A tutorial for fMRI analysis using ANTsR

Introduction

Overview

fMRI issues

- Nuisance signal from CSF and WM [1]
- Bandpass filtering
- Motion correction [2, 3]
- Global signal [4]

ANTsR implementation

The main fMRI-specific functions are:

- `fMRINormalization` — This function leverages structural image processing based on ANTs cortical thickness to implement standard functional image processing recommendations. The function will crop out the first k time frames, do motion correction and produce a variety of nuisance regressors. It will also do spatial and temporal filtering as well as interpolation between time frames that exceed a given framewise displacement. Finally, we return maps to the common coordinate system. Output may be trimmed in the future but currently provides access at different stages: merging versus filtering and normalized, fused BOLD images in both subject and template space. Also accommodates multiple runs.
- `preprocessRestingBOLD` — Preprocess resting fMRI by performing compcor/motion correction, nuisance regression, band-pass filtering, and spatial smoothing.
- `antsMotionCalculation` — Corrects 4D time-series data for motion and returns useful information, e.g., nuisance variables such as framewise displacement and DVARS. Uses `antsMotionCorr` for performing motion correction.
- `timeseries2matrix/matrix2timeseries` — Converts from/to a 4-D `antsImage` object to/from a $n \times p$ matrix where n is the number of time points and p is the number of voxels in the specified mask.
- `antsSpatialICAfMRI` — Perform spatial ICA on group or individual fMRI data.
- `frequencyFilterfMRI` — performs band-pass filtering on BOLD time series data.
- `getfMRInuisanceVariables` — Older, Black box-type function which performs motion correction, runs CompCor and estimates global signal (similar to `preprocessRestingBOLD`). The output is a list with the time series data matrix (time by voxels), motion and other nuisance variables, global signal, the mask and the average time series image.
- `filterfMRIforNetworkAnalysis` — to be deprecated.
- `preprocessfMRI` — to be deprecated (redundant functionality overlap with `preprocessRestingBOLD`).

Helper functions include:

- `matrixToImages/imagesToMatrix` — Converts to/from a $n \times p$ matrix from/to a list of `antsImage` where n corresponds to the number of images and n is equal to the number of voxels inside the specified mask.
- `icaWhiten` — Performs just the ica whitening step. This is useful for testing other decomposition algorithms performance when they use the same prewhitening step as ICA.
- `makePointsImage` – Creates spherical points in the coordinate space of the target image based on the n-dimensional matrix of points and a specified radius. Used with data such as `powers_areal_mni_itk`.

Data:

- `powers_areal_mni_itk` — Defines the set of nodes describing the functional organization of the brain as detailed in [5] and made publicly available.[1]

fMRI papers which use ANTsR

- *The pediatric template of brain perfusion* [6]
- *Subject-specific functional parcellation via prior based eigenanatomy* [7]
- *Unexpected role of interferon-γ in regulating neuronal connectivity and social behaviour* [8]

