AFNI

Introduction, Concepts, Principles

Analysis of Functional NeuroImages

by

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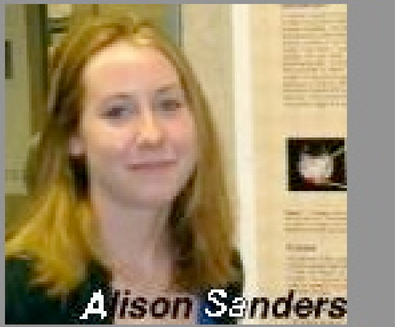


AFNI User

AFNI is a research tool.

Clinical uses are not supported or advised.

http://afni.nimh.nih.gov/afni

AFNI = Analysis of Functional NeuroImages

- Developed to provide an environment for FMRI data analyses
  - And a platform for development of new software
- AFNI refers to both the program of that name and the entire package of programs and plugins (more than 100)
- Important principles in the development of AFNI:
  - Allow user to stay close to the data and view it in many different ways
  - Allow users to assemble pieces in different ways to make customized analyses
  - “Provide mechanism, not policy”
  - Allow other programmers to add features that can interact with the rest of the package

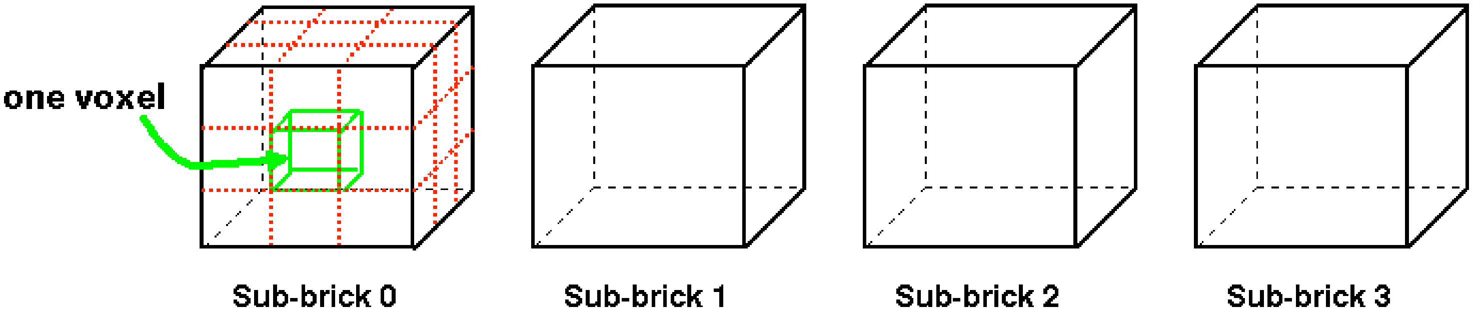
Principles We* Live By

- Fix significant bugs as soon as possible
  - ★ But we define “significant”
- Nothing is secret or hidden (AFNI is open source)
  - ★ But maybe not very well documented or advertised
- Release early and often
  - ★ All users are beta-testers for life
- Help the user
  - ★ Until our patience expires
- Try to anticipate users’ future needs
  - ★ What we think you will need may not be what you actually end up needing

* SSCC NIMH

Fundamental AFNI Concepts

- Basic unit of data in AFNI is the dataset
  - ★ A collection of 1 or more 3D arrays of numbers
    - ✧ Each entry in the array is in a particular spatial location in a 3D grid (a voxel)
    - ✧ Image datasets: each array holds a collection of slices from the scanner
      - ➥ Each number is the signal intensity for that particular voxel
    - ✧ Derived datasets: each number is computed from other dataset(s)
      - ➥ e.g., each voxel value is a *t*-statistic reporting “activation” significance from an FMRI time series dataset, for that voxel
  - ★ Each 3D array in a dataset is called a sub-brick
    - ✧ There is one number in each voxel in each sub-brick



Dataset Contents: Numbers

- Different types of numbers can be stored in datasets
  - 8 bit bytes (e.g., from grayscale photos)
  - 16 bit shorts (e.g., from MRI scanners)
    - Each sub-brick may also have a floating point scale factor α attached, so that the "true" value in each voxel is actually $\alpha\cdot$(value in dataset file)
  - 32 bit floats (e.g., calculated values)
  - 24 bit RGB color triples (e.g., from your digital camera!)
  - 64 bit complex numbers (e.g., for the physicists in the room)
- Different sub-bricks are allowed to have different numeric types

  and I mean this

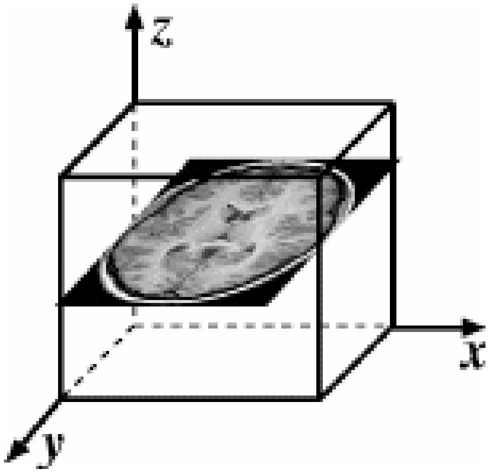
  - But this is **not** recommended
  - Will occur if you "catenate" two dissimilar datasets together (e.g., using 3dTcat or 3dbucket commands)

