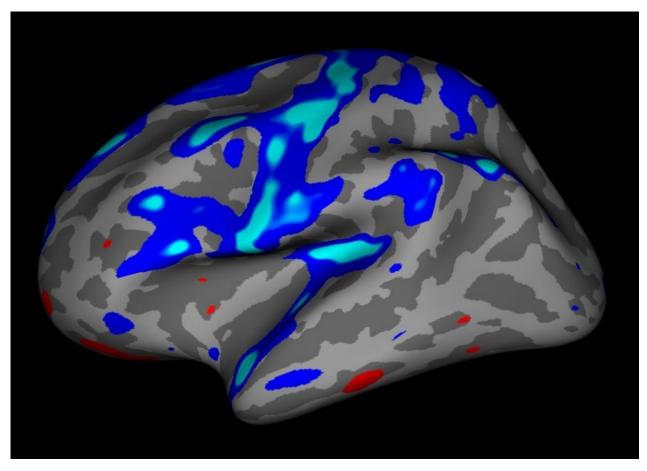
# **1. FreeSurfer Tutorial**



FreeSurfer is brought to you by the Martinos Center for Biomedical Imaging (supported by NCRR/P41RR14075), Massachusetts General Hospital, Boston, MA USA

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# **1. FSL/FreeSurfer Course: FreeSurfer Lectures and Tutorials**

### 1.1. June 23 and 24, 2011

### 1.1.1. Montreal, Canada

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FSL Course Timetable

#### 1.1.2. Thursday, June 23th - Introduction

Time	Title	Туре	Lecturer
5:30 - 5:50	Introduction to Freesurfer	talk	Bruce Fischl

### 1.1.3. Friday, June 24th - Applications / Troubleshooting / Group Analysis / FS-Feat Integration

Time	Title	Туре	Lecturer
9:00 - 9:20	FreeSurfer Applications	talk	David Salat
9:20 - 10:00	Analyzing the Individual Subject	talk	Doug Greve
10:00 - 10:15	Failure Modes	talk	Allison Stevens
10:15 - 10:30	Tkmedit/Tksurfer demonstration	demo	Doug Greve
10:30 - 11:00	tea and coffee		
11:00 - 11:30	Inspection of Freesurfer Output	tutorial	staff
11:30 - 12:30	Troubleshooting	tutorial	staff
12:30 - 2:00	lunch		
2:00 - 2:45	Group Analysis / GLM / Multiple Comparisons	talk	Doug Greve
2:45 - 3:00	QDEC demonstration	demo	Nick Schmansky
3:00 - 3:30	Freesurfer-FEAT Integration	talk	Doug Greve
3:30 - 4:00	tea and coffee		
4:00 - 4:30	Group Analysis / Multiple Comparisons - mri_glmfit	tutorial	staff
4:30 - 5:00	Group Analysis - qdec	tutorial	staff
5:00 - 5:30	Freesurfer-FEAT Integration	tutorial	staff
5:30 - 6:15	Future Directions of FreeSurfer	talk	Bruce Fischl

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# **1. Inspection of Freesurfer Output**

In this exercise you will visualize and inspect correctly processed output data so that you can become familiar with what the end product should look like. The exercise will step you through visual inspection of a variety of output, but is not necessarily the recommended procedure to take when trying to verify each subject. Some steps are only necessary to check when there are problems. However it is a good idea for new users to become familiar with what the expected output should look like and how to view it.

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If you are taking one of the formally organized courses, the tutorial data is already installed on the computer provided to you so please skip ahead to the **Viewing Volumes with Tkmedit** section. If not, then to follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names. If you are using the tutorial data please set the environmental variable TUTORIAL\_DATA to the location that you have downloaded the data to (on the course laptops, it happens to be mapped to /media/disk):

First you need to set your SUBJECTS\_DIR to the appropriate place:

tcsh
setenv SUBJECTS\_DIR \$TUTORIAL\_DATA/buckner\_data/tutorial\_subjs
cd \$SUBJECTS\_DIR

\* This will set your SUBJECTS\_DIR to the location where your tutorial data is if you have defined the variable TUTORIAL\_DATA as indicated at the top of this tutorial. If you are not using the tutorial data you should set your SUBJECTS\_DIR to the directory in which the subject you will use for this tutorial is located. \* Notice the command to open tcsh. If you are already running the tcsh command shell, then the 'tcsh' command is not necessary.

### 1.1. Viewing Volumes with Tkmedit

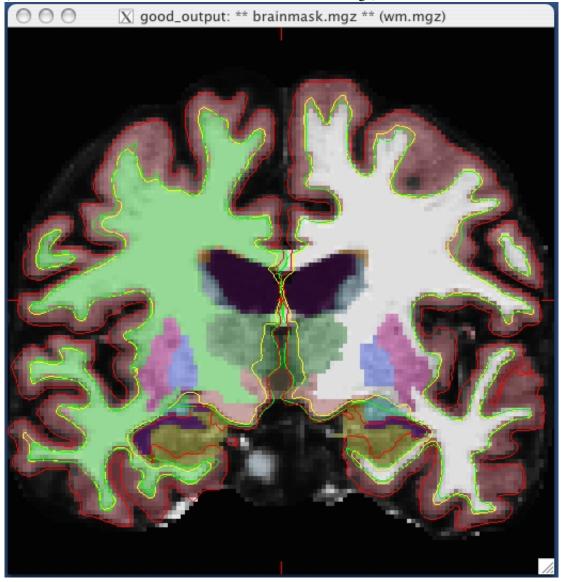
The volumes that are output can be loaded into tkmedit, along with surface outlines and the subcortical segmentation. With one command line you can load in the brainmask.mgz and wm.mgz volumes, the rh.white and lh.white surfaces (outlines), and the subcortical segmentation. Highlight the command in the box below, right click and select copy, then use the middle button of your mouse to click inside the terminal window (this will paste the command). Press enter.

```
tkmedit good_output brainmask.mgz \
    -aux T1.mgz -surfs \
    -segmentation aseg.mgz $FREESURFER_HOME/FreeSurferColorLUT.txt
```

- good\_output : name of subject
- brainmask.mgz : brain mask volume in mgz format (found in \$SUBJECTS\_DIR/good\_output/mri)
- -aux T1.mgz : load a second volume called T1.mgz (also found in \$SUBJECTS\_DIR/good\_output/mri)
- -surfs : load all surfaces (orig, white, and pial, for left and right hemispheres)
- -segmentation aseg.mgz : load automatic volume segmentation called aseg.mgz

You should see a tkmedit window open up to this:

(use the zoom  $\swarrow$  and move  $\clubsuit$  buttons to match this image)



You are currently looking at the brainmask.mgz (loaded as the main volume) with the surfaces displayed and the aseg.mgz (subcortical segmentation) overlayed. The pial (red line), and white (yellow line) surfaces are shown. You can toggle between the brainmask.mgz (main volume) and the T1.mgz (the auxiliary volume or 2nd volume loaded) and you can click on and off the aseg.mgz overlay. To become more familar with the buttons in the Tkmedit Toolbox please read the Freesurfer Tools section of this tutorial.

Here are the things you can look at while this is loaded in tkmedit:

- intensity normalization
- skull strip
- wm.mgz volume
- the final surfaces
- subcortical segmentation

For the first run through you might find it easiest to toggle off the aseg with the  $\square$  button. Using ctrl-g on the keyboard will have the same effect. You may also find it easier if you toggle off all the surfaces with the  $\square$  and  $\square$  buttons.

#### **Intensity Normalization**

Scroll through the brainmask volume and notice that the intensity is all uniform. You should not see any very bright or very dark spots. If you click on any voxel that is in the wm you can see that it has been normalized to an intensity of (or very close to) 110. The voxel intensity is shown in the Tkmedit Toolbox. This check and the following, Skull Strip check, can be done simultaneously since they both require you to look at features on the brainmask volume.

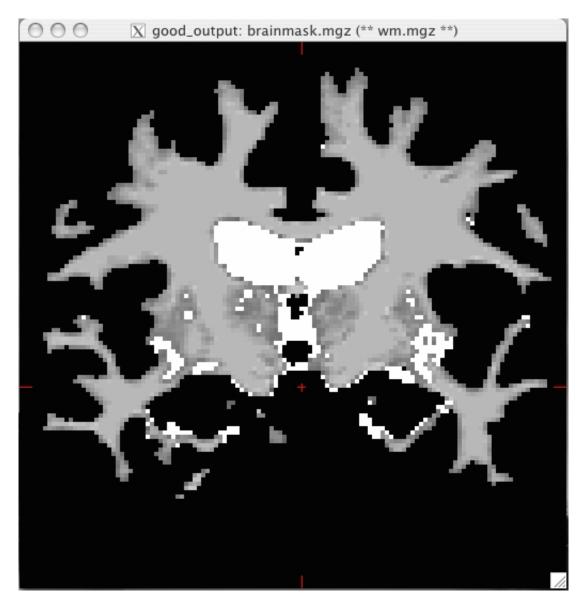
#### **Skull Strip**

Scroll through the brainmask volume and notice that there is no skull left in your image. Notice also that the cerebellum is still included in the brainmask volume. You should not see any large areas of skull left behind, or any areas of cortex or cerebellum removed from this volume. You should compare the brainmask.mgz volume to the T1.mgz volume that is also loaded to ensure that the skullstrip has worked properly. You can switch between views in a number of ways, ctrl-1 will show the main volume and ctrl-2

will show the auxiliary volume, or you can use the buttons and  $\clubsuit$ . Alt-c will allow you to quickly switch back and forth between the two volumes. This check and the previous, Intensity Normalization check, can be done simultaneously since they both require you to look at features on the brainmask volume.

#### White Matter Volume

To check the wm volume you should load it in as a new aux volume. To do this go to **File -> Aux Volume** -> **Load Aux Volume** and browse to wm.mgz. This will open the white matter volume, which will look like this:



This volume is comprised of all the voxels that freesurfer is calling white-matter, shown in shades of gray. These are the voxels that were normalized to an intensity of, or very close to, 110 as described above. The bright white voxels are voxels that have been added to the volume during the automatic editting of the wm volume. These edits fill the entire ventricle and basal ganglian defect. You can alternate between the

wm.mgz volume ( button) and the brainmask.mgz volume ( button) to see how well freesurfer has classified the white matter.

#### **Final Surfaces**

Switch back to the main volume by using the button (Alt-c on the keyboard will also do this), this will show the brainmask.mgz volume. To check your surfaces you will need to toggle them back on with the

F for the pial surface and for the white surface. The surfaces that are overlaid are the pial surface (red line) and white surface (yellow line). The white surface is the surface used in all calculations of thickness so it is important that this surface follows the boundary of the white matter accurately. The pial surface should accurately follow the boundary between the gray matter and the CSF. As you scroll through the slices keep in mind that you are looking at a 2-dimensional rendering of a 3-dimensional image, be sure to look at more than just one view too (i.e., sagittal, coronal and horizontal).

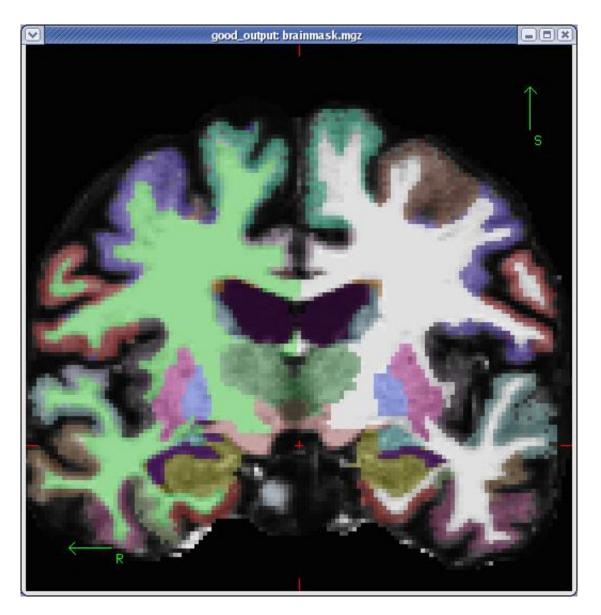
There are regions where the surfaces are not intended to be accurate that you should be aware of. Areas around the hippocampus and amygdala, as well as along the midline cutting plane will often show some inaccuracies. The pial surfaces will not follow the border of the amygdala, instead it will curve inward, mimicking the white surface (see coronal slice 137). Along the midline cut it is possible to see some overlapping of the surfaces from one hemisphere to another.

#### **Subcortical Segmentation**

Toggle on the subcortical segmentation with the button. This will show the complete segmentation of the subcortical structures. Each structure is labeled with a unique color/number distinction. If you click on a voxel the structures name and number label will be shown in the Tkmedit Toolbox. Scrolling through the slices you will be able to see that everything is labeled, and done so accurately. Sometimes it is easier to see the structures and their boundaries looking in either the sagittal or horizontal view, so be sure to check around in all of them.

#### **Aparc+Aseg segmentation**

To load in the aparc+aseg.mgz segmentation you can go to **File --> Load Segmentation**, and browse to the aparc+aseg.mgz, which will look like this:



This segmentation shows the same subcortical structures that are labeled in the aseg.mgz, but uses the cortical parcellation labels around the cortex.

# **1.2. Viewing Surfaces with Tksurfer**

Now that you've checked out everything in the the displays one hemisphere at a time. This exercise will go through visualizing things on the left hemisphere only, but everything works the same on the right hemisphere (except in the initial command you should specify rh instead of lh if you want to look at the right hemisphere). To become more familar with the buttons in the Tksurfer Toolbox, please read the Freesurfer Tools section of this tutorial.

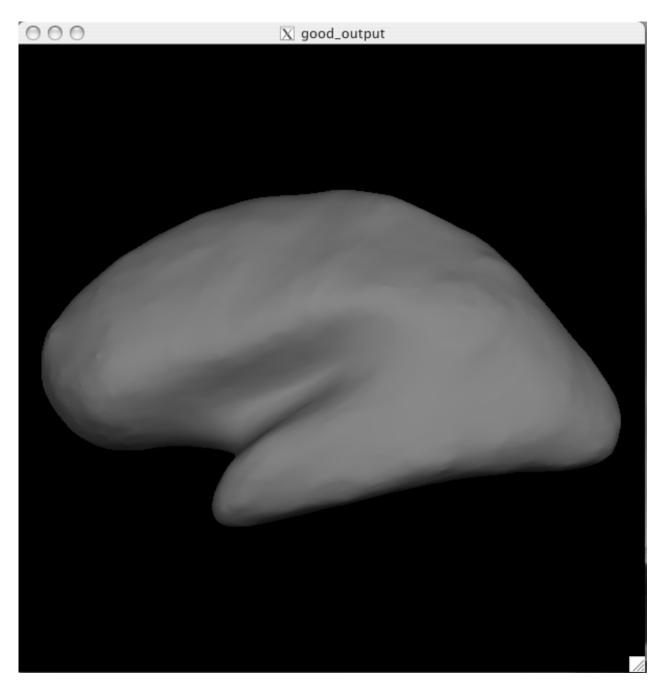
Here are the things you can look at with tksurfer:

- pial, white and inflated surface
- sulc and curv curvature files
- thickness files
- cortical parcellation

To open tksurfer with the left hemisphere inflated surface of your subject, **open a new terminal window**, then use the following command:

setenv SUBJECTS\_DIR \$TUTORIAL\_DATA/buckner\_data/tutorial\_subjs
cd \$SUBJECTS\_DIR
tksurfer good\_output lh inflated

You should see a tksurfer window open up to this:



You are currently looking at the inflated surface. The surface can be rotated using the buttons in the navigation toolbar: **Operation Operation Operation Operation Operation Description Description**

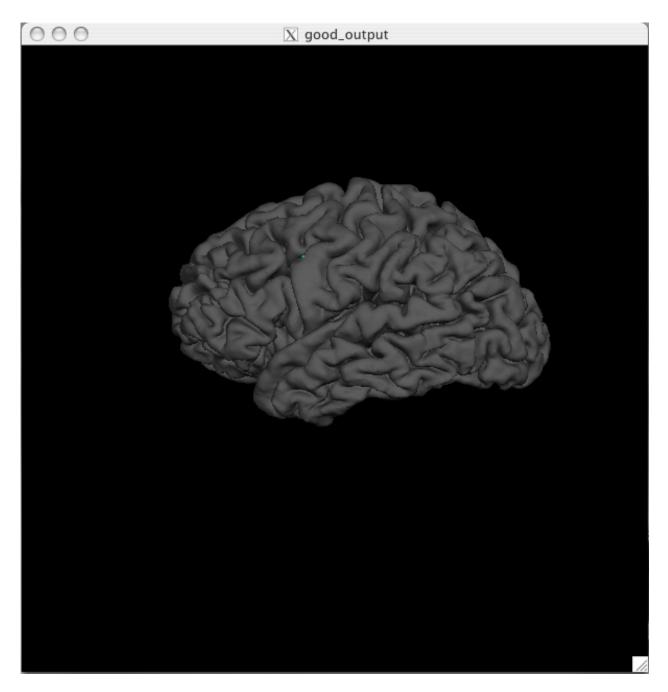
Inflated surface

The inflated surface is good to look at when checking to see if you need to make edits to the wm.mgz volume. You'll notice as you inspect this surface that it is smooth and free from holes, bumps and other defects. If you click on the surface, you will see the coordinates of the vertex you clicked on in the Tools window. To clear the marks made on the surface after clicking on it, you can do Ctrl+Shift+middle click on each mark to erase it.

#### **Pial Surface**

You can load in other surfaces, and tksurfer will then allow you to switch between them all. The easiest way to do this is to hold down CONTROL and click with the right mouse button on the various surface

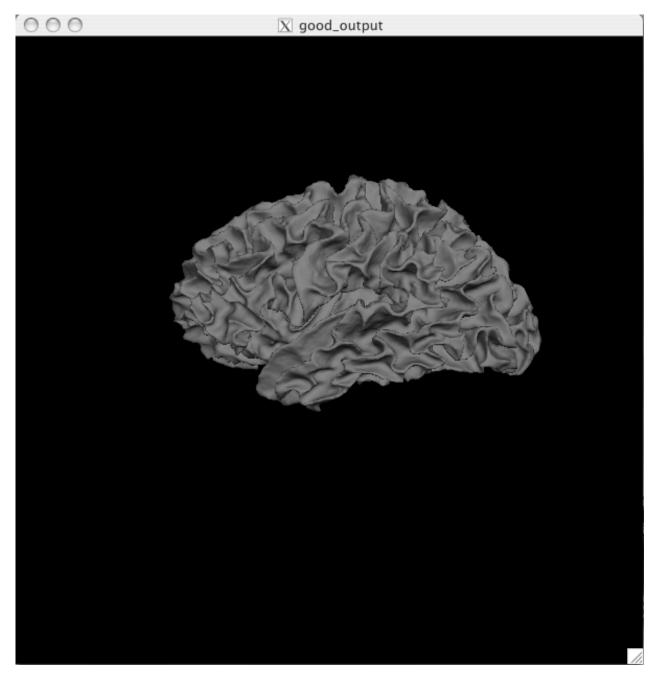
buttons. To load the Pial Surface hold ctrl and right click the pial surface button  $\bigcirc$  a box will pop up where you could browse to the location of the surface you want to load, but it should be already filled in with the path to the lh.pial surface - so you can hit ok. This is what the pial surface will look like:



The pial surface is showing you the outer boundary of the gray matter/CSF. This is the same file that was viewed in tkmedit, just represented as a surface image rather than the red outline on the volume. You can inspect this surface by rotating it around as you wish.

#### White Surface

You can follow the same procedure that you used to load the pial surface for the white surface, except this time hold ctrl and right click the white surface button  $\bigcirc$  You can inspect this surface by rotating it



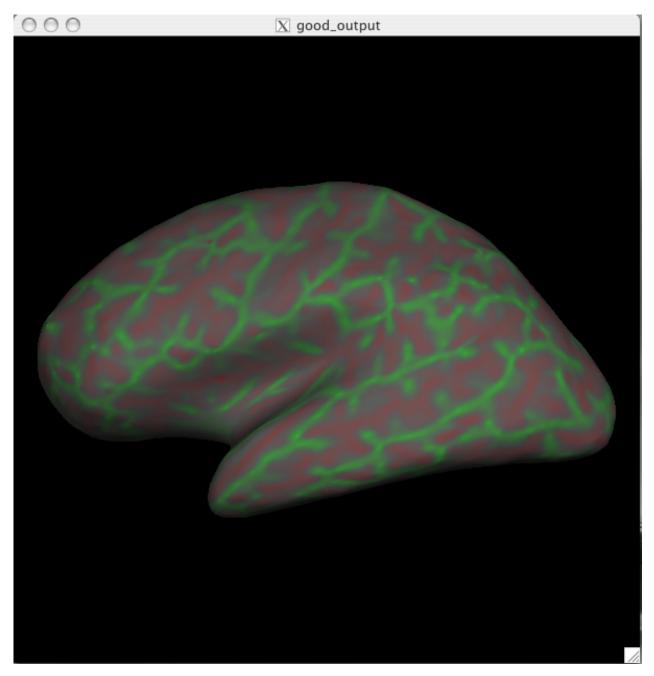
around as you wish. This is what the white surface will look like:

The white surface shows the boundary between the white and the gray matter. Again, this is the same file that was viewed in tkmedit (as the yellow outline) just represented in a 3D manner.

**Curv and Sulc Files** 

Switch back to viewing the inflated surface of the brain by pushing the M button. You can now load in

the curvature file, lh.curv, by holding ctrl and right clicking on the **s** button on the tksurfer Tools window. The box that pops up will automatically have selected the lh.curv file, so click ok. The curvature file will look like this:



This is showing the slightly smoothed mean curvature. It has the units of 1/mm, and with an outward pointing normal vector field. Negative regions are folded-out and shown in green (gyral), and positive regions are folded-in and shown in red (sulcal)

To view the sulc file you could have changed the dialog box to say lh.sulc, or now you can go to **File -> Curvature -> Load Curvature** and browse to the lh.sulc file (found in

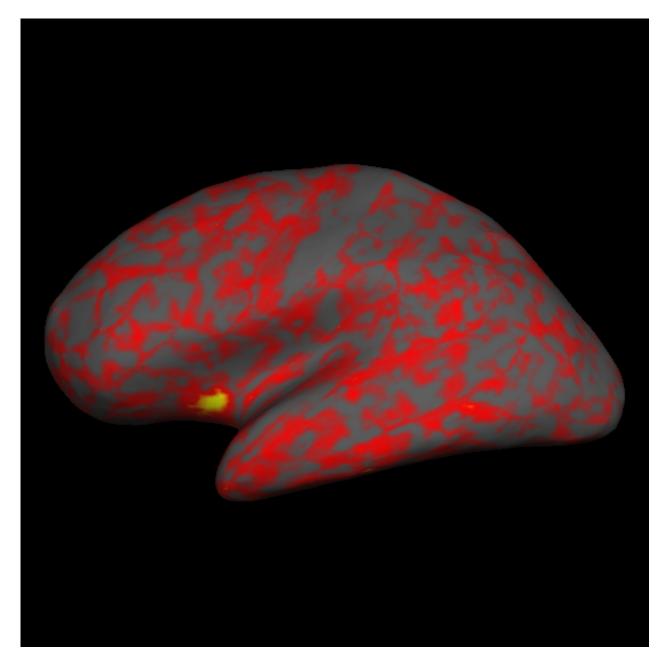
\$TUTORIAL\_DATA/buckner\_data/tutorial\_subjs/good\_output/surf/). Be sure you select the curvature file for the correct hemisphere, if you select the wrong one it will not look right. The lh.sulc file will load a slightly different display of green/red values than the lh.curv. These are showing the sulcal depth and again here the red regions are sulcal and the green regions are gyral. You can view this as you click on the pial and/or white surfaces.

#### **Thickness Maps**

To view the thickness maps it is probably best to toggle off the curvature **C**. You can load the thickness

map, lh.thickness, by holding ctrl and right clicking *on the tksurfer Tools window and browse to the lh.thickness file (you may have to go up a directory - it's found in the two sets to the like the two sets to the two se* 

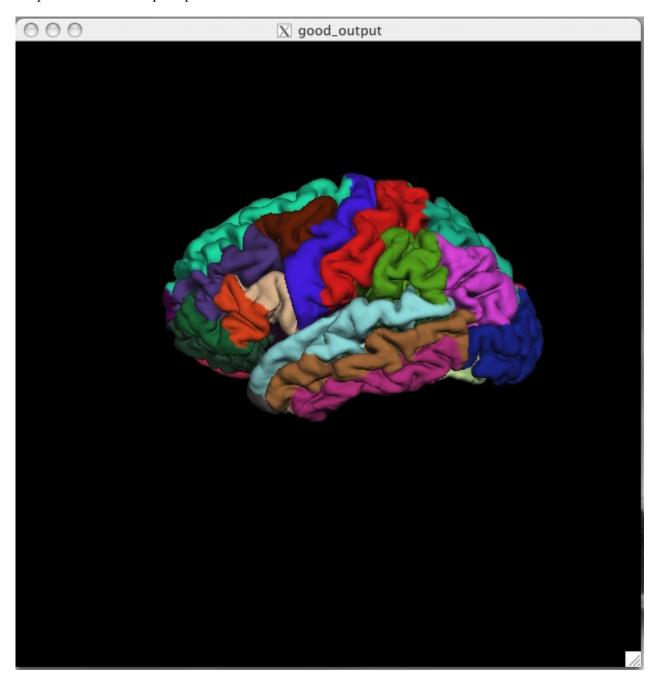
\$TUTORIAL\_DATA/buckner\_data/tutorial\_subjs/good\_output/surf/). Be sure you select the thickness file for the correct hemisphere, if you select the wrong one it will not look right. The lh.thickness map should open up and look like this (after the threshold is changed to Linear):



You can adjust the thresholds of the map by going to **View -> Configure -> Overlay** which will open a new window that will allow you to change the thresholds. First click the **Thresholds: Linear** checkbox to make the image match that shown above. Then try some different settings and see how it affects the display on the map (some options to try min of 1.0 and max of 3.0, min of 2 max of 5, etc). After you change the min and max values be sure to hit apply.

#### **Cortical Parcellations**

To view the cortical parcellation it is probably best to toggle off the curvature and thickness files , and to switch to the pial surface, . You can load the parcellation by going to **File -> Label -> Import Annotation** and browsing to the lh.aparc.annot file (found in \$TUTORIAL\_DATA/buckner\_data/tutorial\_subjs/good\_output/label/). Be sure you select the label file for the correct hemisphere, if you select the wrong one it will not look right. The lh.aparc.annot should open up and like this:



You can inspect the parcellation by rotating the surface to see all sides. You can switch to the inflated view, or turn the labels to outline view, whichever way is most comfortable for you to view the parcellations. The parcellation that is loaded here was created with the Desikan-Killiany atlas. By default there are two parcellations that are made when recon-all is run. The second parcellation, called ?h.aparc.a2005s.annot, is created with the Destrieux atlas. The difference is the number and designation of the areas that are labeled. You can load this second parcellation by first going to **File -> Label -> Delete all Labels**. This will remove the first parcellation. Then you can repeat the steps for loading a parcellation, this time browsing to lh.aparc.a2005s.annot to load the second parcellation.

# **1.3.** Using Tkmedit and Tksurfer together

When you are viewing the same subject in tkmedit and tksurfer at the same time (generally done using two separate terminals, one to launch tkmedit and one to launch tksurfer) you can use some tools to switch from one point on the surface (in tksurfer) to the same point in the volume (in tkmedit). To do this, first

put your cursor at the point you want in tksurfer. Next, click the save point button 🛱 in tksurfer. This

will save the cursor position. Then, in the tkmedit toolbox window, click the goto saved point button This will now bring you to the same point, only in the volume. Look for the red plus sign (it is small and can be hard to find, depending on the saved point, but should be near the surface for this example). This technique is very useful when you see something wrong on the surface of a subject (in tksurfer) and you want to see what is happening in the volume in that same place.

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# 1. Troubleshooting your output

This set of exercises will take you through a few examples of problem outputs, asking you to identify the problems and possible methods to fix the problems. Each example will have a before and after picture, as well as an explanation of how to fix the problems seen.

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If you are taking one of the formally organized courses, the data is already installed on the computer provided to you so please skip ahead to the **Failure Modes** section. If not, then to follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. This set of exercises is not possible without the tutorial data set. Please set the environmental variable TUTORIAL\_DATA to the location that you have downloaded the data to (on the course laptop, it happens to be set to /media/disk):

First make sure you have your SUBJECTS\_DIR set to the correct location:

```
tcsh
setenv SUBJECTS_DIR $TUTORIAL_DATA/buckner_data/tutorial_subjs
cd $SUBJECTS_DIR
```

• Notice the command to open tcsh. If you are already running the tcsh command shell, then the 'tcsh' command is not necessary.

#### Contents

- 1. Troubleshooting your output
  - 1. Failure Modes
  - 2. Edits to the brainmask volume
  - 3. Edits to the wm volume
  - 4. Correcting topological defects
  - 5. Skull strip
  - 6. Adding control points
  - 7. Talairach transformation
  - 8. Editing the Aseg

### **1.1. Failure Modes**

Below, we ask you to look at several subjects with various errors that require manual intervention. Open each subject and try to identify the problem on your own. For each subject, there is a link to instructions on how to fix the problem. While you are trying to identify the problems with the subjects listed below you may find it helpful to refer back to the previous exercise, or open your own instance of the subject good\_output to compare these subjects to a good example.

### 1.2. Edits to the brainmask volume

Open the subject, **pial\_edits\_before**.

tkmedit pial\_edits\_before brainmask.mgz -aux T1.mgz -surfs

The command above will open the brainmask.mgz volume, the T1.mgz loaded as aux, and the surfaces for both hemispheres. (You can feel free to open other volumes in aux or to load in the aseg.mgz if you want or need to.) Scroll through the slices of this subject to check the pial and white surfaces for accuracy, making sure they follow the actual gray/white boundaries and that they do not include anything that should not be included.

If you would like to see the inflated surfaces for each hemisphere of this subject, **open a second and third terminal window**, then open the surfaces in tksurfer:

tksurfer pial\_edits\_before lh inflated

tksurfer pial\_edits\_before rh inflated

Once you have identified the problem (or have given up!), click here for detailed instructions on how you can fix it.

#### **1.3.** Edits to the wm volume

Now, take a look at the next subject, wm1\_edits\_before.

tkmedit wml\_edits\_before brainmask.mgz -aux Tl.mgz -surfs

Again, this will bring up the brainmask.mgz volume, the T1.mgz volume, and the surfaces for both hemispheres.

In your second and third terminal window, if not already open, open the surfaces in tksurfer:

tksurfer wm1\_edits\_before lh inflated

tksurfer wml\_edits\_before rh inflated

This will open the inflated surfaces for both hemispheres in tksurfer.

The trouble with this subject has occurred during the white matter segmentation step. Check the surfaces in the T1 volume to find the spot that does not match the actual gray/white boundary. If you load the wm.mgz volume as the aux volume you will see a hole in this area. If you are looking in tksurfer you will see a hole or dimple in the inflated surface. You can use the "save point" "goto point" commands to find this spot in the volume.

Click here for detailed instructions on how you can fix it. You can also compare the original data to a corrected and rerun version, **wm1\_edits\_after**.

To view this corrected subject (which was corrected by following the detailed instructions provided to you):

```
tkmedit wm1_edits_after brainmask.mgz -aux wm.mgz -surfs
```

### 1.4. Correcting topological defects

Open the subject, topo\_defect\_before, in tkmedit.

tkmedit topo\_defect\_before brainmask.mgz -aux wm.mgz -surfs

This will open the brainmask.mgz volume, the wm.mgz loaded as aux, and the surfaces for both hemispheres.

Take a look at this subject, inspecting the various outputs that were mentioned in the previous exercise and see if you can identify what is wrong.

Hint: It's a very small error in the posterior part of the brain.

Once you spot the error, click here for detailed instructions on how you can fix it. After you have fixed it, you can compare the original data to the a corrected and rerun version, **topo\_defect\_after**. To view this corrected subject:

tkmedit topo\_defect\_after brainmask.mgz -aux wm.mgz -surfs

### 1.5. Skull strip

Now, take a look at the next subject, **skullstrip1\_before**, in tkmedit.

This will open the brainmask.mgz volume, the T1.mgz loaded as aux, and the surfaces for both hemispheres.

The trouble with this subject has occurred in the skull stripping step. Check the brainmask.mgz volume carefully, comparing it to the T1.mgz volume (loaded in aux) to make sure that the skull has been completely stripped away, leaving behind the complete cortex and the cerebellum.

You can feel free to open other volumes in aux or to load in the aseg.mgz if you want or need to.

Click here for detailed instructions on how you can fix it. You can also compare this to the finished version, **skullstrip1\_after**.

### 1.6. Adding control points

Now, take a look at the next subject, cp\_before.

```
tkmedit cp_before brainmask.mgz -aux T1.mgz -surfs
```

Again, this will bring up the brainmask.mgz volume, the T1.mgz volume, and the surfaces for both hemispheres.

In your second terminal window, if not already open, open the surfaces in tksurfer:

```
tksurfer cp_before lh inflated &
tksurfer cp_before rh inflated &
```

This will open the inflated surfaces for both hemispheres in tksurfer.

The trouble with this subject has occured during the intensity normalization. Check the white surface and be sure it is including all of the white matter as it should. If you find an area that is not included as white matter be sure to check to see that the intensity is at (or very close to) 110.

You may need to open other volumes in order to see or fix the problem.

Click here for detailed instructions on how you can fix it.

#### **1.7.** Talairach transformation

Now, take a look at the next subject, **tal\_before**.

tkmedit tal\_before brainmask.mgz -aux T1.mgz -surfs

Again, this will bring up the brainmask.mgz volume and the T1.mgz volume and the surfaces for both hemispheres.

In a second and third terminal window, if not already open, open the surfaces in tksurfer:

tksurfer tal\_before lh inflated

tksurfer tal\_before rh inflated

This will open the inflated surfaces for both hemispheres in tksurfer.

The trouble with this subject is a little tricky to figure out. The midline cutting planes are not actually on the midline, but the real root of this problem is in one of the very initial steps of recon-all. The talairach transform is bad and will need to be fixed.

You may need to open other volumes in order to see or fix the problem.

Click here for detailed instructions on how you can fix it.

### **1.8. Editing the Aseg**

We generally advise against editing the aseg.mgz unless absolutely necessary. Try using an xopts script first to change the intensity thresholds being used. Editing the aseg will add more variance to your data. Instructions on how to edit the aseg are here.

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# **1. Correcting topological defects**

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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

Sometimes, the topological fixer fails which leads the white matter (WM) surface (yellow line) to exclude some voxels that are white matter. Simple edits to wm.mgz volume can correct this defect.