[1] http://www.nil.wustl.edu/labs/petersen/Resources.html

Tutorial

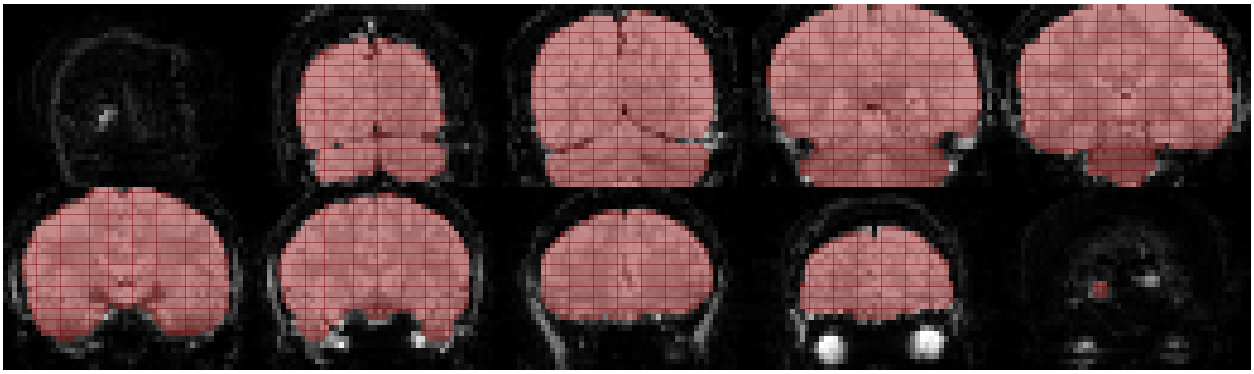
Initialization

```
# We include all the necessary R package dependencies.  We assume that the user
# is running this script (stitchTutorialDocument.R) in the repo directory.  The
# following R packages are required:
#   * ANTsR: image I/O, fMRI processing
#   * ggplot2: plot generation
#   * igraph:  network connectivity measures
#   * psych:  network connectivity measures
#   * corrplot:  correlation plot generation

invisible( suppressMessages( library( ANTsR ) ) )
library( pander )
library( ggplot2 )
library( igraph )
library( psych )
library( corrplot )

rootDirectory <- "./"
knitr::opts_knit$set( root.dir = rootDirectory )
knitr::opts_chunk$set( comment = "" )

figuresDirectory <- paste0( rootDirectory, "Figures/" )
if( ! dir.exists( figuresDirectory ) )
  {
  dir.create( paste0( rootDirectory, "Figures/" ) )
  }
dataDirectory <- paste0( rootDirectory, "Data/" );
```

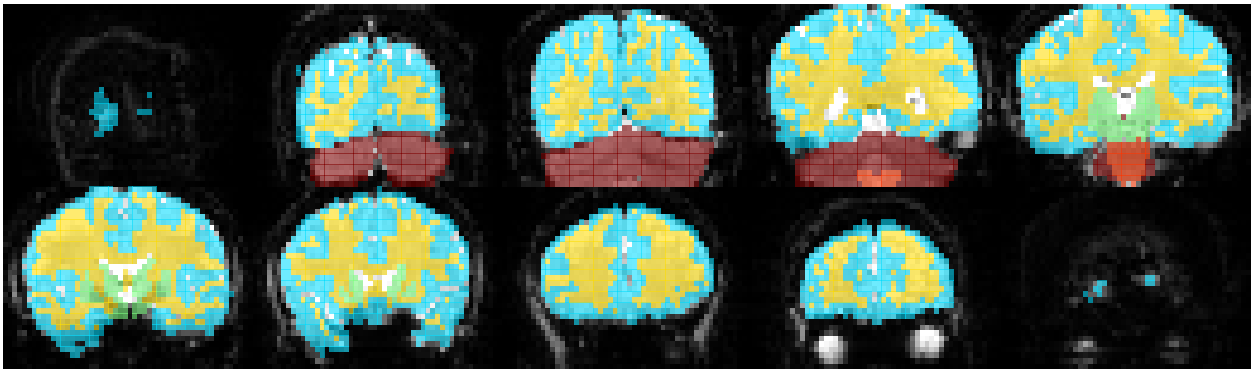
Read in input data

```
# Load the AAL (Automated Anatomical Labeling) data table and the AAL label image.
# These data define the AAL atlas of the human brain often used in fMRI for obtaining
# the anatomical regions of interest:
#    N. Tzourio-Mazoyer; B. Landeau; D. Papathanassiou; F. Crivello; O. Etard; N.
#    Delcroix; Bernard Mazoyer & M. Joliot (January 2002). "Automated Anatomical
#    Labeling of activations in SPM using a Macroscopic Anatomical Parcellation
#    of the MNI MRI single-subject brain". NeuroImage. 15 (1): 273-289.
#
# Also load the individual subject resting state BOLD images:  4-D bold, 3-D bold
# mask image, and 3-D segmentation (csf, gm, wm, etc.) image.

data( aal, package = 'ANTsR' )
aalLabelTable <- aal
aalFileName <- paste0( dataDirectory, "aal.nii.gz" )
aalImage <- antsImageRead( filename = aalFileName, dimension = 3 )

restingStateBoldFile <- paste0( dataDirectory, "rsbold.nii.gz" )
```

```
restingStateBoldImage <- antsImageRead( restingStateBoldFile, dimension = 4 )

restingStateBoldMaskFile <- paste0( dataDirectory, "rsboldmask.nii.gz" )
restingStateBoldMaskImage <- antsImageRead( restingStateBoldMaskFile, dimension = 3 )

restingStateBoldSegFile <- paste0( dataDirectory, "rsboldseg.nii.gz" )
restingStateBoldSegImage <- antsImageRead( restingStateBoldSegFile, dimension = 3 )

# Let's look at the images to make sure things make sense, e.g. masks are aligned.
#  Average of 4-D bold with mask superimposed

restingStateBoldAverage <- getAverageOfTimeSeries( restingStateBoldImage )
invisible( plot.antsImage( restingStateBoldAverage, restingStateBoldMaskImage,
   alpha = 0.75, ncolumns = 5 ) )
```

