Sunnybrook
HEALTH SCIENCES CENTRE

Image Processing Software Guide

Version 1.03

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SUNNYBROOK IMAGE PROCESSING SOFTWARE

OVERVIEW

- Sunnybrook image processing software is a collection of both external (e.g FMRIB's FSL) and in-house (e.g. SABRE) developed tools that have been assembled together.
- The software can be run on Linux, SunOS, or Windows (through Cygwin), wit

CYGWIN INSTALLATION INSTRUCTIONS FOR WINDOWS USERS

- Cygwin can be downloaded from: http://www.cygwin.com/
- You will need to add two additional packages that are typically not selected for installation by default:
    1. perl: Larry Wall's Practical Extracting and Report Language (found under Perl category)
    2. bc: The GNU numeric processing language and reverse polish calculator (found under Utils category)
- If, when running the Sunnybrook software, you encounter "unable to remap XXX.dll to the same address as parent" errors, you will need to rebase all cygwin dlls. This can be done easily using the following procedure:
    1. Close all Cygwin terminal windows that may be open
    2. Click on the Windows Start button, select Run, and type: `C:\cygwin\bin\ash.exe` to open a new Cygwin terminal
    3. In the new Cygwin terminal, type: `/usr/bin/rebaseall`

  If Cygwin can not find the rebaseall command, you will need to install the rebase package (found under Utils category) using the Cygwin setup utility
- If you will be using the software from within the research network at Sunnybrook, you will also need to map the network directory `/home116/egibson` containing the `sb` software directory to a drive letter in Windows. This can be done by mapping `\\netshare\egibson` through Network Connections in Windows and selecting a drive letter.

GENERAL INSTALLATION PROCEDURE FOR WINDOWS, LINUX and SunOS

Setup WITHIN the Sunnybrook Research Computer Network

Configuring

- Add the following to your shell setup file (.bashrc, .profile, .cshrc etc., depending on the particular shell that you use). If you are not sure what shell you use, type:
  `echo ${SHELL}`

For tcsh or csh, add:

```
setenv PATH /home116/egibson/sb/cross_platform/scripts/:${PATH}
setenv SBBINDIR /cygdrive/X/sb/windows/** (OR) setenv SBBINDIR /home116/egibson/sb/linux/ (OR) setenv SBBINDIR /home116/egibson/sb/unix/
setenv PATH ${SBBINDIR}/SB/bin/:${PATH}
```

For bash or sh or ksh, add:

```
PATH=/home116/egibson/sb/cross_platform//scripts/:${PATH}
SBBINDIR=/cygdrive/X/sb/windows/** (OR) SBBINDIR=/home116/egibson/sb/linux/ (OR) SBBINDIR=/home116/egibson/sb/unix/
PATH=${SBBINDIR}/SB/bin/:${PATH}
export PATH SBBINDIR
```

Note for Windows users: "x**" above refers to the drive letter to which the network directory `/home116/egibson` has been mapped to.

Setup OUTSIDE the Sunnybrook Research Computer Network

Unpacking

- Download sb.tar.gz to your home directory, and type:

```
cd
tar zxvf sb.tar.gz
```

Configuring

- Add the following to your shell setup file (.bashrc, .profile, .cshrc etc., depending on the particular shell that you use). If you are not sure what shell you use, type:
  `echo ${SHELL}`

For tsch or csh, add:

```
setenv PATH ${HOME}/sb/cross_platform/scripts/:${PATH}
setenv SBINDIR ${HOME}/sb/windows/ (OR) setenv SBBINDIR ${HOME}/sb/linux/ (OR) setenv SBBINDIR ${HOME}/sb/unix/
setenv PATH ${SBBINDIR}/SB/bin/:${PATH}
```

For bash or sh or ksh

```
PATH=${HOME}/sb/cross_platform/scripts/:${PATH}
SBBINDIR=${HOME}/sb/windows/ (OR) SBBINDIR=${HOME}/sb/linux/ (OR) SBBINDIR=${HOME}/sb/unix/
PATH=${SBBINDIR}/SB/bin/:${PATH}
export PATH SBBINDIR
```

BRAIN PARCELLATION – SABRE

OVERVIEW

- SABRE is a semi-automated procedure that parcellates the brain into different regional areas (steps 5 to 10 below). If required, additional thalamus, cingulate, and cholinergic parcellations can be generated (steps 11 to 13 below).
- SABRE requires a T1 image and a good head-from-brain mask. PD/T2 images are typically used in a semi-automated procedure to generate the head-from-brain mask. This semi-automated head-from-brain procedure ensures that all sulcal CSF voxels are included in the mask. Often, other head-from-brain procedures (e.g. FSL's `bet` or freesurfer's `mri_watershed`) remove significant numbers of sulcal CSF voxels. However, if PD/T2 images are not available, or are not compatible with the Sunnybrook head-from-brain algorithm, any accurate skull-stripping procedure could be substituted, in which case several preprocessing steps could be omitted (steps 1 to 4 below).

IMAGE REQUIREMENTS			
Command/Task	**Step(s)**	**Image Type***	**Processing**
sb_301_coregister	1	a) T1	Unprocessed, uncorrected for intensity inhomogeneities (or corrected, if using the –noic option)
		b) PD (optional)	Unprocessed, uncorrected (or corrected, if using the –noic option)
		c) T2 (optional)	Unprocessed, uncorrected (or corrected, if using the –noic option)
		d) FLAIR (optional)	Unprocessed, uncorrected (or corrected, if using the –noic option)
sb_302_hfb	3, 4	a) PD	Skull-on, uncorrected (or corrected if using the –noic option), in T1 acquisition space
		b) T2	Skull-on, uncorrected (or corrected, if using the –noic option), in T1 acquisition space
		c) Rough brain mask	Skull-off (e.g. bet), in T1 acquisition space
SABRE	5-13	a) T1	Skull-on, uncorrected (or corrected, if using the –noic option)
		b) Good brain mask	"1"s or "8"s for brain, "0"s for background

*All input images must be in 16 bit Analyze format. For legacy 8 bit T1 Sunnybrook data, alternate versions of the `segment_and_acpc` and `parcellate` scripts are available (see steps 7 and 10 below)

SOFTWARE REQUIREMENTS			
Command/Task	**Step(s)**	**Requirements (Within Sunnybrook)**	**Requirements (Outside Sunnybrook)**
sb_301_coregister	1	bc, perl	(bc, perl, N3): optional, if needing inhomogeneity correction FSL's bet & flirt: required
sb_302_hfb	3	perl	(perl, N3): optional, if needing inhomogeneity correction FSL's bet & flirt: required
sb_303_isotropicT1	5	--	--
sb_304_segment_and_acpc	6, 7	Analyze	Analyze
segmentation relabelling	8	Brainkit (or other image editing software)	Brainkit (or other image editing software)
sb_305_parcellate_brain	9, 10	Analyze	Analyze
sb_306_parcellate_thalamus	11	--	--
sb_307_parcellate_cingulate	12	Analyze	Analyze
sb_308_parcellate_cholingergic	13	(Analyze): optional, if performing good parcellation	(Analyze): optional, if performing good parcellation

STANDARD INPUT

P	PD image, unprocessed
T1	T1 image, unprocessed
T2	T2 image, unprocessed
FL	FLAIR Image, unprocessed

Notes

- If there are intensity inhomogeneities in the images, follow the 3T instructions below, otherwise follow the 1.5T instructions.

STEP 1: COREGISTER IMAGES TO THE T1

3T

```
sb_301_coregister -t1 T1 -t2 T2 -pd P -fl FL
```

1.5T

```
sb_301_coregister -t1 T1 -t2 T2 -pd P -fl FL -noic
                  -t1t 0.5 -t2t 0.5 -pdt 0.5 -flt 0.5
```

If running outside Sunnybrook, FSL is required, and the –fslinstalled option should be added to the above command.

Purpose

- sb_301_coregister uses FSL's bet and (optionally) John Sled's N3 to correct for intensity inhomogeneities in the input images and then uses FSL's flirt to coregister and reslice the N3-corrected and uncorrected images in T1 space with 6 dof, a normalized mutual information cost function and sinc interpolation.

Notes

- If the images differ substantially from those acquired on the 1.5T/3T Sunnybrook scanners, the -t2t, -pdt, -t1t and/or -flt bet thresholds may need adjustment.
- For 3T data, the resulting `_nu` images are bias-corrected, but this bias-correction may not be ideal because the correction was done without a proper head-from-brain mask. However, this correction should be adequate for the purposes of image coregistration and generation of the head-from-brain image (step 3). For T1, FLAIR (and possibly PD/T2) images, a second, more complete bias correction is performed later, prior to segmentation.