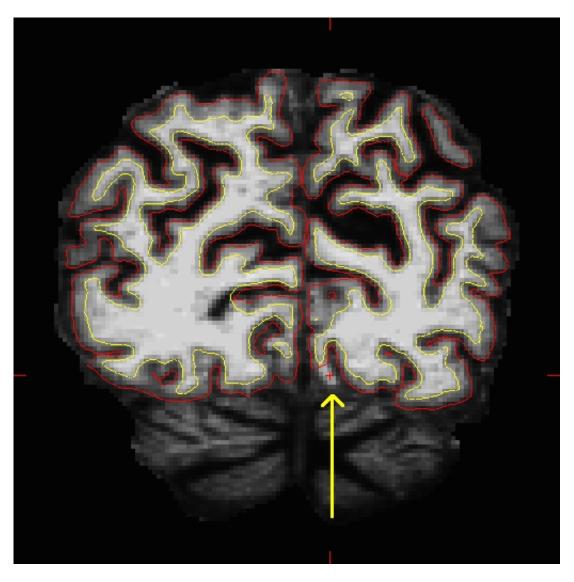
### 1.1. Editing the Volume

Subject topo\_defect\_before is an example of topological defect and this page will walk you through fixing it.

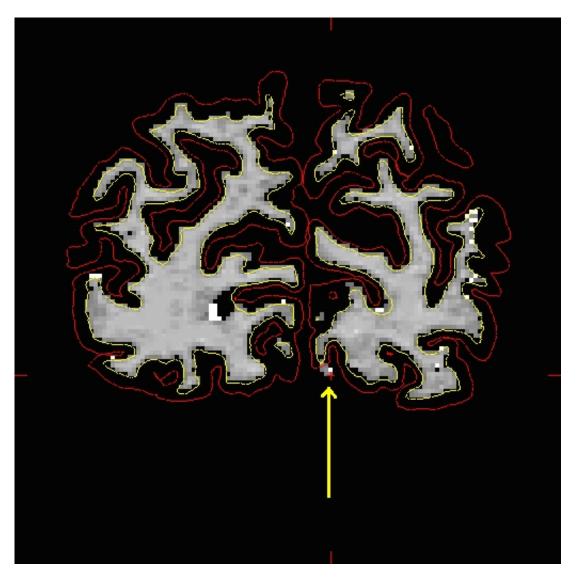
Open topo\_defect\_before in tkmedit.

```
tkmedit topo_defect_before brainmask.mgz -aux wm.mgz -surfs
```

Use the arrow keys to scroll through the volume to find the error in white matter surface. If you look at coronal slice 57, there is a defect in the left hemisphere closer to the midline, where some white matter voxels are segmented incorrectly. This type of topological defect is called a 'hole' which needs to be filled as opposed to a handle which needs to be cut.



As you can see in wm.mgz, some white matter voxels are excluded from the white matter surface.



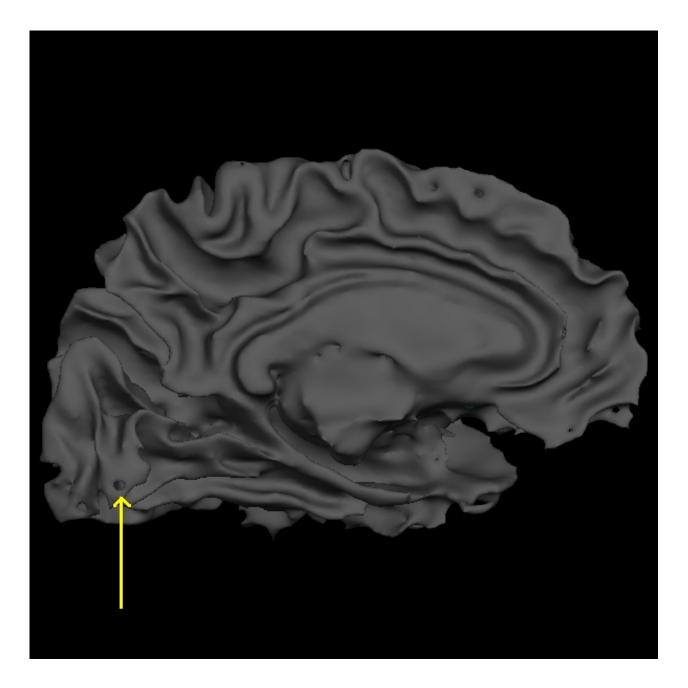
To view the defect in the surface viewer, first open another terminal:

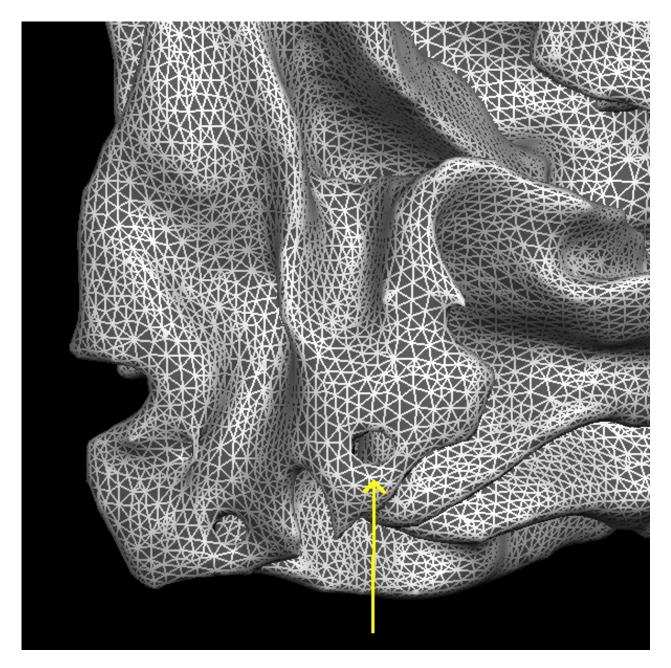
tcsh

and then open the surface viewer:

tksurfer topo\_defect\_before lh smoothwm.nofix

Below are couple of images of lh.smooth.nofix loaded in tksurfer and zoomed in image of lh.smooth.nofix with Wireframe Overlay turned on (**View --> Wireframe Overlay**), respectively. You can see the error as pointed by the yellow arrow, which is a hole that needs to be filled to correct this type of topological defect.





Now back to the tkmedit display, if you scroll through slice by slice, you'll notice that there is a small group of white matter voxels which disconnect and connect back again after a few slices. The white matter surface doesn't include these voxels when they are disconnected.

This area is too thin to add control points to extend the WM surface. You'll need to add some white matter voxels to fix this error. But first scroll through the slices in the brainmask.mgz volume and make a note of where the white surface is just beginning to exclude voxels and where the problem ends. In this case, the problem starts at coronal slice 53 and goes on upto slice 60.

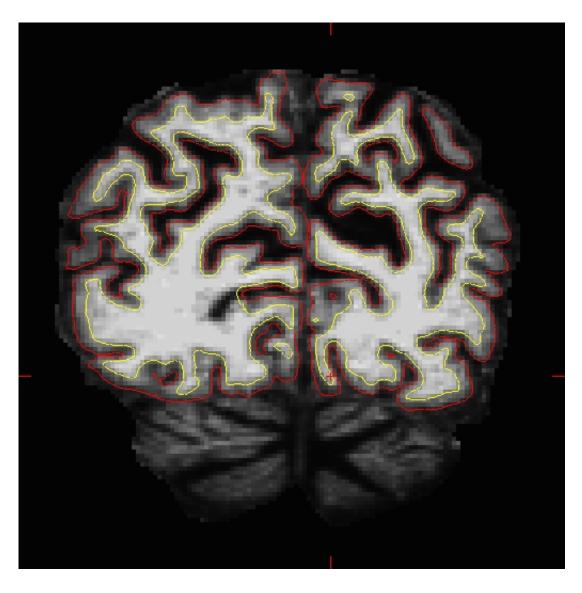
To begin editing, click on the Edit Voxels Tool button on the tkmedit tool bar, then go to **Tools --> Configure Brush Info...** Since wm.mgz is loaded as aux volume, select Aux volume as the Target. Use the middle mouse button to begin painting in the voxels. Use the right mouse button to delete voxels.

Start adding the voxels to wm.mgz volume on coronal slice 53. Toggle between the brainmask.mgz and wm.mgz using Ctrl-1 and Ctrl-2. Use brainmask.mgz as a guide while painting. Only add as many voxels as is needed to correct the error. Switch between the coronal, sagittal and horizontal views to check if you need to add more for the white matter to be labelled correctly. With more practice, it becomes easier to decide where to add voxels to correct the defect.

At any time, you can save the changes you've made to the wm volume by selecting 'Save Aux Volume' in tkmedit's 'File' menu, and clicking 'OK'

You can look at wm.mgz volume of topo\_defect\_after to compare your edits with the tutorial data.

Coronal slice 57 in brainmask.mgz looks like this after correction.



# **1.2. Regenerating the Surface**

Once you have made all the edits to the wm.mgz volume, you can run the following command to regenerate the surfaces:

recon-all -autorecon2-wm -autorecon3 -subjid topo\_defect\_before

#### Do not run this command if you are conducting the tutorial!

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# 1. Making Edits to the White Matter

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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

Sometimes the white matter is not segmented correctly: sometimes voxels that should be white matter are excluded, and other times voxels that should not be white matter are included in error. Either of these occurences can be fixed with simple manual edits. Below you will find examples of a few of the common problems and how to fix them. For some people it is easiest to see these problems when looking at the inflated surface in tksurfer. Others can pick them out while viewing the volumes in tkmedit. Either method is fine for identifying these problems. However they can only be fixed using tkmedit with the wm.mgz volume.

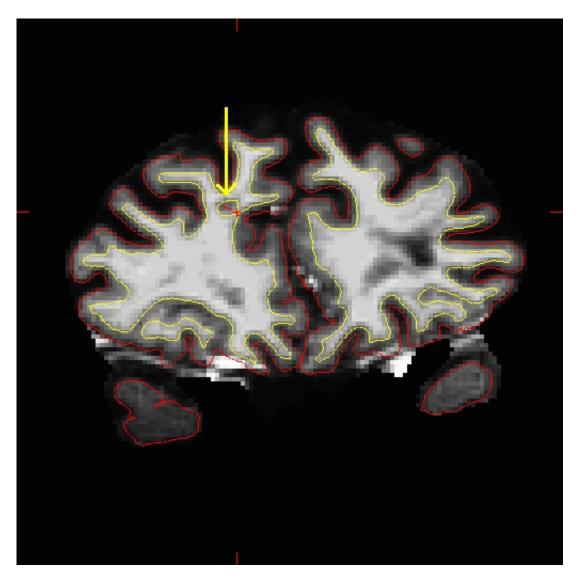
### 1.1. Geometric inaccuracy due to brain lesion

wm1\_edits\_before is an example of white matter being excluded from the wm.mgz volume due to the presence of a brain lesion. This page will walk you through fixing this specific example as well as other common problems with the wm.mgz volume.

First, make sure you have wm1\_edits\_before loaded in tkmedit:

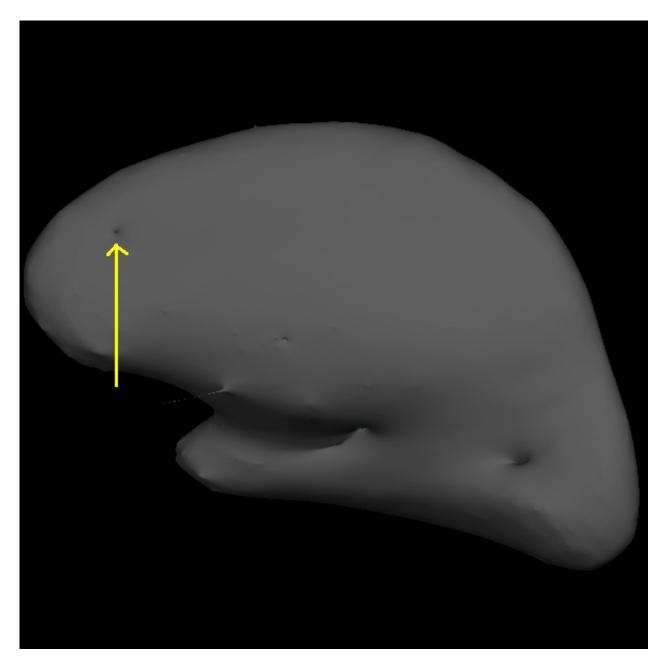
```
tkmedit wml_edits_before brainmask.mgz -aux wm.mgz -surfs
```

If you look at coronal slice 160 you will notice an area on the right hemisphere where the white surface (yellow line) does not follow the surface of the brain, but in fact cuts into it. This geometric inaccuracy is caused by a lesion where white matter has been marked as non-white matter.

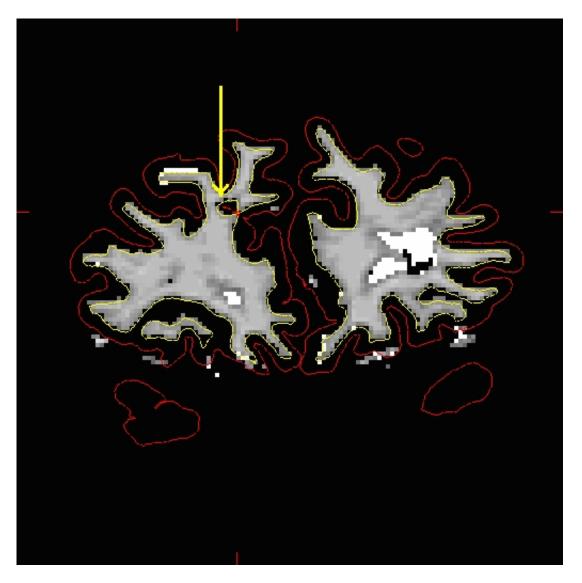


You can see this as a dimple or hole on the inflated surface in tksurfer too (see next image). If not already open, in a second terminal window, open the rh surface in tksurfer and you can see the dimple or hole on the medial side:

tksurfer wml\_edits\_before rh inflated

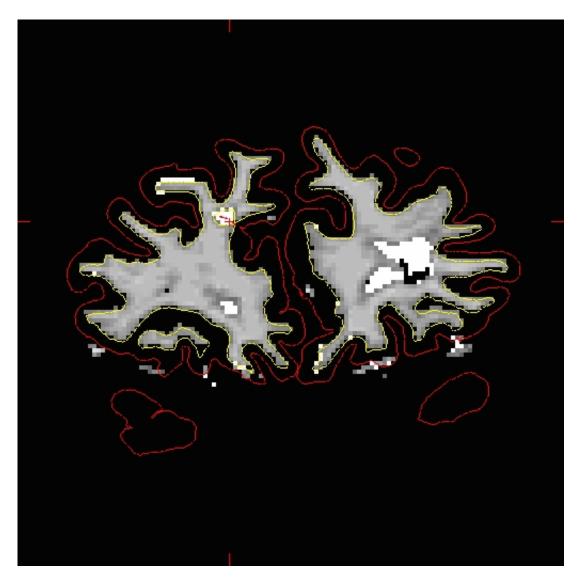


With the wm.mgz volume loaded into tkmedit you can see that this area has been left out of the wm volume completely.



To fix this problem you will need to fill in the missing voxels in the wm.mgz volume. First, it's a good idea to use the arrow keys to scroll through the individual slices in the volume until you get a good idea of where the problem starts and ends. You will want to start filling in voxels when the inaccuracy appears, and keep filling them in slice by slice until the problem is no longer visible. The coronal view and a brush radius of one or two are good settings for painting in voxels. Zoom In and Out either by using the icons in the tkmedit Tool window or by Ctrl-right mouse button (Zoom In) and Ctrl-left mouse button (Zoom Out) to help you see the inaccuracy.

To begin editing voxels, click on the Edit Voxels Tool button  $\square$ , then go to **Tools --> Configure Brush Info...** Set the *radius* to 2 and *Shape* to be *Circle*. By default tkmedit will edit on the main volume loaded, if the wm volume is loaded as your aux volume you will also need to select *Aux volume* as the *Target*. Use the middle button on your mouse to begin painting in the voxels. If you fill in too many voxels, the right mouse button acts as an eraser. When you are done filling the region, slice 160 should look like this:



While filling the hole in slice 160, you will notice that the hole is not completely enclosed. To see where the boundary should be, switch back and forth between the brainmask.mgz and wm.mgz volume with Ctrl-1 and Ctrl-2. Use the brainmask.mgz volume as a guide, and make the boundary of the voxels that you paint in correspond as closely as possible to the boundaries suggested in the brain volume. Go to the next slice, and fill in this slice in the same way. You will need to continue filling in slices in this fashion until the region is completely filled in. If you would like to compare your edits to ones that have been done for you you can open a second tkmedit with the wm1\_edits\_after wm.mgz volume, and for each slice check your results with the corresponding slice in the wm1\_edits\_after volume. At any time, you can save the changes you've made to the wm volume by selecting 'Save Volume' in tkmedit's 'File' menu, and clicking 'OK'. You can verify your results by viewing the changes made to the wm1\_edits\_after wm.mgz volume

**Tips:** 

• Sometimes regions may appear enclosed in some slices (i.e. appear as holes), open in subsequent slices (i.e. no longer appear as holes), then enclosed again as you scroll forward. The rule of thumb when editing these is to keep filling until you reach the slice where they finally open up and are no longer enclosed.

•

When filling in regions that are in both hemispheres, you may opt to fill them in both hemispheres at the same time, when you become more comfortable with editing inaccuracies. To load the white surface for the other hemisphere you will need to go to File --> Aux Surface --> Load Aux Main Surface and select the other hemisphere's white surface (i.e., rh.white). This will load the rh.white surface as the Aux surface and you can now follow the surfaces for both hemispheres at the same time.

- When editing geometric inaccuracies in the future, you may find it easier to edit the first slice and last slice first, thereby 'capping' the slices that need filling. Then you just need to fill all the slices between the endcaps.
- Sometimes there is variation between a subject's left and right hemispheres, so that in a particular slice one hemisphere's region will finally 'open up', but the other hemisphere's region is still enclosed. In such cases, continue to fill the enclosed region only, even though the other is open. This will address the topological problem.
- Problems like the one shown here, due to brain lesions, should always be fixed with edits to the wm.mgz volume. Do not use control points to try and automatically adjust the intensity in these areas.

After you have saved all of your edits, you could recreate the final surfaces with the command:

recon-all -autorecon2-wm -subjid wm1\_edits\_before

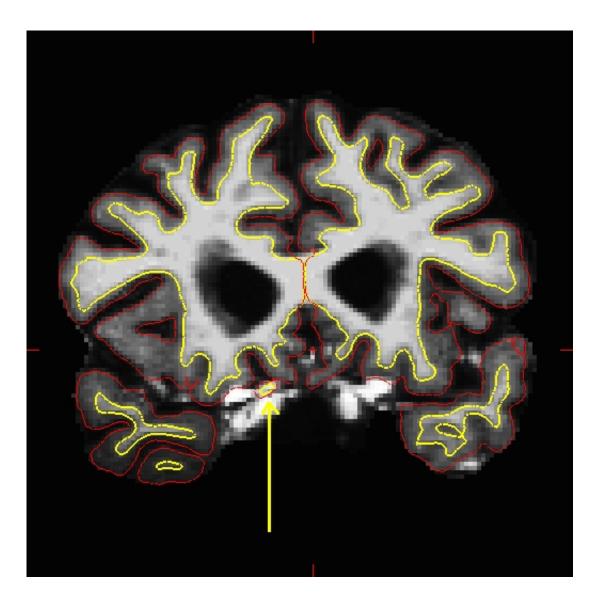
**Do not run this command if you are conducting the tutorial!** This step will take a long time and there is no need to run it for the tutorial purposes.

## 1.2. Non-white matter classified as white matter

Sometimes non-white matter (i.e., skull) is included as white matter. To load in the example use this command:

```
tkmedit wm2_edits_before wm.mgz -aux brainmask.mgz -surfs
```

Scroll through the brainmask.mgz volume slices and as you go posterior you will see a group of voxels included as white matter, around lice 152. They are surrounded by the white surface (yellow line). This group lies outside of the pial surface (red line) and is clearly not white matter. In order to correct this inaccuracy it is necessary to delete the voxels which are not part of the white matter.





Using Ctrl-1 and Ctrl-2 to switch back and forth between the brain and wm volumes, you can clearly see that this region should not be labeled as white matter. To delete these voxels, click the Edit Voxels Tool

button in the tkmedit toolbar, and put your mouse cursor over the voxels you want to delete. Delete them with the right mouse button. This is what slice 152 should look like after you have deleted the errant voxels:



Go through the slices with the up and down arrows, continuing to delete voxels until the inaccuracy is no longer there. (It spans approximately 30 slices.) You can check your results by looking at the wm2\_edits\_after wm.mgz volume in tkmedit. Note that in this volume, only a few of the voxels have been removed from each slice, instead of removing the entire inaccuracy. This is because you only need to remove as many voxels as is necessary for the segmentation to classify this region as non-white matter. Until you are more comfortable with editing inaccuracies, go ahead and remove all of the voxels. More practice will give you a better feel for how many voxels need to be removed.

Once you have made all the edits to the wm.mgz volume you would regenerate the surfaces using the following command:

recon-all -autorecon2-wm -autorecon3 -subjid wm2\_edits\_before

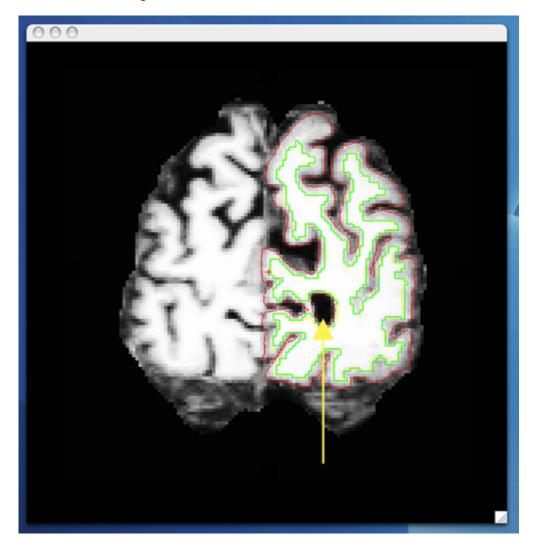
**Do not run this command if you are conducting the tutorial!** This step will take a long time and there is no need to run it for the tutorial purposes.

## **1.3. Lateral Ventricle**

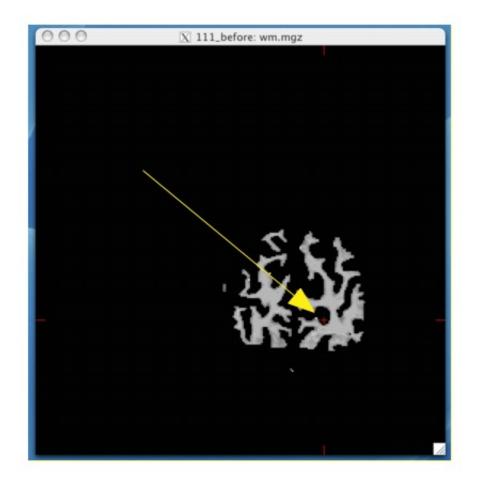
The automatic edits made to to wm volume will fill the lateral ventricles. On occasion they are not filled entirely. Open wm3\_edits\_before in tkmedit.

tkmedit wm3\_edits\_before wm.mgz -aux brainmask.mgz -surfs

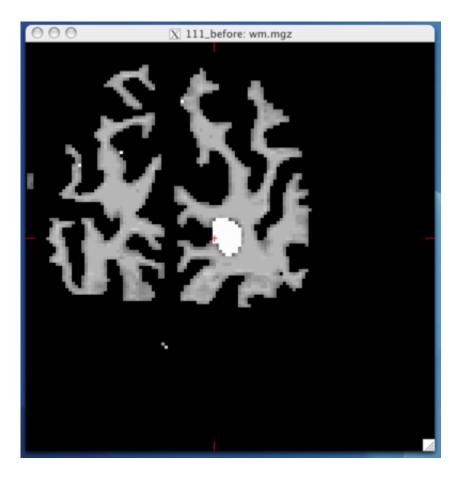
As you scroll to the posterior portions of the brainmask.mgz you will see the white surface (yellow line) start to follow the edge of the ventricle (look around coronal slice 53).



When you toggle between wm and the brain volume, you can see that this portion of the ventricle should be filled in. This inaccuracy can be corrected in the same fashion as the inaccuracies above, using the Edit Voxels tool to fill in the missing voxels.



When you are finished filling in slice 53, it should look like this:



Continue filling in this region until the hole disappears. The geometric inaccuracy is present from slices 50 through 168. Once you have fully edited the wm.mgz volume you would regenerate the surfaces with the following command:

recon-all -autorecon2-wm -autorecon3 -subjid wm3\_edits\_before

**Do not run this command if you are conducting the tutorial!** This step will take a long time and there is no need to run it for the tutorial purposes.

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## **1.** Correcting Pial Surfaces

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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

The pial surface is created by expanding the white matter surface so that it closely follows the gray-CSF intensity gradient as found in the brainmask.mgz volume. Once an accurate white surface is created then you can work on correcting the pial surface if needed. The pial surface boundary and white matter surface boundary should not cross. After the pial surface has been generated, it's a good idea to visually check it for defects that may have been created during automatic topology fixing. To check the pial surface, it may be loaded into tkmedit and viewed along with the brainmask.mgz volume. If the surface appears not to follow the gray-CSF boundary in the volume, edits may be required.

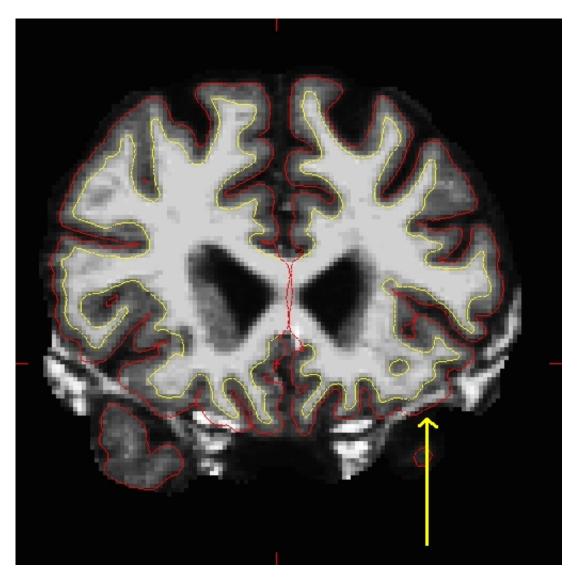
### 1.1. Editing the Volume

Subject pial\_edits\_before is an example of the pial surface including non-cortex within the boundaries. This page will take you through fixing this and other similar problems with the pial surface including non-cortex material.

First, make sure you have pial\_edits\_before loaded in tkmedit:

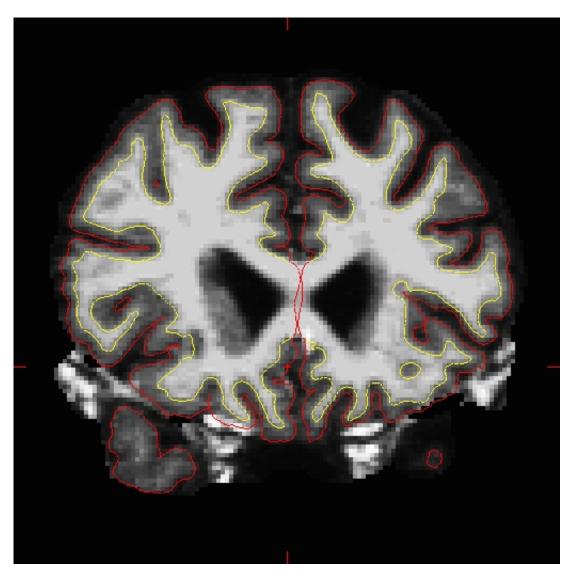
```
tkmedit pial_edits_before brainmask.mgz -surfs
```

Use the arrow keys to go through the volume slice by slice, and view the pial surface (red line) and white matter surface (yellow line). Notice the bright diagonal line in slice 157 that has caused the pial surface to expand past the actual pial boundary. This is the result of a bad segmentation incorporating a piece of the dura within the pial surface.



To fix this type of error you can simply edit away the offending voxels from the brainmask.mgz volume. To do this you will need to select the edit voxels tool and set the brush to a size and shape comfortable for you. A circle brush of radius 2 works well for this edit. In the tkmedit toolbar, go to **Tools -> Configure Brush Info...** Set Radius to 2, and Shape to "Circle". Close the configuration menu, and click the "Edit Voxels" button in tkmedit toolbar.

Find a place in the image where the dura is causing errors in the segmentation. Use the right mouse button to delete the voxels. It is not necessary to completely remove the dura to get an adequate pial surface, but it is good to do so until you are more familiar with manual editing. When you are finished removing the bright diagonal line in slice 157, it should look like this:



Continue on the other slices until the dura is removed.

Ctrl-z and **Edit -> Undo Last Edit** in tkmedit only allow you to go back one edit. If you remove too many voxels, you can use the clone tool to paint areas back in from the original T1 volume. Load T1 as aux volume. To paint voxels, go to **Tools -> Configure Volume Brush...**, in the tkmedit toolbar.

Set Mode to "Clone", and Clone Source to "Aux Volume". To change the size and shape of your brush, go to:

#### Tools -> Configure Brush Info...

Select a radius and shape of the brush. Click the "Edit Voxels" button in tkmedit toolbar and use the middle button on the mouse to paint in voxels from the auxiliary volume

At any time, you can save the changes you've made to the brainmask.mgz volume by selecting 'Save Volume' in tkmedit's 'File' menu, and clicking 'OK'

You can check your result by viewing the brainmask.mgz volume in the pial\_edits\_after directory.

## **1.2. Regenerating the Surface**

When you are finished editing the voxels, you will need to regenerate the surfaces. Since the white matter hasn't been changed, you don't need to resegment the volume. You can regenerate the pial surface with:

recon-all -autorecon2-pial -autorecon3 -subjid pial\_edits\_before

**Do not run this command if you are conducting the tutorial!** This step will take a long time and there is no need to run it for the tutorial purposes.

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## 1. Fixing a bad skull strip

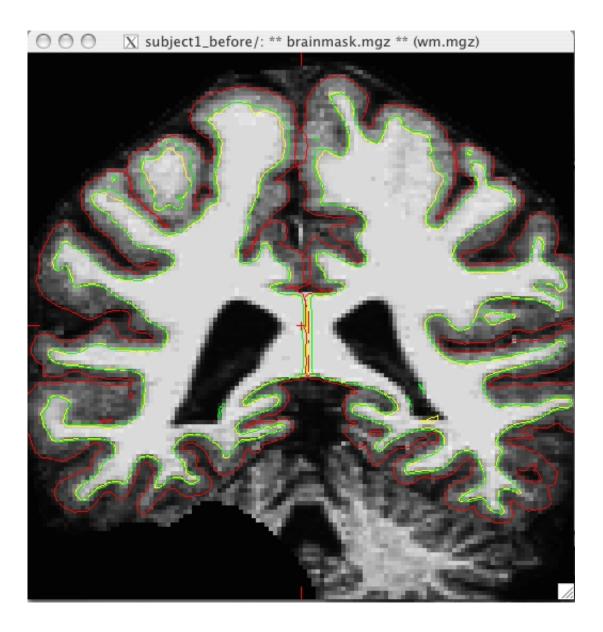
#### •

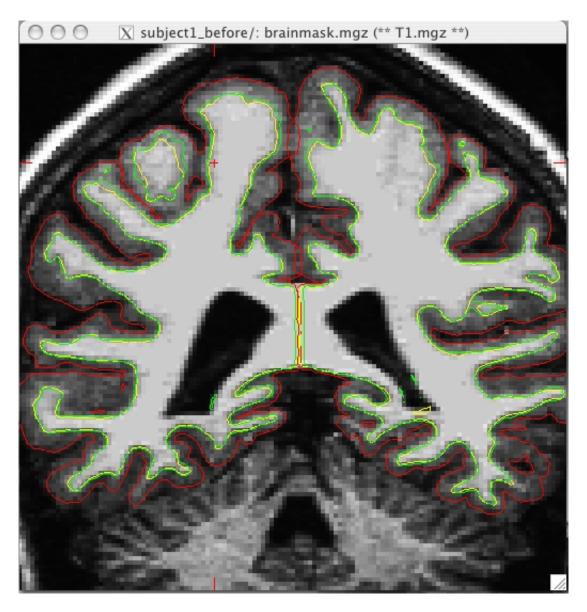
To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

Occasionally, the skull stripping step either removes more than just the skull, causing part of the brain to be removed as well, or too little, leaving behind portions of the skull. Both of these problems need to be corrected before continuing to the next step, either by manually editing the volumes or by adjusting input parameters to the skull stripping step, and running the skull strip again until a good result is obtained. Often the sagittal view reveals skull strip failures. Note that the inflated 2D surface is a less reliable gauge of skull strip failure unless large portions of the brain are missing, or lots of skull is retained.

skullstrip1\_before has a poor skull strip, an entire hemisphere of the cerebellum has been stripped away along with the skull. This page will walk you through the process of fixing this particular subject and also offer suggestions for fixing other common skull stripping problems.

If you look at coronal slice 91 for skullstrip1\_before you can see that the brainmask.mgz volume (the first picture) is missing the right hemisphere of the cerebellum and that it is present in the T1.mgz volume (the second picture):





In general there are two ways to fix a volume when there is something missing from the cortex or cerebellum, you can clone the missing pieces in manually or you can adjust the parameters of mri\_watershed to do it automatically. For this case, because there is such a lot missing on so many slices you should adjust the parameters of mri\_watershed.

## 1.1. Adjusting watershed parameters

The watershed algorithm is used during the skull stripping step to find a boundary between the brain and skull. The mri\_watershed program uses a default preflooding height of 25 percent. If we want the algorithm to be more conservative (i.e. if part of the brain has been removed), you will want to make that number larger than 25. If you want the algorithm to be more aggressive (i.e. part of the skull has been left behind), you will want to make the height less than 25. There aren't any hard and fast rules about how to select your height value. You can adjust the preflooding height by passing the following flag to recon-all:

recon-all -skullstrip -wsthresh <h> -clean-bm -subjid <subject name> where <h> is replaced with the preflooding height you'd like to use and <subject name> is replaced with your subject. The clean-bm flag is used to instruct recon-all to write over the old brainmask.mgz volume with your new edits. If you do not use this flag your changes will not take effect.

