit]
3dTstat -mean -prefix rm.signal.all all_runs.FT+tlrc
++ 3dTstat: AFNI version=AFNI_2011_05_26_1457 (Sep  9 2011) [64-bit]
++ Authored by: KR Hammett & RW Cox
++ Output dataset ./rm.signal.all+tlrc.BRIK
3dTstat -stdev -prefix rm.noise.all errts.FT+tlrc
++ 3dTstat: AFNI version=AFNI_2011_05_26_1457 (Sep  9 2011) [64-bit]
++ Authored by: KR Hammett & RW Cox
++ Output dataset ./rm.noise.all+tlrc.BRIK
3dcalc -a rm.signal.all+tlrc -b rm.noise.all+tlrc -c full_mask.FT+tlrc -expr c*a/b -prefix TSNR.FT
```

exec directory: subject_results/group.horses/subj.FT
exec command: tcsh -xef proc.FT |& tee output.proc.FT
status: process finished: SUCCESS

Goal: run group analysis on single subject response magnitudes

- how do we get there?
  - create beta (response magnitude) maps for each subject
    - should be aligned, probably to a well known template
  - run group analysis program (e.g. **`3dttest++`**, **`3dMEMA`**, **`3dANOVA`***)
    - can use **`uber_ttest.py`** to run single group tests

- how do we create aligned beta maps?
  - write single subject processing script: pre-processing through regression
    - inputs: anat, EPI, stimulus timing
    - controls: processing decisions like blur size and alignment template
    - outputs: beta weights (and t-stats, contrasts, blur estimates, etc.)

- how do we write single subject processing scripts?
  - **`afni_proc.py`** can be used to generate processing scripts
  - **`uber_subject.py`** can be used to generate **`afni_proc.py`** command
    - can also run the **`afni_proc.py`** command (generates proc script)
    - can also run the proc script (i.e. actually analyze the data)

General suggestions

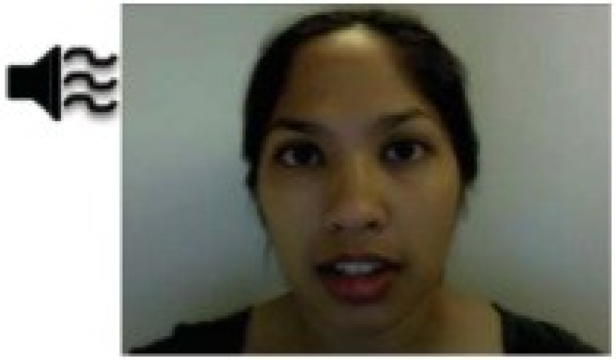
- picture this experiment as your own
  - decisions on processing were made by you (and your colleagues)
    - hopefully before acquiring any data
  - there is no single "correct" way to analyze data, just reasonable ways
- focus on understanding the processing steps
  - in light of your having chosen which steps to perform
- practice the good habit of reviewing results
  - do the initial images look good?
  - review each processing step along with data
  - are the EPI and anat well aligned by the end?
  - do the statistical results look reasonable?
- create scripts for any processing step
  - they are a record of how data was processed
  - easy to apply to any new subjects
  - easy to repeat
    - expect to re-analyze everything (mistake, new decision, etc.)
    - keep original data and all processing scripts

Review of stimulus conditions

- Speech Perception Task: Subjects were presented with audiovisual speech that was presented in a predominantly auditory or predominantly visual modality.
- A digital video system was used to capture auditory and visual speech from a female speaker.
- There were 2 types of stimulus conditions:

(1) **<u>Auditory-Reliable</u>**

Example: Subjects can clearly *hear* the word “cat,” but the video of a woman mouthing the word is degraded.

