# Protecting the Aging Brain – Diet Study

#### **Dataset:**

I have acquired two distinct datasets from OpenNeuro.

The first dataset comprises scans of 12 participants who underwent imaging under three distinct conditions:

- 1. On a standard non-ketogenic diet.
- After following a standard diet and fasting for 12 hours.
- 3. After being on a ketogenic diet for one week.

The second dataset involves scans of 30 participants under three conditions:

- 1. After adhering to a standard diet and fasting for 12 hours (pre-bolus).
- 2. Following the 12-hour fast, with a bolus of glucose drink.
- 3. Following the 12-hour fast, with a bolus of ketogenic drink.

Due to space limitations, I've downloaded only 7 subjects from the second dataset, as the scanned data files are quite large. I've organized the data into separate folders for each dataset. Within each dataset's folder, I've created individual folders for every participant, and within those, I've placed their respective T1 and BOLD images.

# **Dataset Preprocessing:**

Our initial goal is to eliminate any irrelevant noise present in the datasets. To achieve this, we embark on preprocessing the datasets.

#### **Motion Correction:**

As a part of this preprocessing stage, we focus on addressing minor movements, including subtle head or body shifts. These minute movements can result in undesired blurring, distortion, and misalignment within the images. Our aim is to rectify these issues and enhance the quality of the data.

#### Command:

3dvolreg -prefix output;

# Motion Correcting for all subjects:

#!/bin/bash

```
subs=$(ls -d dataset2/sub*)

for sub in $subs; do
    cd ${sub}
    gzs=$(ls *gz)
    for gz in $gzs; do
        3dvolreg -prefix $gz -overwrite $gz;
    done
    cd ../../
Done
```

#### **Extract First Volume:**

Selected from the motion-corrected dataset, the initial volume represents the image captured at the beginning of the scanning session, subsequent to the application of motion correction procedures.

#### Command

```
3dTcat -prefix output mc_data[0]
```

Extracting first volume for all subjects:

```
#!/bin/bash

subs=$(ls -d dataset2/sub*)

for sub in $subs; do
    cd ${sub}
    gzs=$(ls *.gz)
    for gz in $gzs; do
        3dTcat -prefix vol_${gz} ${gz}[0]
    done
    cd ../../
done
```

#### Registration:

During this phase, our objective is to bring the datasets into alignment by mapping them onto a shared coordinate system.

Performing registration across all participants

```
#!/bin/bash
subs=$(ls -d dataset2/sub*)

for sub in $subs; do
    cd ${sub}
    gzs=$(ls vol*)
    for gz in $gzs; do
        antsRegistration --dimensionality 3 --float 0 --output
[to_${gz/.nii/},affine_${gz}] --interpolation Linear --transform
Affine[0.1] --metric MI[../ref.nii.gz,$gz,1,32, Regular, 0.25]
--convergence [1000x500x250x100,1e-6,10] --shrink-factors 8x4x2x1
--smoothing-sigmas 3x2x1x0vox -v
    done
    cd ../../
Done
```

#### **Bandpass Filtering:**

In this stage, our aim is to isolate a targeted frequency range within a signal while reducing the influence of frequencies lying outside this designated range.

#### Command:

```
3dTproject -input input_filename -prefix output_filename -passband
0.01 0.1
```

## Performing bandpass filtering across all participants

```
#!/bin/bash

subs=$(ls -d dataset2/sub*)

for sub in $subs; do
    cd ${sub}
    gzs=$(ls sub*.nii)
    for gz in $gzs; do
        3dTproject -input $gz -prefix bp_${gz} -passband 0.01 0.1
    done
    cd ../../
done
```

## **RSFC Metrics:**

During this phase, these RSFC metrics offer understanding into the coordinated activity and communication patterns within the brain's inherent networks.

#### ReHo:

At this stage, ReHo helps to evaluate how closely the time series of a specific voxel aligns with those of its neighboring voxels.