#### Part of the brain is missing

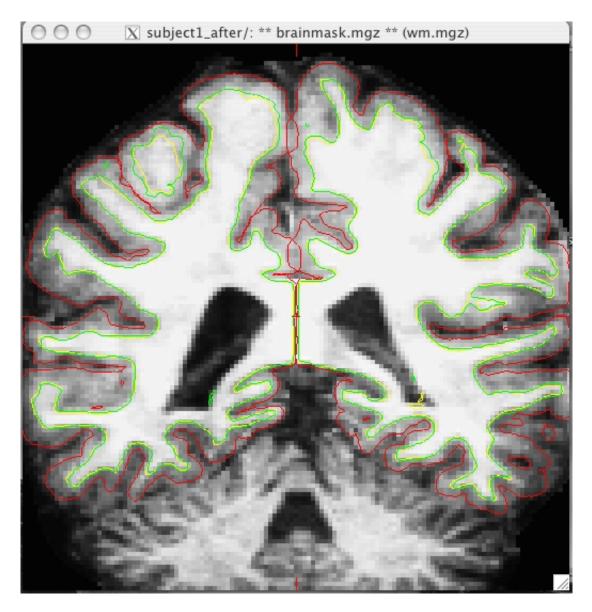
Now we will take another look at the first volume we looked at, where part of the cerebellum had been removed. You can adjust the watershed threshold by passing the -wsthresh flag to recon-all. In this instance, since too much was removed, we want to raise the watershed threshold so use the command:

recon-all -skullstrip -wsthresh 35 -clean-bm -no-wsgcaatlas -subjid skullstrip1\_before

Take a look at your output volume (brainmask.mgz has been changed) along with the original T1 volume (T1.mgz), and verify the result of the new skull stripping is correct.

tkmedit skullstrip1\_before brainmask.mgz -aux T1.mgz -surfs

It should look like this:

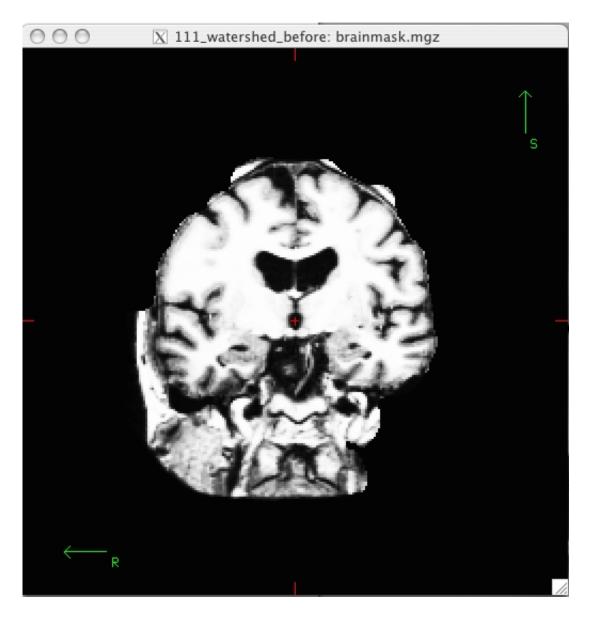


#### Some skull still remains

Sometimes the skull strip will leave pieces of skull in the brainmask volume. Subject skullstrip2\_before is an extreme example of this, it can be the case that there is just a bit of skull left. Open skullstrip2\_before to see what it looks like:

tkmedit skullstrip2\_before brainmask.mgz

and you should see this:



For this example, since there is so much skull remaining, we want to lower the watershed threshold, so you could use the command:

recon-all -skullstrip -wsthresh 5 -clean-bm -no-wsgcaatlas -subjid skullstrip2\_before

Take a look at your output volume (brainmask.mgz has been changed) along with the original T1 volume (T1.mgz), and verify the result of the new skull stripping is correct.

tkmedit skullstrip2\_before brainmask.mgz -aux T1.mgz -surfs

You can compare this to the brain volume in skullstrip2\_after to see that your changes look the same.

#### Reprocessing the data

If you make changes to the brainmask.mgz volume, you can re-start the recon-all process from this point by using the following command:

recon-all -autorecon2 -autorecon3 -subjid <subject name>

#### **Trying Several Thresholds**

If you would like to quickly run several different watershed thresholds simultaneously, you can use the -multistrip flag which will create several orig, T1, and nu volumes stripped with the following preflood heights: 5, 10, 20, 30.

You can change which preflood heights it uses (instead of 5, 10, 20, 30) by first doing:

```
setenv SUBJECTS_DIR /path/to/your/data
setenv WATERSHED_PREFLOOD_HEIGHTS '20 30 40 50'
recon-all -multistrip -clean-bm -s <subjid> -no-isrunning
```

Note that it will launch mri\_watershed for each one for each vol orig, nu and T1, so if you have 12 heights, for example there will be 36 processes.

## 1.2. Using gcut

When the skull stripping has left a small part of dura left, it is quicker to try and rerun the skullstrip step using the -gcut flag than to do manual editing. This flag removes any extra dura that could influence the surfaces.

recon-all -skullstrip -clean-bm -gcut -subjid <subjid>

INFO: Care must be taken to thoroughly inspect your data when using -gcut. In particular, inpsect the edges of gm and cerebellum for over-aggressive cutting. Add -segmentation brainmask.gcuts.mgz to tkmedit to view the voxels which gcut has removed.

## 1.3. Manual editing

When the skull stripping process has left just a few slices with either missing brain regions or too much skull you can edit these manually using tkmedit.

**Part of the brain is missing** Sometimes there will be small regions missing from the pons or cerebellum, or from part of the cortex itself. To fix these you'd need to open the output volume from the skull stripping step (brainmask.mgz) and the original T1 volume (T1.mgz) simultaneously.

For this following example we will use a subject with a missing cerebellum to demonstrate the tools, but in reality this is not an example of something you would want to edit by hand.

Switch back and forth between the two volumes a few times with Ctrl-1 and Ctrl-2, and use the arrow keys to view the different slices. Notice that a large part of the cerebellum has been stripped away along with the skull around slice 91.

In the tkmedit toolbar, go to:

#### **Tools -> Configure Volume Brush...**

Set Mode to "Clone", and Clone Source to "Aux Volume". Click the Close button to close the configuration window. You can also change the size and shape of your brush, to do this go to:

#### Tools -> Configure Brush Info...

Select a radius and shape that you are comfortable using. Close the configuration menu, and click the "Edit Voxels" button in tkmedit toolbar.

Use Ctrl-1 and Ctrl-2 to cycle between the two volumes. Find a place in the image where the cerebellum is missing in the output volume, then use the middle button on the mouse to paint in cerebellum from the auxiliary volume. Cycle back and forth between the volumes frequently so you know where you are.

Continue painting until the slice is no longer missing part of the brain. Repeat for the other slices in the output volume. Go to **File -> Save Main Volume As...** and save your output.

#### Some skull still remains

If there are small pieces of skull still remaining on only a few slices you can manually remove the voxels. To do this make sure that the "Edit Voxels" button is still selected. Removing voxels is very similar to painting in voxels, except you use the right mouse button instead of the middle button. Use Ctrl-1 and Ctrl-2 to cycle between the volumes. Find a place in the image where skull remains. Use the right mouse button to delete the voxels. Continue on the other slices until all skull is removed. Save your volume.

Ctrl-z and **Edit -> Undo Last Edit** in the time of the right mouse button to delete them again. If you remove too many voxels, you can use the right mouse button to delete them again. If you remove too many voxels, you can use the clone tool to paint areas back in from the original T1 volume, similar to painting in voxels as described above.

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# **1. Using Control Points to Fix Intensity Normalization**

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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

Sometimes the intensity normalization step will fail because it cannot determine the proper intensity for white matter. The result is an erroneous white matter segmentation. A control point is a manually selected location in the volume that the user feels sure is inside the white matter boundary, and subsequently should be normalized to an intensity of 110. TkMedit displays the intensity of any voxel your cursor is on as a "value" in the Cursor and Mouse sections of its Tools interface.

#### **Manually Selecting Control Points**

Subject cp\_before is an example of a subject that needs some control points in order to ensure that the voxels are normalized correctly and then included in the wm.mgz volume. This page will walk you through setting control points to fix this type of problem.

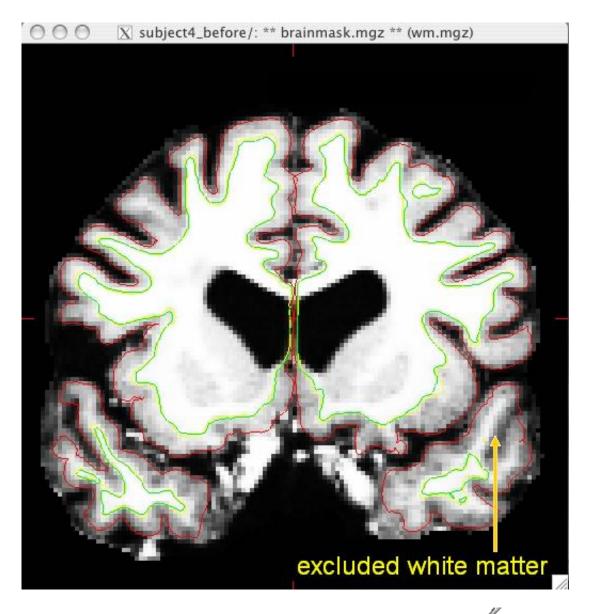
First, make sure you have cp\_before loaded into tkmedit:

tkmedit cp\_before brainmask.mgz -aux T1.mgz -surfs

**Note:** If you are using your own subject data, where surfaces are not yet available (ie. the -autorecon2 stage has not run yet), then don't include the surfaces in the tkmedit command:

tkmedit <my\_subj> brainmask.mgz -aux T1.mgz

Scroll through this subject and find the location where the white matter is being excluded from the surface. In subject cp\_before, this happens around coronal slice 149.



To add control points you will first need to select the Edit Control Points tool  $\checkmark$ . Middle-mouse-button clicking will create a control point; right-button clicking will delete a control point. As you select control points, they will appear as small green crosshairs. Select a few control points around your trouble areas, space them out throughout the brain and on different slices. You want to pick points in a region where the wm intensity is lower than it should be (that is, having a voxel value less than 110).

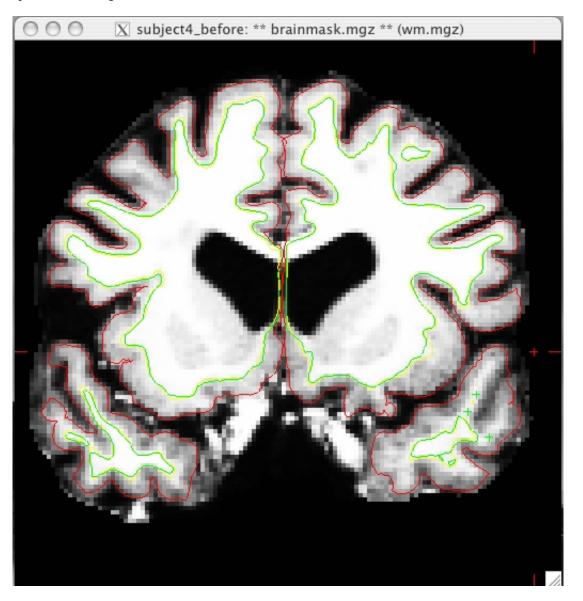
#### General tips for adding control points:

- Control points should only be added in regions that are definitely white matter (i.e., not in the cortex, cerebellum, brainstem, or outside of the skull).
- Control points should also only be added in regions where voxel intensity is not 110. A control point in a region that is already normalized to 110 will be useless.
- ullet

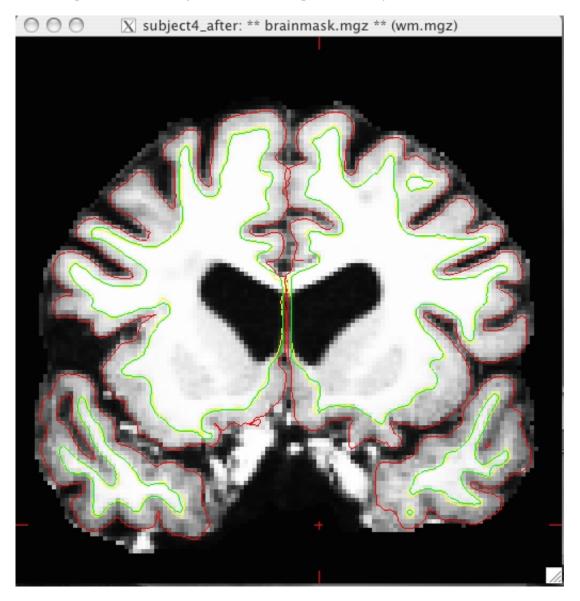
Control points should NOT be used to try and normalize a brain lesion to 110. Such defects should be fixed with white matter edits

- Control points can help recover thin wm strands that are dark by putting some at the base of the strand.
- Control points are also useful in areas of very bright intensity.
- Start off with a few control points spread out in your trouble area. You may need to add more. With experience you will be able to determine how many are appropriate, given your specific subject.

Here is an example of one slice with the control points added. Note that there are other control points spread out through other slices as well.



After adding the control points, go to **File -> Save Control Points**; this will create a file called <subject name>/tmp/control.dat. Using the added control points the subject should now look like this:



Once your control points are saved you can rerun recon-all as follows:

recon-all -autorecon2-cp -autorecon3 -subjid cp\_before

**Do not run this command if you are conducting the tutorial!** This step will take a long time so there is no need to run it for the purposes of this tutorial.

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## **1. Fixing Bad Output From the Talairach Registration**

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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

FreeSurfer computes a linear Talairach transform contained in a 3x4 matrix in a file called talairach.xfm and is located in the <subject name>/mri/transforms directory. Under some circumstances, the alignment can fail the automatic failure detection scheme (-tal-check flag, which can be disabled via -notal-check).

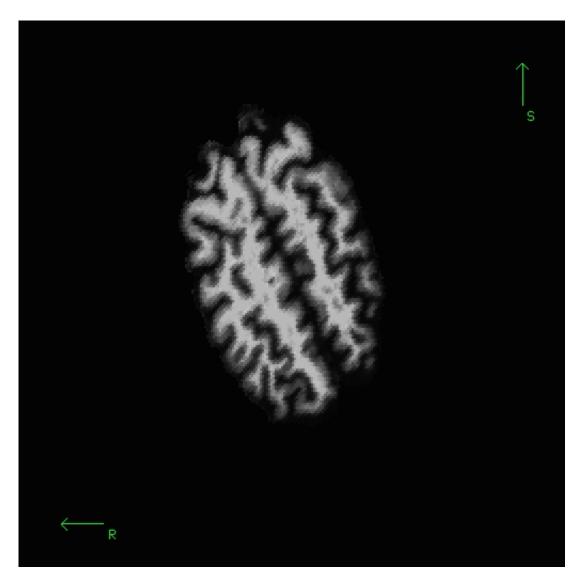
The best way to check the transform is by loading it visually. This can be done directly from tkmedit (File --> Load Transform), however new users sometimes feel more comfortable viewing the transform in tkregister2, where it is possible to view the transform on top of the talairach subject. When viewing in tkmedit you can expect some distortion and stretching from the position of your subject - especially a sagittal tilt, but you want to avoid any changes in orientation (i.e., the coronal view of your subject should be the coronal view of your talairach transform), severe changes in positioning in the window (i.e., your transformation should not show up in the top left corner of the view, while your subject is nicely centered), and rotations or twisting. If any of these are seen, you will need to correct your transform.

Subject tal\_before is an example of a bad talairach. The surface cutting planes are far off center but the problem actually originates from the bad talairach transform. To view this talairach in tkmedit first be sure you have the subject open:

tkmedit tal\_before brainmask.mgz

If the surfaces are also loaded into tkmedit you may find it easier if you toggle them off for this part.

In the tkmedit toolbar, go to **File -> Transforms -> Load Transform for Main Volume...** Click browse, select the file talairach.xfm and click 'OK'. In the coronal view, the transform has resulted in an extremely distorted brain.



To further view and edit this transform we will use tkregister2. To open this you can close the tkmedit window and open tkregister2 with the command:

tkregister2 --mgz --s tal\_before --fstal --surf orig

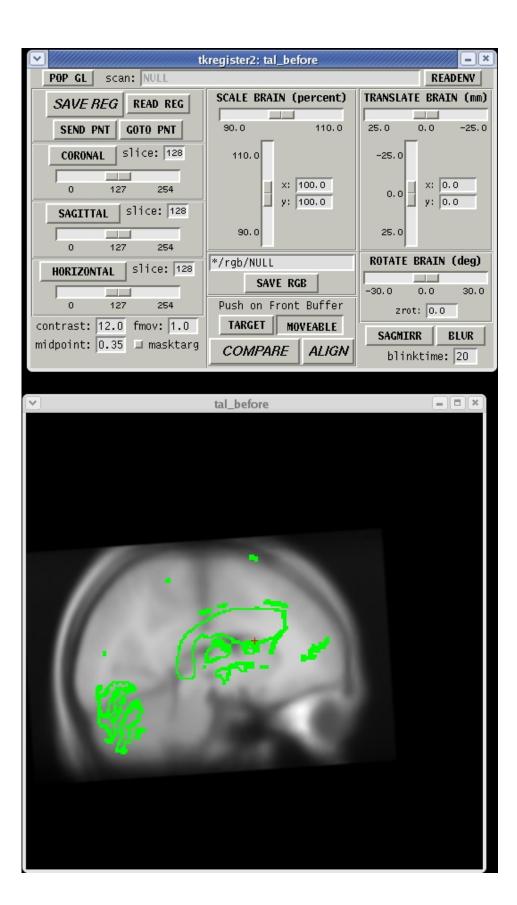
# Note: If your subject surfaces are not yet available, then exclude the --surf origflag from tkregister2.

Brief descriptions on how to operate tkregister2 are given below. More documentation is also available by running "tkregister2 --help".

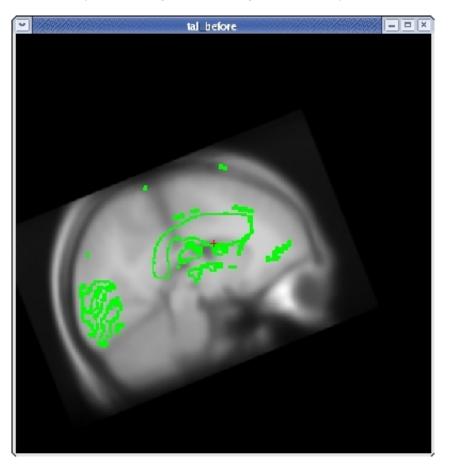
You will see the subject's volume as the TARGET volume and your Talairach volume as a fuzzy MOVEABLE volume. The green lines are the orig surface from the subject. This will be the same in both the TARGET and the MOVEABLE. It can be turned on and off by clicking in the image window and hitting the 's' key.. Find the fmov: box in the tkregister toolbar and make sure it is set to 1.0.

The goal is to stretch, translate, and rotate your MOVEABLE volume so that the two brains look as similar as possible, at least along the key anatomical points (anterior/posterior commissures, the temporal lobes in the coronal plane, and the midline cut).

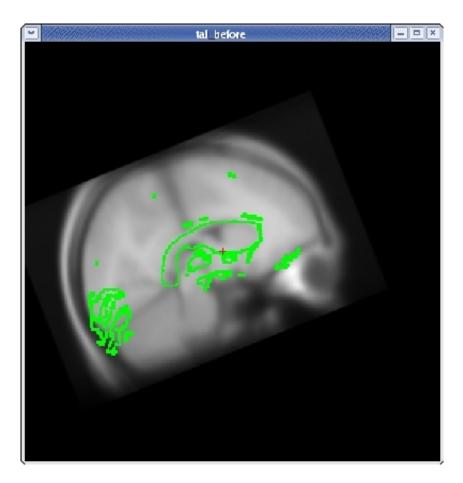
Use Ctrl-1 and Ctrl-2 to switch between the two volumes (or hit the COMPARE button). You will want to do this frequently to check your progress. Click the SAGITTAL button to switch to a sagittal view, and go to slice 128 by using the slider directly below the SAGITTAL button. In this view you have a good view of the corpus callosum in both the moveable and target volumes.



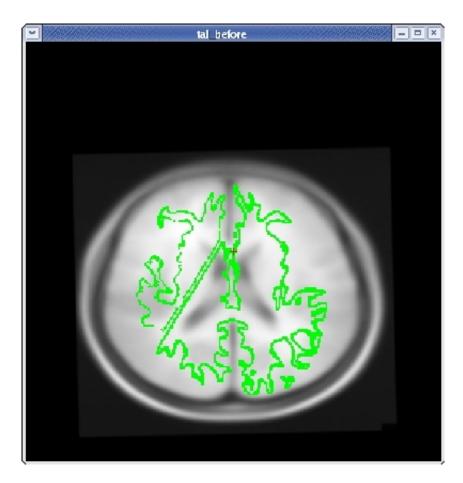
To rotate the moveable volume, use the ROTATE BRAIN slider. You will want to move the slider only a couple of degrees at a time until you achieve the desired effect. You will notice a small red cross icon near the middle of the viewing window. This is the center of rotation. You can change the location of the center of rotation by left-clicking in the viewing window with your mouse.



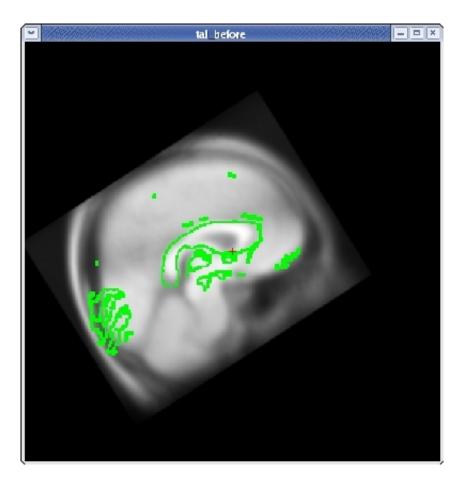
For translation, there are two sliders: one to move the volume left and right, and one to move the volume up and down. Next, translate the moveable brain upwards by using the TRANSLATE BRAIN vertical slider. Again, you only want to move the volume a couple millimeters at a time. You can move the volume left and right in the same way.



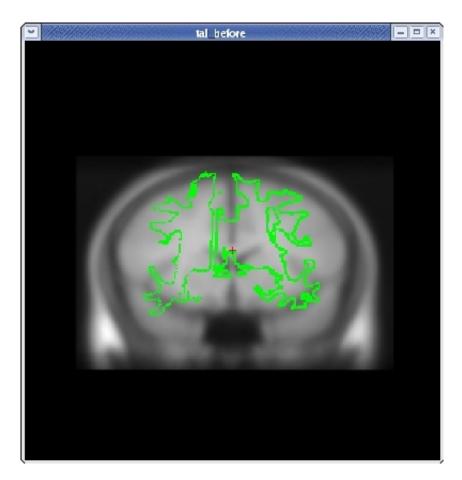
Once you have the corpus callosum aligned as well as possible in the sagittal plane, click the HORIZONTAL button to get a horizontal view, and use the slider directly below the HORIZONTAL button to go to slice 128. Use Ctrl-1 and Ctrl-2 to switch between the two volumes. Use the ROTATE BRAIN and TRANSLATE BRAIN buttons as before to align the midlines of both volumes.



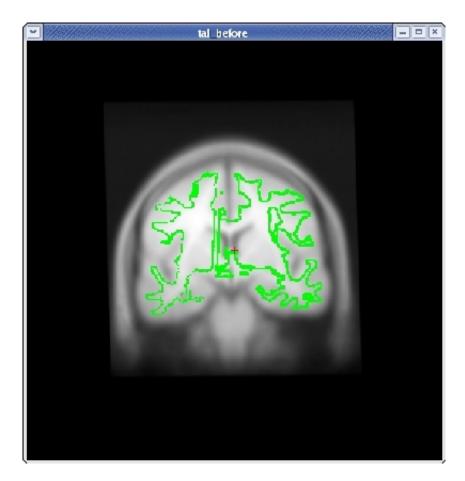
Once you are done aligning the brains in the horizontal view, switch back to slice 128 in the sagittal view. Fine tune your rotation and translation again until the corpus callosum is once again aligned in both volumes.

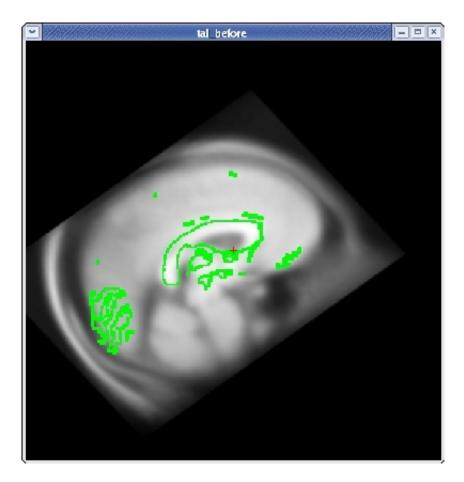


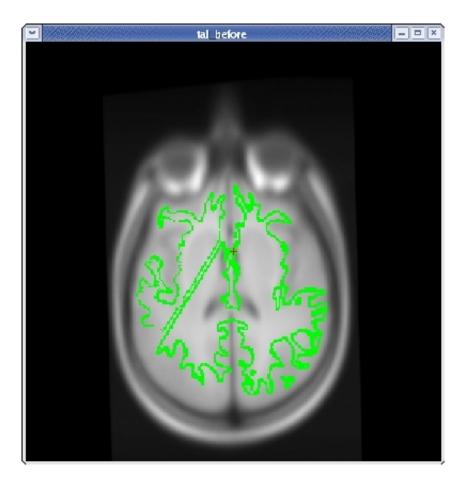
Click the CORONAL button, and go to slice 128. Align the midlines of the brains again in the same way.



Continue this way, switching frequently between the HORIZONTAL, SAGITTAL, and CORONAL views, and align the visible brain structures as much as possible in all of the slices. Use the SCALE BRAIN button as needed to scale the brain in the X and Y direction. Keep in mind that you are working in 3D, not in 2D, so any changes made in one view will affect the other views as well.







When you are satisfied with your registration result, click the SAVE REG button in the tkregister2 toolbar. You can close tkregister2 by clicking on the red close button in the upper left hand corner of the viewing window, or by typing Ctrl-C in in the same terminal window as the new matrix.

Because the talairach transformation affects everything in the stream it is necessary to rerun the whole process using your new talairach, which is a default. You can do this with the command:

recon-all -all -subjid tal\_before

This step will take a long time to run, it is not necessary to run this step for the purposes of this tutorial. You can compare your new talairach to the one in tal\_after.

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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

### 1. Data Conversion

In this exercise, an image from a scanner, in DICOM format, will be converted into mgz format, using recon-all.

You first want to make sure you are working in the appropriate directory and that you have set your SUBJECTS\_DIR variable correctly. You can do this by entering the following directory:

cd \$TUTORIAL\_DATA/buckner\_data/tutorial\_subjs

Set the current directory to be the default subjects directory using this command:

```
setenv SUBJECTS_DIR ${PWD}
```

Two acquisitions of an anonymized volume have been provided for this tutorial with the recognizable face characteristics removed, in the directory buckner\_data/tutorial\_subjs/014-anon and buckner\_data/tutorial\_subjs/015-anon. Using the recon-all script you can specify both the acquisitions to be converted to mgz format as well as the name you wish to give your subject in the following format:

recon-all -all -i <in volume 1> -i <in volume 2> -s <subject name>

where <in volume 1> is the first file that appears in the first acquisition directory, and <in volume 2> is the first file that appears in the second acquisition directory. For this example we will use *anon* as our <subject name>. To convert the two *anon* acquisitions use the command:

recon-all -i 014-anon/001.dcm -i 015-anon/001.dcm -s anon

The output file list can be viewed with the following command:

```
ls ${SUBJECTS_DIR}/anon/mri/orig
```

which will show:

001.mgz 002.mgz

View the output of the data conversion with tkmedit:

tkmedit -f \${SUBJECTS\_DIR}/anon/mri/orig/001.mgz

The full path to the converted volume is specified with the -f option because a file is being specified explicitly. If tkmedit is called without the -f, it will assume that the data is in  $SUBJECTS_DIR/<subject name>/mri, where <subject name> is the first argument on the tkmedit command line (anon in the example below).$ 

You can motion correct your two acquisitions using the -motioncor option of recon-all like this:

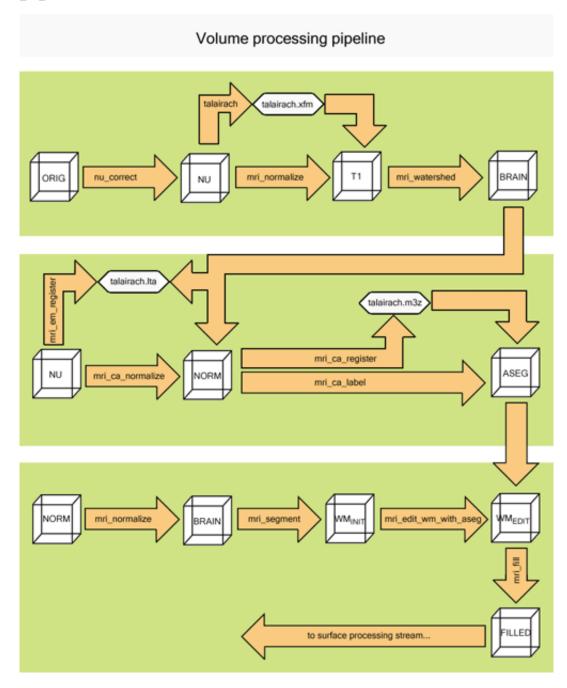
recon-all -motioncor -s anon

Once motion correction has been performed using 001.mgz and 002.mgz, the volume \${SUBJECTS\_DIR}/anon/mri/orig.mgz has been produced, tkmedit will be able to find that volume using the subject directory and the volume name:

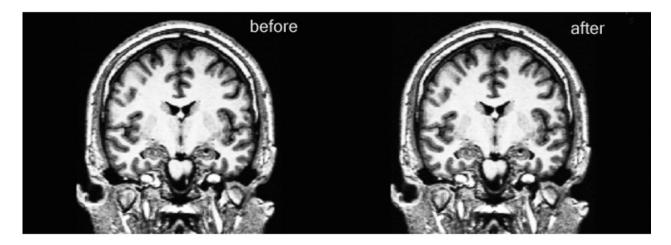
tkmedit anon orig.mgz

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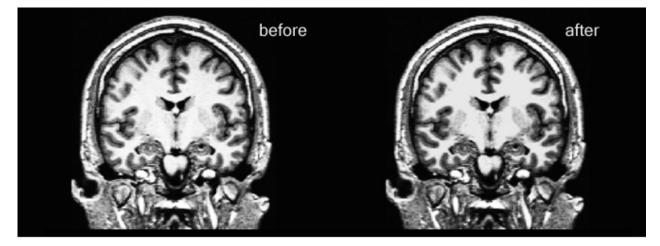
# **1.** Image before and after comparisons (volume processing pipeline)



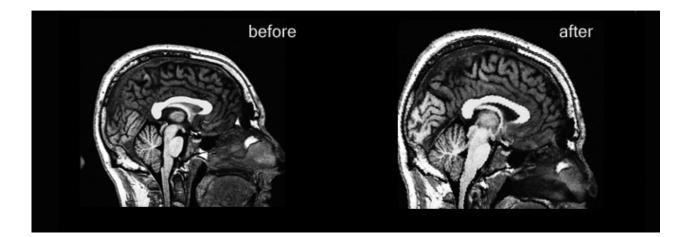
1.1. 1.0 N3 intensity normalization: nu\_correct\_subject <subject name>



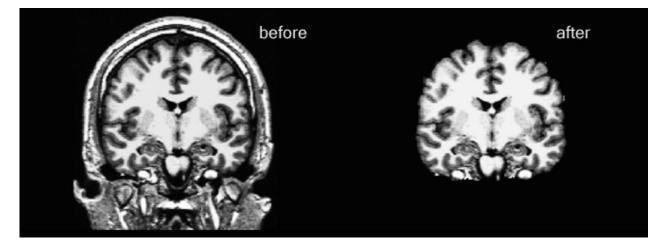
1.2. 2.0 Cortical intensity normalization: mri\_normalize orig T1



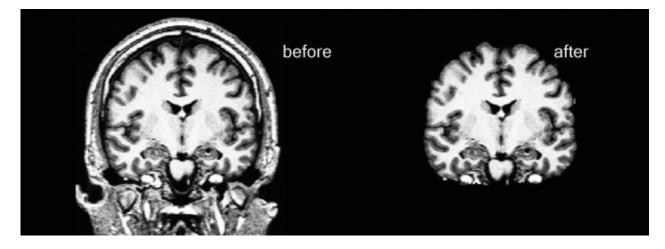
1.3. 3.0 Cortical Talairach: talairach <subject name>



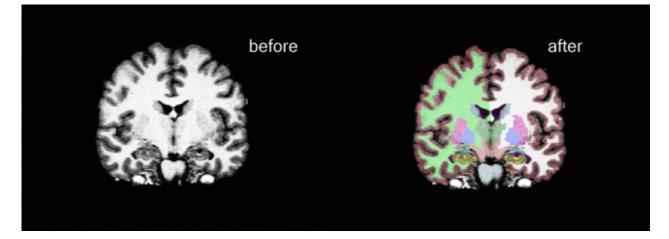
**1.4. 4.0 Cortical skull stripping: mri\_watershed T1 brain** 



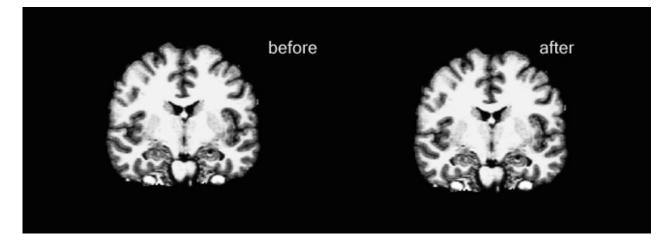
**1.5. 5.0 Subcortical intensity normalization: mri\_ca\_normalize -mask brain nu \$GCA transforms/talairach.lta norm** 



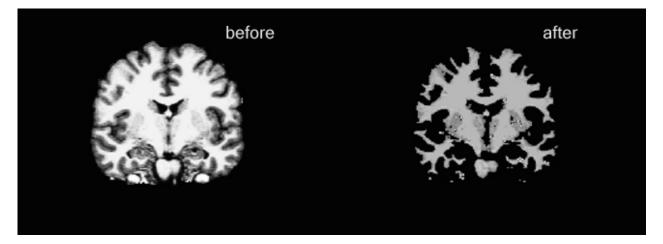
1.6. 6.0 Subcortical segmentation: mri\_ca\_label norm transforms/talairach.m3d \$GCA aseg



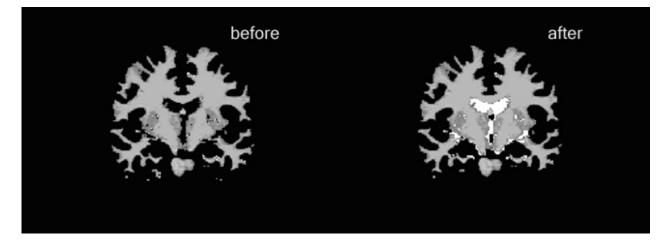
**1.7. 7.0 Subcortical brain intensity normalization: mri\_normalize norm brain** 



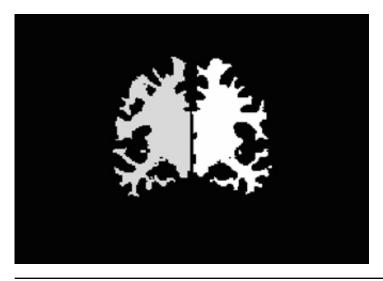
**1.8. 8.0 Subcortical white matter segmentation: mri\_segment brain wm** 



1.9. 9.0 Auto segmentation Editing: mri\_edit\_wm\_with\_aseg wm aseg wm



## 1.10. 10.0 Filling: mri\_fill wm filled



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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

## 1. Visually check the cortical reconstruction output with tkmedit

For each step so far, look at the output generated for the subject 'bert'. As you are performing a cortical reconstruction, it is a good idea to visually examine your data for errors or defects that may have been generated in the reconstruction steps. When using tkmedit, scroll through the volumes with the up and down arrow keys to visually inspect each slice. For a reminder of which inputs and outputs are taken by each step in the reconstruction, refer to this guide First, set your subjects directory environment variable:

```
cd $TUTORIAL_DATA/buckner_data/tutorial_subjs
setenv SUBJECTS_DIR ${PWD}
```

#### **Data conversion**

Before preprocessing is done, data must be converted from native scanner format to mgz format. The output of this step is the bert/mri/orig.mgz. To open it in tkmedit:

tkmedit bert orig.mgz

#### **Intensity normalization**

The intensity normalization step takes the bert/mri/orig volume as input and outputs the bert/mri/T1 volume. Look at these volumes simultaneously to see the difference from the intensity normalization step. Use Ctrl-1 and Ctrl-2 to switch between the volumes.

tkmedit bert T1.mgz -aux orig.mgz

#### **Skull stripping**

The skull stripping step takes the bert/mri/T1 volume as input, strips off the skull, and outputs the bert/mri/brainmask volume. Look at the T1 and brainmask volumes simultaneously with tkmedit. Use Ctrl-1 and Ctrl-2 to switch between the volumes.

tkmedit bert brainmask.mgz -aux T1.mgz

#### Segmentation

The segmentation step takes the /bert/mri/brainmask volume as input, removes the gray matter and CSF, and outputs the /bert/mri/wm volume. Look at the brainmask and wm volumes simultaneously with tkmedit. Use Ctrl-1 and Ctrl-2 to switch between the volumes.

tkmedit bert wm.mgz -aux brainmask.mgz

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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

# **1.** Visually check the cortical reconstruction output with tkmedit and tksurfer

For each step so far, look at the output generated for the subject 'bert'. Each time you perform a cortical reconstruction, it is a good idea to visually examine the final surfaces generated after automatic topology fixing for errors or defects that may have been generated in the reconstruction steps. First, set your subjects directory environment variable:

```
cd $TUTORIAL_DATA/buckner_data/tutorial_subjs
setenv SUBJECTS_DIR ${PWD}
```

#### Generating cutting planes and filling

This step takes the /bert/mri/wm.mgz segmented white matter volume as input, and outputs the bert/mri/filled.mgz volume. To view this volume in tkmedit:

tkmedit bert filled.mgz

#### Tessellate

This step takes the /bert/mri/filled.mgz volume as input, and outputs the surfaces bert/surf/lh.orig and /bert/surf/rh.orig for the left and right hemispheres, respectively. To view these surfaces in tksurfer:

tksurfer bert lh orig tksurfer bert rh orig

#### **Smoothing and Inflation**

The smoothing and inflation step takes the bert/surf/lh.orig and /bert/surf/rh.orig surfaces as input, and outputs the following surfaces in the bert/surf directory: lh.smoothwm, rh.smoothwm, lh.curv, rh.curv, lh.sulc, rh.sulc, lh.inflated, and rh.inflated. To view the smoothed and inflated surfaces in tksurfer:

tksurfer bert lh smoothwm tksurfer bert rh smoothwm tksurfer bert lh inflated tksurfer bert rh inflated

To view the curv files, load the lh inflated surface into tksurfer again. Go to **File** -> **Curvature** -> **Load Curvature...**, click **Browse**, and select the file lh.curv. You will see a red and green pattern overlaid on the inflated surface that represents the curvature of the white matter at that location. Repeat with the right hemisphere inflated and curv files.