Dataset Contents: Header

- Besides the voxel values, a dataset also contains auxiliary information, including (some of which is optional):
  - ★ *xyz* dimensions of each voxel (in mm)
  - ★ Orientation of dataset axes;

    for example, *x*-axis=R-L, *y*-axis=A-P, *z*-axis=I-S

    ⇒ axial slices



  - ★ Location of dataset in scanner coordinates
    - ✧ Needed to overlay one dataset onto another
  - ★ Time between sub-bricks, for 3D+time datasets
    - ✧ Such datasets are the basic unit of FMRI data (one per imaging run)
  - ★ Statistical parameters associated with each sub-brick
    - ✧ e.g., a *t*-statistic has the degrees-of-freedom parameter stored

Dataset Files - I

- AFNI formatted datasets are stored in 2 files
  - The .HEAD file holds all the auxiliary information
  - The .BRIK file holds all the numbers in all the sub-bricks
- Datasets can be in one of 3 coordinate systems (AKA views)
  - Original data or +orig view: from the scanner
  - AC-PC aligned or +acpc view:
    - Dataset rotated so that the anterior commissure and posterior commissure are horizontal, the AC is at (*x*,*y*,*z*)=(0,0,0), and the hemispheric fissure is vertical
  - Talairach or +tlrc view:
    - Dataset has been rescaled to have its 12 sub-regions conform to the Talairach-Tournoux atlas
    - Talairach or Stererotaxic coordinates
    - Not quite the same as MNI coordinates, but close

Dataset Files - II

- AFNI dataset filenames consist of 3 parts
  - ⋆ The user-selected prefix (almost anything)
  - ⋆ The view (one of +orig, +acpc, or +tlrc)
  - ⋆ The suffix (one of .HEAD or .BRIK)
  - ⋆ Example: **BillGates+tlrc.HEAD** and **BillGates+tlrc.BRIK**
  - ⋆ When creating a dataset with an AFNI program, you supply the prefix; the program supplies the rest
- AFNI programs can ***read*** datasets stored in several formats
  - ⋆ ANALYZE (.hdr/.img file pairs); i.e., from SPM, FSL
  - ⋆ MINC (.mnc); i.e., from mnitools
  - ⋆ CTF (.mri, .svl) MEG analysis volumes
  - ⋆ NIfTI-1 (.nii) — a new standard format that AFNI, SPM, FSL, and BrainVoyager have agreed upon
  - ⋆ ASCII text (.1D) — numbers arranged into columns

Dataset Directories

- Datasets are stored in directories, also called sessions
  - All the datasets in the same session, in the same view, are presumed to be aligned in *xyz*-coordinates
    - Voxels with same value of (*x*,*y*,*z*) correspond to same brain location
  - Can overlay (in color) any one dataset on top of any other one dataset (in grayscale) from same session
    - Even if voxel sizes and orientations differ
  - Typical AFNI contents of a session directory are all data derived from a single scanning session for one subject
    - Anatomical reference (SPGR or MP-RAGE volume)
    - 10-20 3D+time datasets from EPI functional runs
    - Statistical datasets computed from 3D+time datasets, showing activation (you hope and pray)
    - Datasets transformed from +orig to +tlrc coordinates, for comparison with datasets from other subjects