#### Command

```
3dReHo -prefix reho_outputfile -inset bp_file
```

#### Generating ReHo for all BP dataset

```
#!/bin/bash

subs=$(ls -d dataset2/sub*)

for sub in $subs; do
    cd ${sub}
    bps=$(ls bp*.nii)
    for bp in $bps; do
        3dReHo -prefix reho_${bp} -inset $bp
    done
    cd ../../

done
```

#### **ReHo Transformation:**

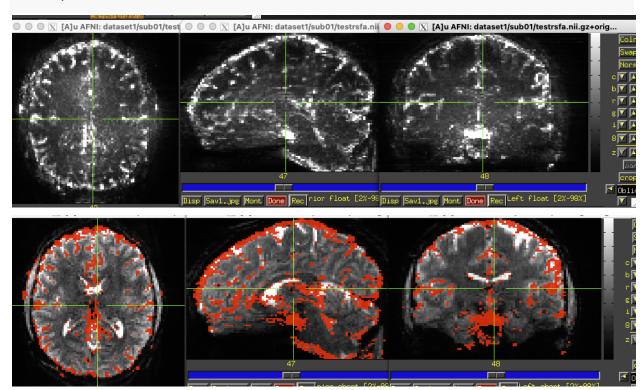
```
subs=$(ls -d dataset1/sub*)

for sub in $subs; do
    cd ${sub}
    rehos=$(ls reho*gz)
    for reho in $rehos; do
        matname=${reho/reho_bp_/to_vol_}
        matname=${matname/.nii.gz/0GenericAffine.mat}
        echo $matname
```

```
antsApplyTransforms -d 3 -e 0 -i $reho ../ref.nii.gz -o
inref_${reho} -t $matname
    done
    cd ../../
done
```

## Stacking 3d to 4d ReHo Image:

3dTcat -prefix all\_reho\_fast.nii.gz sub01/reho sub02/reho ...
sub11/reho



## Generated fALFF and RSFA using 3dRSFC

```
subs=$(ls -d dataset1/sub*)

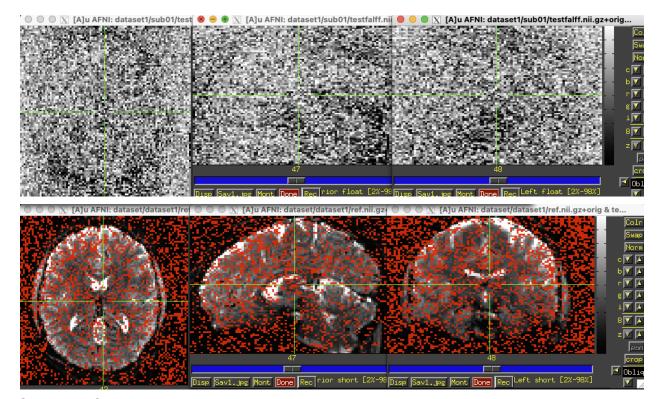
for sub in $subs; do
    cd ${sub}
    bps=$(ls bp*gz)
    for bp in $bps; do
        rsfc_name=rsfc_metric_${bp/.nii.gz/}
        3dRSFC -prefix $rsfc_name 0.01 0.1 $bp
        3dAFNItoNIFTI -prefix rsfc_metric_RSFA_${bp/.nii.gz/}
```

#### **fALFF** Transformation:

```
import os
from nipype.interfaces.ants import ApplyTransforms
dataset dir = 'dataset1'
# List all subdirectories
subs = [subdir for subdir in os.listdir(dataset dir) if
os.path.isdir(os.path.join(dataset dir, subdir))]
# subs = ["sub01"]
for sub in subs:
    print(sub)
    sub dir = os.path.join(dataset dir, sub)
    os.chdir(sub_dir)
    # List reho*gz files
    falffs = [falff for falff in os.listdir('.') if
falff.startswith('rsfc metric fALFF') and falff.endswith('.nii')]
    # rehos = ["reho bp sub-001 ses-ket task-rest run-1 bold.nii.gz"]
    for falff in falffs:
        # print(falff)
        # Generate the matname
        matname = falff.replace('rsfc metric fALFF bp ',
'to vol ').replace('.nii', '0GenericAffine.mat')
        # print(matname)
        # Create an instance of the ApplyTransforms interface
        at = ApplyTransforms()
        at.inputs.dimension = 3
```

```
at.inputs.input_image = falff
at.inputs.reference_image = '../ref.nii.gz'
at.inputs.output_image = f'inref_{falff}.gz'
at.inputs.transforms = [matname]

# Execute the interface
result = at.run()
os.chdir('../../')
```



Somehow fALFF images are a bit weird.