STEP 2: CHECK COREGISTRATION AND FIX IF NECESSARY

User Intervention

- Examine T1acq images to check success of coregistration

To fix a bad coregistration (for example, a bad T1-FL coregistration), do the following:

```
mkdir newcoreg
cd newcoreg
sb_bet_all ../T1_nu ../FL_nu
```

Select best bet and delete others (often it is a poor T1 bet that causes the misregistration)

```
sb_coregister_IN_to_REF -ref T1_bet_X -in FL_bet_X -omat FL_to_T1.omat
sb_reslice_IN_to_REF -ref T1_bet_X -in FL_bet_X -init FL_to_T1.omat -out newcoreg
```

Check newcoreg, if good, replace bad FL-T1 rotation matrix

```
cp T1_to_FL.omat ../
```

Reslice images using good FL-T1 rotation matrix

```
cd ../
```

3T: `sb_301_coregister -t1 T1 -t2 T2 -pd P -fl FL -fixreg`

1.5T: `sb_301_coregister -t1 T1 -t2 T2 -pd P -fl FL -noic -fixreg`

STEP 3: GENERATE HEAD-FROM-BRAIN (HFB) IMAGE

3T

```
sb_bet T1acq_nu_T2 T1acq_nu_T2_bet -f 0.X
```

1.5T

```
sb_bet T1acq_T2 T1acq_T2_bet -f 0.X
```

User Intervention

X = threshold that best maximizes the amount of non-brain removed and minimizes the amount of brain removed)

3T

```
sb_302_hfb -t2 T1acq_nu_T2 -pd T1acq_nu_P -mask T1acq_nu_T2_bet
           -noic -t2reg
```

1.5T

```
sb_302_hfb -t2 T1acq_T2 -pd T1acq_P -mask T1acq_T2_bet
           -pdt 0.37 -t2t 0.35 -t2reg
```

If running outside Sunnybrook, FSL is required, and the –fslinstalled option should be added to the above command.

Purpose

- sb_302_hfb creates 3 head-from-brain images: the new template "`HfBt`" image (minimal edits required), the old auto "`HfBa`" image (extensive edits required), and the combined "`HfBc`" image (a combination of the new and old HfBs), may be the best option for editing.

Notes

- The –noic option is needed with the 3T data because the input T2 and PD images have already been bias corrected. For 1.5T data, a bias-correction is performed prior to generating the head-from-brain images. This generally improves the head-from-brain result for 1.5T data.
- If bet does not produce a good mask using the T2, you can try using the PD or T1 instead, and then run the sb_302_hfb script replacing the -t2reg option with -pdreg or -t1reg.

STEP 4: EDIT HFB (IF NECESSARY)

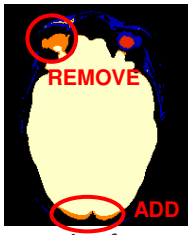
3T

`T1acq_nu_HfBc.img`

1.5T

`T1acq_HfBc.img`

** Or you may decide to use the `HfBa` or `HfBt` image as input for this step, depending on the nature of the editing required.



Example HfBc Image:
Areas to edit are circled

User Intervention

- Remove any non-brain structures and add any brain structures in Brainkit and save.

Notes

- Brain voxels should have a value of either 1 (if editing the HfBt image) or 8 (if editing the HfBc or HfBa image).

STEP 5: CREATE ISOTROPIC T1

3T

```
sb_303_isotropicT1 T1_nu
```

1.5T

```
sb_303_isotropicT1 T1
```

Purpose

Creates a T1 image with isotropic voxel sizes.

STEP 6: GENERATE ACPC ROTATION MATRIX

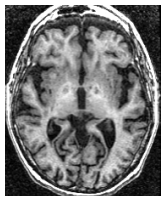
Input

3T

`T1_nu_fc.img`

1.5T

`T1_fc.img`



User Intervention

- ACPC align the isotropic T1 image in analyze (Creating `acpc.mat`).

STEP 7: SEGMENT AND ACPC ALIGN IMAGES

3T

```
sb_304_segment_and_acpc -t1acq T1 -hfb T1acq_nu_HfBd
```

1.5T

```
sb_304_segment_and_acpc -t1acq T1 -hfb T1acq_HfBd -noic
```

Sunnybrook 1.5T with 8 bit T1

```
sb_104_segment_and_acpc -t1acq T1 -hfb T1acq_HfBd
```

** `acpc.mat` and `T1_nu_fc.img` or `T1_fc.img` must be present in dir.

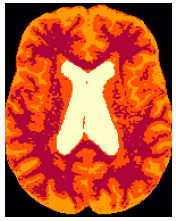
Purpose

- sb_304_segment_and_acpc will segment the T1 (correcting for intensity inhomogeneities using N3 and the good HfB mask prior to segmentation, if requested with the –noic option), and create the ACPC-aligned T1 and T1 erode images.

STEP 8: RELABEL SEGMENTATION

Input

`T1_seg.img`



User Intervention

- Relabel segmentation image by assigning ventricles the value 7 and remove cerebellum in brainkiit or similar image editor program.

STEP 9: SABRE LANDMARKING

Input

`acpc_T1_erode.img`
`acpc_T1.img`

User Intervention

- Perform SABRE landmarking in Analyze (Creating `grid.txt` and `lobtrace.obj`).

STEP 10: PARCELLATE BRAIN

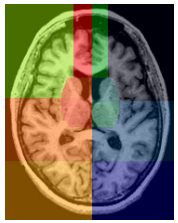
3T/1.5T

```
sb_305_parcellate -t1acpc acpc_T1_erode -seg T1_seg_vcsf_woc
```

Sunnybrook 1.5T with 8 bit T1

```
sb_105_parcellate -t1acpc acpc_T1_erode -seg T1_seg_vcsf_woc
```

** `grid.txt`, `lobtrace.obj` and `acpc.mat` must be present in dir.



Purpose

- Creates lobar mask in T1 acquisition space which parcellates the brain into 26 different regions.

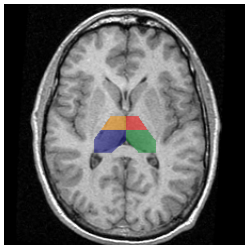
STEP 11: PARCELLATE THALAMUS (OPTIONAL)

3T/1.5T

```
sb_306_parcellate_thalamus -t1acpc acpc_T1
                           -out thalamus
                           -mat acpc_to_T1.mat (or .air)
                           -grid grid.txt
```

Purpose

- Parcellate the thalamus into 4 regions.



STEP 12: PARCELLATE CINGULATE (OPTIONAL)

User Intervention

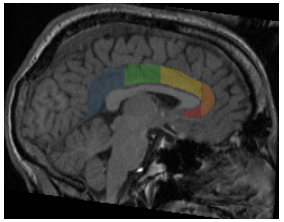
- Perform cingulate landmarking in Analyze (creating cingulate_tracing.obj)

3T/1.5T

```
sb_307_parcellate_cingulate -t1acpc acpc_T1
                            -out cingulate
                            -obj cingulate_tracing.obj
                            -mat acpc_to_T1.mat (or .air)
                            -grid grid.txt
```

Purpose

- Parcellate the cingulate cortex into 5 regions.



STEP 13: PARCELLATE CHOLINGERGIC FIBERS (OPTIONAL)

Method 1: More accurate, but requires cingulate mask from step 12

3T/1.5T

```
sb_308_parcellate_cholinergic -out cholinergic
                              -cing cingulate_acpc
                              -mat acpc_to_T1.mat (or .air)
                              -grid grid.txt
                              -lob acpc_lobmask_filled
```

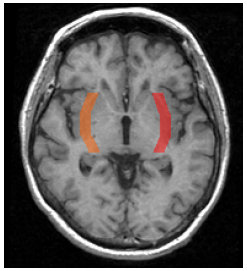
Method 2: Less accurate, but no user intervention required

3T/1.5T

```
sb_308_parcellate_cholinergic -out cholinergic
                              -mat acpc_to_T1.mat (or .air)
                              -grid grid.txt
                              -lob acpc_lobmask_filled
```

Purpose

- Segment cholinnergic fibers into 2 regions.