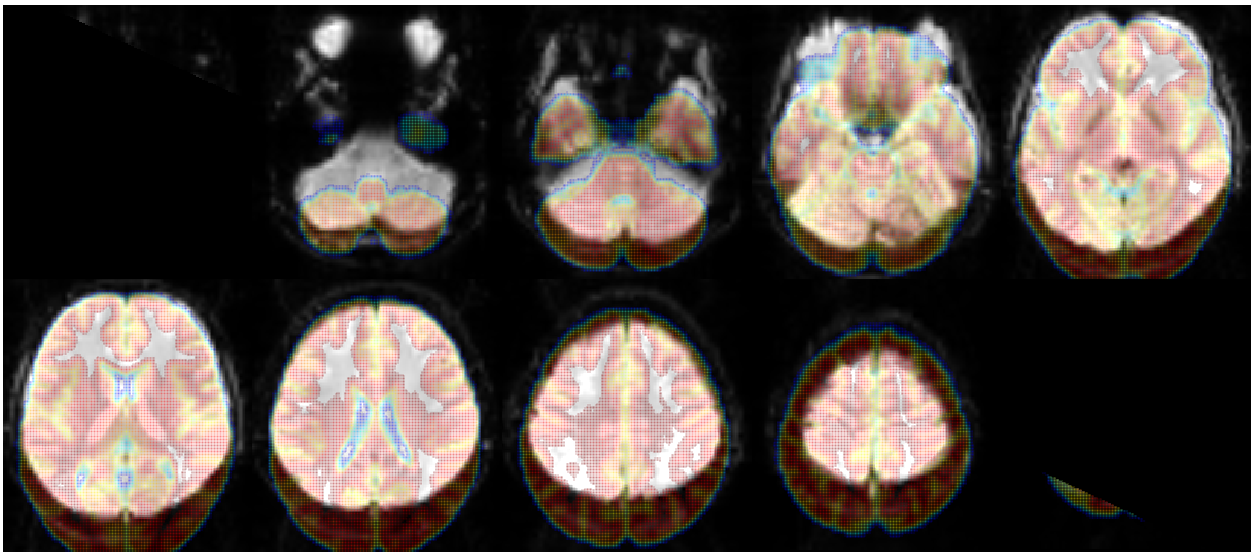


```
# Average of 4-D bold with segmentation mask superimposed
invisible( plot.antsImage( restingStateBoldAverage, restingStateBoldSegImage,
   alpha = 0.9, ncolumns = 5 ) )
```