Next we perform, fALFF, RSFA and Global Signal Transformation Stacking 3d to 4d Image for fALFF, RSFA and Global Signal Image

# Statistical Analysis for DataSet-1

**Generated T-Map** 

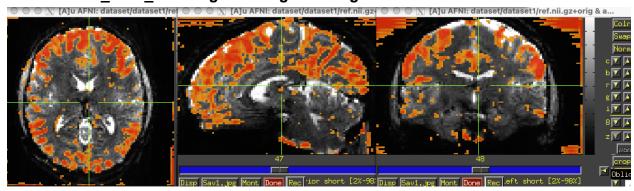
Standard diet minus 12-hour fast

3dttest++ -setA all\_reho\_std.nii.gz[0] all\_reho\_std.nii.gz[1].. -setB all\_reho\_fast.nii.gz[0] all\_reho\_fast.nii.gz[1].. -paired -prefix ttest\_reho\_std\_minus\_fast.nii.gz

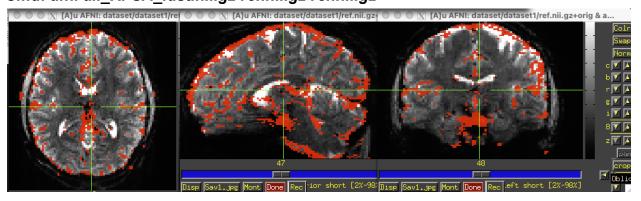
Similarly, create plots for various scenarios such as...

- 1. Standard diet minus ketogenic diet
- 2. 12-hour fast minus ketogenic diet
- 3. No bolus minus glucose drink
- 4. No bolus minus ketogenic drink
- 5. Glucose drink minus ketogenic drink

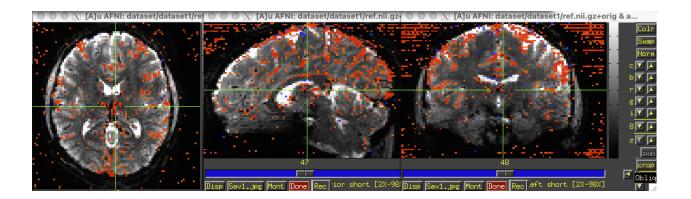
## Cmd: afni all reho fast.nii.gz ref.nii.gz ref.nii.gz



Cmd: afni all\_RFSA\_fast.nii.gz ref.nii.gz ref.nii.gz



Cmd: afni all global signal fast.nii.gz ref.nii.gz ref.nii.gz

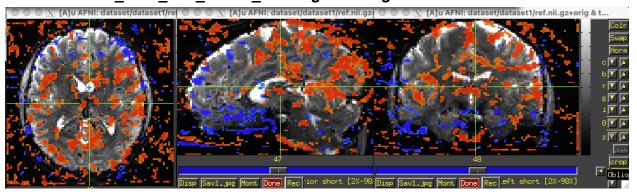


# **Generated Thresholded T-Map for DataSet-1:**

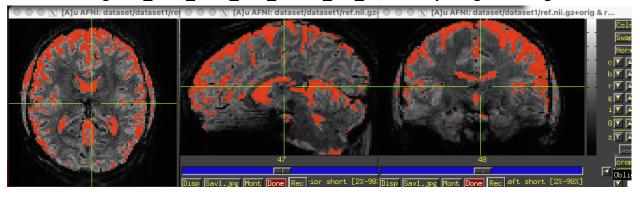
In this step, a threshold of 2 voxels is established, resulting in the separation of positive and negative values.