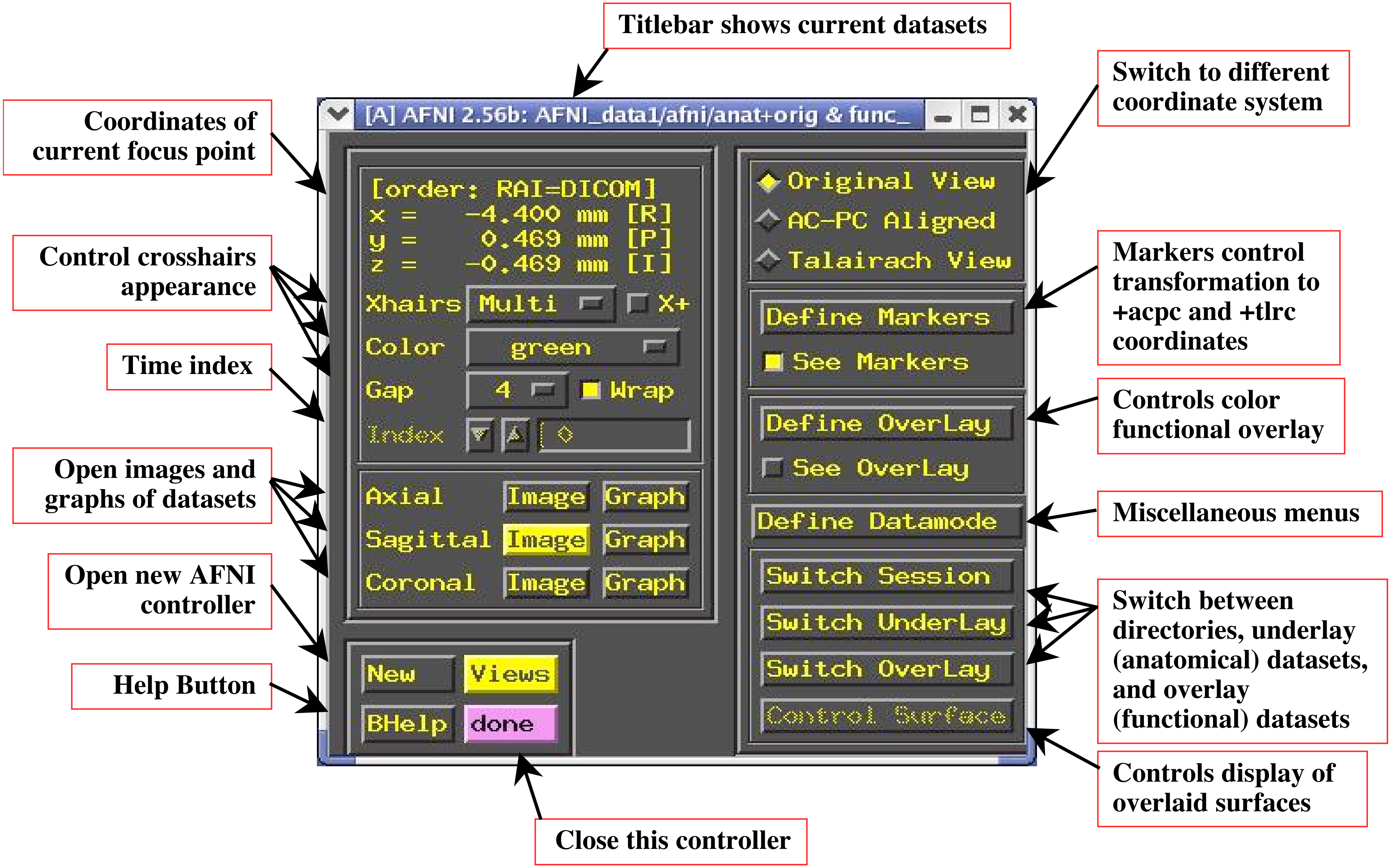
Getting and Installing AFNI

- AFNI runs on Unix systems: Linux, Sun, SGI, Mac OS X
  - ⋆ Can run under Windows with Cygwin Unix emulator
- **If you are at the NIH**: SSCC can install AFNI and update it on your system(s)
  - ⋆ You must give us an account with ssh access
- You can download precompiled binaries from our Website
  - ⋆ **http://afni.nimh.nih.gov/afni**
  - ⋆ Also: documentation, message board, humor, …
- You can download source code and compile it
- AFNI is updated fairly frequently, so it is important to update occasionally

A Quick Overview of AFNI

- Start AFNI from the Unix command line
  - ★ afni reads datasets from the current directory
  - ★ afni dir1 dir2 … reads datasets from directories listed
  - ★ afni -R reads datasets from current directory and from all directories below it
- AFNI also reads a file named .afnirc from your home directory
  - ★ Used to change many of the defaults
    - ✧ Window layout and image/graph viewing setup; popup hints; whether to compress .BRIK files
    - ✧ cf. file README.environment in the AFNI documentation
- Also can read file .afni.startup_script to restore the window layout from a previous run
  - ★ Created from Define Datamode->Misc->Save Layout menu
    - ✧ cf. file README.driver for what can be done with AFNI scripts

AFNI controller window at startup



```
[order: RAI=DICOM]
x =    -4.400 mm [R]
y =     0.469 mm [P]
z =    -0.469 mm [I]
```