SUMMARY OF SABRE REGIONS	
REGION	**#**
Left Superior Frontal	4
Left Middle Frontal	5
Left Inferior Frontal	6
Left Medial Inferior Frontal	7
Left Superior Parietal	8
Left Inferior Parietal	9
Left Occipital	10
Left Anterior Temporal	11
Left Posterior Temporal	12
Left Anterior Basal Ganglia and Thalamus	13
Left Posterior Basal Ganglia and Thalamus	14
Left Medial Superior Frontal	15
Left Medial Middle Frontal	16
Right Superior Frontal	17
Right Middle Frontal	18
Right Inferior Frontal	19
Right Medial Inferior Frontal	20
Right Superior Parietal	21
Right Inferior Parietal	22
Right Occipital	23
Right Anterior Temporal	24
Right Posterior Temporal	25
Right Anterior Basal Ganglia and Thalamus	26
Right Posterior Basal Ganglia and Thalamus	27
Right Medial Superior Frontal	28

SUMMARY OF GENERATED FILES				
FILENAME	**IMAGE/FILE**	**3T/1.5T**	**BIAS-CORRECTION (3T data)**	**SKULL-ON**
input				
*_T1.img	T1 (16 bit Analyze format)	N/A	None	Y
*_P.img	PD (16 bit Analyze format)	N/A	None	Y
*_T2.img	T2 (16 bit Analyze format)	N/A	None	Y
*_FL.img	FLAIR (16 bit Analyze format)	N/A	None	Y
sb_301_coregister				
*_T1_nu.img	T1	3T	N3 (coarse brain mask)	Y
*_P_nu.img	PD	3T	N3 (coarse brain mask)	Y
*_T2_nu.img	T2	3T	N3 (coarse brain mask)	Y
*_FL_nu.img	FLAIR	3T	N3 (coarse brain mask)	Y
*_T1acq_nu_T2.img	T2 in T1 acquisition space, trilinear interpolation	3T	N3 (coarse brain mask)	Y
*_T1acq_T2.img	T2 in T1 acquisition space, sinc interpolation	3T/1.5T	None	Y
*_T1acq_nu_P.img	PD in T1 acquisition space, trilinear interpolation	3T	N3 (coarse brain mask)	Y
*_T1acq_P.img	PD in T1 acquisition space, sinc interpolation	3T/1.5T	None	Y
*_T1acq_nu_FL.img	FLAIR in T1 acquisition space, trilinear interpolation	3T	N3 (coarse brain mask)	Y
*_T1acq_FL.img	FLAIR in T1 acquisition space, sinc interpolation	3T/1.5T	None	Y
*_FL_to_T1.omat	FLAIR to T1 rotation matrix (FSL)	3T/1.5T	N/A	N/A
*_FL_to_T1_inverse.omat	T1 to FLAIR rotation matrix (FSL)	3T/1.5T	N/A	N/A
*_T2_to_T1.omat	T2 to T1 rotation matrix (FSL)	3T/1.5T	N/A	N/A
*_T2_to_T1_inverse.omat	T1 to T2 rotation matrix (FSL)	3T/1.5T	N/A	N/A
user intervention				
*_T1acq_nu_T2_bet	T2 bet in T1 acquisition space	3T	N3 (coarse brain mask)	N (bet)
*_T1acq_T2_bet	T2 bet in T1 acquisition space	1.5T	None	N (bet)
sb_302_hfb				
*_HfBt.img	Head-from-Brain (new template hfb method)	3T/1.5T	N/A	N (hfb)
*_HfBa.img	Head-from-Brain (old auto hfb method)	3T/1.5T	N/A	N (hfb)
*_HfBc.img	Head-from-Brain (combined new/old method)	3T/1.5T	N/A	N (hfb)
*_bin2_x100.img	Head-from-Brain template in T1 space	N/A	N/A	N/A
*_template_to_subject.omat	Head-from-Brain template to T1 rotation matrix	N/A	N/A	N/A
user intervention				
*_HfBd.img	Head-from-Brain after manual edits in brainkit	3T/1.5T	N/A	N (hfb)
sb_303_isotropicT1				
*_fc.img	Isotropic T1	3T/1.5T	N3 (coarse brain mask)	Y
user intervention				
*_acpc.mat	T1-to-ACPC rotation matrix (Analyze)	3T/1.5T	N/A	N/A

SUMMARY OF GENERATED FILES				
FILENAME	**IMAGE/FILE**	**3T/1.5T**	**BIAS-CORRECTION (3T data)**	**SKULL-ON**
*_T1_to_acpc.air or .mat	T1 to ACPC rotation matrix	3T/1.5T	N/A	N/A
*_acpc_to_T1.air or .mat	ACPC to T1 rotation matrix	3T/1.5T	N/A	N/A
*_T1_masked.img	T1	3T/1.5T	None	N (hfb)
*_T1_nu_final.img	T1	3T	N3 (good brain mask)	Y
*_T1_nu_final_masked.img	T1	3T	N3 (good brain mask)	N (hfb)
*_T1_seg	T1 segmentation	3T/1.5T	N/A	N (hfb)
*_acpc_T1	T1 in ACPC space	3T/1.5T	N3 (good brain mask)	Y
*_acpc_T1_erode	T1 erode in ACPC space	3T/1.5T	N3 (good brain mask)	N (erode)
*_T1acq_erode	T1 erode in T1 acquisition space	3T/1.5T	N3 (good brain mask)	N (erode)
user intervention				
*_grid.txt	Text file with SABRE landmark locations (Analyze)	3T/1.5T	N/A	N/A
*_lobtrace.obj	Object map with lobe tracings (Analyze)	3T/1.5T	N/A	N/A
*_T1_seg_vcsf.img	T1 segmentation with ventricles relabelled	3T/1.5T	N/A	N (hfb)
*_T1_seg_vcsf_woc.img	T1 segmentation with ventricles relabelled and cerebellum removed	3T/1.5T	N/A	N (hfb)
sb_305_parcellate_brain				
*_acpc_lobmask_unfilled.img	ACPC aligned lobar mask (may have holes)	3T/1.5T	N/A	N/A
*_acpc_lobmask_filled.img	ACPC aligned lobar mask (no holes)			
*_T1acq_lobmask_unfilled.img	Lobar mask in T1 acquisition space (may have holes)	3T/1.5T	N/A	N/A
*_T1acq_lobmask_filled.img	Lobar mask in T1 acquisition space (no holes)			
*_T1_rotcheck.img	Image used to check rotation from ACPC to T1 space	3T/1.5T	N3 (good brain mask)	N/A
*_stats.txt	Regional statistics for the 26 lobar regions	3T/1.5T	N/A	N/A
sb_306_parcellate_thalamus				
*_thalmus_T1acq	Thalamus mask in T1 acquisition space	3T/1.5T	N/A	N/A
*_thalamus_acpc	Thalamus mask in ACPC space	3T/1.5T	N/A	N/A
user intervention				
cingulate_tracing.obj	Object map with cingulate tracing (Analyze)	3T/1.5T	N/A	N/A
sb_307_parcellate_cingulate				
*_cingulate_T1acq	Cingulate mask in T1 acquisition space	3T/1.5T	N/A	N/A
*_cingulate_acpc	Cingulate mask in ACPC space	3T/1.5T	N/A	N/A
sb_308_parcellate_cholinergic				
*_cholinergic_T1acq	Cholinergic fibers mask in T1 acquisition space	3T/1.5T	N/A	N/A
*_cholingergic_acpc	Cholinergic fibers mask in ACPC space	3T/1.5T	N/A	N/A

FLAIR LESION SEGMENTATION – FLEX

OVERVIEW

- FLEX (Fuzzy Lesion Extractor) segments white matter hyperintensities (WMH) on a FLAIR image using a Fuzzy C-Means clustering technique (step 2 below). The FLAIR image is coregistered to the MNI average T1 image using FSL's flirt and the MNI white matter template is used to minimize false positive voxels.
- If available, the T1 segmentation with VCSF uniquely labelled can be used to reclaim black holes and relabel periventricular and deep white lesions (steps 4 and 5 below).
- sb_flex requires that the FLAIR image be masked and corrected for intensity inhomogeneities. For Sunnybrook 3T data, the **`sb_flair_ic`** script is available for this purpose and uses John Sled's N3 to correct for intensity inhomogeneities (-iterations 150, -stop 0.0001, -distance 55).

IMAGE REQUIREMENTS			
Command/Task	**Step(s)**	**Image Type**	**Processing**
sb_flair_ic	1	a) FLAIR	Unprocessed, uncorrected for intensity inhomogeneities, coregistered to brain mask
		b) Decent brain mask	"1"s or "8"s for brain, "0"s for non-brain
sb_flex	2	a) FLAIR	Skull-off, corrected for intensity inhomogeneities (if necessary)
segment_black_holes	4	a) T1 segmentation	VCSF relabelled, coregistered to T1
relabel_ventricular_wmh	5	a) T1 segmentation	VCSF relabelled, coregistered to T1

SOFTWARE REQUIREMENTS			
Command/Task	**Step(s)**	**Requirements (Within Sunnybrook)**	**Requirements (Outside Sunnybrook)**
sb_flair_ic	1	--	N/A
sb_flex	2	--	FSL (flirt): optional, if needing false positive minimization
edit FLAIR segmentation	3	Analyze (or equivalent)	Analyze (or equivalent)
segment_black_holes	4	--	--
relabel_ventricular_wmh	5	--	--

STANDARD INPUT

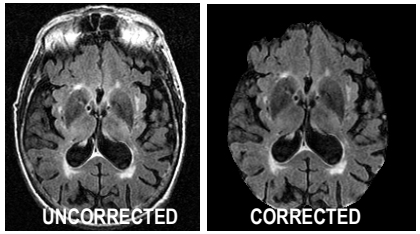
`FL`	Uncorrected FLAIR image, coregistered to HfB brain mask
`HfB`	Edited head-from-brain (HfB) image, coregistered to FLAIR
`T1_seg_vcsf`	T1 segmentation with VCSF relabelled, coregistered to FLAIR, optional but required for steps 4 and 5

STEP 1: FLAIR INHOMOGENEITY CORRECTION AND SKULL REMOVAL

```
sb_flair_ic -fl FL -hfb HfB
```

Notes

- Intensity inhomogeneity correction parameters were optimized for 3T data (VHAD, 2008). Changes may be required for 1.5T data or new 3T data.