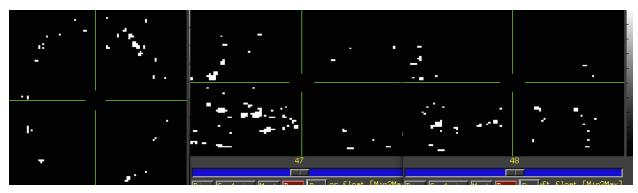
3dcalc -a ttest\_reho\_fast\_minus\_ket.nii.gz[1] -expr 'isnegative(a+2)'
-prefix negitive\_ttest\_reho\_fast\_minus\_ket\_threshmap.nii.gz

# Cmd: afni ttest\_reho\_fast\_minus\_ket.nii.gz ref.nii.gz



#### Cmd: afni negitive\_ttest\_reho\_fast\_minus\_ket\_threshmap.nii.gz ref.nii.gz





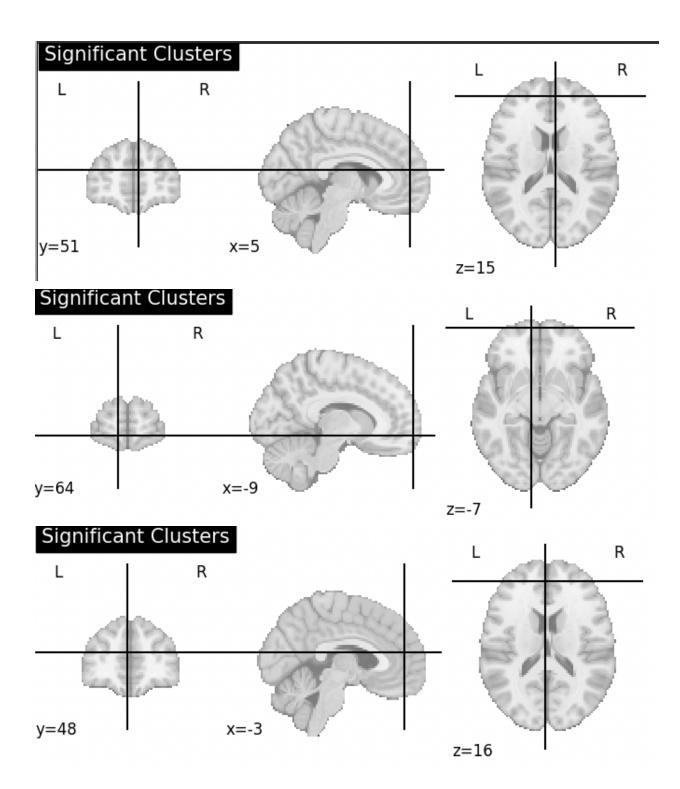
#### Plot the Images:

Images are visualized using the glass brain technique provided by Nilearn.

```
from nilearn import plotting

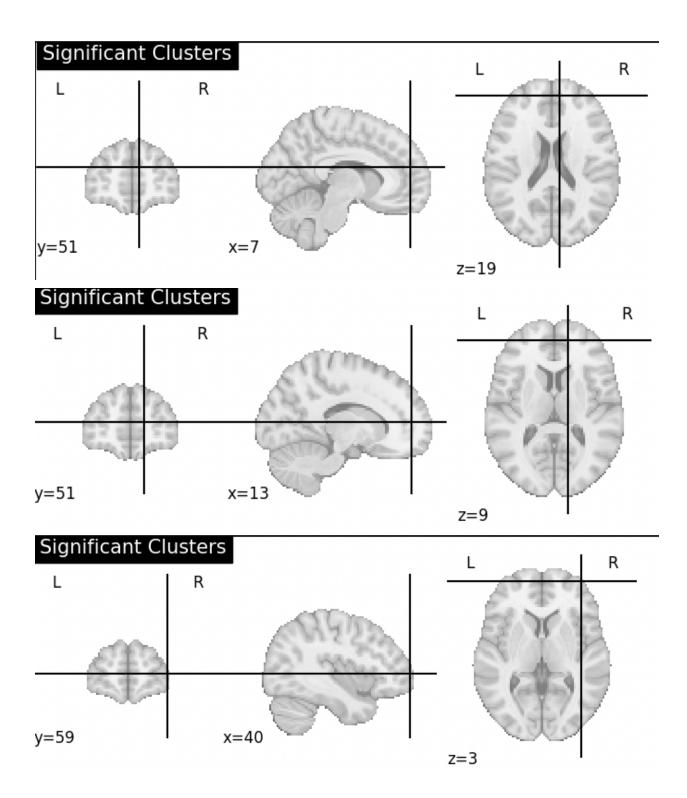
map_filenames =
["negitive_ttest_RSFA_fast_minus_ket_threshmap.nii.gz", ...,
"negitive_ttest_reho_std_minus_ket_threshmap.nii.gz"]
for map_file in map_filenames:
    display = plotting.plot_stat_map(map_file, threshold=3,
colorbar=True, display_mode='ortho', title="Significant Clusters")
    plotting.show()
```