#### **Generate final surfaces**

The final surfaces are generated by taking the bert/surf/lh.orig and bert/surf/rh.orig surfaces as inputs, and outputs the following surfaces in the bert/surf directory: lh.white, rh.white, lh.pial, rh.pial, lh.thickness, rh.thickness. As mentioned previously, the white and pial surfaces are particularly important to check for defects before continuing through the surface processing pipeline. To view the surfaces in tksurfer:

tksurfer bert lh white tksurfer bert rh white tksurfer bert lh pial tksurfer bert rh pial

#### Generate spherical surfaces

The spherical surface is generated by taking the bert/surf/lh.inflated and bert/surf/rh.inflated surfaces as inputs and outputting the surfaces bert/surf/lh.sphere and bert/surf/rh.sphere. To view these files in tksurfer:

tksurfer bert lh sphere tksurfer bert rh sphere

To make these spheres more interesting, view the curv files overlaid on the spherical surfaces. Load the lh sphere surface into tksurfer again. Go to **File -> Curvature -> Load Curvature...**, click **Browse**, and select the file lh.curv. You will see a red and green pattern overlaid on the spherical surface that represents the curvature of the white matter at that location. Repeat with the right hemisphere sphere and curv files.

## **FreeSurfer Tutorial: Process Flow**

This table shows the recon-all steps for the current **dev** version of FreeSurfer. See ReconAllTableStableV5.1 to see a process flow for the latest **stable** version of FreeSurfer.

Click here to see this information presented in a block diagram format and here for a process v. files table.

See also the OtherUsefulFlags for other recon-all options.

recon-all step	Individual Flag	Input	Command Line	Output
recon-all -autorecon1 -subjid <subjid></subjid>	-i <invol1> -i <invol2></invol2></invol1>	invol1.dcm	mri_convert invol1.dcm orig/001.mgz	orig/001.mgz
		invol2.dcm	mri_convert invol2.dcm orig/002.mgz	orig/002.mgz
		orig/001.mgz	mri_robust_templatemov 001.mgz 002.mgzaverage 1template rawavg.mgzsatitinittp 1fixtpnoitiscalesubsample 200	rawavg.mgz
	-motioncor	orig/002.mgz		
	-mononeor	rawavg.mgz	mri_convert rawavg.mgz orig.mgzconform	orig.mgz
		orig.mgz	mri_add_xform_to_header -c transforms/talairach.xfm orig.mgz orig.mgz	orig.mgz
	-talairach	orig.mgz	talairach_avii orig.mgzxfm transforms/talairach.auto.xfm	transforms/talairach.auto.xfm
		transforms/talairach.auto.xfm	cp transforms/talairach.auto.xfm transforms/talairach.xfm	transforms/talairach.xfm
		transforms/talairach.xfm	talairach_afd -T 0.005 -xfm transforms/talairach.xfm	transforms/talairach.xfm
			$awk \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \$	transforms/talairach_avi.log
	-nuintensitycor	orig.mgz, talairach.xfm	mri_nu_correct.mnii orig.mgzo nu.mgzn 2uchar transforms/talairach.xfm	nu.mgz
	-normalization	nu.mgz	mri_normalize -g l nu.mgz T1.mgz	T1.mgz
	-skullstrip	nu.mgz	miemregister -skull nu.mgz SFREESURFER_HOME/average/RB_all_withskull_2008-03-26.gca transforms/alairach_with_skull.lta	transforms/talairach_with_skull.lta
		T1.mgz	mri_watershed -T1 -brain_atlas \$FREESURFER_HOME/average/RB_all_withskull_2008-03-26.gca transforms/talairach_with_skull.lta T1.mgz brainmask.auto.mgz	brainmask.auto.mgz
		brainmask.auto.mgz	cp brainmask.auto.mgz brainmask.mgz	brainmask.mgz

	-gcareg	brainmask.mgz nu.mgz	mri_em_register -mask brainmask.mgz nu.mgz \$FREESURFER_HOME/average/RB_all_2008-03-26.gca transforms/talairach.lta	transforms/talairach.lta
	-canorm	brainmask.mgz nu.mgz transforms/talairach.lta	mri_ca_normalize -mask brainmask.mgz nu.mgz \$FREESURFER_HOME/average/RB_all_2008-03-26.gca transforms/talairach.lta norm.mgz	norm.mgz
	-careg	brainmask.mgz transforms/talairach.lta norm.mgz	mri_ca_register -align-after -nobigventricles -mask brainmask.mgz -T transforms/talairach.lta norm.mgz \$FREESURFER_HOME/average/RB_all_2008-03-26.gca transforms/talairach.m3z	transforms/talairach.m3z
				transforms/talairach.m3z.inv.x.mgz
	-careginv	transforms/talairach.m3z	mri_ca_register -invert-and-save transforms/talairach.m3z	transforms/talairach.m3z.inv.y.mgz
				transforms/talairach.m3z.inv.z.mgz
	-rmneck	nu.mgz transforms/talairach.m3z	mri_remove_neck -radius 25 nu.mgz transforms/talairach.m3z \$FREESURFER_HOME/average/RB_all_2008-03-26.gca nu_noneck.mgz	nu_noneck.mgz
	-skull-lta	transforms/talairach.lta nu_noneck.mgz	mri_em_register -skull -t transforms/talairach.lta nu_noneck.mgz SFREESURFER_HOME/average/RB_all_withskull_2008-03-26.gca transforms/talairach_with_skull.lta	transforms/talairach_with_skull.lta
		norm.mgz	mri_ca_label -align -nobigventricles norm.mgz transforms/talairach.m3z SFREESURFER_HOME/average/RB_all_2008-03-26.gca aseg.auto_noCCseg.mgz	aseg.auto_noCCseg.mgz
	-calabel	transforms/talairach.m3z	mri_cc -aseg aseg.auto_noCCseg.mgz -o aseg.auto.mgz <subjid></subjid>	
		aseg.auto_noCCseg.mgz		aseg.auto.mgz
		aseg.auto.mgz brainmask.mgz	cp aseg.auto.mgz aseg.mgz	aseg.mgz
	-normalization2	norm.mgz	mri_normalize -aseg aseg.mgz -mask brainmask.mgz norm.mgz brain.mgz	brain.mgz
	-normanization2	aseg.mgz	htt_homaniz -aseg aseg.ingz -mask oranimask.ingz hom.ingz orani.ingz	orani.ngz
		brain.mgz		
	-maskbfs	brainmask.mgz	mri_mask -T 5 brain.mgz brainmask.mgz brain.finalsurfs.mgz	brain.finalsurfs.mgz
		brain.mgz	mri_segment brain.mgz wm.seg.mgz	wm.seg.mgz
		wm.seg.mgz		
		aseg.mgz	mri_edit_wm_with_aseg wm.seg.mgz brain.mgz aseg.mgz wm.asegedit.mgz	wm.asegedit.mgz
	-segmentation	brain.mgz		
		wm.asegedit.mgz		
recon-all -autorecon2		norm.mgz	mri_pretess wm.asegedit.mgz wm norm.mgz wm.mgz	wm.mgz
-subjid <subjid></subjid>		wm.mgz		filled.mgz
<subjiu></subjiu>	-fill	aseg.auto_noCCseg.mgz	mri_fill -a/scripts/ponscc.cut.log -xform transforms/talairach.lta -segmentation aseg.auto_noCCseg.mgz wm.mgz filled.mgz	laninte la mana aut la m
		transforms/talairach.lta		/scripts/ponscc.cut.log
		filled.mgz	mri_pretess filled.mgz 255 norm.mgz filled-pretess255.mgz	filled-pretess255.mgz
		norm.mgz	htt_precss meaningz 255 hominingz mea-precss255.nigz	med-pretess255.mgz
		filled-pretess255.mgz	mri_tessellate filled-pretess255.mgz 255 lh.orig.nofix	lh.orig.nofix
	-tessellate	filled.mgz	mri_pretess filled.mgz 127 norm.mgz filled-pretess127.mgz	filled-pretess127.mgz
	tessentite	norm.mgz	nn_preess meaningz 127 normingz mea preess 27 mgz	milea protessi 27 milja
		filled-pretess127.mgz	mri_tessellate filled-pretess127.mgz 127 rh.orig.nofix	rh.orig.nofix
		?h.orig.nofix	mris_extract_main_component ?h.orig.nofix ?h.orig.nofix	?h.orig.nofix
			rm -f filled-pretess255.mgz filled-pretess127.mgz	
	-smooth1	?h.orig.nofix	mris_smooth -nw ?h.orig.nofix ?h.smoothwm.nofix	?h.smoothwm.nofix
	-inflate1	?h.smoothwm.nofix	mris_inflate -no-save-sulc ?h.smoothwm.nofix ?h.inflated.nofix	?h.inflated.nofix
	-qsphere	?h.inflated.nofix	mris_sphere -q ?h.inflated.nofix ?h.qsphere.nofix cp ?h.orig.nofix ?h.orig	?h.qsphere.nofix ?h.orig
		?h.orig.nofix ?h.inflated.nofix	cp ?h.inflated.nofix ?h.inflated	?h.inflated
		?h.qsphere.nofix	mris_fix_topology -mgz -sphere qsphere.nofix -ga <subjid> ?h</subjid>	?h.orig
	-fix			
	-fix		mris euler number ?h.orig	?h.orig
	-fix	?h.orig	mris_euler_number ?h.orig mris remove intersection ?h.orig ?h.orig	?h.orig ?h.orig
	-fix		mris_euler_number ?h.orig mris_remove_intersection ?h.orig ?h.orig rm ?h.inflated	?h.orig ?h.orig
	-fix	?h.orig	mris_remove_intersection ?h.orig ?h.orig	-
	-lix -white	?h.orig ?h.orig aseg.mgz, brain.finalsurfs.mgz,	mris_remove_intersection ?h.orig ?h.orig	?h.white
		?h.orig ?h.orig aseg.mgz, brain.finalsurfs.mgz,	mris_remove_intersection ?h.orig ?h.orig rm ?h.inflated	?h.orig ?h.white ?h.curv
		?h.orig ?h.orig aseg.mgz, brain.finalsurfs.mgz, wm.mgz, filled.mgz	mris_remove_intersection ?h.orig ?h.orig rm ?h.inflated	?h.orig ?h.white ?h.curv ?h.area
	-white	?h.orig ?h.orig aseg.mgz, brain.finalsurfs.mgz, wm.mgz, filled.mgz ?h.orig	mris_remove_intersection ?h.orig ?h.orig rm ?h.inflated mris_make_surfaces -whiteonly -noaparc -mgz -T1 brain.finalsurfs <subjid> ?h</subjid>	<ul> <li>?h.orig</li> <li>?h.white</li> <li>?h.curv</li> <li>?h.area</li> <li>?h.cortex.label</li> </ul>
		?h.orig ?h.orig aseg.mgz, brain.finalsurfs.mgz, wm.mgz, filled.mgz	mris_remove_intersection ?h.orig ?h.orig rm ?h.inflated	?h.orig ?h.white ?h.curv ?h.area
	-white	?h.orig ?h.orig aseg.mgz, brain.finalsurfs.mgz, wm.mgz, filled.mgz ?h.orig	mris_remove_intersection ?h.orig ?h.orig rm ?h.inflated mris_make_surfaces -whiteonly -noaparc -mgz -T1 brain.finalsurfs <subjid> ?h</subjid>	<ul> <li>?h.orig</li> <li>?h.white</li> <li>?h.curv</li> <li>?h.area</li> <li>?h.cortex.label</li> <li>?h.smoothwm</li> </ul>
	-white	<ul> <li>?h.orig</li> <li>?h.orig</li> <li>aseg.mgz,</li> <li>brain.finalsurfs.mgz,</li> <li>wm.mgz, filled.mgz</li> <li>?h.orig</li> <li>?h.white</li> <li>?h.smoothwm</li> </ul>	mris_remove_intersection ?h.orig ?h.orig rm ?h.inflated mris_make_surfaces -whiteonly -noaparc -mgz -T1 brain.finalsurfs <subjid> ?h mris_smooth -n 3 -nw ?h.white ?h.smoothwm mris_inflate ?h.smoothwm ?h.inflated</subjid>	<ul> <li>?h.orig</li> <li>?h.white</li> <li>?h.curv</li> <li>?h.area</li> <li>?h.cortex.label</li> <li>?h.smoothwm</li> <li>?h.inflated</li> </ul>
	-white -smooth2	<ul> <li>?h.orig</li> <li>?h.orig</li> <li>aseg.mgz,</li> <li>brain.finalsurfs.mgz,</li> <li>wm.mgz, filled.mgz</li> <li>?h.orig</li> <li>?h.white</li> </ul>	mris_remove_intersection ?h.orig ?h.orig rm ?h.inflated mris_make_surfaces -whiteonly -noaparc -mgz -T1 brain.finalsurfs <subjid> ?h mris_smooth -n 3 -nw ?h.white ?h.smoothwm</subjid>	<ul> <li>?h.orig</li> <li>?h.white</li> <li>?h.curv</li> <li>?h.area</li> <li>?h.cortex.label</li> <li>?h.smoothwm</li> <li>?h.inflated</li> <li>?h.sulc</li> </ul>

	-sphere	?h.inflated,?h.smoothwm	mris_sphere ?h.inflated ?h.sphere	?h.sphere
	-surfreg	?h.sphere	mris_register -curv ?h.sphere	?h.sphere.reg
	-surreg	-	<pre>\$FREESURFER_HOME/average/?h.average.curvature.filled.buckner40.tif ?h.sphere.reg</pre>	maphete.teg
	-jacobian_white	?h.white ?h.sphere.reg	mris_jacobian ?h.white ?h.sphere.reg ?h.jacobian_white	?h.jacobian_white
	-avgcurv	?h.sphere.reg	mrisp_paint -a 5 \$FREESURFER_HOME/average/?h.average.curvature.filled.buckner40.tif#6 ?h.sphere.reg ?h.avg_curv	?h.avg_curv
		aseg.mgz, ?h.cortex.label	mris_ca_label -1/label/lh.cortex.label -aseg mri/aseg.mgz <subjid> ?h ?h.sphere.reg</subjid>	
	-cortparc	?h.sphere.reg	<pre>\$FREESURFER_HOME/average/?h.curvature.buckner40.filled.desikan_killiany.2007-06-20gcs ?h.aparc.annot</pre>	label/?h.aparc.annot
		aseg.mgz, brain.finalsurfs.mgz, wm.mgz, filled.mgz	nei nele sefere chi MONDITI ner Ti bei fertanf autöb %	?h.pial
	-pial		mris_make_surfaces -white NOWRITE -mgz -T1 brain.finalsurfs <subjid> ?h</subjid>	?h.curv.pial, ?h.area.pial
		?h.orig, label/?h.aparc.annot		?h.thickness
		?h.area		
		?h.area.pial	mris_calc -o ?h.area.mid ?h.area add ?h.area.pial	?h.area.mid
	-surfvolume	?h.area.mid	mris_calc -o ?h.area.mid ?h.area.mid div 2	?h.area.mid
		?h.area.mid	mris_calc -o ?h.volume ?h.area.mid mul ?h.thickness	?h.volume
		?h.thickness	https_cale so https// https// https// https//	.ii.volume
	-parcstats	label/?h.aparc.annot	mris_anatomical_stats -mgz -f stats/?h.aparc.stats -b -a label/?h.aparc.annot -c label/aparc.annot.ctab <subjid> ?h</subjid>	stats/?h.aparc.stats label/aparc.annot.ctab
recon-all	-cortparc2	aseg.mgz, ?h.cortex.label	mris_ca_label -1 ./label/lh.cortex.label -aseg aseg.mgz <subjid> ?h ?h.sphere.reg \$FREESURFER HOME/average/?h.destrieux.simple.2009-07-29.gcs</subjid>	label/?h.aparc.a2009s.annot
-autorecon3 -subjid		?h.sphere.reg	label/?h.aparc.a2009s.annot	noor maparene oo yaamor
<subjid></subjid>	-parcstats2	label/?h.aparc.a2009s.annot	mris_anatomical_stats -mgz -f stats/?h.aparc.a2009s.stats -b -a label/?h.aparc.a2009s.annot -c label/aparc.annot.a2009s.ctab <subjid> ?h</subjid>	stats/?h.aparc.a2009s.stats stats/aparc.annot.a2009s.ctab
	-cortribbon	aseg.mgz		
		?h.white	mris_volmasklabel_left_white 2label_left_ribbon 3label_right_white 41 label_right_ribbon 42save_ribbonsave_distance <subjid></subjid>	?h.ribbon.mgz, ribbon.mgz
		?h.pial		
	-segstats	aseg.mgz	mf_segstatsseg mri/aseg.mgzsum stats/seg.statspv mri/norm.mgzempty -excludeid 0 excl-ctxgmwmsupratentsubcortgraytotalgrayin mri/norm.mgzin-intensity-name normin-intensity-units NRetivsurf-vm-volsurf-ctx-volctab \$FREESURFER_HOME/ASegStatsLUT.txtsubject <-subjid>	stats/aseg.stats
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		label/?h.aparc.a2009s.annot	mri_aparc2asegs <subjid>labelwmhypo-as-wmrip-unknownribbono</subjid>	
	-wmparc	aparc+aseg.mgz	mri/wmparc.mgzctxseg aparc+aseg.mgz	wmparc.mgz
		wmparc.mgz	mri_segstatsseg mri/wmparc.mgzsum stats/wmparc.statspv mri/norm.mgzexcludeid 0 brain-vol-from-segbrainmask mri/brainmask.mgzin mri/norm.mgzin-intensity-name normin-intensity-units MR -etivsubject <-subjid>suff-wm-volctab \$FREESURFER_HOME/FreeSurferColorLUT.txt	stats/wmparc.stats
	-balabels	?h.sphere.reg	mri_label2labelsrcsubject fsaveragesrclabel fsaverage/label/?h.BA*.labeltrgsubject <subjid>trglabel ?h.BA*.labelhemi ?hregmethod surface</subjid>	label/?h.BA*.label
	-label-exvivo-ec	?h.sphere.reg	mris_spherical_average -erode 1 -orig white -t 0.4 -o <subjid> label ?h.entorhinal ?h sphere.reg ?h.EC_average ?h.entorhinal_exvivo.label</subjid>	label/?h.entorhinal_exvivo.label

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## **1. Introduction**

Tutorial data setup:

```
setenv SUBJECTS_DIR $TUTORIAL_DATA/buckner_data/tutorial_subjs/group_analysis_tutorial
cd $SUBJECTS_DIR/glm
```

This tutorial is designed to introduce you to the "command-line" group analysis stream in FreeSurfer (as opposed to QDEC which is GUI-driven), including correction for multiple comparisons. While this tutorial shows you how to perform a surface-based thickness study, it is important to realize that most of the concepts learned here apply to any group analysis in FreeSurfer, surface or volume, thickness or fMRI.

Here are some useful Group Analysis Links: QDEC Tutorial FSGD Format FSGD Examples DODS vs DOSS

# 2. This Data Set

The data used for this tutorial is 40 subjects from Randy Buckner's lab. It consists of males and females ages 18 to 93. You can see the demographics here. You will perform an analysis looking for the effect of age on cortical thickness accounting for the effects of gender in the analysis. This is the same data set used in the QDEC Tutorial, and you will get the same result.

# 3. General Linear Model (GLM) DODS Setup

## 3.1. Design Matrix/FSGD File

For this example, we will model the thickness as a straight line. A line has two parameters: intercept (or offset) and a slope.

- 1. The slope is the change of thickness with age
- 2. The intercept/offset is interpreted as the thickness at age=0.

Parameter estimates are also called "regression coefficients".

To account for effects of gender, we will model each sex with its own line, meaning that there will be four linear parameters (also called "betas"):

- 1. Intercept for Females
- 2. Intercept for Males
- 3. Slope for Females
- 4. Slope for Males

In FreeSurfer, this type of design is called DODS (for "Different-Offset, Different-Slope").

In FreeSurfer, you can either create your own design matrices, or, if you can specify your design in terms of a FreeSurfer Group Descriptor File (FSGD), FreeSurfer will create them for you. The FSGD file is a simple text file. See this page for the format. The demographics page also has an example FSGD file for this data.

Things to do:

1. Create an FSGD file for the above design. One (gender\_age.fsgd) already exists so that you can continue with the exercises.

Information on how to create/view text files can be found here.

## **3.2.** Contrasts

A contrast is a vector that embodies the hypothesis we want to test. In this case, we wish to test the change in thickness with age after removing the effects of gender. To do this, create a simple text file with the following numbers:

0 0 0.5 0.5

Notes:

- 1. There is one value for each parameter (so 4 values)
- 2. The intercept/offset values are 0 (nuisance)
- 3. The slope values are 0.5 so as to average the F and M slopes

4. A file called lh-Avg-thickness-age-Cor.mtx already exists

## 4. Analysis

## **4.1.** Assemble the Data (mris\_preproc)

Assembling the data simply means:

- 1. Resampling each subject's data into a common space, and
- 2. Concatenating all the subjects' into a single file
- 3. Spatial smoothing (can be done between 1 and 2)

This can be done in two equivalent ways:

## 4.1.1. Previously Cached (qcached) Data

When you have run recon-all with the -qcache option, recon-all will resample data onto the average subject (fsaverage) and smooth it at various FWHM (full-width/half-max), usually 0, 5, 10, 10, 20, and 25mm. This can speed later processing. The data for this tutorial have been cached, so run:

```
mris_preproc --fsgd gender_age.fsgd \
    --cache-in thickness.fwhm10.fsaverage \
    --target fsaverage --hemi lh \
    --out lh.gender_age.thickness.10.mgh
```

Notes:

- 1. Only takes a few seconds because the data have been cached
- 2. The FSGD file lists all the subjects, helps keep order.
- 3. The independent variable is the thickness smoothed to 10mm FWHM.
- 4. The data are for the left hemisphere
- 5. The output is lh.gender\_age.thickness.10.mgh (which already exists)

#### Things to do:

1. Run mri\_info on lh.gender\_age.thickness.10.mgh to find its dimensions, ie,

mri\_info lh.gender\_age.thickness.10.mgh

## 4.1.2. Uncached Data

In the case that you have not cached the data, you can perform the same operations manually using the two commands below. There is no need to do this for this tutorial. OPTIONAL: THIS WILL TAKE ABOUT 10 MINUTES

```
mris_preproc --fsgd gender_age.fsgd \
    --target fsaverage --hemi lh \
    --meas thickness \
    --out lh.gender_age.thickness.00.mgh
```

Notes:

- 1. This resamples each subjects data to fsaverage
- 2. Output is lh.gender\_age.thickness.00.mgh, which is unsmoothed

### **OPTIONAL: THIS WILL TAKE ABOUT 5 MINUTES**

```
mri_surf2surf --hemi lh \
    --s fsaverage \
    --sval lh.gender_age.thickness.00.mgh \
    --fwhm 10 \
    --cortex \
    --tval lh.gender_age.thickness.10B.mgh
```

- 1. This smooths by 10mm FWHM
- 2. "--cortex" means only smooth areas in cortex (exclude medial wall). This is automatically done with qcache. You can also specify other labels.
- 3. Output is lh.gender\_age.thickness.10B.mgh.

## 4.2. GLM Analysis (mri\_glmfit)

```
mri_glmfit \
    --y lh.gender_age.thickness.10.mgh \
    --fsgd gender_age.fsgd dods\
    --C lh-Avg-thickness-age-Cor.mtx \
    --surf fsaverage lh \
    --cortex \
    --glmdir lh.gender_age.glmdir
```

Notes:

- 1. Input is lh.gender\_age.thickness.10.mgh
- 2. Same FSGD used as with mris\_preproc. Maintains subject order!
- 3. DODS is specified (it is the default)
- 4. Only one contrast is used (lh-Avg-thickness-age-Cor.mtx) but you can specify multiple contrasts.
- 5. "--cortex" specifies that the analysis only be done in cortex (ie, medial wall is zeroed out). Other labels can be used.

- 6. The output directory is lh.gender\_age.glmdir
- 7. Should only take about 1min to run.

Things to do:

When this command is finished you will have an lh.gender\_age.glmdir. There will be a number of output files in this directory, as well as two other directories. If you did an *ls* in your glmdir there will be:

```
y.fsgd -- copy of input FSGD file (text)
Xg.dat -- design matrix (text)
mask.mgh -- binary mask (surface overlay)
beta.mgh -- all parameter estimates (surface overlay)
rstd.mgh -- residual standard deviation (surface overlay)
sar1.mgh -- residual spatial AR1 (surface overlay)
fwhm.dat -- average FWHM of residual (text)
dof.dat -- degrees of freedom (text)
lh-Avg-thickness-age-Cor -- contrast subdirectory
mri_glmfit.log -- log file (text, send this with bug reports)
```

**Study Questions:** 

- 1. What was the DOF for this experiment?
- 2. What was the FWHM?
- 3. How many frames does beta have? Why?

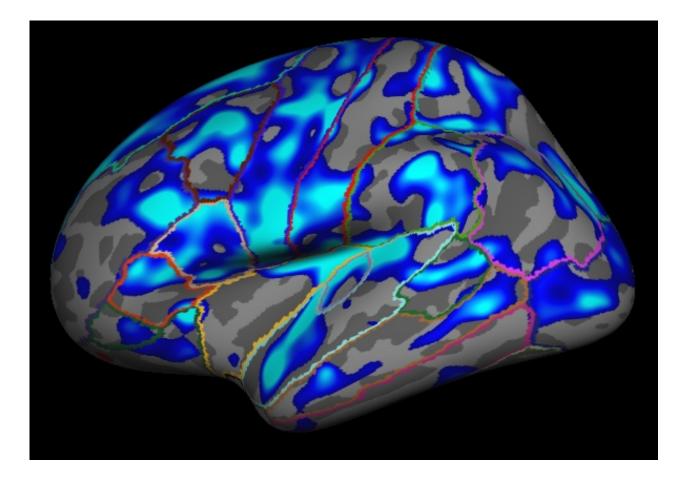
There will be a subdirectory for each contrast that you specify. The name of the directory will be that of the contrast matrix file (without the .mtx extension). The **lh-Avg-thickness-age-Cor** directory will have the following files:

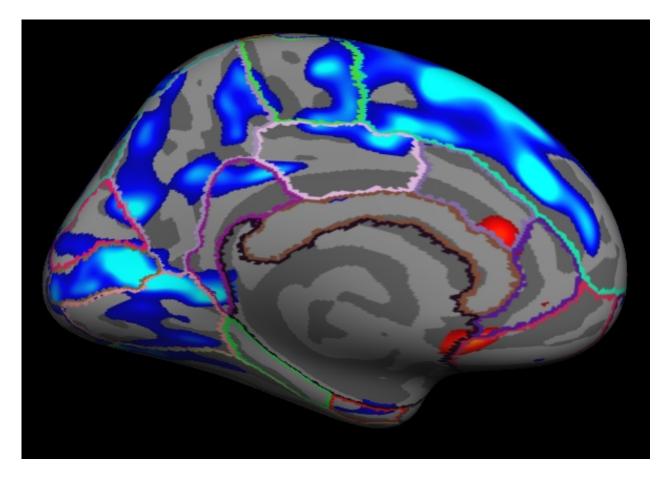
```
C.dat -- original contrast matrix (text)
gamma.mgh -- contrast effect size (surface overlay)
F.mgh -- F ratio of contrast (surface overlay)
sig.mgh -- significance, -log10(pvalue), uncorrected (surface overlay)
```

View the uncorrected significance map with tksurfer:

```
tksurfer fsaverage lh inflated \
    -annot aparc.annot -fthresh 2 \
    -overlay lh.gender_age.glmdir/lh-Avg-thickness-age-Cor/sig.mgh
```

You should see:





Notes:

1.

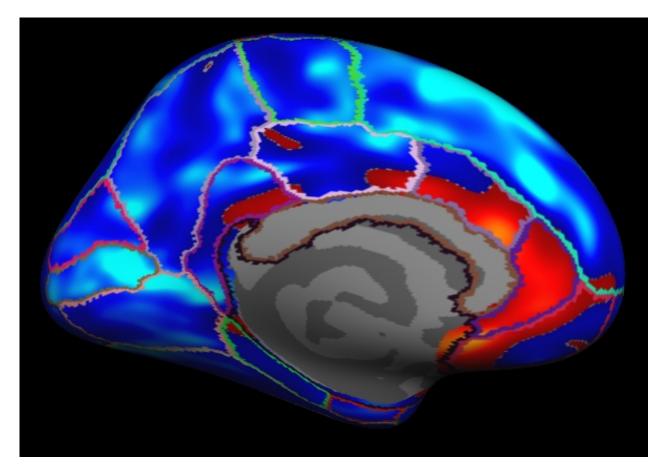
The threshold is set to 2, meaning vertices with p<.01, uncorrected, will have color.

- 2. Blue means a negative correlation (thickness decreases with age), red is positive correlation.
- 3. Click on a point in the Precentral Gyrus. What is it's value? What does it mean?

Viewing the medial surface, change the overlay threshold to something very, very low (say, .01):

```
View->Configure->Overlay, set "Min" .01
```

You should see:



Notes:

- 1. Almost all of cortex now has color
- 2. The non-cortical areas are still blank (0) because they were excluded with --cortex in mri\_glmfit above.

## 4.3. Clusterwise Correction for Multiple Comparisons

To perform a cluster-wise correction for multiple comparisons, we will run a simulation. The simulation is a way to get a measure of the distribution of the maximum cluster size under the null hypothesis. This is done by iterating over the following steps:

- 1. Synthesize a z map
- 2. Smooth z map (using residual FWHM)
- 3. Threshold z map (level and sign)
- 4. Find clusters in thresholded map
- 5. Record area of maximum cluster
- 6.

Repeat over desired number of iterations (usually > 5000)

In FreeSurfer, this information is stored in a simple text file called a CSD (Cluster Simulation Data).

Once we have the distribution of the maximum cluster size, we correct for multiple comparisons by:

- 1. Going back to the original data
- 2. Thresholding using same level and sign
- 3. Finding clusters in thresholded map
- 4. For each cluster, p = probability of seeing a maximum cluster that size or larger during simulation.

## **4.3.1. Run the simulation**

All these steps are performed with the mri\_glmfit-sim:

```
mri_glmfit-sim \
    --glmdir lh.gender_age.glmdir \
    --sim mc-z 5 4 mc-z.negative \
    --sim-sign neg \
    --overwrite
```

Notes:

- 1. Specify the same GLM directory (--glmdir)
- 2. The simulation type is Z Monte Carlo (mc-z)
- 3. Specify the sign (neg for negative, pos, or abs)
- 4.

Vertex-wise threshold of 4 (p < .0001)

5.

Only 5 iterations (usually > 5000)

- 6. CSD files will have mc-z.negative base
- 7. -- overwrite deletes previous CSD files

This has been run this data with 1000 iterations (CSD base of mc-z.neg). This can often take many hours.

## 4.3.2. View CSD File

The CSD files are stored in lh.gender\_age.glmdir/csd. Look at one of them:

less lh.gender\_age.glmdir/csd/mc-z.neg4.j001-lh-Avg-thickness-age-Cor.csd

Or click here.

Notes:

- 1. Rows that begin with '#' are headers or comments.
- 2. Each data row is an iteration.
- 3. The 3rd column is the maximum cluster size for that iteration.
- 4. The CSD file itself is not particularly important, so don't worry too much about it.