STEP 2: SEGMENT LESIONS

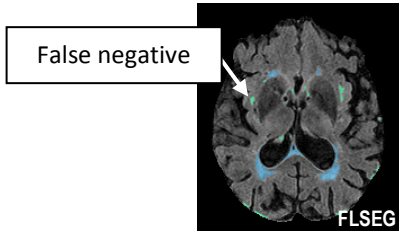
```
sb_flex -fl FL_mc
```

If running outside Sunnybrook, FSL is required (if needing false positive minimization), and the –fslinstalled option should be added to the above command.

See the following FLEX configuration section for details on selecting and customizing options for the `sb_flex` script.

Notes

- Some false positives and false negatives will remain on the segmentation image.
- Alternate false positive minimization strategies can be applied to the `FL_mc_axcor` image (e.g. direct masking with the WM template or possibly the freesurfer subcortical segmentation result).



Blue:
Probable WMH (1s)
Green:
Probable artefact (2s)

STEP 3: EDIT SEGMENATATION

User Intervention
Remove any false positives and reclaim any false negatives.

Input
`FL_mc_flwmt_lesions`

Notes
If using Analyze to perform the editing, the image will need to be converted into an object map. This can be done in Analyze or using the command line tools `obj2img` and `img2obj`.

STEP 4: RELABEL BLACK HOLES

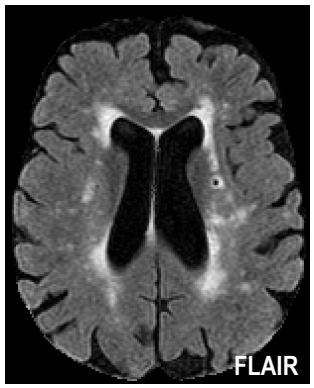
```
segment_black_holes -t1 T1_seg_vcsf.img
                    -fl FL_mc_flwmt_lesions_edit.img
                    -out FL_mc_edit_bh.img
```

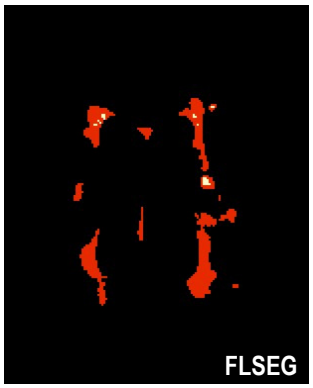
VCSF, CSF and GM must have different values on the T1 segmentation. If the values differ than the default values used at Sunnybrook (7,5,4) use the `-csf`, `-gm` and `-vcsf` options available for the `segment_black_holes` and `relabel_ventricular_wmh` (step 5) commands.

The `-fl` image should contain only lesion voxels. Other voxel types (e.g. probable non-lesion voxels) should be removed. If necessary, removal of non-lesion voxels can be accomplished using the `imgmath` or `mask_img` command.

Notes

- The T1 segmentation is used to find black holes.





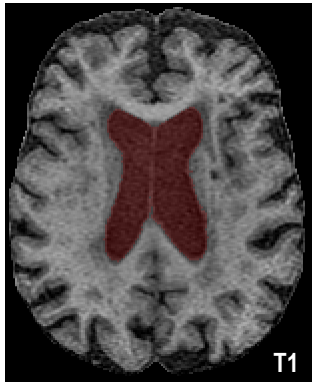
Red:
WMH (1s)
White:
Black holes (2s)

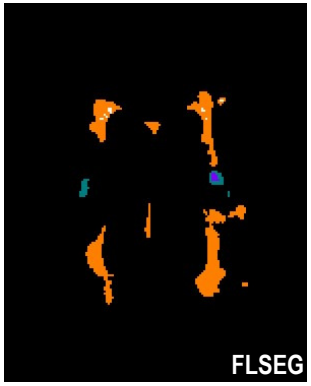
STEP 5: RELABEL VENTRICULAR LESIONS

```
relabel_ventricular_wmh -t1seg T1_seg_vcsf.img
                        -lseg FL_mc_edit_bh.img
                        -out FL_mc_edit_bh_vwmh.img
```

Notes

- Any lesion connected in 3D to VCSF is labelled as a periventricular (PV) lesion.





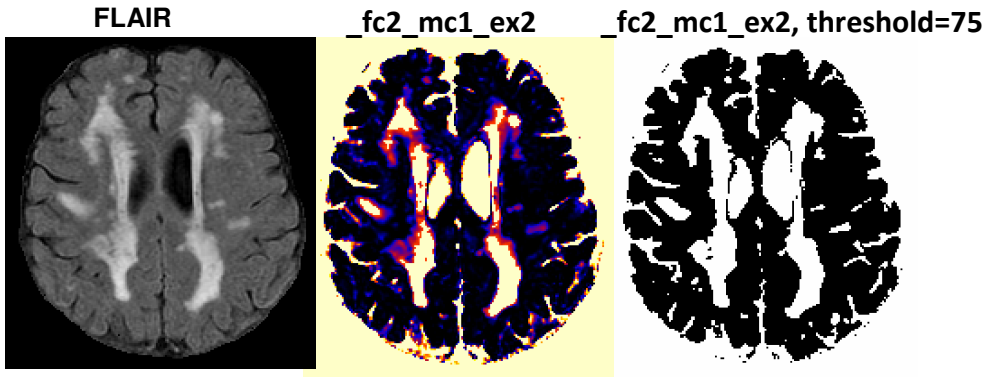
Blue:
Deep WMH (1s)
Purple:
Deep black holes (2s)
Orange:
PV WMH (3s)
White:
PV back holes (4s)

SUMMARY OF GENERATED FILES	
FILENAME	**IMAGE/FILE**
sb_flair_ic	
_mc.img	FLAIR image, masked and corrected for intensity inhomogeneities
sb_flex	
_mc_axcor_flhyper.img	All hyperintense voxels identified; no false positive minimization is performed
_mc_flf.img	Anisotropic diffusion filtered FLAIR
_mc_flf_cor_fc2_mc1_ex2_ax.img	Fuzzy cluster results performed in the coronal orientation, reoriented back to axial plane
_mc_flf_fc2_mc1_ex2.img	Fuzzy cluster results performed in the axial orientation
_mc_flf_norm.img	Intensity normalized, diffusion filtered FLAIR.
_mc_flwmt_lesions.img	Contains “1”s for probable lesions and “2”s for probable artifact voxels
_mc_WMT_to_subject.dof/.omat	Transformation matrix from MNI space to subject space
_mc_WMT_to_subject.img	MNI white matter template transformed to subject space
User Intervention	
_mc_flwmt_lesions_edit	FLAIR segmentation result edited to remove false positives and/or reclaim false negatives
segment_black_holes	
_mc_edit_bh	“1”s for WMH and “2”s for black holes
relabel_ventricular_wmh	
_mc_edit_bh_vwmh	“1s” for deep WMH, “2”s for deep black holes, “3”s for ventricular WMH and “4”s for ventricular black holes

CONFIGURING FLEX

There are 3 thresholds that can be adjusted and passed to the `sb_flex` command. These thresholds should remain fixed across different subjects, but can be modified for images acquired using different acquisition parameters.