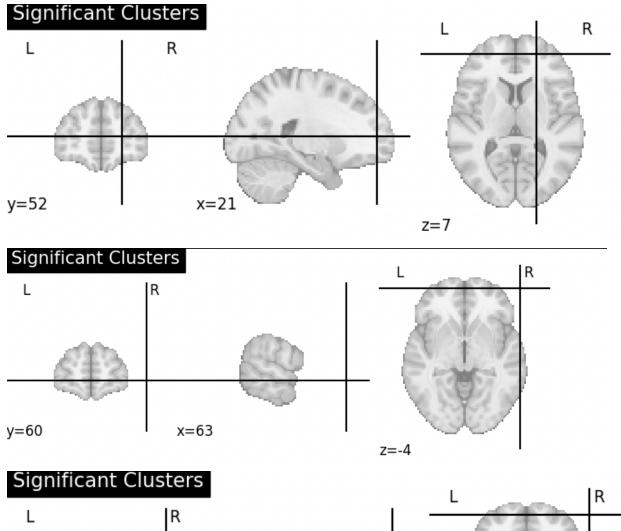
# Significant Cluster Cluster for DataSet-1

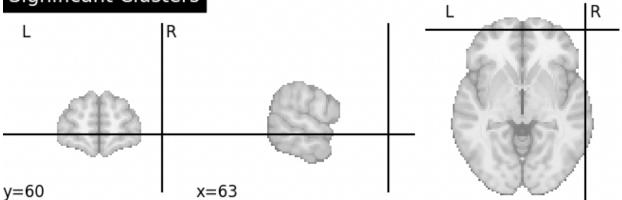


# Significant Clusters L R L R x=-1 y=47 z=26 Significant Clusters L R R L y=49 x=0z=-2 Significant Clusters L R R L y=52 x=17

z=14







z=-4

# **Statistical Analysis for DataSet-2**

# **Generated T-Map**

For following:

- 1. No bolus minus glucose drink
- 2. No bolus minus ketogenic drink
- 3. Glucose drink minus ketogenic drink

```
#!/bin/bash
#Reho
#No bolus minus glucose drink
3dttest++ -setA all reho glc1.nii[0] all reho glc1.nii[1]
all reho glc1.nii[2] all reho glc1.nii[3] all reho glc1.nii[4]
all reho glc1.nii[5] -setB all reho glc2.nii[0] all reho glc2.nii[1]
all_reho_glc2.nii[2] all_reho_glc2.nii[3] all_reho_glc2.nii[4]
all_reho_glc2.nii[5] -paired -prefix ttest_reho_case1.nii.gz
#No bolus minus ketogenic drink
3dttest++ -setA all reho bhb1.nii[0] all reho bhb1.nii[1]
all reho bhb1.nii[2] all reho bhb1.nii[3] all reho bhb1.nii[4]
all reho bhb1.nii[5] -setB all reho bhb2.nii[0] all reho bhb2.nii[1]
all reho bhb2.nii[2] all reho bhb2.nii[3] all reho bhb2.nii[4]
all_reho_bhb2.nii[5] -paired -prefix ttest_reho_case2.nii.gz
#Glucose drink minus ketogenic drink
3dttest++ -setA all_reho_glc2.nii[0] all_reho_glc2.nii[1]
all reho glc2.nii[2] all reho glc2.nii[3] all reho glc2.nii[4]
all reho glc2.nii[5] -setB all reho bhb2.nii[0] all reho bhb2.nii[1]
all reho bhb2.nii[2] all reho bhb2.nii[3] all reho bhb2.nii[4]
all reho bhb2.nii[5] -paired -prefix ttest reho case3.nii.gz
#fALFF
#No bolus minus glucose drink
3dttest++ -setA all_fALFF_glc1.nii.gz[0] all_fALFF_glc1.nii.gz[1]
all fALFF glc1.nii.gz[2] all fALFF glc1.nii.gz[3]
all_fALFF_glc1.nii.gz[4] all_fALFF_glc1.nii.gz[5] -setB
all fALFF glc2.nii.gz[0] all fALFF glc2.nii.gz[1]
all fALFF glc2.nii.gz[2] all fALFF glc2.nii.gz[3]
all fALFF glc2.nii.gz[4] all fALFF glc2.nii.gz[5] -paired -prefix
ttest fALFF case1.nii.gz
#No bolus minus ketogenic drink
3dttest++ -setA all fALFF bhb1.nii.gz[0] all fALFF bhb1.nii.gz[1]
all fALFF bhb1.nii.gz[2] all_fALFF_bhb1.nii.gz[3]
all fALFF bhb1.nii.gz[4] all fALFF bhb1.nii.gz[5] -setB
all fALFF bhb2.nii.gz[0] all fALFF bhb2.nii.gz[1]
```

```
all_fALFF_bhb2.nii.gz[2] all_fALFF_bhb2.nii.gz[3]
all fALFF bhb2.nii.gz[4] all fALFF bhb2.nii.gz[5] -paired -prefix
ttest_fALFF_case2.nii.gz
#Glucose drink minus ketogenic drink
3dttest++ -setA all fALFF glc2.nii.gz[0] all fALFF glc2.nii.gz[1]
all fALFF glc2.nii.gz[2] all fALFF glc2.nii.gz[3]
all_fALFF_glc2.nii.gz[4] all_fALFF_glc2.nii.gz[5] -setB
all_fALFF_bhb2.nii.gz[0] all_fALFF_bhb2.nii.gz[1]
all_fALFF_bhb2.nii.gz[2] all_fALFF_bhb2.nii.gz[3]
all fALFF bhb2.nii.gz[4] all fALFF bhb2.nii.gz[5] -paired -prefix
ttest_fALFF_case3.nii.gz
#RSFA
#No bolus minus glucose drink
3dttest++ -setA all RSFA glc1.nii.gz[0] all RSFA glc1.nii.gz[1]
all RSFA glc1.nii.gz[2] all_RSFA_glc1.nii.gz[3]
all RSFA glc1.nii.gz[4] all RSFA glc1.nii.gz[5] -setB
all RSFA glc2.nii.gz[0] all RSFA glc2.nii.gz[1]
all RSFA glc2.nii.gz[2] all RSFA glc2.nii.gz[3]
all RSFA glc2.nii.gz[4] all RSFA glc2.nii.gz[5] -paired -prefix
ttest RSFA case1.nii.gz
#No bolus minus ketogenic drink
3dttest++ -setA all_RSFA_bhb1.nii.gz[0] all_RSFA_bhb1.nii.gz[1]
all RSFA bhb1.nii.gz[2] all RSFA bhb1.nii.gz[3]
all RSFA bhb1.nii.gz[4] all RSFA bhb1.nii.gz[5] -setB
all_RSFA_bhb2.nii.gz[0] all_RSFA_bhb2.nii.gz[1]
all RSFA bhb2.nii.gz[2] all RSFA bhb2.nii.gz[3]
all RSFA bhb2.nii.gz[4] all RSFA bhb2.nii.gz[5] -paired -prefix
ttest RSFA case2.nii.gz
#Glucose drink minus ketogenic drink
3dttest++ -setA all RSFA glc2.nii.gz[0] all RSFA glc2.nii.gz[1]
all RSFA glc2.nii.gz[2] all RSFA glc2.nii.gz[3]
all_RSFA_glc2.nii.gz[4] all_RSFA_glc2.nii.gz[5] -setB
all RSFA bhb2.nii.gz[0] all RSFA bhb2.nii.gz[1]
all RSFA bhb2.nii.gz[2] all RSFA bhb2.nii.gz[3]
all RSFA bhb2.nii.gz[4] all RSFA bhb2.nii.gz[5] -paired -prefix
```

```
ttest_RSFA_case3.nii.gz
```