## 4.3.3. View the Corrected Results

In the contrast subdirectory, you will see several new files, all beginning with mc-z:

```
mc-z.neg4.sig.cluster.summary - summary of clusters (text)
mc-z.neg4.sig.cluster.mgh - cluster-wise corrected map (overlay)
mc-z.neg4.sig.ocn.annot - annotation of clusters
```

First, look at the cluster summary (or click here):

```
less lh.gender_age.glmdir/lh-Avg-thickness-age-Cor/mc-z.neg4.sig.cluster.summary
```

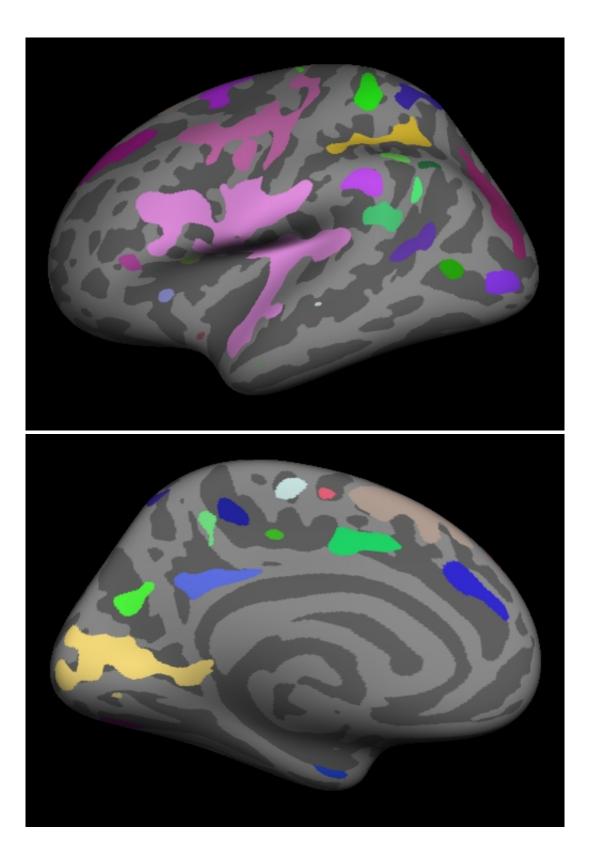
Notes:

- 1. This is a list of all the clusters that were found (38 of them)
- 2. The CWP column is the cluster-wise probability (the number you are interested in). It is a simple p (ie, NOT -log10(p)).
- 3. Note that ALL clusters are found, regardless of their significance.
- 4. Cluster number 1 has a CWP of p=.001.
- 5. Cluster number 21 has a CWP of p=.077.

Load the cluster annotation in tksurfer

```
tksurfer fsaverage lh inflated \
    -annot lh.gender_age.glmdir/lh-Avg-thickness-age-Cor/mc-z.neg4.sig.ocn.annot \
    -fthresh 2 -curv -gray
```

You should see:



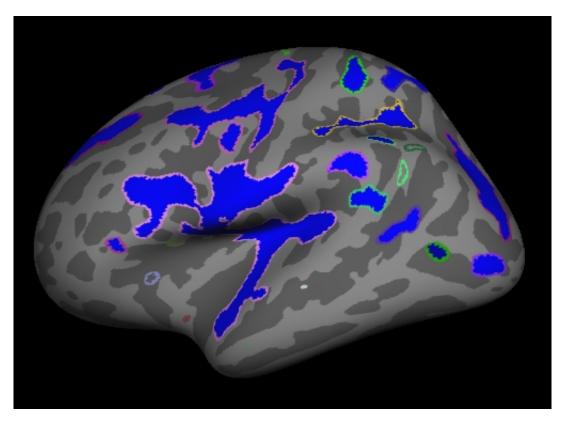
Notes:

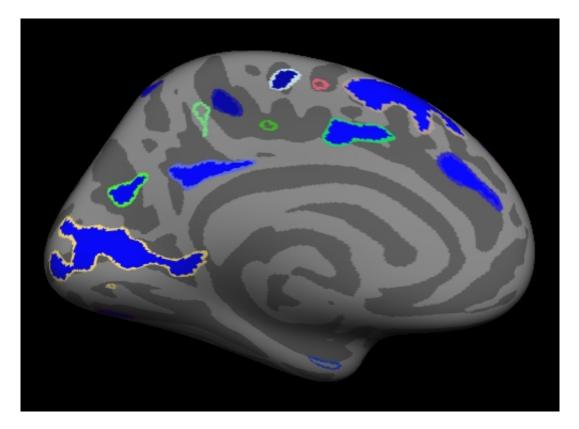
- 1. These are all clusters, regardless of significance.
- 2. When you click on a cluster, the label will tell you the cluster number (eg, cluster-021).

Change the label mode to "Outline" by hitting the outline button

```
File->LoadOverlay
"Browse..."
Select lh.gender_age.glmdir/lh-Avg-thickness-age-Cor/mc-z.neg4.sig.cluster.mgh
OK
OK
```

You should see:





Note:

Things to do:

- 1. Find and click on cluster 1. It is blue and has a value of -3 since this is log10(.001). The -3 is because the correlation is negative.
- 2. Find and click on cluster 21. Its value is -1.11 because this is log10(.077). Note that it is transparent because its significance

is worse than the threshold we set (-fthresh 2, p < .01).

- 3. All vertices within a cluster are the same value (the p-value of the cluster).
- 4. You can changed the cluster-wise threshold by clicking on

View->Configure->Overlay, and setting the "Min" to your desired level. As you do this, some clusters will change transparency.

This page describes the FreeSurfer Group Descriptor (FSGD) File Format (Version 1) as of 11/12/02. FSGD is a way to describe a group of subjects and their accompanying data (like a spreadsheet). This can include class membership and other continuous varibles (eg, age). The nice thing about the FSGD is that various FreeSurfer programs can automatically compute the design matrix from an FSGD file. The FSGDF is used by mri\_glmfit, mris\_preproc, tksurfer, tkmedit. FSGD files are created automatically by qdec, but qdec is currently (12/9/08) limited to two continuous variables and two discrete variables with two levles. The FSGD format uses tags to identify information.

In an FSGD, a **Class** is generally synonymous with a Group. A Class/Group is a specific level of a specific discrete Factor. Eg, if you have Gender as a Factor, then the Factor will have two levels (Male and Female). If the only Discrete Factor in your design is Gender, then there will only be two groups (Males and Females). If you have two Discrete Factors (eg, Gender and Handedness), then you will have four classes/groups (MaleRight, MaleLeft, FemaleRight, and FemaleLeft).

Continuous Factors are referred to as **Variables** in the FSGD. These include things like Age, IQ, Weight, etc.

There are numerous FSGD examples worked out here.

Example of a legal file:

```
GroupDescriptorFile 1

Title MyTitle

Class Class1 plus blue

CLASS Class2 circle green

SomeTag

Variables Age Weight IQ

Input subjid1 Class1 10 100 1000

Input subjid2 Class2 20 200 2000

#Input subjid3 Class2 20 200 2000

DefaultVariable Age
```

Notes:

- \* The first line must be "GroupDescriptorFile 1".
- \* Title is not necessary. This will be used for display.
- \* CLASS only needs the class name, the next two items (eg, "plus blue)" will be used in the display.
- \* The third input is ignored because of the hash (#).
- \* DefaultVariable is the default variable for display.
- \* SomeTag is not a valid tag, so it will be ignored.

General Rules:

\* Each subject is represented by an Input tag.

\* The order of the subjects is not important but you must make sure it is consistent with the input to mri\_glmfit (--y).

\* Each subject must have values for all variables (no empty cells)

- \* Tags are NOT case sensitive.
- \* Labels are case sensitive.
- \* When multiple items appear on a line, they can be separated by any white space (ie, blank or tab).

\* Any line where # appears as the first non-white space character is ignored (ie, it is treated as a comment).

- \* The Variables line should appear before the first Input line.
- \* All Class lines should appear before the first Input line.
- \* Variable label replications are not allowed.
- \* Class label replications are not allowed.
- \* If a class label is not used, a warning is printed out.
- \* The DefaultVariable must be a member of the Variable list.
- \* No error is generated if a tag does not match an expected keyword.
- \* Empty lines are OK.
- \* A class label can optionally be followed by a class marker.
- \* A class marker can optionally be followed by a class color.

These are examples of FreeSurfer Group Descriptor (FSGD) files and contrasts for testing various hypotheses. FreeSurfer can **automatically** construct design matrices from an FSGD file. The FSGD file is usually easier to construct than the design matrix. The FSGD file is used as input to several FreeSurfer programs (mainly mri\_glmfit and mris\_preproc). Note that you can always specify you own matrix using the --X option to mri\_glmfit.

These examples are not meant to be exhaustive, nor are they meant to teach you statistics. See these slides for more of the statistics background on group analysis and how it applies to FreeSurfer.

An alternative to constructing your own FSGD files is to use QDEC, though QDEC cannot be used with more than four groups and/or more than two continuous variables. In the examples below, the term "QDEC-able" appears after those examples that can be analyzed in QDEC.

Below, DODS refers to "Different Offset, Different Slope" and DOSS refers to "Different Offset, Same Slope". Refer to the DODS vs DOSS page for more information.

•

One Group, Zero Covariates (OSGM = One-Sample Group Mean). DODS or DOSS. QDEC-able.

One Group, One Covariate. DODS or DOSS. QDEC-able.

•

Two Groups (1 Factor, Two Levels), Zero Covariates. DODS or DOSS. QDEC-able.

•

Two Groups (1 Factor, Two Levels), One Covariate. DODS only. QDEC-able.

•

Two Groups (1 Factor, Two Levels), Two Covariates. DODS only. QDEC-able.

•

Three Groups (1 Factor, Three Levels), Zero Covariates. DODS or DOSS.

•

Four Groups (2 Factors, Two Levels), Zero Covariates. DODS or DOSS. QDEC-able.

•

Four Groups (2 Factors, Two Levels), One Covariate. DODS only. QDEC-able.

•

Six Groups (2 Factors, Three Levels), Zero Covariates. DODS or DOSS.

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- 1. Introduction
- 2. DODS
- 3. DOSS
- 4. DODS or DOSS: which one should I use?
- 5. SODS?
- 6. I want more flexibility than DODS and DOSS!

# **1. Introduction**

This page describes the differences and similarities between DODS and DOSS. DODS stands for "Different Offset, Different Slope". DOSS stands for "Different Offset, Same Slope". These refer to methods for **automatically** creating a design matrix from an FSGD file and is used by mri\_glmfit and qdec. The terms DODS and DOSS are unique to FreeSurfer (ie, you won't find them in a stats textbook:). For examples of FSGD files see here.

There are only differences between the two in cases where there are more than one group (or class) and at least one continuous covariate (variable).

Consider the simplest case, in which there are two groups (eg, Male and Female) and one continuous covariate (eg, Age), and we are interested in how cortical thickness relates to these variables using a linear model.

A linear model implies two parameters: an offset and a slope. The offset is also called an intercept. The offset/intercept is the thickness at a covariate of 0 (eg, birth) and is measured in the same units as thickness (eg, mm). The slope is the rate of change of the thickness and is measured in mm/years.

# 2. DODS

Generally speaking, we would probably use a separate linear model for each group, meaning that each group would have its own offset and its own slope (thus DODS) for a total of four parameters. The model here would imply that Males and Females could start at different thicknesses and the thicknesses could grow/shrink at different rates. The design matrix would have four columns (one for each parameter). As always, the design matrix has a row for each subject. In the automatic construction, the columns (regressors) would have the following format and interpretation:

•

Regressor 1: has a value of 1 if the subject (row) is a Male, 0 otherwise. This regressor codes for membership in the Male group.

•

Regressor 2: has a value of 1 if the subject (row) is a Female, 0 otherwise. This regressor codes for membership in the Female group.

•

Regressor 3: has a value of the subject's age if the subject is a Male, 0 otherwise. This regressor codes for the slope of the Males' thickness vs age.

•

Regressor 4: has a value of the subject's age if the subject is a Female, 0 otherwise. This regressor codes for the slope of the Females' thickness vs age.

Generally speaking, the number of DODS regressors will be:

```
NregressorsDODS = Nclasses*(Nvariables+1) = 2^{*}(1+1) = 4
```

The order will be intercept, slope for variable 1, slope for variable 2, etc.

# 3. DOSS

In the DOSS model, we still allow for the different groups to start at different places (different offset), but we constrain them to evolve at the same rate (same slope). In a scatter plot of the data and best fit lines, the two lines would be constrained to be parallel. The assumption is that the rate (slope) is the same across groups.

The design matrix would have three columns. As always, the design matrix has a row for each subject. In the automatic construction, the columns (regressors) would have the following format and interpretation:

•

Regressor 1: has a value of 1 if the subject (row) is a Male, 0 otherwise. This regressor codes for membership in the Male group.

•

Regressor 2: has a value of 1 if the subject (row) is a Female, 0 otherwise. This regressor codes for membership in the Female group.

٠

Regressor 3: has a value of the subject's age regardless of the group. This regressor codes for the slope of the thickness vs age.

Generally speaking, the number of DODS regressors will be:

NregressorsDOSS = Nclasses + Nvariables = 2+1 = 3

The order will be the Nclasses intercepts followed by the Nvariables slopes.

# 4. DODS or DOSS: which one should I use?

The choice of DODS and DOSS usually comes down to two things:

• Truth

Power

"Truth" refers to how you believe the data actually behave. If you truly believe that the groups evolve at different rates, then your model should reflect this, and thus you should use DODS.

"Power" refers to the fact that DOSS has fewer regressors than DODS and so will have a larger degree-of-freedom (DOF). The DOF is the number of subjects minus the number of regressors. In the above example, the DODS only has one fewer DOF than DOSS, which probably won't be that important if you have more than 20 subjects. On the other hand, if you have 4 groups and 3 variables, then DODS will have 16 regressors whereas DOSS will have only 7, meaning that DODS will have 8 fewer DOFs than DOSS, which could be important.

You can, of course, drive yourself crazy deciding how to model your data. I refer you to the words of the famous statistician George Box: "All models are wrong, but some are useful". Make sure your models are useful.

# 5. SODS?

We could have taken the extra step of automatically generating models that have the same offset but allow for different slopes. But we didn't.

# 6. I want more flexibility than DODS and DOSS!

To be sure, there are a lot more ways to model your data than DODS and DOSS, however, it becomes increasingly difficult to **automatically** generate design matrices. The FreeSurfer tools (namely mri\_glmfit) allows you as much flexibility as you want in modeling, but you have to construct the design matrix yourself and pass it to mri\_glmfit with the --X flag.

Author: Doug Greve

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# **1. FreeSurfer Tutorial: Surface Group Analysis with Qdec**

•

If you are taking one of the formally organized courses, the tutorial data is already installed on the computer provided to you. If not, then to follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names. If you are using the tutorial data please set the environmental variable TUTORIAL\_DATA to the location to which you have downloaded the data (on the course laptops, it happens to be in /media/disk).

In this tutorial, you will learn how to perform statistical analysis of group surface-based data, including:

•

Preprocessing the group data

•

Constructing a qdec.table.dat file of subject demographics

•

Using Qdec to design and execute your analysis

- Interacting with the Qdec display
- Creating Regions of Interest (ROIs) for further analysis and a final check of your data

Assuming that all 'recon-all -all' processing has been completed for all subjects in the study, FreeSurfer's Qdec application can be used to perform inter-subject/group averaging and inference on the cortical surface. Qdec permits statistical inferences to be made about effects of interest in relation to error variance. The mri\_glmfit command is used to model the data as a linear combination of effects related to variables of interest, confounds and errors. Qdec also allows for certain permutation testing and other means for correcting for multiple comparisons. For group analysis, this technique fits a general linear model (GLM) at each surface vertex to explain the data from all subjects in the study. In this section, a brief overview of linear modeling is presented and can be skipped if this material is already familiar. Other software packages have similar types of programs (e.g., FSL's GFEAT). Note that Qdec wraps the mri\_glmfit utility, which has more extensive GLM capabilities than QDEC, but is a command-line-only tool. It has its own tutorial.

## **1.1. Introduction**

## Qdec

Qdec is a single-binary application included in the FreeSurfer distribution. QDEC is an acronym for Query, Design, Estimate, Contrast. It is intended to aid researchers in performing inter-subject / group averaging and inference on the morphometry data (cortical surface and volume) produced by the FreeSurfer processing stream. Qdec is a GUI front-end to a 'statistics engine' (the mri\_glmfit binary, included in FreeSurfer, currently fills this role) intended to:

- 1. select the subjects meeting the criteria under study
- 2. generate the necessary input to the stats engine, which, for mri\_glmfit, includes: 1.

a **Design** matrix (called **X** in the GLM equation) containing the explanatory variables, 2.

a parameter Estimate matrix (called A in the GLM equation), and

3.

the Contrast vector(s)

3. generate and optionally display the output data and/or images

### Linear Modeling overview

Linear modeling describes the observed data as a linear combination of explanatory factors plus noise, and determines how well that description explains the data being analyzed. In order to understand how to perform group analysis in FreeSurfer, you need to understand the general linear model (GLM) and how to construct a GLM in matrix notation. You can click here for a review of this material. The notation we use here is:**y=X\*beta**, where **y** is the vector observed data (e.g., thicknesses for each subject at a vertex), **X** is the known design matrix (e.g., gender, age), and **beta** is the vector of unknown parameter estimates (PEs). The interpreted as a slope indicating the change of thickness with age. The analysis/estimation is then the process of computing **beta** given the data **y** and the design matrix **X**. A Null Hypothesis (H0) is constructed with a contrast matrix **C**. Inferences are drawn by testing whether the value **gamma=Cb** is zero.

## **1.2. Preprocess Group Data**

### set SUBJECTS\_DIR

If you are using the tutorial data you will need to set your SUBJECTS\_DIR to the directory with the group analysis data in it:

setenv SUBJECTS\_DIR \$TUTORIAL\_DATA/buckner\_data/tutorial\_subjs/group\_analysis\_tutorial
cd \$SUBJECTS\_DIR

\* this will set your SUBJECTS\_DIR to the location where your tutorial data is located (the directory containing the group analysis subjects, assuming you have defined the variable TUTORIAL\_DATA as indicated at the top of this tutorial). If you are not using the tutorial data you should set your SUBJECTS\_DIR to the directory in which the subjects you will use for this tutorial are located. For the organized course, TUTORIAL\_DATA is set for you.

#### recon-all

Prior to using the qdec application, your group subject data must be processed by the standard FreeSurfer processing stream, via the recon-all script. A freesurfer tutorial is available. This processing stream supplies the surfaces and morphometry data on each subject. **The data in the tutorial set has been processed for you.** 

#### pre-smoothed fsaverage surfaces

Qdec needs each subject to have pre-computed smoothed data for the target surface (fsaverage is the default) for each measure (thickness, sulc, area, curv, etc.). Your SUBJECTS\_DIR should contain either a link or a copy of the 'fsaverage' subject found in your \$FREESURFER\_HOME/subjects directory. Presmoothing the data onto the target surface is not part of the normal recon processing stream, but you can easily create this data with recon-all, using the command:

recon-all -s <subjid> -qcache

For the purposes of this tutorial, the -qcache command has been run for all of the subjects. The -qcache flag will run numerous back-to-back mris\_preproc processes on your machine, so be prepared for it to run for about an hour. The help text of recon-all -help contains a section on other -qcache otpions.

#### qdec.table.dat

The primary input to Qdec is a text file, named qdec.table.dat, containing the subject IDs, and discrete and continuous factors, in table format. This is essentially a table of demographics for your subject including all the variables and factors that you wish to consider. Here is an example file:

```
fsidgenderagediagnosisLeft-Cerebral-White-Matter011121_vc8048Female70Demented202291021121_62313-2Female71Demented210188010607_vc7017Female73Nondemented170653021121_vc10557Male75Demented142029020718_62545Male76Demented186087020322_vc8817Male77Nondemented149810
```

For each discrete factor, there should exist a file named <factor>.levels which lists all possible levels. For example, accompanying the example qdec.table.dat file must be a file named 'gender.levels' containing these lines:

Female Male

and there must be a file named 'diagnosis.levels' containing these lines:

#### Demented Nondemented

Of course you may have different discrete factor names and levels (or even no discrete factors, in which case all column data are assumed to be continous factors). Continuous factors do not need a <factor>.levels file to define them.

For organizational purposes it is best to make a directory called qdec within your \$SUBJECTS\_DIR. You can save the qdec.table.dat and <factor>.levels files in there. When Qdec runs it will also save your analyses to this directory. A qdec subdirectory, with a qdec.table.dat has been made for you. Here is a sample of what that looks like:

fsid	gender	age	Left-Hippocampus	Right-Hippocampus
140	Female	18	4214	4190
049	Male	19	4543	4153
141	Female	20	3896	3741
084	Male	21	4804	4722
021	Male	22	4021	3969
093	Female	22	3603	3597

This file contains 40 subject IDs, their gender, age, left hippocampus volume, and right hippocampus volume. To accompany this file you will need to create a <factor>.levels text file to define the discrete factor names. You can do this using the text editor of your choice (emacs, vi, pico...). For this tutorial, the qdec.table.dat file already exists for you.

Optionally, your qdec.table.dat file can explicitly specify the SUBJECTS\_DIR, by including it in the first non-commented line:

```
# This is a comment-line in the qdec.table.dat file.
# This explicitly specifies the SUBJECTS_DIR:
SUBJECTS_DIR /my/path/to/subject/data
```

•

•

#### fsaverage

For display purposes, you will need to have an average subject included in your SUBJECTS\_DIR. !Freesurfer's fsaverage, made in MNI305 space, will do fine:

```
cd $SUBJECTS_DIR
if (! -e fsaverage) ln -s $FREESURFER_HOME/subjects/fsaverage
```

This will add a copy of fsaverage into your SUBJECTS\_DIR. For the purposes of this tutorial, fsaveragehas already been linked to your \$SUBJECTS\_DIR

If you wish to make your own average subject from your set you can do so using make\_average\_subject.

# **1.3. Usage**

To start qdec, from your \$SUBJECTS\_DIR, simply type qdec:

setenv SUBJECTS\_DIR \$TUTORIAL\_DATA/buckner\_data/tutorial\_subjs/group\_analysis\_tutorial
cd \$SUBJECTS\_DIR

qdec &

It may take a few seconds for Qdec to open. The ampersand directs the terminal to run this process in the background, so you may see your command prompt return before Qdec opens.

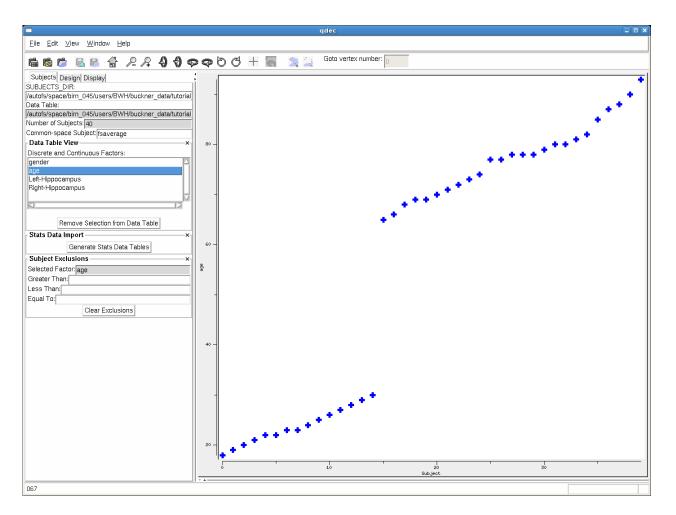
### 1.3.1. Subjects

When Qdec opens you are looking at the Subjects tab. The first thing you will need to do is to load

your qdec.table.dat file. Click **File -> Load Data Table**, or you can use the button, and traverse to your subjects directory and select the qdec.table.dat file. When you click Open, it should load the file, the contents scrolling-by in the terminal window. If the data is loaded correctly, you should see in the terminal window a summary, like this example:

```
gender discrete 2
1
   1 Female
   2 Male
 age continuous 0
2
3 Left-Hippocampus continuous 0
4 Right-Hippocampus continuous 0
               Continuous Factors:
                                         Mean:
                                                     StdDev:
               _____
                                         ____
                                                     _____
                                        57.175
                                                     26.940
                             age
                 Left-Hippocampus
                                      3645.850
                                                     579.214
                 Right-Hippocampus
                                      3725.325
                                                     555.961
Number of subjects:
                    40
                   4 (1 discrete, 3 continuous)
Number of factors:
Number of classes:
                    2
Number of regressors: 8
```

The factors (gender, age, Left-Hippocampus, and Right-Hippocampus) should appear in a list under 'Data Table View' on the control panel. If you choose a factor from this list a scatter plot of your data will appear in the window. Choose 'age', and the display should like this:



The x-axis has the subject number (taken from the order the subjects are listed in the qdec.table.dat), and the y-axis has the value of the variable you've selected. In the example shown you can see a plot of the ages of all 40 subjects. You can use this to visually check your data for outliers. In Qdec if you roll your cursor over one of the points on the plot you can find out which subject it is, the ID will be shown in the lower left corner of the Qdec interface.

# 1.3.2. Stats Data Import

It is possible to import the aseg and aparc data from the FreeSurfer-processed group data. To do so, press the 'Generate Stats Data Tables' button. This will run the FreeSurfer utilities asegstats2table and aparcstats2table on your group data. Upon completion (you should see the progress in the terminal screen), then the 'Generate Stats Data Tables' button becomes a menu-selection list. Within that menu, you can select 'aseg.volume'. Upon doing so, the volumetric data for various subcortical structures are displayed. You could choose for example 'Left-Lateral-Ventricle', then click 'Add Selection to Data Table'. Upon doing so, it will appear in the 'Discrete and Continuous Factors' display of the 'Data Table View'. 'Left-Lateral-Ventricle' is now available as a possible factor in your analysis ('Left-Hippocampus' and 'Right-Hippocampus' were imported from aseg.volume in this manner). If you select 'Left-Lateral-Ventricle' in the 'Data Table View', then the volumetric data for all the subjects is plotted. You can look for patterns or outliers. If you suspect an outlier, right-click on that point in the scatter-plot.

You can then either exclude that subject, or examine it in tkmedit or tksurfer, to check for possible problems in the original scan data. Lastly, you can save this newly imported data (in this case, 'Left-Lateral-Ventricle') to your qdec.table.dat file by selecting 'Save Data Table' from the File menu (but dont do this for the tutorial).

# 1.3.3. Design

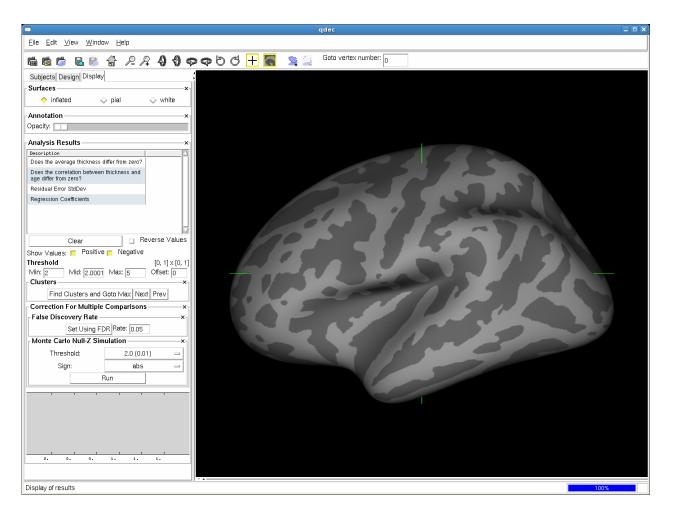
When you click-over to the Design tab your discrete (gender) and continuous (age, Left-Hippocampus, Right-Hippocampus and Left-Lateral-Ventricle) factors should appear.

		qdec	
<u>Eile Edit View W</u> indow <u>H</u> elp			
	🗣 D G 🕂 🔳 🛸 🔅	Goto vertex number:	
	♥ ♥ ♂ + ■         ■	Goto vertex number:	
Help			

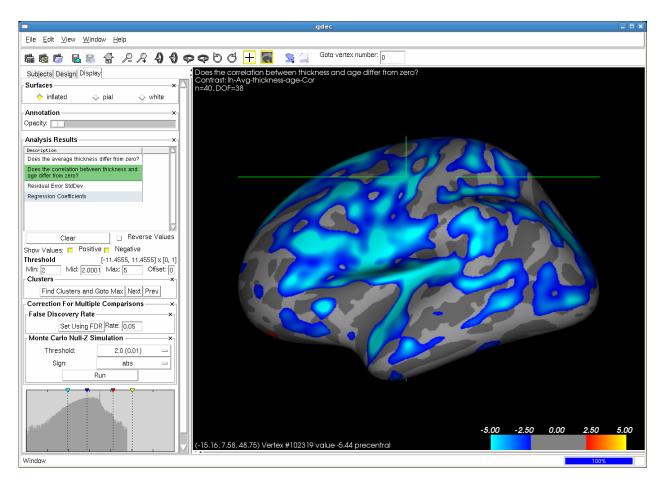
You can select up to four factors in the Design tab to regress against. For the tutorial data, you could select 'gender' and 'age' (or 'Left-Hippocampus', as long as just one Continuous Factor is selected). Nuisance Factor menu in Design tab allows selection of any number of continuous variables of which will be treated as 'nuisance variables' in the glm (contrast matrix will have value '0' for these). For simplicity in this example, choose only 'age' in the Continuous variables menu, leaving the 'Measure', 'Hemisphere' and 'Smoothing' at their defaults (thickness, lh and 10mm). Before you click the 'Analyze' button you will want to name your Design, something like 'lh-thickness-age-fwhm10', and enter that into the "Design Name" text entry box at the top of the window. Now click the 'Analyze' button and the stats-engine will begin processing, executing the mri\_glmfit executable. Upon clicking 'Analyze', the terminal will display the output of this processing. Also, progress information is shown in the bottom bar of the Qdec application. An analysis make take a minute or two.

# 1.3.4. Display

Once the analysis is complete (taking up to several minutes for a large subject set), you can click the Display tab and the fsaverage inflated surface will appear in the display window. You will see a list of questions summarizing the various analyses that were completed.

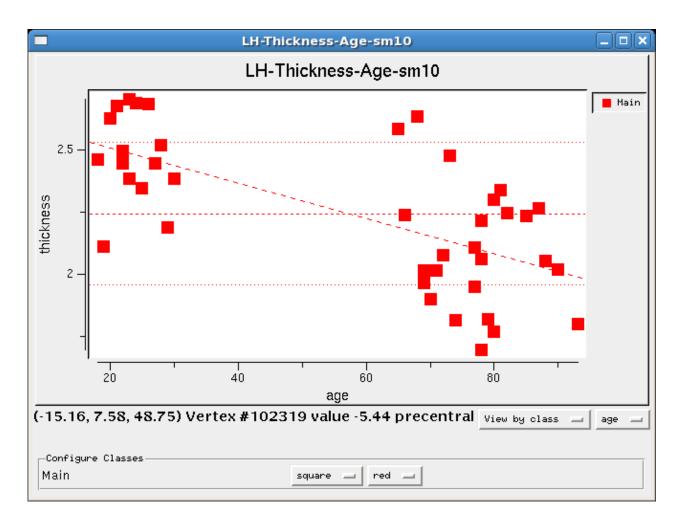


You can click on one of these questions to load the results. If you click on **Does the correlation between thickness and age differ from zero** it will display the statistically significant regions where age and thickness are correlated. Here is an example display:



'FindClustersAndGotoMax' button runs mri\_surfcluster on a selected result, which will find clusters based on currently selected display threshold, output a table of results to the terminal, and move the cursor to the max vertex in the #1 cluster. 'Next' and 'Prev' buttons allow cycling movement of the cursor through the found clusters. On the displayed image, notice the green cross-hairs that indicate the vertex you have currently selected. You can change vertices and display a plot of the data for a particular vertex by left-clicking on a point while holding-down the Ctrl key. If you'd like to turn off the cursor display you

can use the <u>use</u> button. Here is an example plot that corresponds to the shown selected vertex:



The plot shows your measure on the y-axis (vertical) - in this case, cortical thickness - and the variable on the x-axis (horizontal) - in this case, age. Each data point on the plot is representative of an individual subject, denoting their age and cortical thickness at the vertex you have selected. For this example at this vertex, we can see that the cortex is thinning with age. The information at the bottom of the both the plot window and the Qdec window shows that this vertex has surface coordinates (-15.16, 7.58, 48.75) and is Vertex# 102319. The significance value is -5.44 and it is in the precentral region. The significance in this display is a -log(10)p value, and not a straight p value.

In addition to this you can also do correction for multiple comparisons by Monte Carlo cluster-wise simulation making use of pre-run data, making run-time near instantaneous.

# 1.3.5. Interacting with your data

**Rotating, Panning and Zoom** You can rotate the display, hold-down the left mouse button and move the mouse. Holding-down the middle button while moving the mouse will move the display in the window. Holding-down the right mouse button while moving will zoom the display.

There are buttons at the top of the Qdec display that will rotate and zoom as well:



If you get it rotated too far, the home button is will reset it.

**Parcellation Display** The cortical parcellation is loaded into Qdec upon opening. On the Display tab you can adjust the annotation opacity. There may be a slight delay while the display updates, be patient! Sliding the button to the right will begin to show the parcellation annotation underneath the overlay. You can bring the opacity to a level that is useful in your interaction with the data. When you have selected a point, which is accomplished by holding down the ctrl key and left-clicking the mouse, the information at the bottom of the window will tell you what region, or parcellation unit, the point is found.

#### **Significance Thresholds**

You can also adjust the threshold levels for the overlay on the Display tab.

When setting a color scale, you're interested in two things: the threshold (i.e., the value below which the vertex will be transparent - Min), and the saturation point (i.e., the value beyond which the color will not change - Max). In Qdec you can also specify the point where the color will reach the midpoint, with Mid on the control panel (by default Mid is set to just higher than Min. The meaning of these thresholds depends upon the nature of the data you have loaded as the overlay. The map you are currently viewing is  $-\log 10(p)$ , where p is the significance, so a Min of 2 will display all vertices with p<.01 and a Max of 5 will show vertices of p<.00001 as the same color. You can lower the threshold to 1.3, 2, 3, to show all vertices with p<.05. You could raise the threshold to 4, 5, 6 to show all vertices with p<.0001.

# 1.3.6. Variations on Design

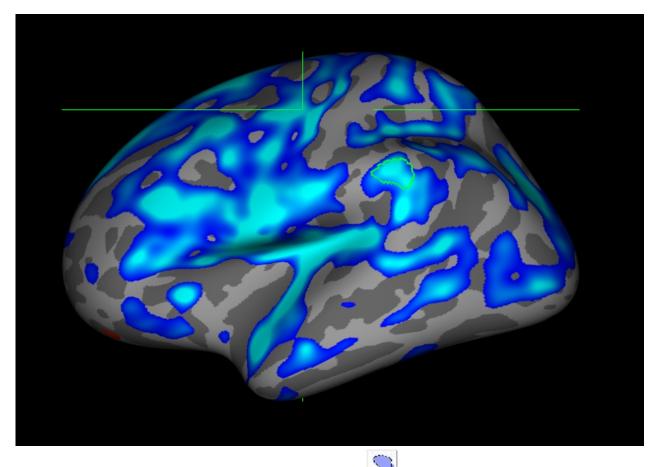
With Qdec it is easy to design and run a variety of different analyses. For the first example we looked simply at age and thickness in our subjects. Click back to the Design tab and select gender, to add it to the design. You will want to change the name of the design, call it 'lh-thickness-age-gender-fwhm10', and click Analyze. When the analysis is done running, click the Display tab and see that there are

additional questions in the list summarizing the various analyses that were completed. Among the questions displayed now are **Does the Thickness--age correlation differ between male and female?** and **Does the average thickness differ between male and female?** Click on one of these questions to display the statistically significant regions where the age and thickness correlation are different in men and women, or the average thickness is different in men and women (respectively). Similarly, you can add in one of the other continous variables - hippocampal volume - and run that design.