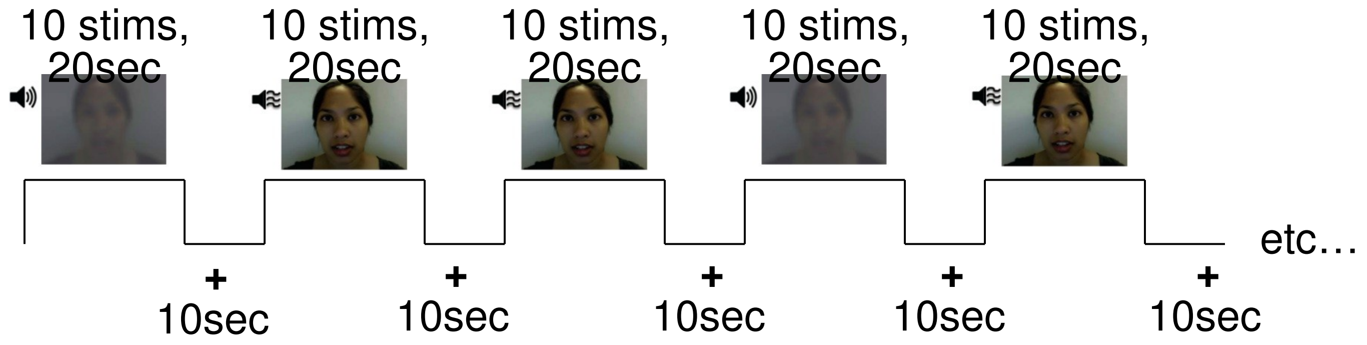


(2) **<u>Visual-Reliable</u>**

Example: Subjects can clearly *see* the video of a woman mouthing the word “cat,” but the audio of the word is degraded.

❖ Experiment Design:

- There were 3 runs in a scanning session.
- Each run consisted of 10 blocked trials:
  - 5 blocks contained Auditory-Reliable (*Arel*) stimuli, and
  - 5 blocks contained Visual-Reliable (*Vrel*) stimuli.
- Each block contained 10 trials of *Arel* stimuli OR 10 trials of *Vrel* stimuli.
  - Each block lasted for 20 seconds (1 second for stimulus presentation, followed by a 1-second inter-stimulus interval).
- Each baseline block consisted of a 10-second fixation point.



❖ Data Collected:

- 2 Anatomical datasets for each subject, collected at 3 tesla.
  - 175 sagittal slices
  - voxel dimensions = 0.938 x 0.938 x 1.0 mm
- 3 Time Series (EPI) datasets for each subject.
  - 33 axial slices x 152 volumes = 5016 slices per run
  - TR = 2 sec; voxel dimensions = 2.75 x 2.75 x 3.0 mm
- Sample size, n = 10 (all right-handed subjects)

afni_proc.py
uber_subject.py

- What is **afni_proc.py**?
  - a program used to generate processing scripts for single subject analysis
    - command-line program
  - generated scripts are in **tcsh** syntax
  - scripts are written to be easily read and modified

- What is **uber_subject.py**?
  - for running single subject analysis
  - a graphical user interface to **afni_proc.py**
  - quickly create processing scripts
  - can analyze all subjects from GUI
  - good for learning
    - FMRI processing, shell scripting, AFNI commands
    - can compare against manually generated scripts
      - for sanity, bug detection, quick evaluation

Overview of remaining steps

- `cd AFNI_data6/FT_analysis`
  - review directory contents and note subject data under directory **FT**
- **from the home directory**, run `uber_subject.py` and analyze subject **FT**
  - set subject ID, group ID
  - specify inputs: anat, EPI, stimulus timing files (all under **`FT_analysis/FT`**)
  - controls: BLOCK(20,1), init GLTs, remove first 2 TRs
  - peruse other options, e.g. multiple CPUs for 3dDeconvolve?