Titlebar shows current datasets

Coordinates of current focus point

Control crosshairs appearance

Time index

Open images and graphs of datasets

Open new AFNI controller

Help Button

Close this controller

Switch to different coordinate system

Markers control transformation to +acpc and +tlrc coordinates

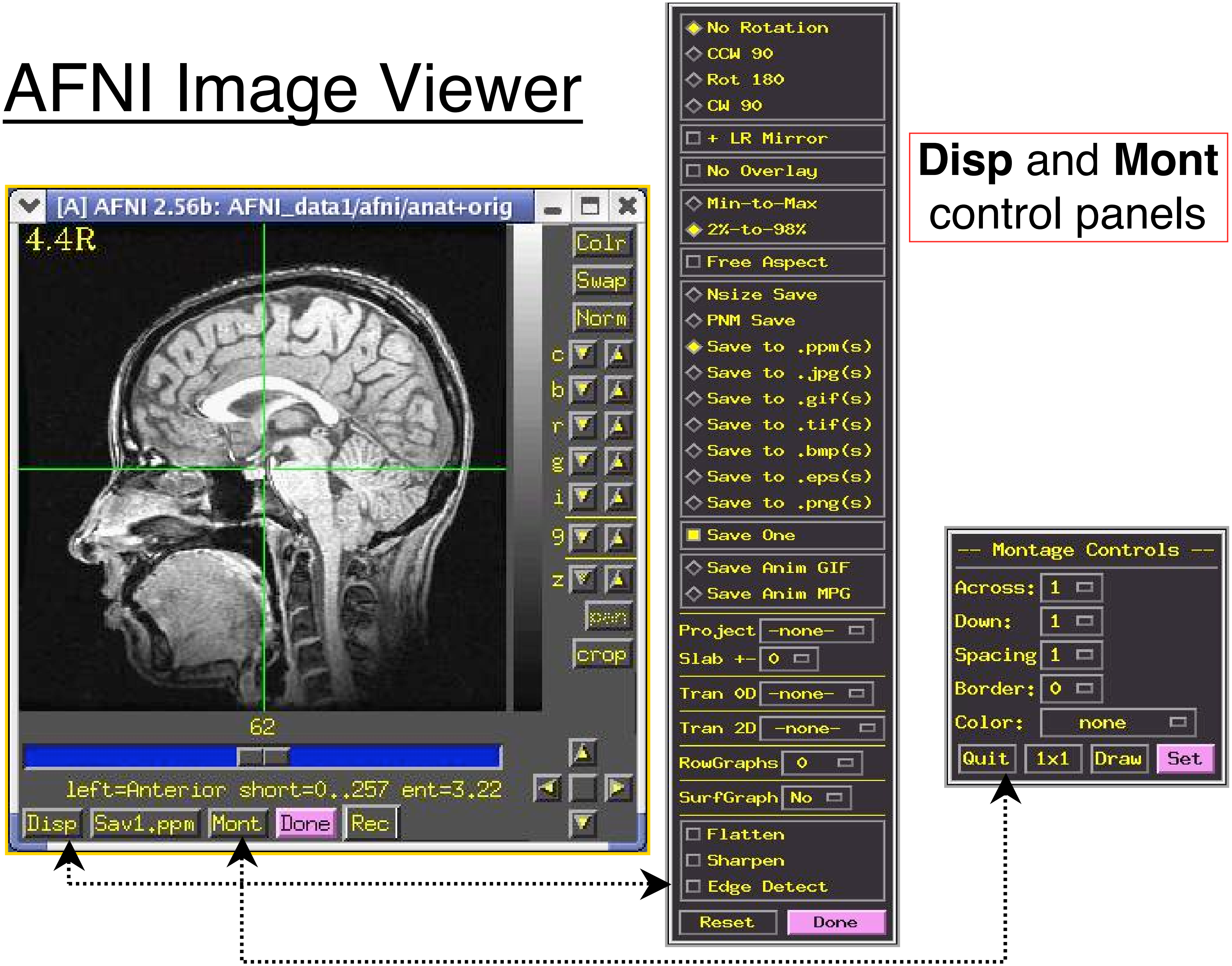
Controls color functional overlay

Miscellaneous menus

Switch between directories, underlay (anatomical) datasets, and overlay (functional) datasets

Controls display of overlaid surfaces

AFNI Image Viewer



[A] AFNI 2.56b: AFNI_data1/afni/anat+orig

4.4R

62

left=Anterior short=0..257 ent=3.22

● No Rotation
○ CCW 90
○ Rot 180
○ CW 90

□ + LR Mirror

□ No Overlay

○ Min-to-Max
● 2%-to-98%

□ Free Aspect

○ Nsize Save
○ PNM Save
● Save to .ppm(s)
○ Save to .jpg(s)
○ Save to .gif(s)
○ Save to .tif(s)
○ Save to .bmp(s)
○ Save to .eps(s)
○ Save to .png(s)