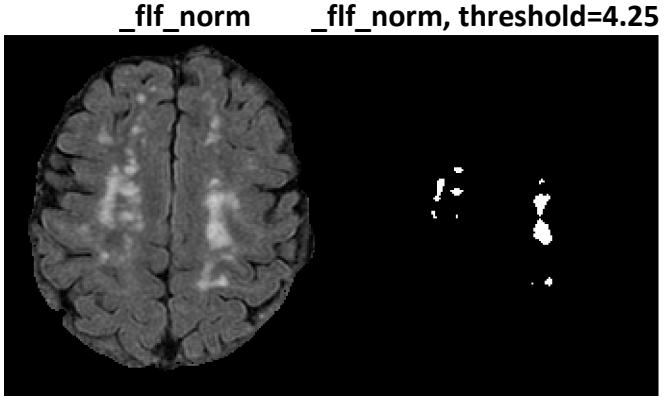
These thresholds have been selected for 3T Sunnybrook data (VHAD, 2008) and are the default values for the `sb_flex` command (-at = 100, -ct = 100, -wt = 160, -nt = 4.6). For other data, appropriate threshold values can be selected by running the FLEX.sh script on 2 or 3 test FLAIR volumes with varying degrees of lesion load and inspecting the result as follows:



1. **`flthresh_ax (-at) / flthresh_cor (-ct)`**: These thresholds should be selected by referring to `the _flf_fc2_mc1_ex2` and `_flf_cor_fc2_mc1_ex2_ax` images. Any voxel greater than this threshold should correspond to either hyperintense or CSF/background voxels (see example images on the right). These thresholds may need to be changed if the image contrast or disease state is substantially different than the test volumes used at Sunnybrook Health Sciences Centre.

2. **`wmt_threshold (-wt):`** This threshold should be selected by referring to the `_WMT_to_subject` image. This threshold only affects the classification of hyperintense voxels as probable lesion or probable artifact in the `_flwmt_lesions` image.

   Note: The `_WMT_to_subject` image can be thresholded and used to mask the `_axcor_flhyper` image directly for an alternative false positive minimization strategy.



3. **`norm_thresh (-nt)`**: This threshold should be selected by referring to the `_flf_norm` image. Any voxel greater than this threshold should be obviously hyperintense for all test subjects (see example images on the right). This threshold will have little effect at small lesion loads, but can prevent underestimation of the true lesion volume at high lesion loads (WMH>~5% total slice volume). This threshold should be set conservatively; it should not define the boundary of the hyperintensities.

PD/T2 LESION SEGMENTATION – LESION EXPLORER

OVERVIEW

- Lesion Explorer segments white matter hyperintensities (WMHs) on PD/T2 images. The T1 segmentation image is used to remove ventricular CSF (VCSF) false positives. Sulcal CSF and GM false positives are removed either by using an edge cleaning technique or, optionally, by using a CSFGM mask created from the T1 image. Any remaining false positives or false negatives are manually excluded or included. The T1 segmentation is then used to relabel black holes and segment periventricular and deep white lesions.

IMAGE REQUIREMENTS			
Command/Task	**Step(s)**	**Image Type***	**Processing**
sb_le	1	a) T2	Corrected for intensity inhomogeneities (if necessary)
		b) PD	Corrected for intensity inhomogeneities (if necessary)
		c) T1 (optional)	Corrected for intensity inhomogeneities (if necessary), required if using the –csfgm option
		d) T1 segmentation	VCSF relabelled, cerebellum removed (removal of cerebellum is optional, but advantageous, especially if using –edge option)
relabel_black_holes	3	d) T1 segmentation	VCSF relabelled
relabel_ventricular_wmh	4	d) T1 segmentation	VCSF relabelled

* The T1, T2 and PD input images must be 16 bit Analyze format. The T1 segmentation image can be 8 or 16 bit but the maximum value in the image must be < 255. Input images may be unmasked as they will be masked by the T1 segmentation prior to lesion segmentation.

SOFTWARE REQUIREMENTS			
Command/Task	**Step(s)**	**Requirements (Within Sunnybrook)**	**Requirements (Outside Sunnybrook)**
sb_le	1	--	--
edit LE segmentation	2	Analyze or Lesion Explorer GUI or equivalent	Analyze or Lesion Explorer GUI or equivalent
relabel_black_holes	3	--	--
relabel_ventricular_wmh	4	--	--

STANDARD INPUT

`T1`	T1 image
`T1_seg_vcsf_woc`	T1 segmentation, VCSF relabelled, cerebellum removed
`T1acq_T2`	Coregistered T2
`T1acq_P`	Coregistered PD

Notes

- All input images must be coregistered and corrected for intensity inhomogeneities (if present).

STEP 1: SEGMENT LESIONS

```
sb_le -pd T1acq_P -t2 T1acq_T2 -t1 T1 -out LE -seg T1_seg_vcsf_woc
```

the above command is equivalent to:

```
sb_le -pd T1acq_P -t2 T1acq_T2 -t1 T1 -out LE -seg T1_seg_vcsf_woc
      -vdilateball 1 -csfgm -minobjectsize 5 -vcsf 7 -ct 150
      -pdt 0.05 -t2t 0.02
```

See the following Lesion Explorer configuration section for details on selecting and customizing options for the `sb_le` script.

Typically the T1_seg_vcsf_woc is created during the SABRE process. If this is not the case, the T1 segmentation can be created using the following command, and subsequently edited to relabel VCSF and (optionally) remove the cerebellum. The T1 should be masked to remove non-brain and corrected for intensity inhomogeneities (if present) prior to segmentation.

16 bit T1

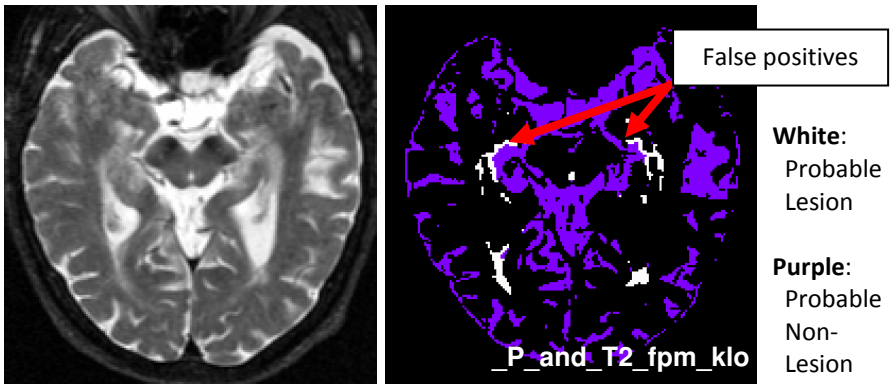
```
T1seg_3T T1_masked T1_seg -bfgw 0 5 4 3 -win_size 10
```

8 bit T1

```
T1seg T1_masked T1_seg -bfgw 0 5 4 3
```

Notes

- Some false positives and/or false negatives will remain on the final segmentation. Alternative false positive minimization procedures can be applied to the `_P_and_T2_lsegment` image if desired.



STEP 2: EDIT SEGMENTATION

User Intervention

- Remove any false positives and/or reclaim any false negatives.

Input

`LE_P_and_T2_lsegment_fpm_klo` **(or)** `LE_P_and_T2_lsegment_fpm_masked_klo`

The choice of input will depend on the nature of the edits required and the software package used for performing the edits.

STEP 3: RELABEL BLACK HOLES

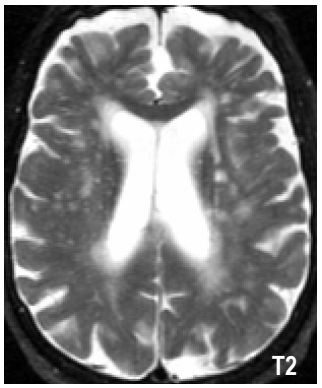
```
relabel_black_holes -lseg LE_P_and_T2_lsegment_edit.img
                    -t1seg T1_seg_vcsf_woc.img
                    -out LE_P_and_T2_lsegment_edit_bh.img
```

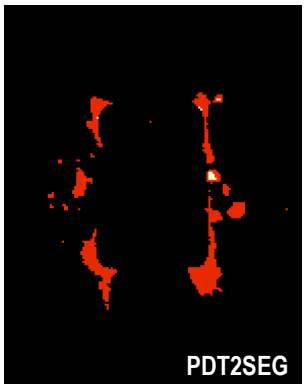
The `-lseg` image should contain only lesion voxels. Other voxel types (e.g. probable non-lesion) should be removed. If necessary, removal of non-lesion voxels can be accomplished using the `imgmath` or `mask_img` command.

VCSF, CSF and GM must have different values on the T1 segmentation. If the values differ than the default values used at Sunnybrook (7,5,4) use the `-csf`, `-gm` and `-vcsf` options when running the `segment_black_holes` and `relabel_ventricular_wmh` (step 4) commands.

Notes

- The T1 segmentation is used to find black holes.