  - create afni_proc.py command
  - execute afni_proc.py command (to create proc script)
  - execute proc script (analyze subject data)
- briefly review processing script
- review proc script in modest detail, while viewing processed data
  - run `afni` from **`FT.results`** directory and follow script
  - run resulting **`@ss_review_driver`** script
- run group analysis (**`3dttest++, 3dMEMA or 3dANOVA2`**)
  - run **`uber_ttest.py`** on data under **`AFNI_data6/group_results`**
  - or run existing **`s1.3dANOVA2`** script

Single Subject Analysis: FT

- from home directory (use the **cd** command), run **uber_subject.py &**
  - subject ID ***FT***, group ID ***horses***
  - browse anat: choose ***AFNI_data6/FT_anaysis/FT/FT_anat+orig.HEAD***
  - browse EPI: choose ***FT_epi_r1+orig.HEAD*** (and ***epi_r2*** and ***epi_r3***)
    - select all 3 data files with ctrl or shift keys, then hit ***Open***
  - browse stim: choose ***AV1_vis.txt*** and ***AV2_aud.txt***
    - init basis funcs: choose ***BLOCK(5,1)***
    - change ***5*** to ***20*** (i.e, apply ***BLOCK(20,1)***) and hit Enter
  - symbolic GLTs: ***init with glt examples***
  - expected options: removed first ***2*** TRs per run
  - (optional) extra regress options: ***2*** CPUs (or 12 or whatever is appropriate)
  - ***generate afni_proc.py command*** (left action button)
  - ***generate proc script*** (middle action button)
  - ***process this subject*** (right action button)
    - scripts/results are under `subject_results/group.horses/subj.FT`
  - review the `proc.FT` script while looking at the results under `FT.results`
    - `cd subject_results/group.horses/subj.FT`
    - `cd FT_results ; afni &`
  - after script and data review, run `./@ss_review_driver`
    - **minimal** data review (should run on each subject)

Group Analysis: paired t-test (Vrel-Arel)

- from home directory, run `uber_ttest.py &`
    - using data from under `AFNI_data6/group_results`
  - program ***3dttest++***, script ***script.V-A***, prefix ***ttest.V-A***, mask dset: ***skip***
  - fill "datasets A" table with datasets for `Vrel` betas
    - get subj dsets
      - ✓ choose file : `OLSQ.FP.betas+tlrc.HEAD`
      - ✓ alter name into wildcard pattern: replace "***FP***" with "*******"
      - ✓ press Enter or ***apply pattern*** button (should have 10 datasets)
      - ✓ press ***OK*** (at bottom)
    - set name ***Vrel***, data index ***Vrel#0_Coef*** (or index 0), t-stat index: ***skip***
  - fill "datasets B" table with datasets for **Arel** betas
    - identical steps, except for the text fields at the bottom:
      - ✓ set ***Arel***, data index ***Arel#0_Coef***, t-stat index: ***skip***
  - 3dttest++ options: ***-paired***
  - ***generate processing script***: press left action button (note hint)
  - ***execute processing script***: press green action button (note hint)
- script/output/results are under: **group_results/test.001.3dttest++**
- script is **script.V-A**, results are under **ttest.results**
- **cd group_results/test.001.3dttest++/ttest.results** (practice **`<tab>`** key)
- **afni &**

Additional comments

- inputs for subject **`FT`** are under **`AFNI_data6/FT_analysis/FT`**

- results from **`uber_*.py`** go where the program was run
  - **`uber_subject.py`**: **`subject_results/group.GROUP/subj.SUBJECT`**
  - **`uber_ttest.py`**: **`group_results/test.INDEX.PROGRAM`**
  - so in class, these directory trees should end up under the home directory

- it is not necessary to master all shell script details
  - but want to understand processing steps

- when analyzing data, run **`@ss_review_driver`** for every subject
  - script represents the **minimum** of what to look at for each subject
  - for the first few subjects analyzed, look at all results in detail
    - in more detail than the level of this class
    - before acquiring many subjects

Note what is under **AFNI_data6/FT_analysis**

FT	- subject data directory
s01.ap.simple	- basic **afni_proc.py** script
s02.ap.align	- more advanced script
s09.cleanup	- remove analysis results
s11.proc.FT	- result of **s01.ap.simple**
s12.proc.FT.align	- result of **s01.ap.align**

under **FT**

AV1_vis.txt	- visual reliable timing
AV2_aud.txt	- auditory reliable timing
FT_anat+orig.BRIK/HEAD	- anatomical dataset
FT_epi_r1+orig.BRIK/HEAD	- EPI run 1
FT_epi_r2+orig.BRIK/HEAD	- EPI run 2
FT_epi_r3+orig.BRIK/HEAD	- EPI run 3

AV1_vis.txt:

```
60 90 120 180 240
120 150 180 210 270
0 60 120 150 240
```

AFNI Start to Finish
(the horror continues...)

- To continue reviewing the data on your own, please see the corresponding tutorial that continues under the data directory:
  - ➢ **`AFNI_data6/FT_analysis/tutorial`**
- Alternatively, this can be viewed from the AFNI web site:

  http://afni.nimh.nih.gov/pub/dist/edu/data/CD.expanded/AFNI_data6/FT_analysis/tutorial
- or from the Help menu of **`uber_subject.py`**
  - ❖ Help --> Browse --> web: tutorial-single subject analysis