■ Save One

○ Save Anim GIF
○ Save Anim MPG

Project -none-
Slab +- 0
Tran 0D -none-
Tran 2D -none-
RowGraphs 0
SurfGraph No

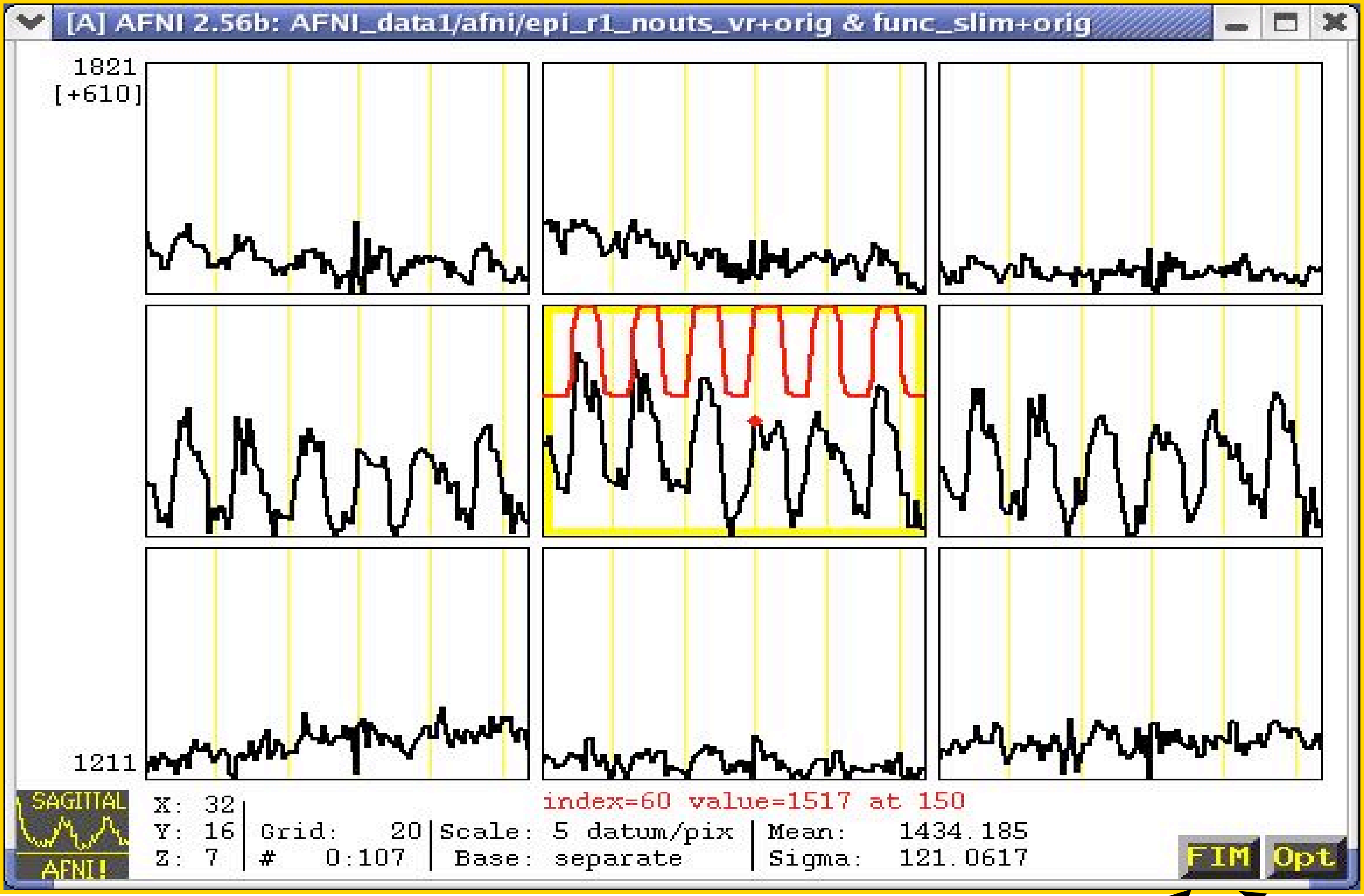
□ Flatten
□ Sharpen
□ Edge Detect

-- Montage Controls --

Across: 1
Down: 1
Spacing 1
Border: 0
Color: none

Disp and **Mont** control panels

AFNI Time Series Graph Viewer



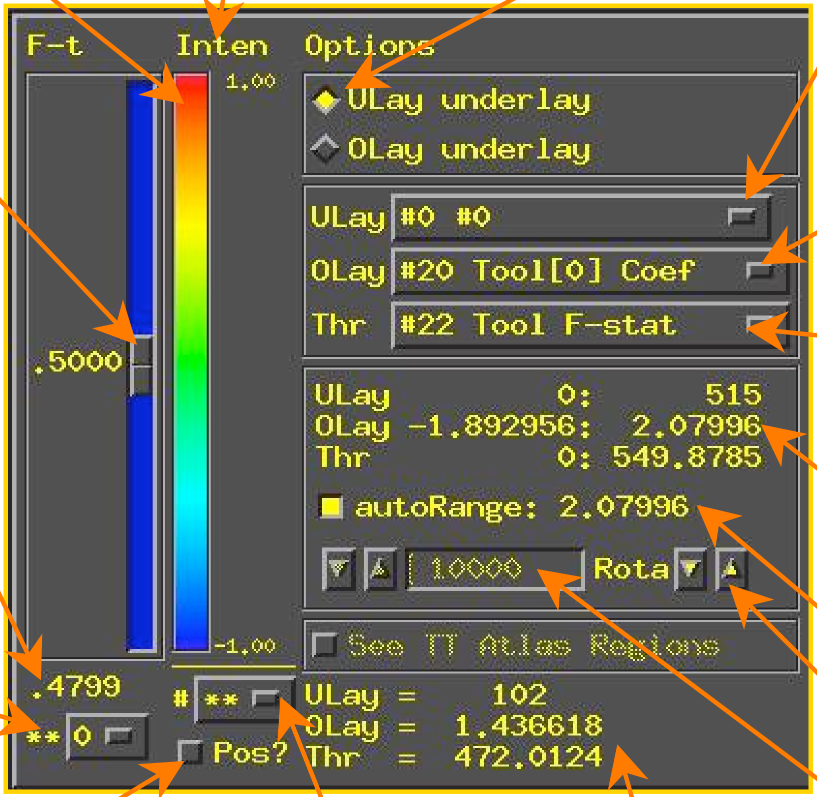
[A] AFNI 2.56b: AFNI_data1/afni/epi_r1_nouts_vr+orig & func_slim+orig

```
                  index=60 value=1517 at 150
X:  32
Y:  16 | Grid:     20 | Scale:  5 datum/pix | Mean:    1434.185
Z:  7  | #     0:107  |  Base:  separate    | Sigma:   121.0617
```

Data (black) and Reference waveforms (red)