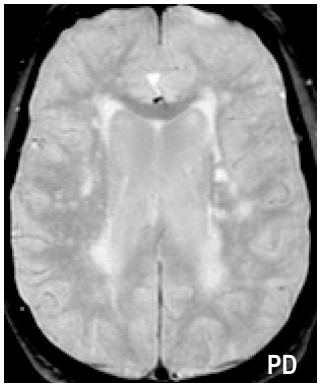
Red:
WMH (1s)
White:
Black Holes (2s)

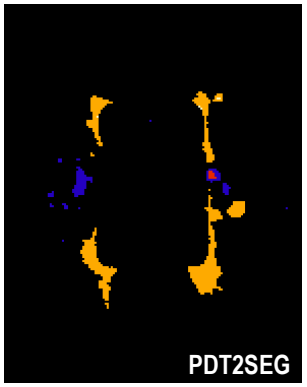
STEP 4: RELABEL VENTRICULAR LESIONS

```
relabel_ventricular_wmh -lseg LE_P_and_T2_lsegment_edit_bh.img
                        -t1seg T1_seg_vcsf_woc.img
                        -out LE_P_and_T2_lsegment_edit_bh_vmh.img
```

Notes

- Any lesion connected in 3D to VCSF is relabelled as a periventricular (PV) lesion.





Blue:
Deep WMH (1s)
Red:
Deep Black Holes (2s)
Orange:
PV WMH (3s)
White:
PV Black Holes (4s)

SUMMARY OF GENERATED FILES	
FILENAME	**IMAGE/FILE**
sb_le	
_P_and_T2_lsegment_fpm_klo	LE segmentation result, false positive minimized, lesions greater than or equal to minobjectsize retained as probable lesion, "1"s for probable lesion, "2"s for probable non-lesion
_P_and_T2_lsegment_fpm_masked	LE segmentation result, false positives minimized, "1"s for probable lesion
_P_and_T2_lsegment_fpm_masked_klo	LE segmentation result, false positives minimized, lesions greater than or equal to minobjectsize retained as probable lesion, "1"s for probable lesion
_P_and_T2_lsegment	LE segmentation result, no false positive minimization performed
_T1_csfgm	CSFGM mask
_masked_fc4_mc2_ex2	T1 FCM results (CSF class) used to create CSFGM mask
_P_lsegment	LE segmentation result for PD image *(debug mode only)*
_T2_lsegment	LE segmentation result for T2 image *(debug mode only)*
_T2_masked	Dilated VCSF removed from T2 *(debug mode only)*
_P_masked	Dilated VCSF removed from PD *(debug mode only)*
_dilated	T1 segmentation result with dilated ventricles *(debug mode only)*
User Intervention	
_P_and_T2_lsegment_edit	LE segmentation result, edited to remove false positives and/or reclaim false negatives, "1"s for lesions
relabel_black_holes	
_P_and_T2_lsegment_edit_bh	"1"s for WMH and "2"s for black holes
relabel_ventricular_wmh	
_P_and_T2_lsegment_edit_bh_vwmh	"1s" for deep WMH, "2"s for deep black holes, "3"s for ventricular WMH and "4"s for ventricular black holes

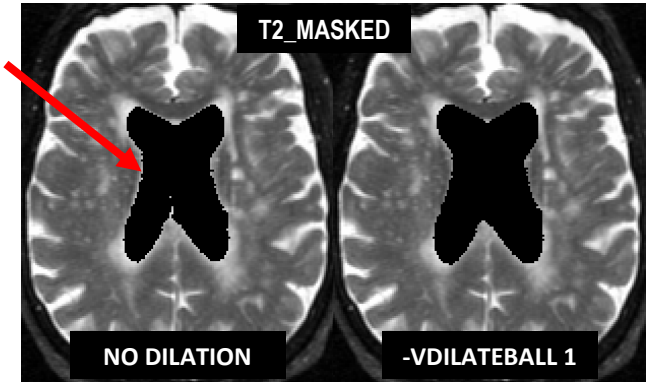
CONFIGURING LESION EXPLORER

- There are several options that can be adjusted for different datasets. These options should be kept fixed for a given set of acquisition parameters and disease state.
- These options have been selected for Sunnybrook 3T data (VHAD 2008) and are the default values for the `sb_le` script (`-vdilateball 1 -csfgm -minobjectsize 5 -vcsf 7 -pdt 0.05 -t2t 0.02 -ct 150`).

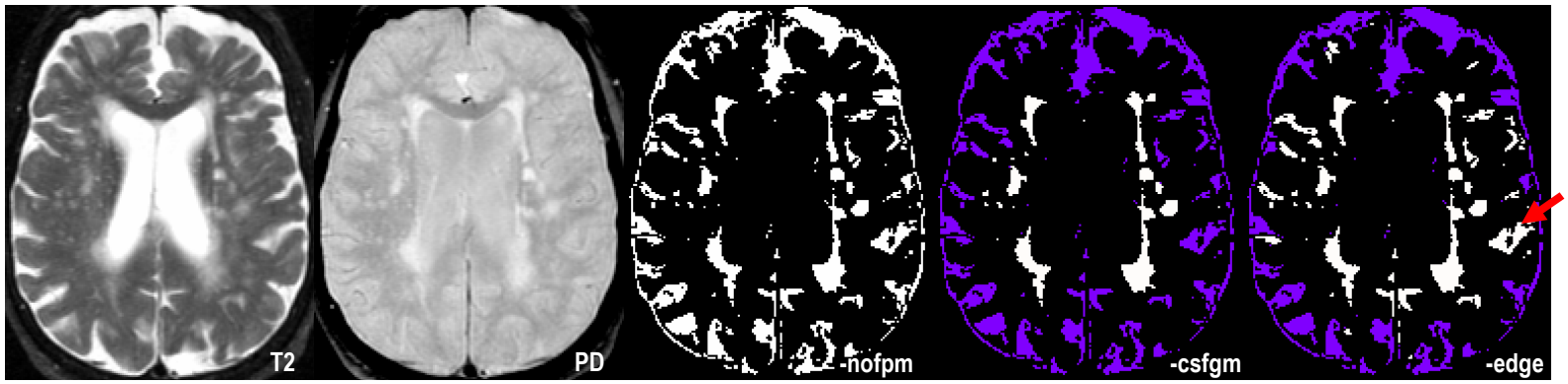
SUMMARY OF OPTIONS	
OPTION	**FUNCTION**
-vdilatecross <int>:	Dilate ventricular CSF with a cross structuring element of radius <int>
-vdilateball <int>:	Dilate ventricular CSF with a ball structuring element of radius <int>
-csfgm:	Use new CSFGM mask for false positive minimization, requires T1
-edge:	Use old edge cleaner for false positive minimization, does not require T1
-nofpm:	No false positive minimization performed, does not require T1
-minobjectsize <int>:	Keep lesions that are greater than or equal to <int> voxels
-vcsf <int>:	Value assigned to ventricular CSF on the T1 segmentation
-debug:	Keeps intermediary files, useful for initial parameter selection
-pdt <float>:	PD threshold for Lsegment (>0 and <1)
-t2t <float>:	T2 threshold for Lsegment (>0 and <1)
-ct <int>:	Threshold for `fc4_mc2_ex2` image used to create CSFGM mask (>0 && <1000)
-forcefcm:	Forces generartion of the `fc4_mc2_ex2` image if it already exists (if rerunning the script a second time)

1. **`-vdilatecross <int> / -vdilateball <int>`**: Specifies the 2D dilation operation to be performed on VCSF. The `-vdilatecross` option is a more conservative dilation option than the `-vdilateball` option. The dilation radius should be chosen to maximize both the number of VCSF partial volume voxels removed and the number of lesion voxels retained. To select an appropriate dilation radius, run the `sb_le` command using the `-debug` and `-nofpm` options and experiment with values for the `-vdilatecross` and `-vdilateball` options. The effect of the dilation operation can be assessed by inspecting the `T2_masked` and/or `P_masked` images.

Partial volume voxels removed by dilating ventricles



2. **`-csfgm / -edge / -nofpm`**: Specifies the false positive minimization strategy to be performed. The `-csfgm` option creates a sulcal CSFGM mask from the T1 image which is then used as a mask to remove false positives. The `-edge` option uses an edge cleaning operation to remove false positives. It is not as effective as the `-csfgm` option, but requires less processing time and does not require a T1 image. The `-nofpm` option can be used if no false positive minimization strategy is needed (other than the standard first step removal of VCSF).