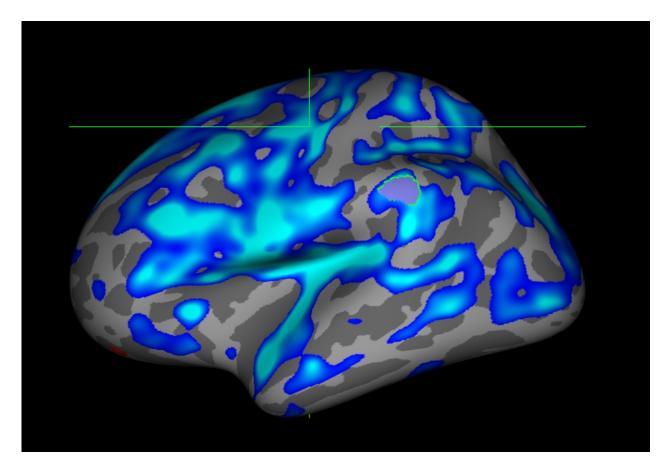
You can change your design even more, if you click back to the Design tab, you can change your measure from thickness to something else - area, area.pial, volume, sulc, curv, and jacobian\_white are your choices. You can also change your level of smoothing - 0, 5, 10, 15, 20, and 25 are your choices. And you can perform any of these on the left (lh) or right (rh) hemispheres. Take a few minutes to select a new design to run, remember to call it something new before you hit Analyze so that the directory of results can be saved.

# **1.3.7.** Define a Region of Interest

FreeSurfer has the ability to compute statistics averaged over a defined region of interest (ROI), which is another popular way to test statistical hypotheses and a good way to check your data. To define a label that marks a region of interest (ROI) on the surface hold down shift then left click and drag to draw your ROI. When drawing your ROI, draw slowly, allowing the display to catch up with you if necessary. There is no need to worry about closing the ROI precisely, when you are done and release the mouse button QDEC will automatically close the ROI for you. You should then see a green outline of the ROI you drew, like this:



You can then select the *add the selection to the ROI* button , and your label should now be filled in with purple, like this:



If you do not add your label to the ROI and you start to draw again, QDEC will erase your first label and begin a second. If you have added something to the ROI and want to remove it you can use the *remove* 

selection from ROI button . When you are finished you can save your label by selecting File -->

**Save Label** or clicking the save label button. A dialog box will pop up, and you can choose the location and name to save your label. For this example you can call your label **lh.supramarg.label**, since it is a label of the supramarginal gyrus, and click **Save**.

It may then be useful to map this label to all of the individual subjects in your group study, to either extract statistical values from this region or to visualize the area on each subject to check the integrity of your results. You can do this automatically by selecting **File** --> **Map Label to Subjects...**, a dialog box will pop up asking for the label name, you can enter everything before the *.label* of the name, so for this example enter **lh.supramarg**, and click **Ok** (OPTIONAL as it takes a few minutes for this to finish). This will use mri\_label2label to map this label from your average surface onto all the subjects in your study. When it is complete (this can take several minutes) each subject will have a file named lh.supramarg.label in its label subdirectory. But for tutorial purposes, you can run the following command to map the label onto a subject.

mri\_label2label --srclabel lh.supramarg --srcsubject fsaverage --trgsubject 004 --trglabel lh.supramarg --regmethod surface --hemi lh

The above command creates the label lh.supramarg.label in 004/label/ directory. This label can be loaded onto each subject's surface in tksurfer, or opened on the volume in tkmedit (not shown in this tutorial).

**mris\_anatomical\_stats** You can use mris\_anatomical\_stats to get a set of statistics on each individual label you've created. The command to run this on the label lh.supramarg.label that you generated for subject 004 is:

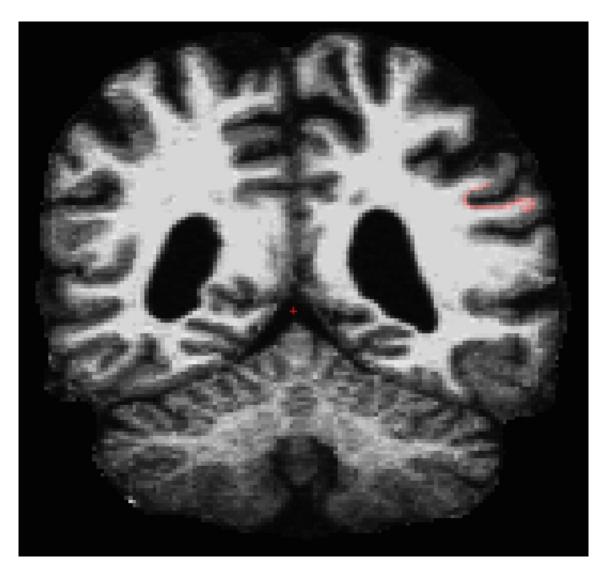
```
cd $SUBJECTS_DIR
mris_anatomical_stats -1 lh.supramarg.label \
    -t lh.thickness -b -f 004/stats/lh.supramarg.stats 004 lh
```

This will output a stats file to 004/stats/lh.supramarg.stats, which looks like this. This file gives the number of vertices, surface area, gray matter volume, average thickness and st. deviation, mean curvature, gaussian curvature, folding index, and curvature index for this region only. You can run this same command on all your subjects to generate these statistics. You could then use aparcstats2table to generate one space delimited table of all these measures for your subjects.

In addition, it might be a good idea to visualize this label on each subject, to be sure there is not a defect in the surface causing your result. You can look at your label in tkemdit on the volume with the command:

```
tkmedit 004 brainmask.mgz -label lh.supramarg.label
```

If you scroll towards the posterior end of the brain (coronal slice 80), you will find your label. Typically you would load the surfaces as well, but for this tutorial, it is easier to find the label without them.



# **1.3.8.** Correction for Multiple-Comparisons

See this page for information on support for correction for multiple comparisons in Qdec. Note that it is not a tutorial. A more in-depth tutorial on this subject is found here.

# 1.3.9. Miscellaneous

Qdec supports some customizations via the .Qdecrc file.

(end of qdec tutorial)

# **1. FreeSurfer Tutorial: Applying FreeSurfer Tools to FSL fMRI Analysis (FEAT)**

The purpose of these series of exercises is to give you some familiarity with integrating FreeSurfer and FSL's functional analysis. The main challenge in the integration is getting a subject's anatomical data properly registered with their functional data. Once registered, you can use FreeSurfer display tools to visualize your FSL functional activation maps on the subject's anatomical volume and on the surface. You can also convert to the common surface space for group analysis with mri\_glmfit or FSL's flame or randomise.

For structural data, we will use \${SUBJECTS\_DIR}/bert and \${SUBJECTS\_DIR}/fsaverage. For functional data, we will use fbert1.nii.gz and fbert2.nii.gz. The functional data set consists of two runs, 85 volumes each, 64 x 64 x 35 voxels, with size 3.4375 x 3.4375 x 4.0 mm^3. TR = 3 sec. The experiment is a periodic block design with 15 sec ON blocks of simultaneous finger tapping, flashing checker board, and auditory tone. The OFF blocks are rest periods. The paradigm starts with an OFF block. The FEAT functional analysis has already been done, and the runs have been combined with GFEAT (Fixed-effects, one-sample group mean). This data was smoothed in the volume at 5mm, but when preparing for a group surface-based analysis, we recommend that you smooth on the surface prior to group analysis and not smooth in the volume at all (or less than the voxel size) You can download the tutorial data from here.

Throughout the tutorial, it will be assumed that you are in the fbert-feat directory. If you list the directory (ls), you will see:

```
fbert1.nii.gz -- run 1 raw functional data
fbert1.design.fsf -- run 1 FEAT design
fbert1.feat -- run 1 FEAT directory design
fbert2.nii.gz -- run 2 raw functional data
fbert2.design.fsf -- run 1 FEAT design
fbert2.feat -- run 2 FEAT directory design
fbert.gfeat -- runs 1 and 2 combined in FFx model
run-fsfeat-tut -- script to run through all the non-interactive components
```

You will see some other files/directories there too, but these are the most important.

### 1.1. 1.0 Registration

The registration process computes a matrix that maps the FEAT example\_func to the subject's anatomical using FLIRT. This matrix can then be used in later steps to display functional maps on the anatomical volume and the surface.

•

Exercise A. Registering FSL Feat output to the anatomical

### 1.2. 2.0 Overlaying onto Same-Subject Anatomical

The statistical maps from Feat may be overlaid onto the subject's anatomical volume, the surface derived from the anatomical volume, or the FSL's standard volume. All these options are described in the following exercise.

•

Exercise B. Overlaying FSL Feat statistical maps

# 1.3. 3.0 Surface-based Group Functional Analysis

The statistical maps from Feat may be overlaid onto the subject's anatomical volume, the surface derived from the anatomical volume, or the FSL's standard volume. All these options are described in the following exercise.

•

Exercise C. Surface-based Group Analysis

### 1.4. 4.0 Mapping automatic segmentations

FreeSurfer automatically generates cortical and subcortical segmentations from the subject's anatomical data. These segmentations can be mapped into the functional space for performing region of interest (ROI) analysis. Then, the segmentation for a particular structure can be extracted to create a binary mask. Go through the following exercise for details.

•

Exercise D. Mapping automatic segmentations to the functional space

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•

To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

# 1. Registering FSL Feat output to the anatomical

The registration is a multi-step process. Each step is described in detail below. To begin the exercises, first enter the following:

```
tcsh
setenv SUBJECTS_DIR $TUTORIAL_DATA/buckner_data/tutorial_subjs
cd $SUBJECTS_DIR/fbert-feat
ln -s $FREESURFER_HOME/subjects/fsaverage
ln -s $FREESURFER_HOME/subjects/bert
```

The subjects 'fsaverage' and 'bert' may already exist in the subjects directory, so ignore any warnings that might be issued that say 'File exists'.

### 1.1. 1.1 Automatic registration

The program that performs the registration is called reg-feat2anat (which uses FLIRT to do the actual computations). Information about how to run this program and what this program is doing can be obtained with:

reg-feat2anat --help

Now, run the registration for bert's first fMRI data:

reg-feat2anat --feat fbert1.feat --subject bert

Verify that this created directory fbert1.feat/reg/freesurfer. There are several files with matrices in them. The most important one is anat2exf.register.dat.

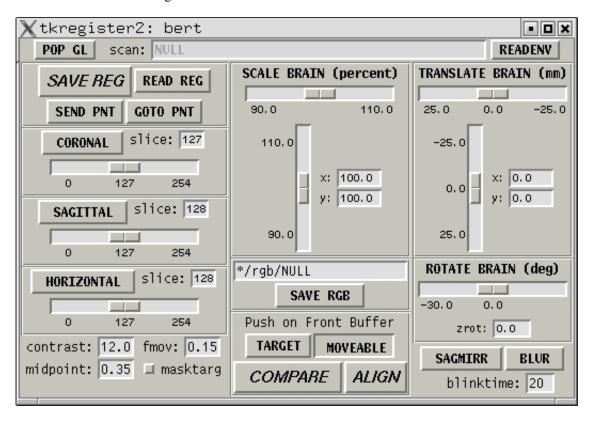
### 1.2. 1.2 Manual Checking/Editing the registration

You should ALWAYS visually check your registration. This is done with tkregister2 through reg-feat2anat. This program brings up a GUI which allows you to inspect and interactively edit the registration matrix. tkregister2 is a complicated with lots of features. You can get a full help on it with:

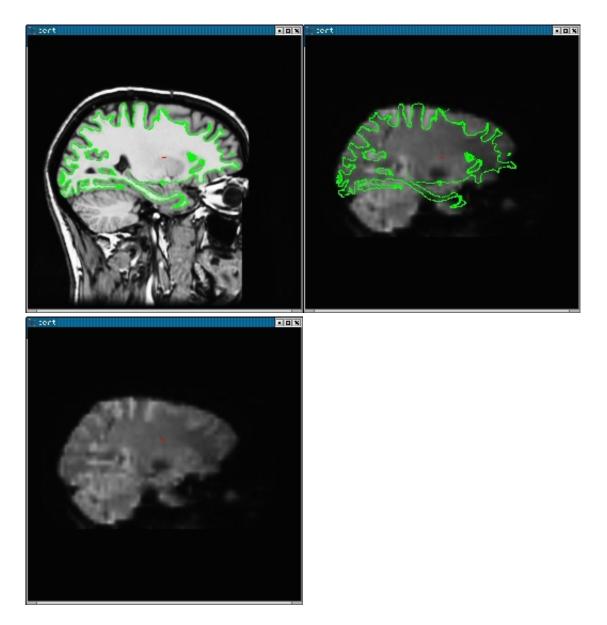
To check the registration computed in the previous step, run:

```
reg-feat2anat --feat fbert1.feat --manual
```

You will see the following GUI window:



You will also see an anatomical similar to the one on the left below. Pressing the "COMPARE" button will allow you to flip between the anatomical (left) and functional (middle). The green line is the orig surface. It is the same in both the anatomical and functional. The right is the same image as the middle without the surface.



# 1.3. 1.3 Navigating through tkregister2

tkregister2 is controlled through the interface and through keypress commands. When using a keypress command, the image window must have control of the cursor; this can be accomplished by clicking in the image window. Here are some useful commands.

A. To switch between the functional and anatomical, press the "COMPARE" button.

B. To change orientation, press the CORONAL, SAGITTAL, or HORIZONTAL button.

C. To change slice, operate the slider.

D. To change the functional brightness, change the value in the "fmov" entry box. Or you can use the 'i' keypress command to turn on automatic intensity normalization.

E. To toggle the surface on and off, use the 's' keypress command.

You can get more information on keypress commands and general usage with:

tkregister2 --help.

# 1.4. 1.4 Evaluating the registration

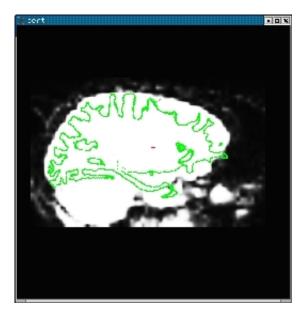
The first step in evaluation is to make sure that there was no catastrophic failure, e.g., the functional is rotated by 90 degrees with respect to the anatomical, or is shifted by a large amount. Such failures are usually caused by failures in the registration to standard space (see section on troubleshooting).

The second step is to make sure there was not a left-right flip. This is tricky because the brain is very symmetrical. However, the cortical folding patterns often have subtle asymmetries. In general, you cannot see the folding patterns in the functional, but you can see the CSF which will also follow the surface (see the images above). Make sure that asymmetries in the (green) surface are present in the CSF.

The final step is to check the alignment. This is done by making sure that green surface follows the contour of the CSF.

# 1.5. 1.5 Pitfalls

There are several ways in which the registration may be good but can appear bad. First, don't evaluate the registration in areas known to have B0 distortion (e.g., orbital frontal, medial temporal lobe). Second, be careful using the ventricles for alignment. The anterior portion is often susceptible to B0 distortion. Third, if it looks like the surface cuts through a CSF fold, look at adjacent slices to see if it lines up better. This can happen because functional voxels are so big. Finally, be careful trying to use what looks like the edge of the brain in the functional. For example, in the middle image above, it looks like the surface extends beyond the top of the brain. However, if we adjust the brightness (set fmov to 1; see image below), one can clearly see that the surfaces are actually in brain.



### 1.6. 1.6 Editing the registration

If for some reason you do not believe that the automatic registration is sufficiently close, you can edit the registration manually. tkregister allows you to translate, rotate, and scale the brain using the sliders on the right side of the GUI. All the modifications happen in the viewing plane. The rotations are about the red cross. Note: you should not need to adjust the scale (ie, stretch the brain) because both brains belong to the same person. This is equivalent to contraining the registration to be rigid (ie, 6 DOF). When done, press the "SAVE REG" button. Note that if you modify the registration, don't re-run reg-feat2anat as that will overwrite your edits. As an exercise, try translating the functional. First, view the functional and then move the "TRANSLATE" slider. You will see the functional brain translate (the green surface will be fixed). DON'T save the registration (if you do, then just re-run reg-feat2anat). You can get more information on editing the registration with: tkregister2 --help.

# 1.7. 1.7 Check FEAT registration to Standard Space

This registration tool can be used to check and adjust the FEAT registration to standard space with:

reg-feat2anat --feat fbert1.feat --manxfm func2std

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# 1. Overlaying FSL Feat statistical maps

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To follow this exercise exactly be sure you've downloaded the tutorial data set before you begin. If you choose not to download the data set you can follow these instructions on your own data, but you will have to substitute your own specific paths and subject names.

In this exercise, the data set of subject **bert** is used for demonstration. Type the following prior to beginning the exercises:

```
tcsh
setenv SUBJECTS_DIR $TUTORIAL_DATA/buckner_data/tutorial_subjs
cd $SUBJECTS_DIR/fbert-feat
ln -s $FREESURFER_HOME/subjects/fsaverage
ln -s $FREESURFER_HOME/subjects/bert
```

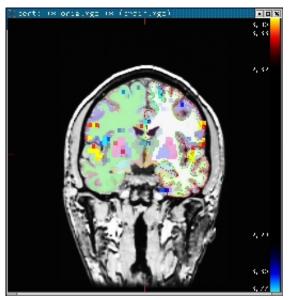
The subjects 'fsaverage' and 'bert' may already exist in the subjects directory, so ignore any warnings that might be issued that say 'File exists'.

### 1.1. 1.0 Overlaying the statistical map onto the bert's orig volume

Use the following command to display the zmap (zstat1.nii.gz) from the first run overlaid onto the bert's orig volume. It will display the automatic segmentation, and will also set the threshold at z = 2.3:

```
tkmedit bert orig.mgz lh.white -aux brain.mgz \
    -overlay ./fbert1.feat/stats/zstat1.nii.gz \
    -overlay-reg ./fbert1.feat/reg/freesurfer/anat2exf.register.dat \
    -segmentation aparc+aseg.mgz -fthresh 2.3 -fmax 4.3
```

You should see the image below:



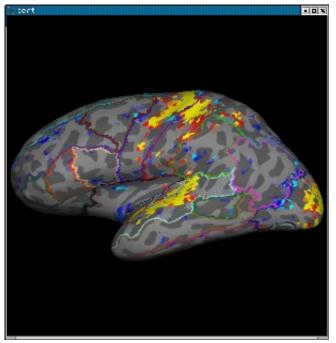
When you click or mouse over a voxel, the cortical or subcortical structure that that voxel belongs to will be displayed in the control panel. You can view any of the volumes in the stats dir in this way as well as the clustered maps in the feat directory.

### 1.2. 2.0 View statistical maps on bert's surface

To view any of the statistical maps on bert's surface, close the tkmedit GUI (or open a new terminal window) and run:

```
tksurfer bert lh inflated \
    -overlay ./fbertl.feat/stats/zstatl.nii.gz \
    -overlay-reg ./fbertl.feat/reg/freesurfer/anat2exf.register.dat \
    -fthresh 2.3 -fmid 3.3 -fslope 1 -annot aparc.annot
```

Change the cortical parcellation to outline mode with View->LabelStyle->Outline. You should see the image below:



When you click or mouse over a vertex, the control panel will display the name of the cortical structure. You can view any of the volumes in the stats dir in this way as well as the clustered maps in the feat directory. You can also run the tkmedit and tksurfer commands above in separate shells and use the Save-Point/Goto-Point functionality to navigate through the volume and surface.

# 1.3. 3.0 Displaying Same-Subject, Cross-Run GFEAT Results

Typically, one collects more than one run/series of functional data for each subject. The individual runs are analyzed separately, then combined in standard space with GFEAT using a fixed-effects model. Since the data are no longer in the subject's native functional space, a different registration matrix is needed to map the GFEAT results to the individual. Each run of reg-feat2anat will create a reg/freesurfer/anat2std.register.dat. Any one of these can be used to map the GFEAT data to the subject's anatomy.

First, verify that the registration is good with:

```
tkregister2 --mov fbert.gfeat/mean_func.nii.gz --surf \
    --reg fbert1.feat/reg/freesurfer/anat2std.register.dat
```

mean\_func.nii.gz is the mean of the example\_func's in standard space. Since this is the mean func, it will be blurry if you have spatially smoothed. Note: if there is a problem with this registration, you need to repair the registrations for each individual run.

Now show gfeat results on anatomical volume:

```
tkmedit bert orig.mgz -seg aparc+aseg.mgz \
  -ov fbert.gfeat/copel.feat/stats/zstat1.nii.gz \
  -ovreg fbert1.feat/reg/freesurfer/anat2std.register.dat \
  -fthresh 2.3 -fmax 4.3
```

Here we've used the anat2std.register.dat from the first run.

Now show gfeat results on the surface:

```
tksurfer bert lh inflated -annot aparc.annot \
  -ov fbert.gfeat/copel.feat/stats/zstatl.nii.gz \
  -ovreg fbert1.feat/reg/freesurfer/anat2std.register.dat \
  -fthresh 2.3 -fmid 3.3 -fslope 1
```

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Type the following prior to beginning the exercises:

```
tcsh
setenv SUBJECTS_DIR $TUTORIAL_DATA/buckner_data/tutorial_subjs
cd $SUBJECTS_DIR/fbert-feat
ln -s $FREESURFER_HOME/subjects/fsaverage
ln -s $FREESURFER_HOME/subjects/bert
```

The subjects 'fsaverage' and 'bert' may already exist in the subjects directory, so ignore any warnings that might be issued that say 'File exists'.

### 1. 1.0 Resampling COPEs to common surface

#### 2. 1.1 Individual Runs for a Subject

In order to perform surface-based group analysis, you need to have all the copes sampled into a common space. This is done with mris\_preproc:

```
mris_preproc --target fsaverage --hemi lh --fwhm 5 \
    --out xrun/lh.copel.mgh \
    --iv fbert1.feat/stats/copel.nii.gz fbert1.feat/reg/freesurfer/anat2exf.register.dat \
    --iv fbert2.feat/stats/copel.nii.gz fbert2.feat/reg/freesurfer/anat2exf.register.dat
```

In this case, the common space is the left hemisphere of fsaverage. Surface-based smoothing of 5mm FWHM is used. The output lh.cope1.mgh looks like a volume because it is in mgh format, but it is really a surface stored in a volume format (note it's dimensions are  $1974 \times 1 \times 83 = 163842 =$  number of vertices in fsaverage's surface). It has 2 frames, one for each run.

Note that each volume (cope1.nii.gz) is accompanied by a registration matrix (anat2exf.register.dat). The above example extracts data from each run of the same subject. If you have multiple subjects, you can specify the gfeat/cope1/stats/cope1.nii.gz paired with the anat2std.register.dat matrix.

You can also extract the cope variances (varcopes) in the same way:

```
mris_preproc --target fsaverage --hemi lh --fwhm 5 \
    --out xrun/lh.varcopel.mgh \
    --iv fbert1.feat/stats/varcopel.nii.gz fbert1.feat/reg/freesurfer/anat2exf.register.dat \
    --iv fbert2.feat/stats/varcopel.nii.gz fbert2.feat/reg/freesurfer/anat2exf.register.dat
```

#### 3. 1.2 Runs combined in a GFEAT within a Subject

Once the runs are combined in a GFEAT analysis, they are no longer in the native functional space but rather in the standard space. You can still use the surface-based analysis as long as the subjects have not been combined together. To do this, you just need the appropriate registration matrix that takes the data from standard space to the subject's anatomical. This is computed when you run reg-feat2anat. Each run will have this matrix, but they should all be the same. Eg:

```
mris_preproc --target fsaverage --hemi lh --fwhm 5 \
    --out xsubject/lh.copel.mgh \
    --iv fbert.gfeat/copel.feat/stats/copel.nii.gz fbert1.feat/reg/freesurfer/anat2std.register.dat \
    --iv fnick.gfeat/copel.feat/stats/copel.nii.gz fnick1.feat/reg/freesurfer/anat2std.register.dat \
    ...
```

The cope is specified from the gfeat, but the registration matrix is specified from the feat from the first run of each subject. The output can be used in the surface-based group analysis below.

### 4. 2.0 Surface-based Group Analysis

Once you have the copes in the common surface space, the (nearly) same as you would with a thickness study. With a functional study, you also have the flexibility to run fixed- and weighted random-effects analysis.

The command below can be used to run a fixed-effects analysis across both runs. Normally, you would probably run a random effects or weight random effects analysis, but since we only have two runs here, we'll use a fixed-effects. The dof=163 is the sum of the dofs from fbert?.feat/stats/dof. The model here is a simple one-sample group mean (OSGM) in which the design matrix is simply a column of 1s. For more elaborate designs, use an FSGD file instead of --osgm.

This will create a directory called xrun/lh.osgm.ffx:

```
ls xrun/lh.osgm.ffx
ar1.mgh -- spatial AR1 map
beta.mgh -- regression coefficients (same as PEs)
fwhm.dat -- estimate of spatial FWHM (not valid with 2 data points)
mask.mgh -- binary mask (if used)
mri_glmfit.log -- log file
osgm -- contrast directory
rstd.mgh -- residual standard deviation
rvar.mgh -- residual standard variance
Xg.dat -- design matrix (text)
X.mat -- design matrix (matlab)
```

The most important results are in the contrast directory:

```
ls xrun/lh.osgm.ffx/osgm
C.dat - contrast matrix
cnr.mgh - contrast-to-noise ratio
F.mgh - F ratio
gamma.mgh - contrast size (same as COPE)
maxvox.dat - info about the vertex with max F stat
sig.mgh - significance map (-log10(p))
```

To view the higher-level results, run:

tksurfer fsaverage lh inflated \
 -overlay xrun/lh.osgm.ffx/osgm/sig.mgh \
 -fthresh 2 -fmid 3 -fslope 1 -annot aparc.annot

There are several differences between this invocation of tksurfer and the one used during the individual analysis before:

- The subject is fsaverage, not bert
- The input is a sig map, which is -log10(p)
- •

The thresholds are significance thresholds, not z. So, 2 means p < .01.

#### 5. 3.0 Alternative method to resample COPEs to common surface

This command will resample the copes to the common surface space, but does not concatenate them together as mris\_preproc does. Rather, it stores the result in fbert1.feat/reg\_surf-lh-fsaverage/stats for the left (reg\_surf-rh-fsaverage for right).

feat2surf --feat fbert1.feat --cope-only

# **1. FreeSurfer Tutorial: FreeSurfer Tools**

Tkmedit and tksurfer are two important tools in the FreeSurfer package.

Tkmedit displays volumetric anatomical data and allows the user to navigate through that data and view it from different orientations. Tkmedit also displays other data types such as functional data and surfaces as overlays onto this anatomical data.

Tksurfer displays surface data and allows the user to navigate through that data and view it from different orientations. Tksurfer also displays other data types such as functional data and curvature as overlays onto this surface data.

Tkmedit and tksurfer are described in detail below.

# 1.1. 1.0 Tkmedit

```
Interface
General usage
Working with data
Quick reference
```

# 1.2. 2.0 Tksurfer

•

Interface

•

General usage

•

Working with data

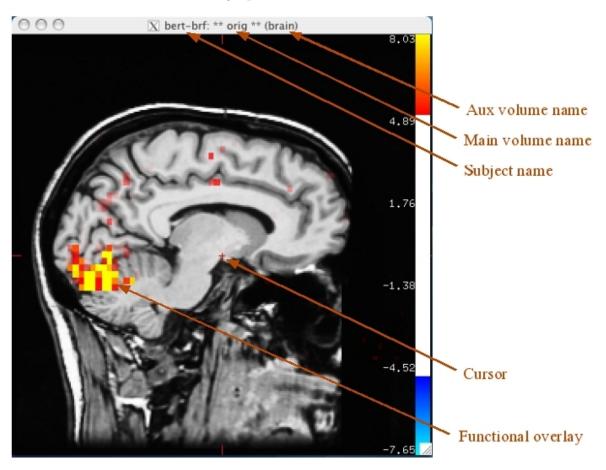
•

Quick reference

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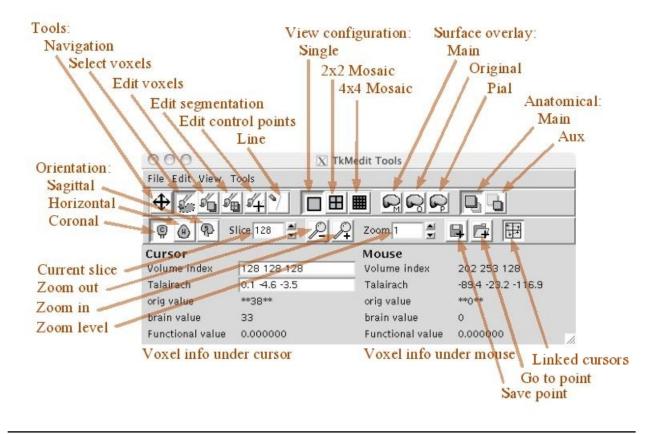
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# 1. Tkmedit interface



The display window

#### The tools window



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# 1. General usage

# 1.1. 1.0 Quick start

Tkmedit is started by one of two methods: using the tkmedit-sess script, or invoking it from the command line with the tkmedit command. Depending on which method you use, you may see different types of data loaded, but all methods require an anatomical data set to be loaded. Other types of data can be loaded from the File menu.

To explore your data, you can move the mouse over the picture of the volume (the Display Window) and see information about the area under the mouse in the window on the bottom (the Tools Window). You can change the view with the orientation buttons (the ones shaped like cross-sections of the head labeled 'C,' 'H,' and 'S,' which stand for Coronal, Horizontal and Sagittal) and the slice number field in the Navigation Toolbar. To save pictures of your data, go to the Tools menu and select the Save RGB item near the bottom.

### 1.2. 2.0 Viewing area

Tkmedit consists of two windows. The top window, called the Display Window, shows the currently displayed slice. Multiple panes can show multiple orientations and configurations. The bottom window, called the Tools Window, contains the controls and feedback information. Here is the interface.

Information about the currently displayed data can be obtained by moving the mouse over the Display window or by clicking to set the cursor. As the mouse is moved in the display window, the mouseover section of the Tools window will display information about the voxel (or volume element) under the tip of the mouse arrow. Alternatively, most tools set the cursor upon clicking with the left button. The Cursor section of the Tools window displays information about the voxel under the cursor. By default, the cursor is drawn as a red cross. Guidelines are drawn on the sides of the Display Window to help locate the cursor.

It is possible to display multiple orientations of data simultaneously in the Display Window. This is done by splitting the window up into multiple viewing panes, each with a different slice, orientation, or other viewing characteristics. The cursor can be linked between so that a click in one pane to set the cursor will update the cursor in all the other panes. There are three built-in configurations: Single View, Multiple Views, and Mosaic View. These can be activated by clicking on the appropriate icon in the Navigation

Toolbar ( Single, H Multiple, Mosaic) or by choosing the appropriate item in the View menu.

In the latter two views, there is more than one pane in the Display Window. A green border is drawn around the active pane. To make another pane active, click in it to set the cursor. The mouseover section of the Tools Window will only display information about the voxel under the mouse if it is in the active pane.

#### 2.1 Single view

This is the default view. It is the simplest and fastest view for tkmedit to render.

#### 2.2 Multiple views

This viewing mode divides the window into four panes. Each of the three orientations is shown, with one orientation duplicated. This configuration is optimal for viewing all planes around a single point. By default, the cursor is linked so that setting it in one view moves the current slice in all other views to the cursor's location in that view's orientation.

#### 2.3 Mosaic view

When mosaic view is first activated, the volume is sliced up in the orientation of the last active pane. Sixteen panes are created, each showing a different slice in the same orientation. Note that this mode is slow on most machines and is not optimal for 'live' navigation. It is mainly provided as a way to set up a view to be printed or saved to an image file.

### 1.3. 3.0 Navigation

#### 3.1 Changing slices

The current slice can be changed with the controls on the right side of the slice label in the Navigation Toolbar, with the mouse button 2 with the Navigation Tool (), or with the arrow keys. The current orientation can be changed by clicking on the appropriate button in the Navigation Toolbar ( coronal, horizontal, or sagittal) or by pressing the 'X,' 'Y,' and 'Z' keys.

With the Navigation Tool active, clicking with button 2 in the top half of the screen increases the slice number by 1, and clicking in the bottom half decreases it by 1. Clicking in dragging will continuously change the slice number; dragging up will increase the slice number, dragging down will decrease it.

#### **3.2 Zooming**

The view in any pane may be zoomed in to magnify the size of a voxel. The current zoom level is displayed both in the Navigation Toolbar and on the Navigation Toolbar. The zoom level may be changed in three ways:

- 1. By holding down the control key and clicking with button 1 to set the cursor and zoom in, or button 3 to zoom out. This works with any tool active.
- 2. By clicking on the zoom buttons in the toolbar or the zoom slider in the display area, zooming in or out around the cursor but not set the cursor.
- 3. By activating the Navigation Tool and clicking with button 3. Clicking in the top half of the Display window zooms in, and clicking in the bottom half zooms out. Clicking and dragging up continuously zooms in, and dragging down zooms out. This operation does not change the cursor.

#### 3.3 Panning

While zoomed in, you will not be able to see the entire slice. To pan around in the current slice, you may hold down the control key and click with button 2 to set the cursor and re-center, or activate the Navigation Tool and click and drag with button 1 Using that Navigation Tool will not change the cursor.

As you can see, navigation is primarily down in a modal fashion with the Navigation Tool or at any time with the control-click combinations, although the Navigation Tool is more intuitive and also allows slice changing. You will probably benefit from using both methods.

### 1.4. 4.0 Cursor

The cursor serves two main purposes. It acts as a central focus point when multiple viewing panes are open, and it selects a voxel whose information will be displayed in the Tools Window. It also serves as a kind of bookmark for sharing cursors with tksurfer, as a centroid for rotation, selecting a voxel to be graphed in the Time Course Window, and other various functions, explained later.

#### 4.1 Linked cursors

When the viewing area is split into multiple panes, each panel may have a separate cursor location. If cursor linking is active, setting the cursor in one panel will set the cursor in all panes. Cursor linking can

be activated with the button on the Main Toolbar.

For example, in the Multiple Views configuration, setting the cursor in the coronal pane will change the slice in the horizontal and sagittal planes. So the cursor becomes a way of navigating through the volume using the three orientations.

Depending on the viewing configuration, with Linked Cursors active, other attributes are linked as well, including zooming and display flags. In Mosaic View mode, only the in-plane cursor coordinates will be linked, so the slice in each pane will be the same. However, if the slice number is increased in one pane, the slice number will be increased in all other panes.

#### 4.2 Cursor and mouse information

The bottom of the Tools Window displays information about the voxel under the cursor (on the left) and the mouse (on the right). The information displayed depends on the kind of data loaded. If only an anatomical volume is loaded, only the volume index coordinates, volume Talairach coordinates, and volume value will be displayed. (If for some reason a Talairach transform could not be found, RAS (Right Anterior Superior) coordinates will be displayed instead.)