Menus for controlling graph displays

Define Function: Color Overlay Panel



```
F-t          Inten   Options
                1.00  ◆ ULay underlay
                      ◇ OLay underlay
                      ULay #0 #0
                      OLay #20 Tool[0] Coef
                      Thr  #22 Tool F-stat
.5000                 ULay           0:      515
                      OLay -1.892956:   2.07996
                      Thr            0: 549.8785
                      ■ autoRange: 2.07996
                      10000 Rota
               -1.00  □ See TT Atlas Regions
.4799        # **     ULay =      102
** 0         □ Pos?   OLay =   1.436618
                      Thr  =   472.0124
```

Color map

Hidden popup menu here

Choose which dataset makes the underlay image

Choose which sub-brick from Underlay dataset to display (usu. Anat - has only 1 sub-brick)

Threshold slider

p-value of current threshold

Choose range of threshold slider, in powers of 10

Choose which sub-brick of functional dataset makes the color

Choose which sub-brick of functional dataset is the Threshold

Shows ranges of data in Underlay and Overlay dataset

Shows automatic range for color scaling

Rotates color map

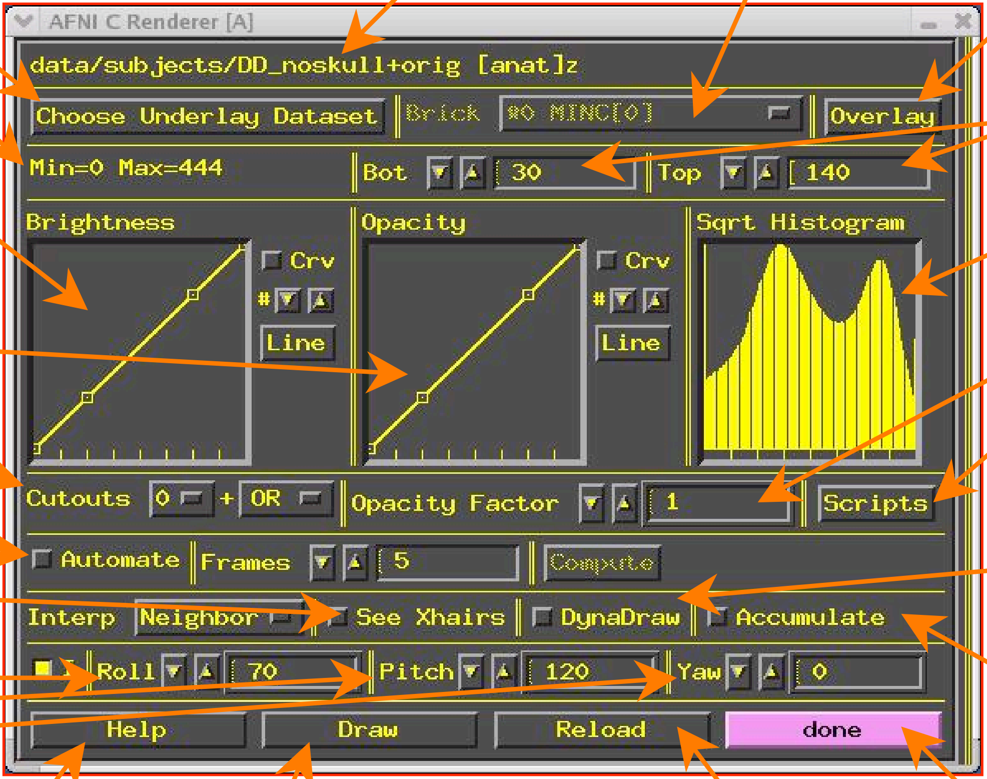
Positive-only or both signs of function?

Number of panes in color map

Shows voxel values at focus

Lets you choose range for color scaling

Volume Rendering: an AFNI plugin



data/subjects/DD_noskull+orig [anat]z

Brick #0 MINC[0]

Min=0 Max=444

Bot 30 Top 140

Cutouts 0 + OR

Opacity Factor 1

□ Automate Frames 5

Interp Neighbor □ See Xhairs □ DynaDraw □ Accumulate

Roll 70 Pitch 120 Yaw 0

Pick new underlay dataset

Name of underlay dataset

Sub-brick to display

Open color overlay controls

Range of values in underlay

Change mapping from values in dataset to brightness in image

Mapping from values to opacity

Cutout parts of 3D volume

Compute many images in a row

Show 2D crosshairs

Control viewing angles

Range of values to render

Histogram of values in underlay dataset

Maximum voxel opacity

Menu to control scripting (control rendering from a file)

Render new image immediately when a control is changed

Accumulate a history of rendered images (can later save to an animation)