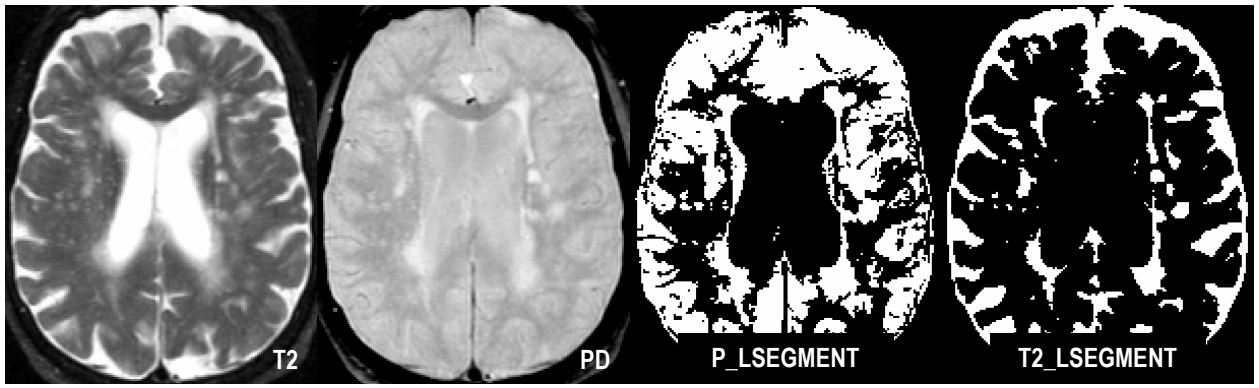
Purple:
Probable non-lesion
White:
Probable lesion

With the `-edge` option, many non-lesion voxels remain classified as probable lesion and therefore more manual editing of the `-edge` output is required.

3. **`-minobjectsize <int>`**: Removes small objects that typically correspond to non-lesion voxels. To retain all objects, set this option to 1.

4. **`-vcsf <int>`**: Specifies the value (between 0 and 255) that has been assigned to VCSF on the T1 segmentation.

5. **`-pdt <int> / -t2t <int>`**: Specifies the threshold (between 0 and 1) used to segment lesions on the PD and T2 images respectively. To select an appropriate value, run the `sb_le` command with the `-debug -t2t 0.05 -pdt 0.05 -nofpm -vdilatecross 0` options. Inspect the output. If lesions voxels are excluded on the `T2_lsegment` image, lower the `-t2t` threshold. If non-lesion voxels are included, increase the `-t2t` threshold. If lesion voxels are excluded on the `P_lsegment` image, increase the `-pdt` threshold. Typically, non-lesion voxels will be included on the PD image and this is OK.



6. **-ct <int>:** Specifies the threshold used for the creation of the CSFGM mask. An appropriate value is an integer between 0 and 1000 that thresholds the `_fc4_mc2_ex2` image so that primarily CSF voxels remain. These voxels will undergo a series of morphological operations to produce the CSFGM mask. Ideally, all voxels shown in purple on the CSFGM mask below will correspond to gray matter and sulcal CSF voxels on the T1. If too many white matter voxels are included in the CSFGM compartment, increase the `-ct` threshold. If not enough CSFGM voxels are included in the CSFGM compartment, lower the `-ct` threshold.

7. **-debug <int>:** Keeps all intermediary files.

8. **-forcefcm:** To save time, if the `fc_mc2_ex2` image already exists (i.e. if the script has already been run) the `fc_mc2_ex2` image will not be recreated. The `-forcefcm` option can be used to override this behaviour. Typically, this option is not necessary because the `fc_mc2_ex2` image does not change unless the input T1 image has been modified. Therefore, any required changes to the CSFGM mask can usually be accomplished using the `-ct` option.

PD/T2 LESION SEGMENTATION – PDT2SEG

OVERVIEW

- pdt2seg segments white matter hyperintensities (WMHs) on PD/T2 images. The T1 segmentation image (with ventricular CSF labelled) can be used to remove ventricular CSF false positives. The T1 image can be used to create a CSFGM mask to remove sulcal CSF and GM false positives. Any remaining false positives or false negatives are manually excluded or included. The T1 segmentation can then used to relabel black holes and segment periventricular and deep white lesions.
- pdt2seg

IMAGE REQUIREMENTS			
Command/Task	**Step(s)**	**Image Type***	**Processing**
sb_pdt2seg_preprocess	1	a) PD	Skull-on, uncorrected
		b) T2	Skull-on, uncorrected
		c) HfB	Head-from-brain mask (0's for non-brain, and 1's or 8's for brain)
sb_pdt2seg	2	a) PD	Masked, corrected for intensity inhomogeneities
		b) T2	Masked, corrected for intensity inhomogeneities
		c) T1 (optional)	Masked, corrected for intensity inhomogeneities (required for false positive minimization)
		d) T1 segmentation (optional)	VCSF relabelled (required for false positive minimization)
relabel_black_holes	4	d) T1 segmentation	VCSF relabelled
relabel_ventricular_wmh	5	d) T1 segmentation	VCSF relabelled

* The input images must be coregistered and in Analyze format.

SOFTWARE REQUIREMENTS			
Command/Task	**Step(s)**	**Requirements (Within Sunnybrook)**	**Requirements (Outside Sunnybrook)**
sb_pdt2seg_preprocess	1	--	N/A
sb_pdt2seg	2	--	--
edit LE segmentation	3	Analyze or equivalent	Analyze or equivalent
relabel_black_holes	4	--	--
relabel_ventricular_wmh	5	--	--

STANDARD INPUT

`T1_masked_nu_final`	T1 image, masked, corrected for intensity inhomogeneities
`T1_seg_vcsf`	T1 segmentation, VCSF relabelled, cerebellum removed
`T1acq_T2`	Coregistered T2, unmasked, uncorrected
`T1acq_P`	Coregistered PD, unmasked, uncorrected
`HfB`	Head-from-brain mask image (1's or 8's for brain, 0's for nonbrain)

Notes

- All input images must be coregistered.

STEP 1: PREPROCESS IMAGES

```
sb_pdt2seg -pd T1acq_P -t2 T1acq_T2 -hfb T1acq_HfB
```

Notes

- PD/T2 images are masked and then corrected for intensity inhomogeneities with John Sled's N3.

STEP 2: SEGMENT LESIONS

```
sb_pdt2seg -pd T1acq_P_mc -t2 T1acq_T2_mc -t1 T1_masked_nu_final -out
SUB -seg T1_seg_vcsf_woc
```

the above command is equivalent to:

```
sb_pdt2seg -pd T1acq_P_mc -t2 T1acq_T2_mc -t1 T1_masked_nu_final
           -out sub -seg T1_seg_vcsf_woc -vdilateball 1 -csfgm
           -minobjectsize 5 -vcsf 7 -ct 150 -pdt 0.02 -t2t 0.05
```

See the following configuration section for details on selecting and customizing options for the `sb_le` script.

Typically the T1_seg_vcsf_woc is created during the SABRE process. If this is not the case, the T1 segmentation can be created using the following command, and subsequently edited to relabel VCSF and (optionally) remove the cerebellum. The T1 should be masked to remove non-brain and corrected for intensity inhomogeneities (if present) prior to segmentation.

16 bit T1

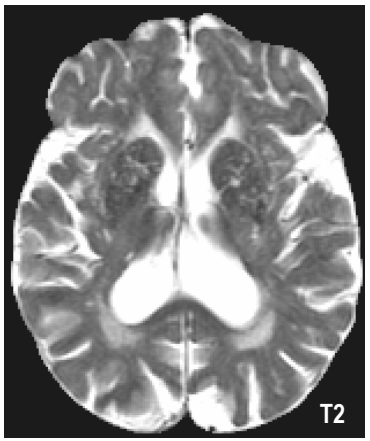
```
T1seg_3T T1_masked T1_seg -bfgw 0 5 4 3 -win_size 10
```

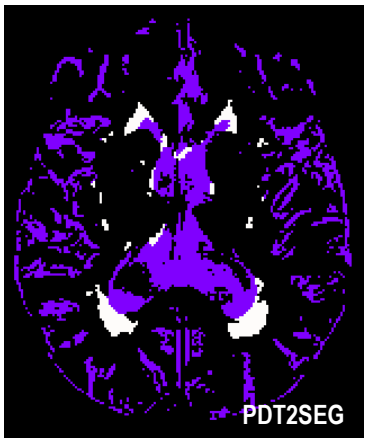
8 bit T1

```
T1seg T1_masked T1_seg -bfgw 0 5 4 3
```

Notes

- Some false positives and/or false negatives will remain on the final segmentation. Alternative false positive minimization procedures can be applied to the `_pdt2seg` image if desired.





White: Probable Lesion

Purple: Probable Non-Lesion

STEP 3: EDIT SEGMENTATION

User Intervention

- Remove any false positives and/or reclaim any false negatives.

Input

`SUB_pdt2seg_pdt2seg_fpm_klo`

STEP 4: RELABEL BLACK HOLES

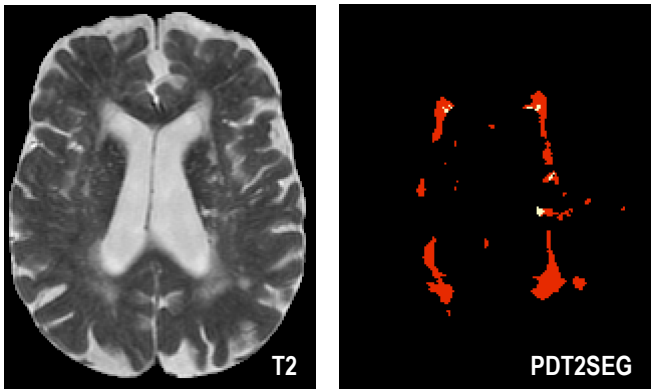
```
relabel_black_holes -lseg SUB_pdt2seg_fpm_klo_edit.img
                    -t1seg T1_seg_vcsf.img
                    -out SUB_pdt2seg_fpm_klo_edit_bh.img
```

The `-lseg` image should contain only lesion voxels. Other voxel types (e.g. probable non-lesion) should be removed. If necessary, removal of non-lesion voxels can be accomplished using the `imgmath` or `mask_img` command.