#### **Generated Thresholded T-Map for DataSet-2:**

In this step, a threshold of 2 voxels is established, resulting in the separation of positive and negative values.

```
#!/bin/bash
#ReHo Metric
3dcalc -a ttest reho case1.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive ttest reho case1 threshmap.nii.gz
3dcalc -a ttest reho case1.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive ttest reho case1 threshmap.nii.gz
3dcalc -a ttest reho case2.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive ttest reho case2 threshmap.nii.gz
3dcalc -a ttest reho case2.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive_ttest_reho_case2_threshmap.nii.gz
3dcalc -a ttest_reho_case3.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive ttest reho case3 threshmap.nii.gz
3dcalc -a ttest_reho_case3.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive ttest reho case3 threshmap.nii.gz
#fALFF Metric
3dcalc -a ttest fALFF case1.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive ttest fALFF case1 threshmap.nii.gz
3dcalc -a ttest fALFF case1.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive ttest fALFF case1 threshmap.nii.gz
3dcalc -a ttest_fALFF_case2.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive ttest fALFF case2 threshmap.nii.gz
3dcalc -a ttest_fALFF_case2.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive_ttest_fALFF_case2_threshmap.nii.gz
3dcalc -a ttest_fALFF_case3.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive ttest fALFF case3 threshmap.nii.gz
```

```
3dcalc -a ttest_fALFF_case3.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive_ttest_fALFF_case3_threshmap.nii.gz

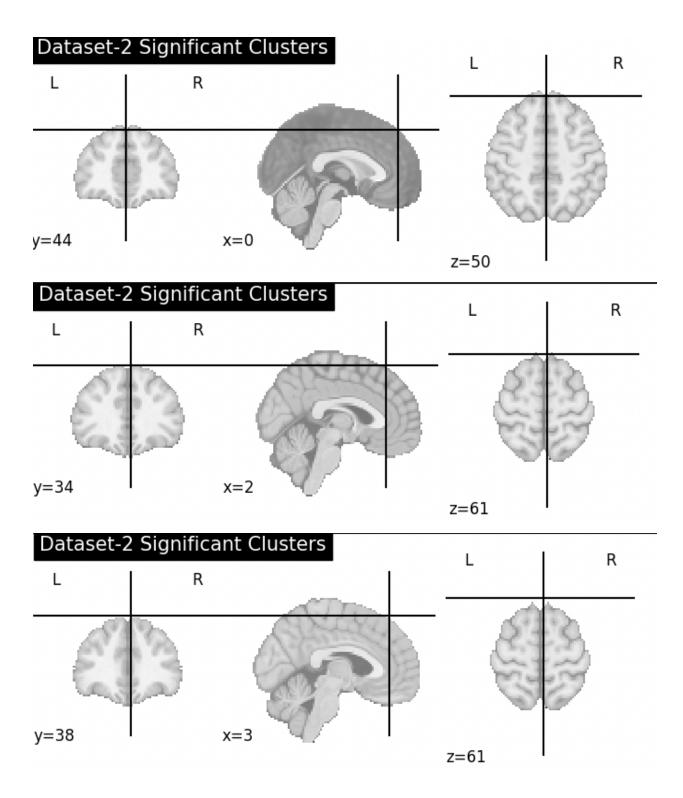
#RSFA Metric
3dcalc -a ttest_RSFA_case1.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive_ttest_RSFA_case1_threshmap.nii.gz
3dcalc -a ttest_RSFA_case1.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive_ttest_RSFA_case1_threshmap.nii.gz

3dcalc -a ttest_RSFA_case2.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive_ttest_RSFA_case2.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive_ttest_RSFA_case2.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive_ttest_RSFA_case3.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive_ttest_RSFA_case3.nii.gz[1] -expr 'isnegative(a+2)' -prefix
negitive_ttest_RSFA_case3.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive_ttest_RSFA_case3.nii.gz[1] -expr 'isnegative(a-2)' -prefix
positive_ttest_RSFA_case3.nii.gz[1] -expr 'isnegative(a-2)' -prefix
```

# Significant Cluster Cluster for DataSet-2

# Dataset-2 Significant Clusters L R R L y=52 x=-2 z=38 Dataset-2 Significant Clusters L R L R x=9 y=37 z=68 Dataset-2 Significant Clusters L R L R y=41 x=4

z = 74



# Dataset-2 Significant Clusters L R R L y=31 x=0z=62 Dataset-2 Significant Clusters L R L R x=10 y=37 z=71 Dataset-2 Significant Clusters L R L R x=19 y=28 z=59