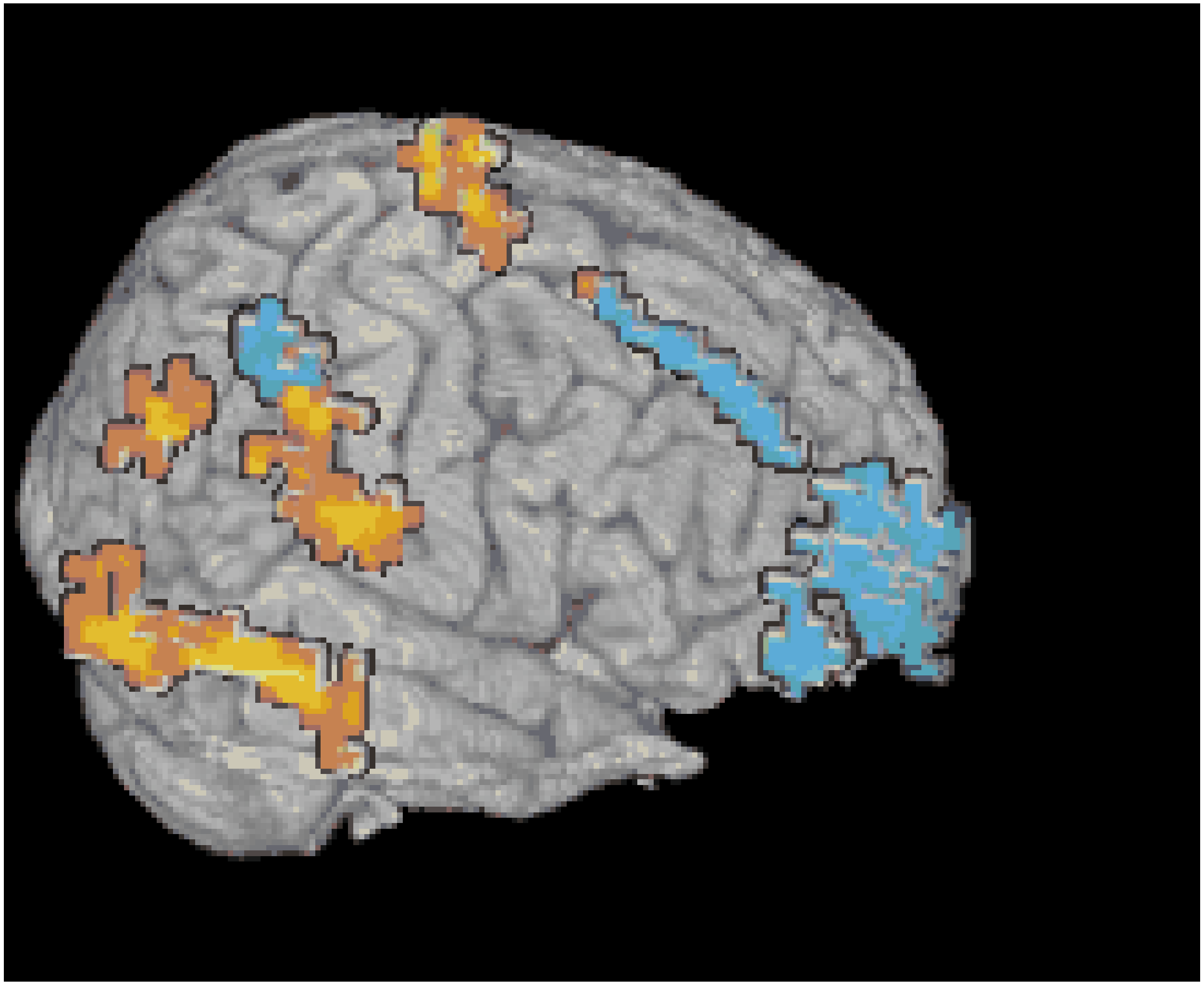
Detailed instructions

Force a new image to be rendered

Reload values from the dataset

Close all rendering windows

Staying Close to Your Data!



"ShowThru" rendering of functional activation:
animation created with Automate and Save:aGif controls

Other Parts of AFNI

- Batch mode programs
  - ⋆ Are run by typing commands directly to computer, or by putting commands into a text file (script) and later executing them

☺ Good points about batch mode

- ⋆ Can process new datasets exactly the same as old ones
- ⋆ Can link together a sequence of programs to make a customized analysis (a personal pipeline)
- ⋆ Some analyses take a long time

☹ Bad points about batch mode

- ⋆ Learning curve is "all at once" rather than gradual
- ⋆ If you are, like, under age 35, you may not know how to type commands into a computer
  - ✧ At least we don't make you use punched cards

AFNI Batch Programs

- Many important capabilities in AFNI are **only** available in batch programs
  - ★ A few examples (of more than 100, from trivial to complex)
- 3dDeconvolve = multiple linear regression on 3D+time datasets, to fit each voxel's time series to a model and then test fits for significance
- 3dvolreg = 3D+time dataset registration, to correct for small subject head movements
- 3dANOVA = 1-, 2-, 3-, and 4- way ANOVA layouts, for combining datasets in Talairach space
- 3dcalc = general purpose voxel-wise calculator
- 3dclust = find clusters of activated voxels
- 3dresample = resample a dataset to a new grid orientation and/or voxel size