VCSF, CSF and GM must have different values on the T1 segmentation. If the values differ than the default values used at Sunnybrook (7,5,4) use the `-csf`, `-gm` and `-vcsf` options when running the `segment_black_holes` and `relabel_ventricular_wmh` (step 4) commands.

Notes

- The T1 segmentation is used to find black holes.



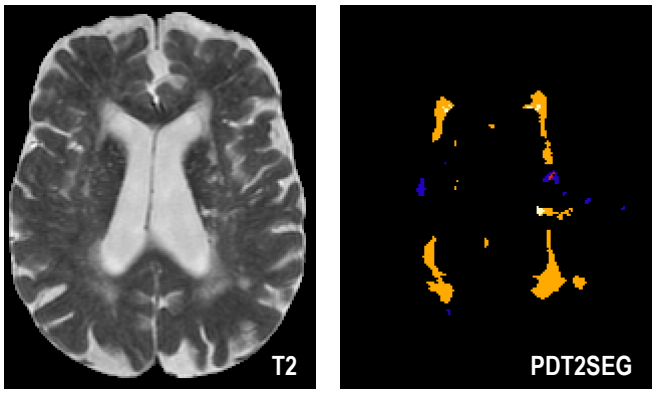
Red:
WMH (1s)
White:
Black Holes (2s)

STEP 5: RELABEL VENTRICULAR LESIONS

```
relabel_ventricular_wmh -lseg SUB_pdt2seg_fpm_klo_edit_bh.img
                        -t1seg T1_seg_vcsf_woc.img
                        -out SUB_pdt2seg_fpm_klo_edit_bh_vwmh.img
```

Notes

- Any lesion connected in 3D to VCSF is relabelled as a periventricular (PV) lesion.



Blue:
Deep WMH (1s)
Red:
Deep Black Holes (2s)
Orange:
PV WMH (3s)
White:
PV Black Holes (4s)

SUMMARY OF GENERATED FILES	
FILENAME	**IMAGE/FILE**
sb_pdt2seg_preproces	
_T2_mc	T2 image, masked and corrected for intensity inhomogeneities
_P_mc	PD image, masked and corrected for intensity inhomogeneities
sb_pdt2seg	
_pdseg	Segmentation result for PD image, no false positive minimization
_t2seg	Segmentation result for T2 image, no false positive minimization
_pdt2seg	Combined PD/T2 segmentation result, no false positive minimization
_fc4_mc2_ex2	Fuzzy C-Means result for T1 CSF class (T1/T1seg input required)
_pdt2seg_fpm_klo	PD/T2 segmentation result, small objects removed, 1's for probable non-lesion, 2's for probable lesion (T1/T1seg or T1seg input required)
_pdt2seg_fpm_masked_klo	PD/T2 segmentation result, small objects removed, 1's for probable lesion (T1/T1seg or T1seg input required)
_dilated	T1 segmentation with dilated ventricles (T1seg input required)
User Intervention	
_pdt2seg_fpm_masked_klo_edit	PD/T2 segmentation result, edited to remove false positives and/or reclaim false negatives, "1"s for lesions
relabel_black_holes	
_pdt2seg_fpm_masked_klo_edit_bh	"1"s for WMH and "2"s for black holes
relabel_ventricular_wmh	
_pdt2seg_fpm_masked_klo_edit_vwmh	"1s" for deep WMH, "2"s for deep black holes, "3"s for ventricular WMH and "4"s for ventricular black holes

CONFIGURING PDT2SEG

- There are several options that can be adjusted for different datasets. These options should be kept fixed for a given set of acquisition parameters and disease state.

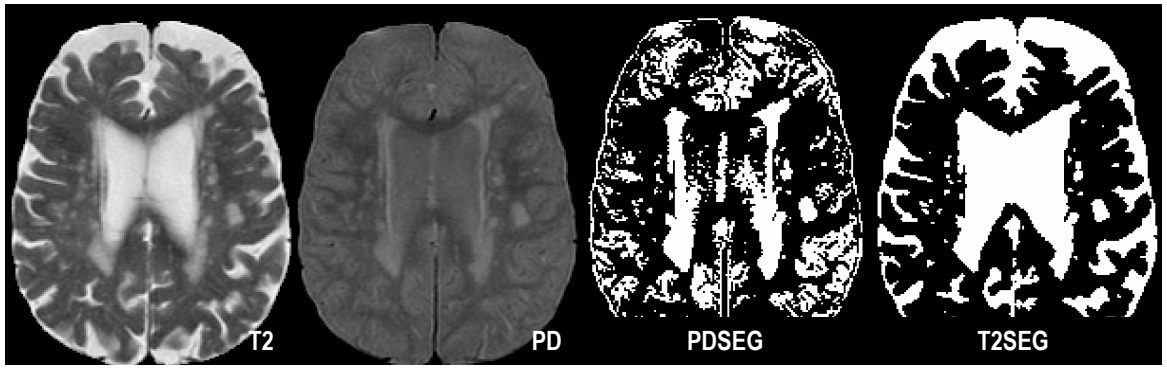
SUMMARY OF OPTIONS	
OPTION	**FUNCTION**
-vdilatecross <int>:	Dilate ventricular CSF with a cross structuring element of radius <int>
-vdilateball <int>:	Dilate ventricular CSF with a ball structuring element of radius <int>
-minobjectsize <int>:	Keep lesions that are greater than or equal to <int> voxels
-vcsf <int>:	Value assigned to ventricular CSF on the T1 segmentation
-pdt <float>:	PD threshold for Lsegment (>0 and <1)
-t2t <float>:	T2 threshold for Lsegment (>0 and <1)
-ct <int>:	Threshold for `fc4_mc2_ex2` image used to create CSFGM mask (>0 && <1000)
-forcefcm:	Forces generartion of the `fc4_mc2_ex2` image if it already exists (if rerunning the script a second time)

1. **-vdilatecross <int> / -vdilateball <int>**: Specifies the 2D dilation operation to be performed on VCSF. The `-vdilatecross` option is a more conservative dilation option than the `-vdilateball` option. The dilation radius should be chosen to maximize both the number of VCSF partial volume voxels removed and the number of lesion voxels retained.

2. **-minobjectsize <int>**: Removes small objects that typically correspond to non-lesion voxels. To retain all objects, set this option to 1.

3. **-vcsf <int>**: Specifies the value that has been assigned to VCSF on the T1 segmentation.

4. **-pdt <int> / -t2t <int>**: Specifies the user defined thresholds used to segment lesions on the PD and T2 images respectively. To select an appropriate value, run the `sb_pdt2seg` command with the default options and inspect the output. If lesions voxels are excluded on the `t2seg` image, lower the `-t2t` threshold. If non-lesion voxels are included, increase the `-t2t` threshold. If lesion voxels are excluded on the `pdseg` image, increase the `-pdt` threshold. Typically, non-lesion voxels will be included on the PD image and this is OK.



T2 lesion threshold = T2 histogram peak value + (T2 histogram peak value FWHM – T2 histogram peak value) * user defined T2 threshold
PD lesion threshold = PD mean of estimated normal WM + (adjusted PD max – adjusted PD min) * user defined PD threshold

5. **`-ct <int>:`** Specifies the threshold used for the creation of the CSFGM mask. An appropriate value is an integer between 0 and 1000 that thresholds the `_fc4_mc2_ex2` image so that primarily CSF voxels remain. These voxels will undergo a series of morphological operations to produce the CSFGM mask. Ideally, all voxels shown in purple on the CSFGM mask below will correspond to gray matter and sulcal CSF voxels on the T1. If too many white matter voxels are included in the CSFGM compartment, increase the `-ct` threshold. If not enough CSFGM voxels are included in the CSFGM compartment, lower the `-ct` threshold.

6. **`-forcefcm:`** To save time, if the `fc_mc2_ex2` image already exists (i.e. if the script has already been run) the `fc_mc2_ex2` image will not be recreated. The `-forcefcm` option can be used to override this behaviour. Typically, this option is not necessary because the `fc_mc2_ex2` image does not change unless the input T1 image has been modified. Therefore, any required changes to the CSFGM mask can usually be accomplished using the `-ct` option.