

AFNI Start to Finish: How to Analyze Data with AFNI

The image displays a collage of screenshots from the AFNI (Analysis of Functional NeuroImages) software interface, illustrating the workflow for analyzing data.

Top Left: AFNI Website
A screenshot of the AFNI website, showing the "Welcome to the AFNI and NIFTI Server at the NIMH in Bethesda, MD USA" message. The navigation menu includes Home, AFNI, SUMA, Download, and Documentation. The "History of AFNI updates" section lists various updates and resources.

Top Right: AFNI Message Board
A screenshot of the AFNI Message Board, showing a list of topics and a search bar. The topics include "New AFNI paper", "AFNI on groupware", "AFNI on groupware", "SUMA workshops in Libano", "2012 AFNI workshop at NIH", "Feature request (long shot)", "Merging Stimulus files", "Using BOLD as a predictor in regression", "3dinfo vs. 3dinfo discrepancy?", "Neurological clustering of ROI timeseries", "ROI Cor_Mat_v04 and my output matrix labels", "BRI plugin compile error?", and "3dinfo vs. 3dinfo".

Bottom Left: uber_subject.py
A screenshot of the `uber_subject.py` script window. It shows the "general subject info" section with fields for "subject ID" (FT) and "group ID" (horses). The "EPI datasets" section lists three datasets: "FT_epi_r1+orig.HEAD", "FT_epi_r2+orig.HEAD", and "FT_epi_r3+orig.HEAD". The "stimulus timing files" section lists two files: "AV1_vis.txt" and "AV2_aud.txt".

Bottom Center: afni_proc.py
A screenshot of the `afni_proc.py` script window. It shows the "set data directories" section with fields for "set top_dir" and "set sub_dir". The "set subject and group identifiers" section shows "set subj = FT" and "set group = horses". The "run afni_proc.py" section shows the command `afni_proc.py -subj_id $subj`.

Bottom Right: process command
A screenshot of the "process command" window. It shows the command `tsh -xef proc.FT |& tee output.proc.FT` and the 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 (i.e. feel responsibility)
 - 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) **Auditory-Reliable**

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

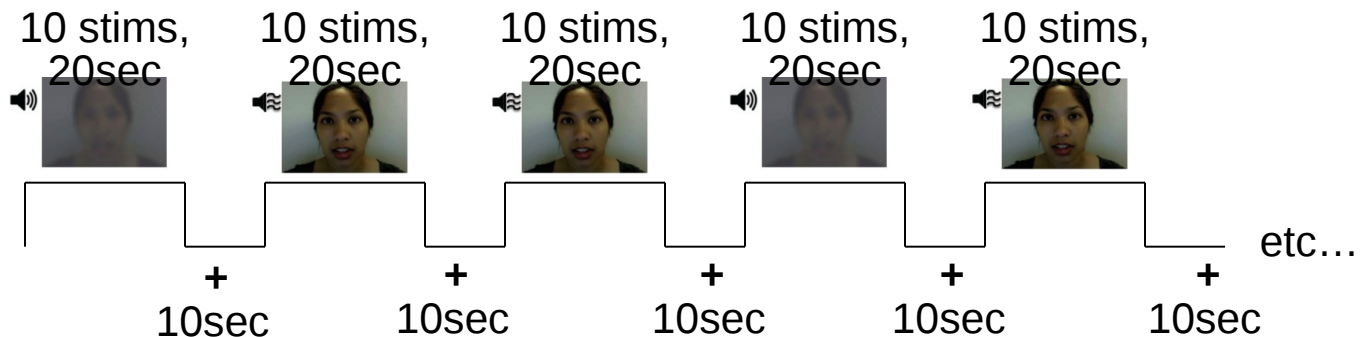


(2) **Visual-Reliable**

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 (*AreI*) stimuli, and
 - 5 blocks contained Visual-Reliable (*VreI*) stimuli.
- ◆ Each block contained 10 trials of *AreI* stimuli OR 10 trials of *VreI* 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

Note what is under **AFNI_data6/FT_analysis**

FT

s01.ap.simple

s02.ap.align

s09.cleanup

s11.proc.FT

s12.proc.FT.align

- subject data directory
- basic **afni_proc.py** script
- more advanced script
- remove analysis results
- result of **s01.ap.simple**
- result of **s01.ap.align**

under **FT**

AV1_vis.txt

AV2_aud.txt

FT_anat+orig.BRIK/HEAD

FT_epi_r1+orig.BRIK/HEAD

FT_epi_r2+orig.BRIK/HEAD

FT_epi_r3+orig.BRIK/HEAD

- visual reliable timing
- auditory reliable timing
- anatomical dataset
- EPI run 1
- EPI run 2
- EPI run 3

AV1_vis.txt:

60 90 120 180 240

120 150 180 210 270

0 60 120 150 240

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**
 - stimuli: set both basis functions to **BLOCK(20,1)**
 - init basis funcns to **BLOCK(5,1)**, change 5 to 20 and hit Enter
 - symbolic GLTs: click to view option frame and **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`
 - choose *paired test*
 - fill "datasets A" table with datasets for **Vrel** betas
 - get subj dsets
 - ✓ choose file : `OLSQ.FP.betas+t1rc.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 table of datasets, so choose *copy other table*
 - ✓ set **Arel**, data index **Arel#0_Coef**, t-stat index: `skip`
 - *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

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