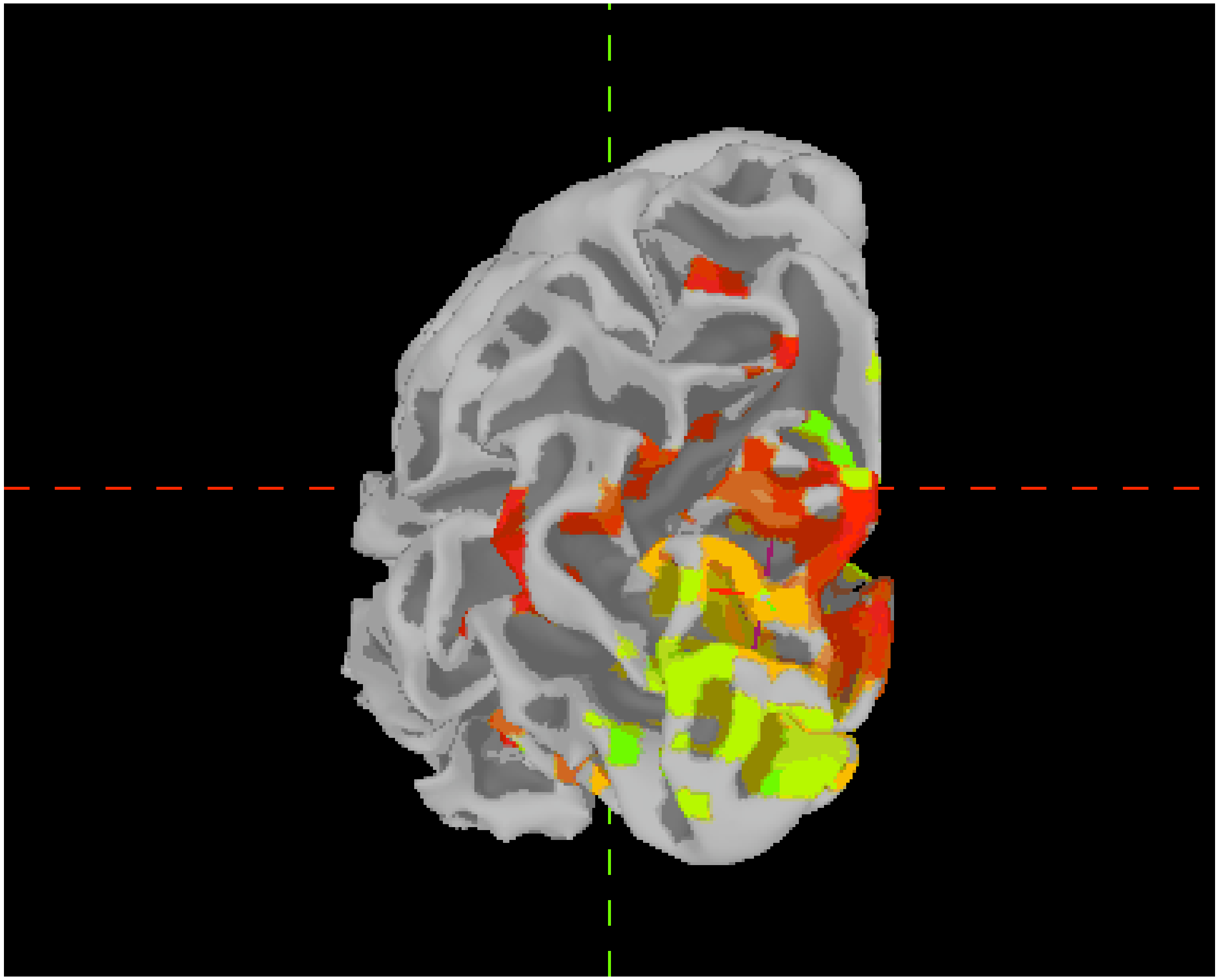
AFNI Plugins

- A plugin is an extension to AFNI that attaches itself to the interactive AFNI GUI
  - ★ **Not** the same as a batch program
  - ★ Offers a relatively easy way to add certain types of interactive functionality to AFNI
  - ★ A few examples:
- Draw Dataset = ROI drawing
- Render [new] = volume renderer
- Dataset#N = lets you plot multiple 3D+time datasets as overlays in an AFNI graph viewer
- Histogram = plots a histogram of a dataset or piece of one
- Edit Tagset = lets you attach labeled “tag points” to a dataset (e.g., as anatomical reference markers)

SUMA, et alii

- SUMA is the AFNI surface mapper
  - ★ For displaying surface models of the cortex
    - ✧ Surface models come from FreeSurfer (MGH) or SureFit (Wash U)
  - ★ Can display functional activations mapped from 3D volumes to the surface
  - ★ Can draw ROIs on the cortical surface
- SUMA is a separate program from AFNI, but can “talk” to AFNI so that volume and surface viewing are linked
  - ★ Click in AFNI or SUMA to change focus point, and the other program jumps to that location at the same time
  - ★ Functional overlay in AFNI can be sent to SUMA for simultaneous display
- And much more — stayed tuned for the SUMA talks to come!

SUMA Teaser Movie



Color from AFNI, Images from SUMA

Images captured with the 'R' recorder function, then saved as animation with Save:aGif control

Other Educational Presentations

- How to get images into AFNI
- Detailed hands-on with using AFNI for data viewing
- Signal modeling and analysis: theory and hands-on
- Image registration
- Volume rendering hands-on
- ROI drawing hands-on
- Transformation to Talairach hands-on
- Group analysis: theory and hands-on
- SUMA hands-on and Surface-based analysis
- Miscellaneous AFNI utility programs and plugins
- A tour of the AFNI Website