As other data are loaded, more information will appear in this area. For example, if a functional overlay is loaded, the functional overlay value will appear in this area. Alternatively, the data shown can be changed in the **View->Information** menu.

# 1.5. 5.0 Saving images

You can save the contents of the Display Window to a picture file. Tkmedit uses the RGB format, a simple format readable by most image programs. To save a single picture, choose **Tools->Save RGB...** You may want to turn off the cursor or other view items in the **View** menu before saving your picture.

You can also save a series of images with the **Tools->Save RGB Series...** command. This command steps through slices in the current orientation and saves an image of each. You choose a directory and prefix for the file names as well as the range of slices to step through. For example, if you choose /tmp for the directory, "image" for the prefix, and 5 and 8 for the start and end, it will create /tmp/image005.rgb, /tmp/image006.rgb, /tmp/image007.rgb, and /tmp/image008.rgb.

# 1.6. 6.0 Tips

• Most volume input operations can take many different kinds of volumes. If the volume is a bfloat or bshort volume, select one of the .bfloat or .bshort files. If the volume is a COR volume, select the COR-.info file or just enter the directory without any file names.

•

Tkmedit uses the Tk toolkit (http://www.tcl.tk) for its interface (the 'Tk' in 'tkmedit'). A nice feature of this toolkit is that all the menus are detachable and can become a separate window that stays up all the time. This is particularly useful when using the View menu. Just click on the top line of the menu when it comes down from the menu bar, and a window will take its place.

- Learn the keyboard shortcuts for volume navigation.
  - The 'x', 'y', and 'z' keys will change orientations.
  - The arrow keys will change slices.
  - You can zoom in with Ctrl + (Ctrl-Shift =) and out with Ctrl -.
  - You can also jump to the Navigation Tool by hitting the 'n' key and click around the volume, then return to the tool you were using previously.
- If you are using tkmedit with the expected directory structure at the NMR-MGH center, tkmedit will automatically look in certain places for files. See the File Name Substitution section.
- Find yourself using certain toolbars? You can set environment variables to automatically hide or show without using the View menu. Set TKMEDIT\_TOOLBAR\_MAIN, TKMEDIT\_TOOLBAR\_NAV, or TKMEDIT\_TOOLBAR\_RECON to 0 or 1 to hide or show the main, navigation, or reconstruction toolbars, respectively.
- All file dialog boxes have a menu at the top which you can use to jump to specific directories to find data quickly. Additionally, you can specify your own shortcut directory by setting the environment variable FREESURFER\_DATA.

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# 1. Working with data

### 1.1. 1.0 Anatomical volumes

#### 1.1 Main and aux volumes

Tkmedit can hold two anatomical volumes in memory at the same time, called the Main volume and the Aux volume. Although it can not display them together at the same time, clicking on a voxel in one volume will display that value at that voxel in both volumes.

The Main volume is considered the central data object in tkmedit. All other data is displayed relative to the Main volume. The subject name and Main volume name are displayed in the title bar of the Display Window. If an Aux volume is loaded, its name is displayed in parentheses next to the Main volume name in the title bar.

If both volumes are loaded, you may select which volume is displayed by choosing Main Volume or Aux

Volume in the View menu, or by pushing in the appropriate button ( for Main and for Aux) on the Main Toolbar. The currently shown volume will be surrounded by double asterisks in the title bar, and its value in the Tool WIndow is shown in double asterisks. If only the Main volume is loaded, asterisks will not be displayed.

You can go to a specific location in the volume by using the Tools, Goto Point... command, or by typing the coordinates into one of the fields in the Tools Window where the coordinates are displayed and pressing the return key.

#### **1.2 Display transform**

Display transforms can be applied to each volume to transform its position. This transform only affects the way the volume is drawn in the Display Window. All coordinates displayed in the Tools Window will not be affected by this transform.

The file must be an LTA (\*.lta) or XFM (\*.xfm) transform type. To load a transform for the Main volume, choose File, Transforms, Load Transform for Main Volume... from the menu and enter the file name of the transform. To unload it, choose File, Transforms, Unload Transform for Main Volume. There are corresponding items for the Aux volume.

#### **1.3 Display options**

Volume display in the tis pretty basic, but there are options for configuring the brightness and contrast of each volume. Choose View, Configure, Brightness / Contrast... to open a dialog with four sliders. The top two control the brightness and contrast of the Main volume and the bottom two the Aux volume.

You may also turn off the volume display by unchecking View, Anatomical Volume. This is useful if you want to look at other types of data with the anatomical volume in the background. Neither the Main nor Aux volume will be displayed if this item is not checked.

You can display a maximum intensity projection by checking the View, Maximum Intensity Projection item. This displays the voxel with the highest value in the current orientation, and is the same for every slice in an orientation.

### 1.4 Editing

Editing is done with the Edit Tool which can be activated by pushing the <sup>4</sup> button in the Main Toolbar, by choosing Tools, Edit Voxels, by pressing the 'a' key.

There are settings for two 'colors' for this tool, each bound to a button when this tool is active. These settings can be choosing Tools, Configure Volume Brush.... This tool, along with the other editing tools, uses the global brush settings available in Tools, Configure Brush Shape... and in the Reconstruction

Toolbar. These include the shape of the brush ( so circular or square), its radius, and whether or not

it acts in all three dimensions ( ). You can also select whether your edits affect only the Main volume or both the Main and Aux volumes. (Most users will only want to edit one volume.) If the Main volume is the only target and even if the Aux volume is visible, the Main volume will still be edited.

This brush uses a threshold that allows you to specify which voxel values to be affected. This allows you to change ranges of values in an area to a new value, useful in cleaning up large anatomical areas.

There are a few other operations you can use to edit the volume. In the Tools, Volume menu there are commands for Thresholding, Flipping, and Rotating the volume. These are self-explanatory and can be used to fix misoriented data. These operations only affect the Main volume only.

The Smart Cut command is designed for removing entire chunks of volume based on the position of the cursor. It will create a cutting plane perpendicular to the current viewing orientation based on the closest edge of the window and remove everything from that plane to the edge. For example, if you place the cursor near the bottom-middle of the screen and activate the command, it will remove everything from the cursor to the bottom edge.

#### 1.5 Undoing

There are three ways to undo an edit. All of them only work with the Main volume, as it is the only one that can be edited. The first is with the standard Edit, Undo Last Edit menu item. This action will undo the last edit, and is the quickest and easiest way to undo single edits.

Another way is to use the Snapshot Volume tool in the Tools, Volume menu. Take a snapshot of the volume to save a copy of its contents in memory by choosing Edit, Take Snapshot of Volume. To restore the volume to this state, use the Edit, Restore Volume to Snapshot tool. This method is ideal for making 'milestone' versions of the the volume state. There are buttons for these commands on the Reconstruction

Toolbar:  $\blacksquare$  for taking a snapshot, and  $\blacksquare$  for restoring to it.

The third way is to use the Undo Volume. This can be envisioned as a separate volume containing all the voxels that have been changed. You can view these voxels by checking View, Undoable Voxels; they will be drawn with a blue highlight. By holding down the shift key and clicking with mouse button 2 with the Edit Voxel tool active on an undoable voxel in Display Window, you can undo all contiguous undoable voxels, like an undo flood fill. This is best suited for undoing contiguous areas of voxels.

### 1.6 Saving

After all your careful edits, you will want to save the changes to your data. tkmedit writes anatomical volumes out as COR- files. Choose File, Save Main Volume to overwrite the original source volume or File, Save Main Volume As... to specify a new location. You can save the Aux volume with corresponding commands in the File, Aux Volume submenu.

# 1.2. 2.0 Control points

Control points are used in surface construction. Every subject has an associated control.dat file in their tmp directory; this is the list of control points, in TalairachSpace, that is read in to tkmedit automatically upon start.

### **2.1 Display options**

Control points appear as green cross hairs in the Display Window. They can be turned off by unchecking **View->Control Points**.

#### 2.2 Making and deleting control points

Control points can be edited with the Edit Control Points tool, which can be activated by pushing the <sup>4</sup>+ button on the Main Toolbar, by choosing **Tools->Edit Control Points**, or by pressing the 'c' key when the mouse is in the Display Window.

To make a new control point, click anywhere in the Display Window with mouse button 2. To remove a control point, click near it with mouse button 3. Button 3 will remove the closest control point to the voxel clicked, within 3 voxels.

# 2.3 Saving

Whenever a new control point is made, it is automatically written to the control.dat file. However, removed control points are not removed from the file until you choose **File->Save Control Points** or quit normally with the **File->Quit** command.

# 1.3. 3.0 Selections

Selections are also called labels or ROIs. You can read or write selections from and to label files or create them from within tkmedit. Note that there is only one selection in tkmedit, even though it doesn't have to be contiguous.

# 3.1 Display options

Selections are drawn as a green overlay. This overlay can be turned off by unchecking **View**, **Selection** / **Label**.

### 3.2 Selecting and deselecting

You can select and deselect voxels with the Select Voxels Tool, which can be activated by pushing the select voxels, or by pressing the 's' key.

This tool is a brush tool, so it uses the global brush settings available in **Tools, Configure Brush Shape**... and in the Reconstruction Toolbar. You can think of selecting and deselecting voxels as painting your selection. To select voxels, use mouse button 2 to draw your selection and button 3 to erase it.

You can also use a flood fill to select and deselect contiguous areas. Open the configuration dialog box with the **Tools, Configure Flood Select**...command. Here, you have options for the making the fill 2D or 3D, settings the source, and the fuzziness and distance parameters.

•

**Source Volume**: Specifies which volume to use when looking for similar voxels, so that the Main, Aux, or segmentation volumes can be used to look for regions.

**Fuzziness**: Determines the level of similarity to consider when finding regions. If it is 0, only voxels with exactly the same value as the voxel clicked will be set. If it is greater than 0, this is the maximum value difference from the clicked voxel that a contiguous voxel can have to be considered in the same region.

**Distance**: Determines the maximum size of the region to fill from the clicked voxel. If it is 0, there will be no distance limit.

**3D**: Determines if the fill is in-plane only or will fill in three dimensions.

Once you have configured your fill, perform it with shift-mouse button-2 or 3; 2 will select an area, and 3 will deselect an area.

#### 3.3 Saving and loading

Tkmedit reads and writes label files. This file format is basically a list of points in RAS coordinate space with some header information. They can be loaded into tksurfer if they intersect with a surface.

# 1.4. 4.0 Surfaces

Tkmedit can read and display 3D surface files. The surfaces are intersected with the displayed slices and drawn overlaid onto the anatomical data.

### 4.1 Surface configurations

There are three possible surface configurations: Main, Original, and Pial. When you first load a surface, it is read into the Main configuration, and tkmedit looks for the other two configurations with similar names.

You can manually load in another surface as the Original or Pial configuration. All three configurations can be displayed at the same time, so you can compare how a surface looks in relation to another one.

### 4.2 Loading and unloading

Use the **File**, **Load Main Surface...** command to load in the Main configuration. This will also try and load Original and Pial versions of this surface. Use the **File**, **Load Surface Configuration...** submenu to load the **Original Vertices** or **Pial Vertices** manually.

Surfaces take up a lot of memory and they some time to intersect and draw. To release these resources when you are done with a surface, use the File, Unload Surface command.

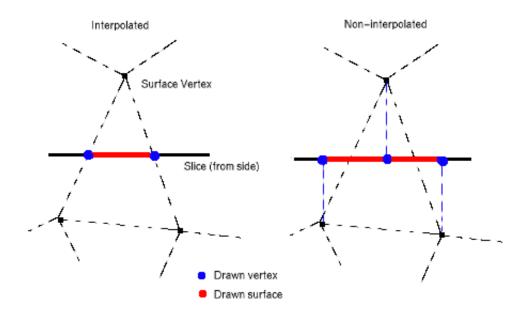
### 4.3 Display options

Each configuration is drawn in a different color. By default, the Main surface is drawn in yellow, the Original surface in green, and the Pial surface in red. You can change these colors in the **View**, **Configure...**, **Surface...** dialog box. You can also configure the width of the line used to draw the surface.

You can turn off surface display by unchecking the **View**, **Main Surface**, **View**, **Original Surface**, and **View**, **Pial Surface** items. You can also hide and show the surfaces with the buttons on the Reconstruction Toolbar ( $\bigcirc_{M}$  Main surface,  $\bigcirc_{D}$  Original surface, and  $\bigcirc_{P}$  Pial surface). You can display the actual

Toolbar (M Main surface, O Original surface, and P Pial surface). You can display the actual vertices of the surface by checking the **View**, **Surface Vertices** option.

There are two ways of determining which points to draw when intersecting the surface with the current slice; by using interpolation, and by using projection. Using interpolation will produce a smoother picture and a truer view of the surface intersection. However, the vertices that are drawn will be the interpolated intersection points of an edge and the slice, not the actual vertex. When projection is used, the vertices drawn will appear in their actual place in-plane. This diagram illustrates the methods better:



When **View**, **Interpolate Surface Vertices** is checked, the interpolation method is used. When it is unchecked, the projection method is used.

#### 4.4 Finding and showing vertices

Tkmedit has a few tools for finding and displaying specific surface vertices. If you know the vertex number of the vertex you are interested in, you can use the **Tools, Surface, Find Main Vertex**... command to enter the number and go to that vertex. Note that if the edge width is smaller than the slice width, the vertex may be 'embedded' in the slice and the cursor will not appear right over a vertex. There are corresponding commands for the Original and Pial surfaces as well.

You can also have tkmedit find the surface vertex closest to the cursor with the **Tools, Surface, Show Nearest Main Vertex** command. The vertex information will be printed to the console. There are corresponding commands for the Original and Pial surfaces as well.

# 1.5. 5.0 Functional volumes

Functional data can be viewed in tkmedit as an overlay onto the anatomical data (or any other volumetric data) or in a graph, as in a time course. Tkmedit can hold one volume for each purpose in memory at the same time. The volumes can be the same, i.e. a volume can be viewed as an overlay and as a time course, or separate.

# 5.1 Loading data

Loading Overlay and Time Course data is similar. Use the **File, Load Overlay Data**... or **File, Load Time Course Data**... command to load in a functional volume.

You can specify a registration file to use. If you do not, tkmedit will look in the same directory as the bfile data for a file called register.dat. A registration file is a matrix that defines the translation between anatomical RAS space and functional RAS space. Tkmedit uses this to align the functional volume with the anatomical volume.

The Time Course volume can also have an offset volume. This must be specified from the command line. See the reference section for details.

### 5.2 Overlay

### 5.2.1 Display options

The Overlay is displayed, as implied in its name, as a translucent overlay in the Display Window. Its default state uses a heat color scale, in which negative values are drawn in blue and positive values are drawn in red. A threshold is used to limit the values shown and define a color ramp so that more values close to the maximum or negative maximum are red or blue and values close to the minimum or negative minimum are yellow or teal.

The Overlay display can be turned off by unchecking the **View**, **Functional Overlay** option. Other aspects of the Overlay display are configured in the dialog box available in **View**, **Configure...**, **Functional Overlay**.... The following options are shown:

**Time Point / Condition**: Changes the currently shown time point or condition in the Display Window. The range is shown in parentheses next to the label. Note that a header file must be present in order to specify the division of data between time points in conditions, otherwise tkmedit assumes multiple time points and no conditions.

Truncate negative / positive values: Check to turn off the display of negative or positive values.

Reverse values: Check to display negative values in red and positive values in blue.

**Grayscale**: Check to use a grayscale color scale instead of the normal blue to red heat color scale. The lowest, most negative values will be shown in black and the highest, most positive values in white.

**Opaque**: Check to make the Overlay opaque.

**Ignore threshold**: Check to display all the values in the volume with a linear threshold, disabling the threshold values.

**Threshold minimum / midpoint / slope**: Configures the heat scale display. The minimum is the lowest value shown. The maximum is defined by ( (1/slope) + midpoint ).

Note that the Apply button must be clicked for changes to take effect.

The **View**, **Mask to Functional Overlay** option is available to only draw areas of the anatomical volume that are in the valid space of the Overlay volume. This gives you a better sense of the entire Overlay volume and how it relates to the anatomical volume.

### 5.2.2 Registration

If enabled, you can use the to register the Overlay volume, or align the Overlay volume with the anatomical data. This changes the registration file. Use the **Tools, fMRI, Register Functional Overlay** command to bring up a dialog box containing arrow buttons. Use these buttons along with the associated parameters to move the Overlay volume.

You can save your changes to the registration file with the **File, Save Overlay Registration** item. You can restore the registration to its original state with the **Tools, fMRI, Restore Overlay Registration**. Use the **Tools, fMRI, Set Registration to Identity** command to set the registration matrix to the identity matrix.

Note that since registration can affect the validity of any data analysis you perform, the MGH-NMR processing stream only allows registration at a specific point in the stream. Registration may not be available at all times. It can be enabled from the command line at start-up time. See the reference section for details.

### 5.3 Time course

# 5.3.1 Graph

The Time Course is displayed in a separate graph window. The graph will only be shown if the Time Course volume has more than one time point. Clicking on a voxel in the Display Window finds the corresponding functional voxel based on the registration and displays the values at that voxel for all time points in the graph window.

If there are multiple conditions defined in the header file, each condition will show up as a separate line in the graph. The legend on the right side of the graph window displays what line corresponds to what condition. By moving the mouse over a condition label in the legend, the corresponding line in the graph will be highlighted. This is useful for picking out one condition in a crowded graph.

You can also zoom into the graph. Click and drag with button 2 to draw a box around the area of interest. The graph will be resized around that box. Click with button 3 to zoom back out.

If the same volume is being used for the Overlay and the Time Course, clicking in the graph window will change the time point in the Overlay display to what x point was clicked. You can use this method to navigate the Overlay volume.

The selection / label can be used to specify an average area to graph. To graph a selection / label as an average, draw or load a selection and choose **Tools, fMRI, Graph Current Selection**. The title at the top of the graph will change to show that a selection is being graphed instead of a single voxel. The contents of a selection can be printed to a text file in a chart form with the **Tools, fMRI, Print Time Course Summary to File**... command. You can use this data in a separate graphic program.

The contents of the graph window can be written to a Postscript file with the **Tools, fMRI, Save Time Course Graph to Postscript File**... command. It can then be included in another document or printed.

### 5.3.2 Display options

The graph can be configured with the **View, Configure..., Time Course Graph** dialog box. The top part of this dialog is a list of mappings between colors and conditions. You will see as many rows as the Time Course volume has conditions. You can assign a color to each condition with the number field on the right or hide colors / conditions with the checkbox on the left. By default, condition 0 is hidden. You can also enter a name for the color in the column on the left. This name will show up in the graph window in the legend on the right.

The bottom part has the following options:

**Show error bars**: If this box is checked and error data is present, vertical errors bars will be drawn at each data point, signifying the possible error for each value.

**Automatically size graph**: If checked, the graph axes will automatically resize according to the range of values. If unchecked, the axes will stay static.

**Subtract pre-stim average**: If checked, the average value of the values before the stimulus (time second = 0) for each condition is subtracted from all values. This effectively adjusts the y axis to normalize for pre-stim values.

**Show percent change**: If an offset volume is available, this will show the percent change for each value instead of the raw value itself.

**Number of pre-stim points**: Originally defined by the header file, this value lets you adjust the time = 0 value.

**Time resolution**: This is defined by the header file and is used to calculate how many seconds are in each time point.

Note that the Apply button must be clicked for changes to take effect.

# 1.6. 6.0 Segmentations

A segmentation is a volume whose values represent an index of an anatomical structure or label to which the corresponding voxel in the main anatomical volume belongs. The structures are listed in a lookup table. This table also specifies the colors the structures should appear in, and is more commonly called a color table.

# 6.1 Loading

Both a volume and a color table must be loaded to view a segmentation. A segmentation data file is treated the same as a normal anatomical volume, so it can be a COR- volume or any other supported file type. The color table assigns a name and color to each index. You can load both items by choosing **File, Load Segmentation...** 

You can create a new segmentation with the **File**, **New Segmentation...** command. You will still need to enter a color table to use.

You can import a surface annotation with the **File, Import Surface Annotation as Segmentation...** command. A surface annotation is a segmentation of the surface, so that every vertex in the surface has an associated label. Tkmedit will look up the corresponding voxel for each vertex and assign the label value to that voxel in a new segmentation.

Like the anatomical volumes, tkmedit can have a Main or Aux segmentation. Each can have its own color table. To load Aux segmentations use the commands in **File**, **Aux Segmentation**. As with the anatomical volumes, only the Main segmentation volume can be edited.

### 6.2 Display options

Segmentations are drawn as a colored overlay in the Display Window. The colors are defined in the color lookup table file specified at load time. The opacity of the overlay can be configured in the dialog box brought up by choosing **View**, **Configure...**, **Segmentation Display...** overlay can be hidden by unchecking View, Segmentation Overlay. You can toggle between the Main and Aux segmentations with the **View**, **Aux Segmentation Overlay** command.

If the **Segmentation Label Volume Count** option in the **View** menu is checked, when the Edit Segmentation tool is active and the left button is used to set the cursor, tkmedit will count the number of contiguous voxels in the label clicked and display the result in the Cursor Information area in the Tools Window. This operation may take anywhere from 1 to 10 seconds depending on the size of the label clicked, so be aware that tkmedit may seem unresponsive for this length of time after setting the cursor with this option activated.

#### 6.3 Editing

A segmentation can be edited with the Edit Segmentation tool, which can be activated by pushing the

button on the Main Toolbar, by choosing **Tools, Edit Segmentation**, or by pressing the 'g' key.

By clicking with button 2 with this tool, you can 'paint' a label in the segmentation. The color used can be set in the dialog box brought up by choosing **Tools, Configure Segmentation Brush...** This tool is a brush tool, so it uses the global brush settings available in **Tools, Configure Brush Shape...** and in the Reconstruction Toolbar. Clicking with button 3 will 'erase' segmentation labels (by setting the label values to 0).

The painting function is similar to the Edit Voxels tool: the clicked voxel, and those around it depending on the brush radius, are set to the value specified in the **Configure Segmentation Brush** dialog. Clicking on a voxel with button 2 and the control and shift key held down will set the current painting color to the color of the clicked voxel. Edits can be undone with the **Edit, Undo Last Edit** menu item.

A flood fill is also available, activated by shift-clicking mouse button 2 or 3. This tool fills areas with values equal to or similar to the voxel clicked. It looks for contiguous areas in the source volume to fill in the segmentation with the selected color. It is configured in the **Configure Segmentation Brush** dialog. It has parameters for 3D, the source volume, fuzziness, distance.

**Source Volume:** Specifies which volume to use when looking for similar voxels, so that the Main, Aux, or segmentation volume can be used to look for regions. **Fuzziness:** Determines the level of similarity to consider when finding regions. If it is 0, only voxels with exactly the same value as the voxel clicked will be set. If it is greater than 0, this is the maximum value difference from the clicked voxel that a contiguous voxel can have to be considered in the same region. **Distance:** Determines the maximum size of the region to fill from the clicked voxel. If it is 0, there will be no distance limit. **3D:** Determines if the fill is in-plane only or will fill in three dimensions.

A fill can be undone with the Edit, Undo Last Edit menu item.

# 6.4 Saving

To save changes to a segmentation, choose **File, Save Segmentation** to overwrite the original segmentation volume or File, Save Segmentation As... to specify a new directory in which to write the COR- volume.

# 1.7. 7.0 DTI

Diffusion Tensor Images can be viewed as an overlay. The red, blue, and green colors represent the direction of the vectors.

# 7.1 Loading

DTI volume data sets consist of two volume: a three frame eigenvector volume (EV volume) and a fractional anisotropy volume (FA volume). These must be specified in the **File, DTI, Load DTI Volumes...** dialog box. There will be a considerable delay while loading DTI volumes as tkmedit does some pre-processing.

The eigenvector volume describes the directionality of diffusion in a particular direction (with the first EV being the direction of greatest mean diffusion within each voxel), and the FA volume describes how homogeneous the diffusion is in a particular direction (high values mean that diffusion is primarily occurring in a single direction- and FA is calculated from something like the variance of the three eigenvalues).

# 7.2 Display options

Head points are displayed in full color. The red, green, and blue spectrums represent the directionality of the EV volume. The opacity of the overlay can be configured in the dialog box brought up by choosing **View, Configure..., DTI Display...** The overlay can be hidden by unchecking **View, DTI Overlay**.

# 1.8.8.0 Head points

Head points data represent the location sensors during an MEG/EEG scan.

# 8.1 Loading

Head points data consist of two files: a list of points and their labels, and a transform file. These must be specified in the **File, Load Head Points...** to load head points data.

#### 8.2 Display options

Head points are displayed as green diamonds in the Display Window. They can be hidden by unchecking **View, Head Points**. You can select a head point by clicking near it. The selected head point is drawn in red and its label is shown in the Tools Window.

When you are viewing the maximum intensity projection (by checking **View**, **Maximum Intensity Projection**), you will see all the head points. Note that in this view you can select head points in all planes, not just the current one.

### 8.3 Editing

You can edit the label of the currently selected head point by choosing **Tools, Head Points, Edit Current Head Point Label....** Enter the new name in the dialog box. You will need to click the point again in the Display Window to see the new name in the Tools Window.

You can also modify the registration transform. Use the **Tools, Head Points, Register Head Points...** command to bring up a dialog box containing arrow buttons. Use these buttons along with the associated parameters to move the head points. It is easiest if you do this in the maximum intensity projection view.

### 8.4 Saving

Save changes to the head points labels by choosing **File**, **Save Head Points**. Save changes to the registration transform by choosing **File**, **Save Head Points Transform**. Both commands overwrite the original files.

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# 1. Reference

# 1.1. 1.0 Mouse commands

# **Mouse Controls for Tools**

	Key Shortcut	Button 1	Button 2	Button 3
Navigation Tool	n	Clicking and dragging pans the view across the current slice. No effect when the zoom level is 1.	Clicking once in the top half of the window increases the slice by 1, clicking in the bottom decreases the slice. Dragging up increases the slice, dragging down decreases it.	Clicking once in the top half of the window zooms in, clicking in the bottom zooms out. Dragging up zooms in continuously, dragging down zooms out.
Select Voxel Tool	S	Clicking sets the cursor.	Clicking adds a voxel to the selection. Sets the cursor when the button is released. Shift+click performs a flood select.	Clicking removes a voxel from the selection. Sets the cursor when the button is released.Shift+click performs a flood deselect.
Edit Voxel Tool	a	Clicking sets the cursor.	Clicking edits a voxel with the Button 2 settings. Shift+click undoes a undoable voxel region. Sets the cursor when the button is released.	Clicking edits a voxel with the Button 3 settings.
Edit Segmentation Tool	g	Clicking sets the cursor. If the ROI Volume Count display option is on, displays the ROI volume in the Tools Window.	Clicking paints a segmentation voxel. Shift+click performs a flood fill. Shift+Ctrl+click sets the color to the color of the voxel clicked. Sets the cursor when the button is released.	Clicking erases a segmentation voxel (sets to label 0). Sets the cursor when the button is released. Shift+click performs a flood erase.
Edit Control Points Tool	с	Clicking sets the cursor.	Clicking makes a new control point. Sets the cursor when the button is released.	Clicking removes nearest control point. Sets the cursor when the button is released.

# **Mouse Controls for Zooming**

Control+Button 1:	Zoom in, doubling the zoom level
Control+Button 2:	When zoomed in, sets the cursor and re-centers the view around the new cursor without changing the zoom level.
Control+Button 3:	Zoom out, halving the zoom level

# 1.2. 2.0 Menus

# **2.1 File**

Load Main Volume	Loads the Main volume from a directory of COR- files.
Load Aux Volume	Loads the Auxiliary volume from a directory of COR- files.
Import Main Volume from File	Reads a specified file as the Main volume.
Import Aux Volume from File	Reads a specified file as the Aux volume.
Load Transform for Main Volume	Loads an LTA or XFM file to use as the display transform for the Main volume.
Load Transform for Aux Volume	Loads an LTA or XFM file to use as the display transform for the Aux volume.
Unload Transform for Main Volume	Removes the display transform for the Main volume.
Unload Transform for Aux Volume	Removes the display transform for the Main volume.
Save Main Volume	Saves the Main volume in its original COR- file directory.
Save Main Volume As	Saves the Main volume in a new directory.
Load Main Surface	Loads a surface overlay (e.g. orig.?h, white.?h, pial.?h), looking for other configurations and loading them available.
Load Surface Configuration	Loads an alternate (Original or Pial) surface configuration.
Unload Surface	Removes the surface and all configurations.
Load Overlay Data	Reads in coregistered functional data for in-plane overlay.

Load Time Course Data	Loads in coregistered functional data for graphing over multiple time points.
Save Overlay Registration	If registration has been enabled, this will save the registration to the register.dat file.
Load Segmentation	Loads a segmentation, requiring a COR- file directory and a color lookup table file.
Save Segmentation	Saves the segmentation in its original COR- file directory.
Save Segmentation As	Saves the segmentation in a new directory.
Load Label	Reads in a cortical label file as the selection.
Save Label As	Saves the selection as a cortical label file.
Load Head Points	Loads a head points transform file and a list of MEG/EEG sensor points.
Save Head Point Transform	Saves an edited head points transform file.
Save Head Points	Saves head points.
Save Control Points	Saves control points to control.dat file. (Done automatically.)
Quit	Exits the program.

# 2.1.1 Load surface configuration

Original Vertices	Load the Original vertex set for an existing surface
Pial Vertices	Load the Pial vertex set for an existing surface

# 2.1.2 Transforms

Load Transform for Main Volume	Load a display transform for the Main anatomical volume.
Load Transform for Aux Volume	Load a display transform for the Aux anatomical volume.
Unload Transform for Main Volume	Unload a display transform from the Main anatomical volume.
Unload Transform for Aux Volume	Unload a display transform from the Aux anatomical volume.

# 2.1.3 Label

Load Label	Load a label file and add it to the current selection.
Save Label As	Save the current selection as a label file.

# 2.1.4 GCA

Load GCA	Load GCA volumes.
Save GCA	Save GCA volumes.
Unload GCA	Unload GCA volumes.

# 2.1.5 Head points

Load Head Points	Load a list of head points.
Save Head Point Transform	Save a modified head point transform (overwrites existing).
Save Head Points	Save list of head points.

# 2.1.6 DTI

Load DTI Volumes... Load DTI volumes.

# 2.1.7 Edit

Undo Last Edit	Undoes the last volume or segmentation edit.
Take Snapshot of Volume	Makes a copy of the current Main volume.
Restore Volume to Snapshot	Restores the Main volume to the saved copy.
Clear Label / Selection	Clears the current selection.
Clear Undo Volume	Clears the Undo Volume.

# 2.2 View

View Configurations	Shows a submenu of view configurations.
Tool Bars	Shows a submenu of available toolbars. Check or uncheck the toolbar in this submenu to show or hide the toolbar.
Information	Shows a submenu of available information area items such as coordinate labels. Check or uncheck the item in this submenu to show or hide the item.
Configure	Shows a submenu of viewing characteristics that can be configured.
Anatomical Volume	Check to display the anatomical volume.
Main Volume	Toggle between showing the Main or Aux volume.

# Aux Volume

Maximum Intensity Projection	Check to display the maximum intensity projection of the current volume, or the highest values in the current plane for each voxel.
Main Surface	Check to display the Main surface configuration.
Original Surface	Check to display the Original surface configuration.
Pial Surface	Check to display the Pial surface configuration.
Surface Vertices	Check to display the surface vertices on top of the surface overlay.
Interpolate Surface Vertices	Check to use interpolation to draw surface vertices. Uncheck to use projection.
Functional Overlay	Check to show the functional overlay volume.
Functional Scale Color Bar	Check to show the color scale bar for the overlay.
Mask to Functional Overlay	Check to mask the anatomical volume to valid functional overlay space.
Show Histogram Percent Change	Check to show the window displaying changes in VLI label volumes.
Segmentation Overlay	Check to show the segmentation overlay.
Segmentation Label Volume Count	Check to count the volume of a label in the segmentation when clicking with the left mouse button when the Edit Segmentation Tool is selected.
Selection / Label	Check to show the selection.
Head Points	Check to show the head points.
Control Points	Check to show the control points.
Cursor	Check to show the cursor.
Axes	Check to show the coordinate space axes.
Undoable Voxels	Check to show the undoable voxels in the Undo Volume.

# 2.2.1 View configurations

Single View	Displays one pane with a single orientation.
Multiple Orientations	Displays four panes, allowing all orientations to be viewed at the same time around the cursor.
Mosaic	Displays a 4x4 grid of different slices in the same orientation.

# 2.2.2 Tool bars

Main	Check to display the Main toolbar, which has the following controls: Tool Selector (Navigation, Select Voxels, Edit Voxels, Edit Segmentation, Edit Control Points), View Configuration (Single, Multiple, Mosaic), Surface Display (Main, Original, Pial), Current Volume (Main, Aux)
Navigation	Check to display the Navigation toolbar, which has the following controls: Orientation (Coronal, Horizontal, Sagittal), Slice number, Zoom Out, Zoom In, Zoom Level, Save Point, Go To Saved Point, Linked Cursors
Reconstruction	Check to display the Reconstruction toolbar, which has the following controls: Brush shape (Circular, Square), 3D brush, Radius, Make Snapshot, Restore to Snapshot, TkTimer on

### 2.2.3 Information

Volume Index Coordinates	Check to show the volume index coordinates (0255).
Volume RAS Coordinates	Check to show the RAS coordinates. The origin is at the center of the volume.
Volume Scanner Coordinates	Check to show the scanner coordinates, usually the RAS coordinates.
MNI Coordinates	Check to show the MNI Talairach coordinates. This was the old Talairach coordinate system, but has been replaced by a slightly modified version.
Talairach Coordinates	Check to show the Talairach coordinates, determined by the talairach.xfm file for a subject. May not be available if this file is missing.
Volume Value	Check to show the value of the Main volume voxel.
Aux Volume Value	Check to show the value of the Aux volume voxel.
Functional Overlay Index Coordinates	Check to show the functional overlay volume index coordinates.
Functional Overlay RAS Coordinates	Check to show the functional overlay RAS coordinates. The origin is at the center of the functional volume.
Functional Overlay Value	Check to show the value of the functional overlay volume voxel.
Segmentation Label	Check to show the name of the ROI in the Main segmentation that the voxel is a part of.
Aux Segmentation Label	Check to show the name of the ROI in the Aux segmentation that the voxel is a part of.
Head Point Label	Check to show the name of the head point nearest the cursor or mouse.
Surface Distance	Check to show the distance from the last cursor location to the present mouse or cursor location. (Used in setting surface distance values.)

# 2.2.4 Configure...

Brightness / Contrast	Opens a dialog box in which you can set the brightness and contrast of the Main and Aux volumes.
Cursor	Opens a dialog box in which you can change the color and shape of the cursor.
Surface	Opens a dialog box in which you can change the color and line width of each surface configuration.
Functional Overlay	Opens a dialog box in which you can change various characteristics of the functional overlay.
Time Course Graph	Opens a dialog box in which you can change various characteristics of the time course graph.
Segmentation Display	Opens a dialog box in which you can change the overlay opacity of the segmentation display.
DTI Display	Opens a dialog box in which you can change the overlay opacity of the DTI display.

