# AFNI Start to Finish: How to Analyze Data with AFNI



#### 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 resulting statistical maps look reasonable?
- create scripts for any processing steps
  - they are records of how data was processed
  - they are easy to apply to any new subjects
  - they are 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.

-4-

# ✤ 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.



### ✤ <u>Data Collected:</u>

- 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, <u>n</u> = 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
  - sood 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
  - $\succ$  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)
- String script script \* 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

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

#### <u>AV1\_vis.txt</u>:

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

- subject data directory
- basic afni\_proc.py script
- more advanced script
- remove analysis results
- result of **s01.ap.simple**
- result of **s01.ap.align**
- visual reliable timing
- auditory reliable timing
- anatomical dataset
- EPI run 1
- EPI run 2
  - EPI run 3

# Single Subject Analysis: FT

- \* from home directory (use the cd command), run uber\_subject.py &
- uber\_subject.py setup:
  - 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 funcs 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: remove first 2 TRs per run
  - > optional: see "extra regress options"
    - jobs for regression: **2** (if computer has many CPUs, use more)
    - run cluster simulation: *no* (if 1 thread, this can take a long time)
    - compute fitts dataset: set (reduces needed RAM)

# Single Subject Analysis: FT

- uber\_subject.py: generate scripts and process data
  - 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
- \* (alternate analysis method, for those who cannot run uber\_subject.py)
  - > (only do this if you were not able to use uber\_subject.py)
  - > cd AFNI\_data6/FT\_analysis
  - > tcsh s04.cmd.usubj
  - > results are under ~/subject\_results/group.horses/subj.FT
- review processing script and results
  - 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
    - considered a **minimal** data review (run for every 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+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 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