# 2.3 Tools

Navigate	Changes the current tool to the Navigation Tool.
Select Voxels	Changes the current tool to the Select Voxels Tool.
Edit Voxels	Changes the current tool to the Edit Voxels Tool.
Edit Segmentation	Changes the current tool to the Edit Segmentation Tool.
Edit Control Points	Changes the current tool to the Edit Control Points Tool.
Configure Brush Info	Opens a dialog box in which you can change the size, shape, and depth of the brush.
Configure Volume Brush	Opens a dialog box in which you can change the threshold and color values for the Edit Voxels Tool.
Configure Segmentation Brush	Opens a dialog box in which you can change the color and fill settings for the Edit Segmentation Tool.
Configure Flood Select	Opens a dialog box in which you can change the parameters for the flood action of the Select Voxels Tool.
Save Point	Saves the current cursor so that tksurfer can use its Go To Point function to go to that point.
Goto Saved Point	Sets the cursor to the one last saved in tksurfer for this subject.
Goto Point	Opens a dialog box in which you can enter specific numerical values in multiple coordinate systems and go to that point.
Volume	Opens a submenu of tools and commands for anatomical data.
Surface	Opens a submenu of tools and commands for surface data.
fMRI	Opens a submenu of tools and commands for to functional data.
Head Points	Opens a submenu of tools and commands for EEG/MEG head point data.
Save RGB	Saves the contents of the Display Window to an RGB file.
Save RGB Series	Opens a dialog box in which you can tell tkmedit to automatically scroll through a series of slices and save their images to RGB files.

# 2.3.1 Volume

Threshold Volume	A tool for changing all Main volume values above or below a specific value to a new value.
Flip Volume	A tool for flipping the Main volume, creating a mirror image.
Rotate Volume	A tool for rotating the Main volume any number of degrees around a main axis.
Smart Cut	A tool for setting large sections of the Main anatomical volume to 0. See the Working With Data section for details.

# 2.3.2 Surface

Show Nearest Main Vertex	Sets the cursor to the point of the nearest vertex on the Main surface configuration.
Show Nearest Original Vertex	Sets the cursor to the point of the nearest vertex on the Original surface configuration.
Show Nearest Pial Vertex	Sets the cursor to the point of the nearest vertex on the Pial surface configuration.
Show Nearest Main Surface Edge	Sets the cursor to the point of the nearest interpolated vertex on the Main surface configuration.
Show Nearest Original Surface Edge	Sets the cursor to the point of the nearest interpolated vertex on the Original surface configuration.
Show Nearest Pial Surface Edge	Sets the cursor to the point of the nearest interpolated vertex on the Pial surface configuration.
Find Main Vertex	Opens a dialog in which you can enter a vertex index on the Main surface. The cursor will be set to that index.
Find Original Vertex	Opens a dialog in which you can enter a vertex index on the Original surface. The cursor will be set to that index.
Find Pial Vertex	Opens a dialog in which you can enter a vertex index on the Pial surface. The cursor will be set to that index.
Set Vertex Distance at Cursor	Sets the value in the Surface Distance information field for the closest vertex in the Main surface.
Average Vertex Positions	Opens a dialog in which you can execute a command to average surface vertex positions in the Main surface.

# 2.3.3 fMRI

Select Contiguous Voxels by Func Value	Selects contiguous voxels to the cursor whose corresponding functional values are greater than or equal to the functional value at the cursor.
Select Contiguous Voxels by Threshold	Selects contiguous voxels to the cursor whose corresponding functional values are greater than or equal to the minimum functional threshold value.
Select Functional Voxel	Selects an entire functional voxel.
Register Functional Overlay	A tool for aligning (registering) the functional overlay data with the anatomical data.
Restore Overlay Registration	Restores the overlay registration to its original value.
Set Registration to Identity	Sets the overlay registration matrix to the identity matrix, removing all transformations.
Graph Current Selection	Graphs the average of all voxel values in the selection in the Time Course graph window.
Print Time Course Summary to File	Creates a file containing a table version of the time course information for the current voxel or selection.
Save Time Course Graph to Postscript File	Saves the contents of the Time Course graph window to a Postscript file suitable for printing or including in other documents.

# 2.3.4 Segmentation

Select Current Label	Selects all the anatomical voxels in the segmentation label under the cursor.
Recompute Segmentation	Recomputes the Main segmentation using the Main anatomical volume and the GCA volume.
Graph Current Label Average	Graphs the average time course for all the anatomical voxels in the segmentation label under the cursor. Only works with a time course loaded.

# 2.3.5 Head points

Restore Head Points	Restores the head points registration to its original value.
Edit Current Head Point Label	Edits the label of the head point closest to the cursor.
Register Head Points	A tool for aligning the head point data with the anatomical data.

# 1.3. 3.0 File name substitution

Whenever a file name is required to load or save data, the following system is used. First, on startup, tkmedit attempts to acquire a User Home Directory and a Subject Home Directory. The User Home Directory is always the current directory from which tkmedit was launched. The Subject Home Directory is based on the way in which the anatomical data was loaded. If the default tkmedit loading method is used, it is the concatenated contents of \$SUBJECTS\_DIR and the subject name. If the -f switch is used, it is the value of that parameter.

If the first character of the file name is ~ (tilde) or . (period), it is substituted with the Subject Home Directory or User Home Directory, respectively. If the first character is another alphanumeric character, a default location directory is prepended to the file name. If the first character is / (slash), it is left alone. This behavior is summarized below:

If the first character of FILENAME is	The final file name is
~ (tilde)	SubjectHomeDir/FILENAME
. (period)	UserHomeDir/FILENAME
/ (slash)	FILENAME
other character	SubjectHomeDir/Subdirectory/FILENAME

...where FILENAME is the file name that was input.

The subdirectory used in the last substitution depends on the file type being loaded, and is appropriate for the standard NMR center directory structure. These are:

Data Type	Subdirectory
Functional data	fmri
Head Points	bem
Surface	surf
Anatomical	mri
ROI/Label	label
RGB	image/rgb
Control Points (control.dat)	tmp
Edit (edit.dat)	tmp
Segmentation	mri
Color Table	\$CSURF_DIR
Label	label
Anatomical Transform	mri/transforms

# 1.4. 4.0 Command line options

# 4.1 Script

To automatically run a script, use the -tcl option, passing a script name. Tkmedit still requires anatomical data to be loaded before doing anything.

tkmedit -tcl SCRIPT\_NAME

# 4.2 Anatomical volume

There are two ways to load anatomical data from the command line. The first is the default tkmedit mode for loading COR- volumes.

tkmedit SUBJECT IMAGE\_TYPE

Where SUBJECT is a subject directory relative to the value of the SUBJECTS\_DIR environment variable, and IMAGE\_TYPE is a subdirectory of  $SUBJECTS_DIR/<$ subject name>/mri. i.e.

tkmedit <subject name> T1

looks in  ${SUBJECTS_DIR}/<subject name>/mri/T1. If there is a COR- volume in this directory, or another kind of readable data, it will be loaded as the Main anatomical volume.$ 

Another way to load anatomical data is with an explicit path or file name using the -f switch:

tkmedit -f PATH/TO/DATA/[FILENAME]

This method looks in the given directory or file for any recognizable data. It will be loaded as the Main volume.

The Aux volume can be loaded with the -aux switch, i.e.

tkmedit <subject name> T1 -aux wm

This looks in \${SUBJECTS\_DIR}/<subject name>/mri/wm for a COR- volume.

Note that tkmedit will not load without specifying a Main anatomical volume with the subject/image type method or the -f method.

### 4.3 Surface

To load a surface from the command line, add the name after the subject:

tkmedit <subject name> IMAGE\_TYPE SURFACE

or

tkmedit -f /path/to/volume/data SURFACE

#### 4.4 Functional volume

Overlay data is loaded with the -overlay switch and time course data with the -timecourse option, with the concatenated path and stem as the argument i.e.:

tkmedit -overlay DATA\_DIRECTORY/STEM -timecourse DATA\_DIRECTORY/STEM

where STEM is the portion of the bfloat file data that comes before the \_xxx slice number. i.e. the stem of a file set named minsig\_000.bfloat through minsig\_015.bfloat is minsig.

To enable functional registration, pass -register on the command line with no arguments.

To specify a registration file that is not in the same directory as the functional volume, use the -overlay-reg or -timecourse-reg option with the registration file as the argument, i.e:

tkmedit -overlay DATA\_DIRECTORY/STEM -overlay-reg /PATH/TO/REGISTRATION\_FILE.DAT

To load an offset volume for the Time Course volume, specify it the same way as the Time Course volume but with the -timecourse-offset switch:

tkmedit -timecourse-offset DATA\_DIRECTORY/STEM

### 4.5 Segmentation

Use the -segmentation switch to load a COR- volume representing a segmentation. A color file is also necessary.

tkmedit -segmentation COR\_DIRECTORY COLOR\_FILE

The COR\_DIRECTORY is the directory of the COR- files and the COLOR\_FILE is the file to use as a color lookup table.

You can specify the opacity of the segmentation overlay with the -segmentation-opacity option:

tkmedit -segmentation COR\_DIRECTORY COLOR\_FILE -segmentation-opacity 0.6

# 4.6 Head Points

Use the -headpts switch to load a head points file. This is a file of points indicating MEG/EEG sensor placement. A transform file is also required and can be specified on the command line.

tkmedit -headpts POINTS\_FILE TRANSFORM\_FILE

# 1.5. 5.0 Crash recovery

If the test of the Main volume, the volume will be saved in the /tmp directory. This can be reloaded by loading the volume from that directory, i.e.

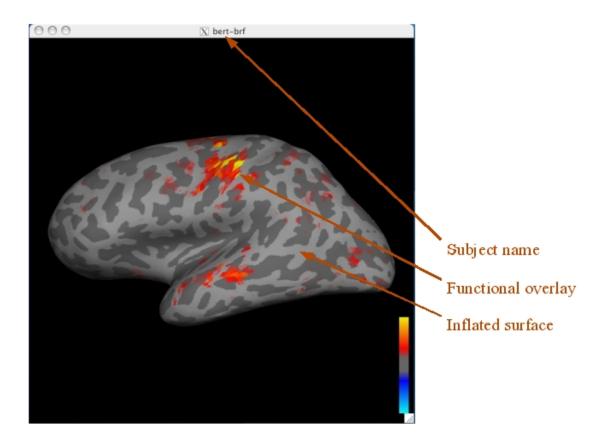
# tkmedit -f /tmp

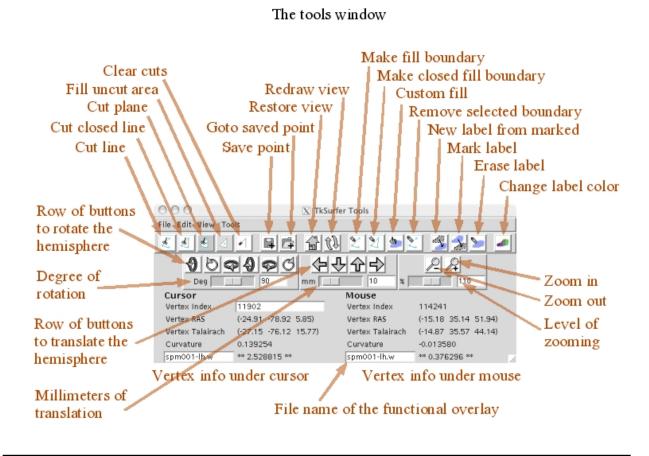
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# **1. Tksurfer interface**

The display window





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# 1. General usage

# 1.1. 1.0 Quick start

Tksurfer can be started in any of three ways: launching it from FreeSurfer with the Surface button, using the tksurfer-sess script, and calling it from the command line. Depending on which method you use, you may see different types of data loaded, but all methods require a surface data set to be loaded. Other types of data can be loaded from the File menu.

To explore your data, you can move the mouse over the picture of the surface (the Display Window) and see information about the area under the mouse in the window on the bottom (under the Mouse section of the tksurfer Tools Window). You can change the view with the navigation buttons (under the toolbar, with icons of various arrows). To save pictures of your data, go to the Tools menu and select the Save RGB As... item near the bottom.

By default, tksurfer does not redraw the display window unless you perform a command that changes the view. This means that if another window obscures the display window and then is moved, the display window will not automatically update. This because of the time it takes to redraw an average surface. To explicitly redraw the view, just click on the Redraw button.

# 1.2. 2.0 Viewing area

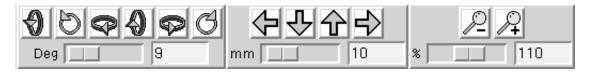
Tksurfer consists of two windows. The top window, called the Display Window, shows the currently loaded surface and its overlays. The bottom window, called the Tools Window, contains controls and feedback information. Here is the interface.

Information about the currently displayed data can be obtained by moving the mouse over the Display window or by clicking to select a vertex. As the mouse is moved in the display window, the Mouse section of the Tools window will display information about the vertex under the tip of the Mouse arrow. Information about the vertex clicked on is displayed in the Cursor section.

The view is configured with various dialog boxes and options available from the **View** menu. From this menu, you can configure the toolbars and labels available in the Tools window, various aspects of the surface, curvature, and overlays, and the time course graph window.

# 1.3. 3.0 Navigation

Tksurfer displays the surface in a 3D environment. You may move the surface with the buttons in the movement area of the Tools window.

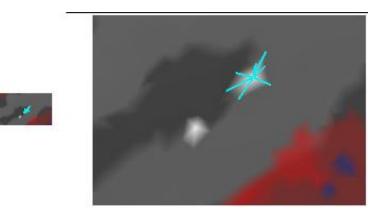


The buttons in the left section rotate the hemisphere, the ones in the middle translate it, and the ones on the right zoom in and out. The sliders adjust the number of degrees of each movement. You can also zoom in and out by holding down the control key and clicking with button 1 or 3. Button 1 zooms in, and button 3 zooms out. The vertex you click becomes the center vertex in the new view.

# 1.4. 4.0 Cursor and marked vertices

Clicking on a vertex sets the cursor. The edges touching the vertex are drawn in a cyan color.

The cursor is drawn every time the mouse is clicked without redrawing the whole window. However, clicking also marks vertices, adding each clicked vertex to a list of marked ones. These are drawn in a white color, but only show up after the screen is redrawn. This screen shot shows two marked vertices and the cursor, at normal zoom level and zoomed in.



To see this behavior, start tksurfer with any subject and surface. Click with button 1 on the surface and see the cyan cursor appear. Now press the Redraw button (or type Alt+R while the mouse is in the display window) and see the white highlight under it. Now click around some more without hitting redraw and see

the cyan cursor move. Now press Redraw (0, 0, 0), and see how all the points you previously clicked are drawn in white.

Most tools require at least two marked vertices, and some as many as four. The icons for the cut tools give hints as to how many you need. To clear the marked vertices (unmark them), click with button 3. Note that the white highlights will not disappear until Redraw is pressed.

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# 1. Working with data

# 1.1. 1.0 Surfaces

Tksurfer holds one main surface. This surface structure has the surface vertex information as well as other information related to the surface, such as additional values for each vertex. Tksurfer can also hold four more surface configurations. These are additional sets of vertex information and locations, but not the other surface information. These configurations are called Inflated, White, Pial, and Original Vertices.

# 1.1 Loading surfaces

The main surface must always be loaded. To load more configurations, use the **File->Load Surface Configurations...** submenu. You can also load a different main surface configuration in this menu. This will replace the main vertex information but not the other surface information.

# 1.2 Surface display options

You can select the currently shown surface configuration with the **View->Surface Configuration** submenu. Only loaded configurations will be available.

You can enable a wireframe overlay with the **View->Wireframe Overlay** option. This will draw the surface polygon edges in blue, so you can see exactly where the edges and vertices are.

# 1.3 Surface related tools

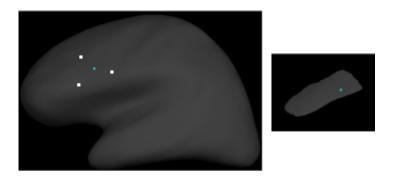
There are four tools available to cut surfaces. These tools are available in the Tools menu and in the Main

toolbar. The Cut Line tool ( $\overset{\bigstar}{\leq}$ ) calculates a line along all the marked vertices in the order that they were

marked and cuts the vertices on that line. The Cut Closed Line tool ( $\checkmark$ ) does the same, but closes the line from the last marked vertex to the first. In the graphics below, the first image shows some marked vertices. The second shows the results after a Cut Line action, and the third shows the results after a Cut Closed Line action.



The Cut Plane tool requires exactly four marked vertices. The first three define three points on the plane and the fourth defines a point inside the plane. This image shows this process. The surface image has been scaled down to show that the entire surface has been cut away in the second image.



The Cut Area tool is used to cut away one incontiguous area from another. Click in the area you wish to keep and perform the action. The other area will be cut. All cut vertices can be restored with the Clear Cuts tool.

# **1.4 Saving surfaces**

Some tools and other functions change the surface vertex information. Other tools change the vertex values and other information. To save these changes to the surface, use the **File->Save Surface** option to overwrite the original surface file and **File->Save Surface** As... to save it a new location. Note that this does not save the extra surface configurations; these are not part of the main surface structure.

# **1.2. 2.0 Curvature**

### 2.1 Loading curvature files

Use the File->Curvature->Load Curvature... item to load a curvature file.

# 2.2 Curvature display options

The curvature is displayed as either a green/red color scale or a binary gray background. When no overlay is displayed, the green/red color scale is used. When an overlay is active, the simpler gray is used.

The **View->Configure->Configure Curvature Display** dialog box allows you to set the midpoint and slope of the curvature color scale. This affects both the green/red and the gray display.

# 2.3 Saving curvature files

Use the **File->Curvature->Save Curvature** to save the curvature values and overwrite the original file, or **File->Curvature->Save Curvature As...** to save a new curvature file in a new location.

# 1.3. 3.0 Overlay

An overlay is a set of values, 0 or 1 per vertex, displayed with a color scale. It is normally used as a functional activation overlay, but can really be any floating point value per voxel. The source volume can be a .w file, a file containing one value per vertex, previously registered with the volume. It can also be a binary volume or bfile. These are volumes of short integers (.bshort) or floating point values (.bfloat) with a corresponding registration file. Binary volumes can contain multiple time points and conditions.

Overlays can be loaded into any of five layers. Tksurfer can display one overlay layer at a time.

### 3.1 Loading overlay files

To load an overlay file, use the **File->Load Overlay...** item. You will be prompted for the overlay file name and the layer in which to load the file. Chose the .w file or a .bshort or .bfloat file if you are loading a binary volume.

If you are loading a binary volume, you will be prompted for the stem of the volume and a registration file. You may leave the registration file field blank if there is a registration file in the same directory as the binary volume named "register.dat."

### 3.2 Overlay file display options

To select which overlay to show, use the **View->Overlay Layer** submenu. The field names are automatically set to the file name loaded. You can change this name by typing a new one into the information area in the Tool window.

The display for the current overlay can be configured in the **View->Configure->Configure Overlay Display** dialog. If the data you have loaded has multiple time points or conditions, you can select which one to show with the **Time Point** and **Condition** fields. You can select the color scale to use with the radio buttons in the top area of the dialog. The **Truncate** option can be checked to turn off the display of negative values. Check the **Reverse** option to reverse the sign of the values as they are drawn in the color scale. **Inverse and Complex are reserved for future upgrades.** Also see SettingTheOverlayColorScale.

Click the **Apply** button to see your changes.

# 3.3 Saving overlay files

An overlay can only be written out to a .w file; binary volumes cannot be written from overlays. To write a .w file, choose **File->Save Overlay As...**, and choose a file name and layer from the dialog box.

# 1.4. 4.0 Time course

The Time Course is a binary volume with multiple time points or conditions. The data is displayed in a graph so that all time points can be seen for a given vertex. When a vertex is clicked, the corresponding voxel is found in the binary volume. Tksurfer graphs the data for all time points at the chosen vertex.

Loading Time Course Volumes Loading binary volume Overlays and Time Course data is similar. Use the **File->Load Time Course...** command to load a time course volume. Tksurfer requires functional volumes to be in binary volume format. It will look for a header file with the same stem as the slice data or just guess the dimensions from the bfile. You need to specify the directory of the data, the stem, and optionally, a registration file. The format of a bfile name looks like this:

/path/to/data/stem\_000.bfloat /path/to/data/stem\_000.bshort The numbers go from 0 to the number of slices minus one. The 'stem' represents any text that appears before the underscore before the number in the bfile.

You can specify a registration file to use. If you do not, tksurfer will look in the same directory as the bfile data for a file called register.dat. A registration file is a matrix that defines the translation between anatomical RAS space and functional RAS space. Tksurfer uses this to align the functional volume with the anatomical volume.

# 1.5. 5.0 Time course graph

The Time Course is displayed in a separate graph window. The graph will only be shown if the Time Course volume has more than one time point. Clicking on a vertex in the Display Window finds the corresponding functional voxel based on the registration and displays the values at that voxel for all time points in the graph window.

If there are multiple conditions defined in the header file, each condition will show up as a separate line in the graph. The legend on the right side of the graph window displays what line corresponds to what condition. By moving the mouse over a condition label in the legend, the corresponding line in the graph will be highlighted. This is useful for picking out one condition in a crowded graph.

You can also zoom into the graph. Click and drag with button 2 to draw a box around the area of interest. The graph will be resized around that box. Click with button 3 to zoom back out.

You can specify an average area to graph in two ways. You can click the vertices you want, marking them, and then choose **Tools->Graph Marked Vertices Avg** to graph the average. (Note that loading a patch marks vertices, so you can also graph the average of a path this way.) You can also load a label and graph the average of it with the **Tools->Graph Label Avg**.

The contents of the graph window can be written to a Postscript file with the **Tools->Save Graph to Postscript File...** command. It can then be included in another document or printed.

# 1.6. 6.0 Patches

# 6.1 Loading patches

Use the File->Patch->Load Patch... item to load a patch file.

# 6.2 Saving patches

Use the **File->Patch->Save Patch** command to overwrite the original file, or **File->Patch->Save Patch As...** to save a new patch file in a new location.

# 1.7. 7.0 Labels

# 7.1 Loading labels

Use File->Label->Load Label... item to load a label file.

# 7.2 Saving labels

Use **File->Label->Save Label** command to overwrite the original file, or **File->Label->Save Label As...** to save a new label file in a new location. the current dipole values.

# 1.8.8.0 Field sign

### 8.1 Loading field sign files

Use the File->Field Sign->Load Field Sign... command to load a field sign file.

### 8.2 Saving field sign files

Use the **File->Field Sign->Save Label** command to overwrite the original file, or **File->Field Sign->Save Field Sign As...** to save a new field sign file in a new location.

# 1.9. 9.0 Field mask

### 9.1 Loading field mask files

Use the File->Field Mask->Load Field Mask... item to load a field mask file.

#### 9.2 Saving field mask files

Use the **File->Field Mask->Save Field Mask** command to overwrite the original file, or **File->Field Mask->Save Field Mask As...** to save a new field mask file in a new location.

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# 1. Reference

#### 1.1. 1.0 Mouse commands

#### 1.1 Mouse clicks

- **Button 1** Sets the cursor, drawing it in the new location, and marks the clicked vertex.
- Button 2 Sets the cursor without drawing it in the new location and unmarks the clicked vertex.

Button 3 Clears marked vertices.

#### **1.1 Mouse controls for zooming**

<b>Control+Button 1</b>	Zooms in, doubling the zoom level
Control+Button 3	Zooms out, halving the zoom level

#### 1.2. 2.0 Menus

2.1 File

Load Surface	Loads the main surface information, including main surface configuration.
Load Surface Configuration	Shows a submenu with commands for loading vertex configurations.
Save Surface	Saves the contents of the entire surface structure, overwriting the original file.
Save Surface As	Saves the contents of the entire surface structure to a new file.
Load Overlay	Loads a .w or binary volume file into an overlay layer.
Save Overlay As	Saves an overlay layer as a new .w file.
Load Time Course	Loads a binary volume as a time course.
Curvature	Shows a submenu with options for loading and saving.
Patch	Shows a submenu with options for loading and saving.
Label	Shows a submenu with options for loading and saving.
Field Sign	Shows a submenu with options for loading and saving.
Field Mask	Shows a submenu with options for loading and saving.
Quit	Quits tksurfer.

#### 2.2 Load surface configuration

Main VerticesLoads the specified vertices from another surface file into the selected surface<br/>configuration.Inflated<br/>VerticesVerticesWhite VerticesPial VerticesOriginal<br/>VerticesLoads a curvature file.Load Curvature...Loads a curvature file.SaveSaves changes to the curvature, overwriting the original file.

**Save Curvature As...** Saves the curvature into a new file.

#### 2.4 Patch

Load Patch	Loads a patch file.
Save Patch	Saves changes to the patch, overwriting the original file.
Save Patch As	Saves the patch into a new file.
2.5 Label	
Load Label	Loads a label file.
Save Label	Saves changes to the label, overwriting the original file.
Save Label As	Saves the label into a new file.
2.6 Field sign	
Load Field Sign	Loads a field sign file.
Save Field Sign	Saves changes to the field sign, overwriting the original file.
Save Field Sign As	S Saves the field sign into a new file.
2.7 Field mask	
Load Field Mask	Loads a field mask file.
Save Field Mask	Saves changes to the field mask, overwriting the original file.
Save Field Mask A	Saves the field mask into a new file.
2.8 Edit	
Undo	Undoes the cut.
Unmark All Vertices	Unmarks all vertices. Same as clicking with button 3 in the Graphics window.
Clear Label	Clears the vertices selected by loading a label.
2.9 View	

Tool Bars	Shows a submenu of available toolbars. Check or uncheck the toolbar in this submenu to show or hide the toolbar.
Information	Shows a submenu of available information area items such as coordinate labels. Check or uncheck the item in this submenu to show or hide the item.
Configure	Shows a submenu of viewing characteristics that can be configured.
Surface Configuration	Shows a submenu of surface configurations. Check one to make the currently shown configuration.
Overlay Layer	Shows a submenu of overlay layers. Check one to make the currently shown layer.
Overlay	Check to display the current overlay layer.
Scale Bar	Check to display a scale bar in the window, showing the scale of 10 mm.
Color Scale Bar	Check to display a color bar in the Display window, showing the range of colors in the current overlay color scale.
Wireframe Overlay	Check to display a wireframe view of the surface edges.

#### 2.10 Tool bars

Check to make the Main toolbar visible. This toolbar contains the following controls: CutMain Line, Cut Closed Line, Cut Plane, Cut Area, Clear Cuts, Save Point, Goto Saved Point, Restore Home View, Redraw View.

#### 2.11 Information

Vertex Index	Check to show the nearest vertex index.
Distance	Check to show the distance in mm to the reported index.
Vertex RAS	Check to show the RAS coordinates of the vertex.
Vertex Talairach	Check to show the Talairach coordinates of the vertex.
MRI Index	Check to show the anatomical index of the vertex in the anatomical volume for this subject.
Vertex Normal	Check to show the normal vector of the vertex.
Spherical X, Y, Z	Check to show the spherical coordinates.
Spherical Rho, Theta	Check to show the spherical rho and theta.
Curvature	Check to show the curvature value at the vertex.
Field Sign	Check to show the field sign value at the vertex.
<b>Overlay Layer 1-5</b>	Check to show the overlay layer value at the vertex.
Amplitude	Check to show the value at the vertex.
Angle	Check to show the value at the vertex.
Degree	Check to show the value at the vertex.
Annotation	Check to show the value at the vertex.
MRI Value	Check to show the MRI value at the associated anatomical location for this vertex.
Parcellation	Check to show the name of the parcellation label of this vertex.

2.12 Configure

Lighting	Opens a dialog box in which you can set the lighting parameters for the scene.
Overlay	Opens a dialog in which you can change various characteristics of the overlay display.
Time Course	Opens a dialog in which you can change various characteristics of the time course graph.
Curvature Display	Opens a dialog in which you can change various characteristics of the curvature display.
Phase Encoded Data Display	Opens a dialog in which you can change various characteristics of phase encoded data display.

#### 2.13 Surface configuration

Main	Check to make this surface configuration the currently displayed one.	
Inflated		
White		
Pial		
Original		
2.14 Overlag	y layer	
Overlay L	ayer 1-5 C	check to make this overlay layer the currently displayed one.
2.15 Tools		
Save Point	t	Saves the current cursor so that tksurfer or tkmedit can use its Goto Saved Point function to go to that point.
Goto Save	d Point	Sets the cursor to the one last saved in tksurfer or tkmedit for this subject.
Run Scrip	t	Executes a tcl script.
Cut Line		Calculates a line between marked vertices and cuts the vertices in that line.
Cut Close	d Line	Same as above, but closes the line between the first and last marked vertex.
Cut Plane		Requires exactly four vertices to be marked. Cuts the surface but not in the plane defined by the four vertices.

Cut Area	Requires one marked vertex and two incontiguous surface areas. Cuts the area that the marked vertex is not in.
Clear Cuts	Removes all cut vertex, restoring the surface to whole.
Send to Subject	Same as Save Point, but allows you to enter a subject name explicitly.
Write Decimation	Not functional, reserved for future upgrade.
Write Dipoles	Writes a file containing the dipole values at each vertex.
Set Background Midpoint to Average	In the curvature colorscale, sets the point at which red transitions to green to the mean curvature across the whole surface, making the data more interpretable.
Fill Stats	Marks vertices based on the overlay values surrounding the cursor.
Fill Curvature	Marks the region around each selected vertex that has a similar curvature to that of the selected curvature. Used for labeling anatomical regions.
Smooth	Iteratively smooths scalar for a user-selected number of iterations. Equivalent to convolving with a surface-based gaussian.
Inflate	Applies the inflation force for a user-specified number of iterations, to smooth the surface.
Swap Surface Fields	Intended for use by those who know the internal data structure of the surface. Opens a dialog box allowing you to specify two surface vertex fields to swap.
Clear Curvature	Sets the curvature value at each vertex to 0.
Graph Marked Vertices Avg	Graphs the average of marked vertices in the time course.
Graph Label Avg	Graphs the average of a label in the time course.
Save Graph to Postscript File	Saves the contents of the time course graph to a Postscript file.
Save RGB As	Saves the contents of the Display window to an RGB file.

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# 1. anatomically derived defect

• A topological defect in the cortical surface that arises from a feature of normal neuroanatomy to be distinguished from defects arising entirely from segmentation errors. See topological defect

# 2. artifact

• A feature that appears in an image but is not actually present in the imaged object.

# 3. average convexity

• The signed distance that a vertex moves during the inflation process.

### 4. brain volume

• The T1 volume after the skull and other non-brain structures have been removed. This volume can be viewed using tkmedit.

#### 5. canonical surface

• Surface-based atlas constructed from the cortical surfaces of 40 normal individuals (used for inter-subject averaging).

# 6. conversion/averaging

•

Process of converting and averaging multiple structural acquisitions from the native magnet format into the native FreeSurfer format (see COR files).

# 7. COR files

•

The native file format used by FreeSurfer to store 3D structural image data.

#### 8. Euler number

•

After Leonhard Euler (1707-83). A topological invariant of a surface that can be computed from the number of edges, vertices and faces in a polygonal tessellation (command mris\_euler\_number). The Euler number of a sphere will equal 2; the Euler number of a surface with n handles is 2 Å 2n.

### 9. filled volume

• The wm volume after separation of the left and right hemispheres and filling of each hemisphere. This volume can be viewed using tkmedit.

# 10. flattening

• Producing a planar (flat) representation of a patch of the cortical surface that has minimal metric distortion.

# 11. gyrus

• A fold or convolution of brain tissue (an outward folded region).

# 12. hypointensities (wm)

• Dark white matter on a T1-weighted image.

# 13. hypointensities (non-wm)

• Dark gray matter on a T1-weighted image.

# 14. inflated surface

• The smoothwm surface after inflation. This surface can be viewed using surfer.

# 15. inflation

• The process of smoothing the cortex while minimizing metric distortion, so that all sulci are fully visible and surface distances are apparent to visual inspection.

# 16. intensity

• Measured amount of magnetic field at a given spatial location, represented by a voxel (higher SNR signal to noise ratio means voxels will have a higher intensity relative to the background noise, and appear brighter).

# 17. label

• A particular region of interest. e.g. in tksurfer the label would be a region of interest in the surface. In tkmedit a label is a region of interest in the volume.

#### 18. morphing

• Computer graphics technique whereby a mapping is computed that smoothly transforms one image or surface into another.

#### **19. morphometrics**

• The study of geometric properties of the human brain.

#### 20. motion correction

• Processing multiple structural volumes so that the effects of subject movement are minimized. This is typically done by aligning multiple images/volume to an initial image/volume (see conversion/averaging).

# 21. MRI volume

• The three dimensional volumetric data set collected from a MRI scanner.

# 22. orig volume

• The original MRI volume. This volume can be viewed using tkmedit.

# 23. orig surface

• The first surface constructed by covering the labeled voxels in the filled volume. This surface can be viewed using surfer.

# 24. pial

• Pertaining to the delicate pia mater which envelops the brain (gray matter). Also, the model of the pial surface (?h.pial).

# 25. pial surface

• The refined estimate of the gray/CSF boundary (pial surface). This surface can be viewed using surfer.

#### 26. region growing process

• An algorithm that groups voxels or sub-regions into larger regions.

# 27. RF-field inhomogeneities

• Spatial variations in the Radio Frequency (RF) excitation pulse. These variations result in changes in the measured intensity for a given tissue class that are related to the spatial location of the voxel.

#### 28. segmentation

• Labeling of tissue classes from MRI data (e.g. white matter).

#### 29. smoothing

• Process of producing a relatively even and regular cortical surface.

#### 30. smoothwm surface

• The orig surface after smoothing. This surface can be viewed using surfer.

#### **31. sulcus**

• A groove or furrow in brain tissue (an inward folded region).

#### 32. supertessellated icosahedron

• Polygonal approximation to a sphere.

### **33.** T1

• Longitudinal relaxation constant.

### 34. T1 volume

• The MRI volume after intensity normalization. This volume can be viewed using tkmedit.

### 35. T1 Weighted Image

• A magnetic resonance image where the contrast is predominantly dependent on T1.

### 36. T2 Weighted Image

• A magnetic resonance image where the contrast is predominantly dependent on T2.

### **37.** T2

• Transverse relaxation constant.

#### 38. Talairach coordinate

• The corresponding location in the Talairach atlas for a given point in a brain that has been coregistered with the atlas (Talairach et al, 1967).

# **39. tessellation**

• Covering of a surface by repeated use of a single shape.

#### 40. topology

• The properties of a surface related to its connectivity that are unaffected by geometric (i.e. rubber sheet) transformations.

# 41. topological defect

• A portion of a surface that results in the surface topology differing from that of a sphere.

#### 42. volume

• A 3-D data set that typically contains either intensity information derived from the original MRI, or the results of segmenting this data into tissue classes.

#### 43. voxel

•

The basic element of an MRI volume (analogous to a pixel in a 2-D image). The volume of a structural voxel is approximately 1 mm<sup>3</sup>.

### 44. white surface

• The refined estimate of the gray/white boundary. This surface can be viewed using surfer.

### 45. wm volume

• The brain volume after white matter segmentation. This is also the volume that is manually edited. This volume can be viewed using tkmedit.

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