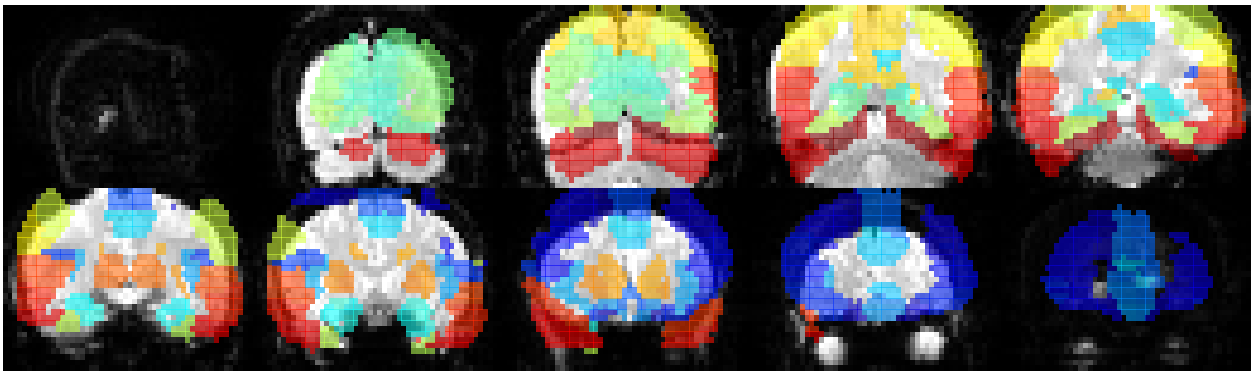
```
# Another possible use of ``plot.antsImage`` is to map the input image to another
# image of known/standard orientation, e.g., MNI space.  To do this, we read in the
# MNI template (see http://www.lead-dbs.org/?p=1241) and do a quick registration.

mniFile <- paste0( dataDirectory, "/mni.nii.gz" )
mniImage <- antsImageRead( mniFile, dimension = 3 )

mniRegistration <- antsRegistration( fixed = mniImage,
                      moving = restingStateBoldAverage, typeofTransform = "QuickRigid",
```

```
                        outprefix = paste0( dataDirectory, "mnixrs" ) )

invisible( plot( restingStateBoldAverage, mniImage, ncolumns = 5, axis = 3, alpha = 0.25,
   domainImageMap = c( mniImage, mniRegistration$fwdtransforms ) ) )
```

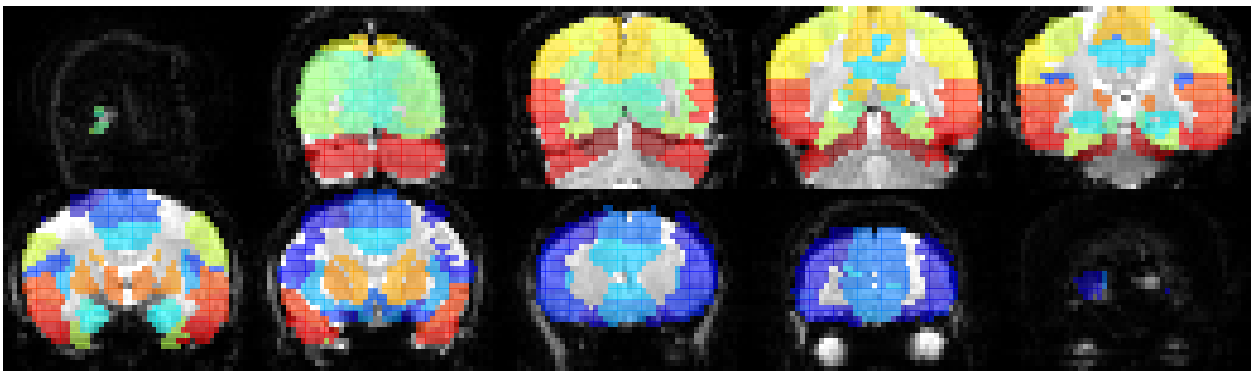


Spatially normalize AAL image

```
# The AAL image (which will be used later in the tutorial) is not in the space of the
# BOLD image so we do a quick registration of the AAL labels to the bold mask.  We
# first do an "AffineFast" transform to see if that has sufficient degrees of freedom.

aalRegistration <- antsRegistration( fixed = restingStateBoldMaskImage,
                        moving = aalImage, typeofTransform = "AffineFast",
                        outprefix = paste0( dataDirectory, "rsboldxaal" ) )

aalWarpedImage <- antsApplyTransforms( fixed = restingStateBoldMaskImage,
                        moving = aalImage, interpolator = 'genericLabel',
                        transformlist = aalRegistration$fwdtransforms )

invisible( plot.antsImage( restingStateBoldAverage, aalWarpedImage,
   alpha = 0.9, ncolumns = 5 ) )
```



```
# Clearly the alignment is not as good as we would like so we redo the registration
# using an "ElasticSyN" transform which looks much better.

aalRegistration <- antsRegistration( fixed = restingStateBoldMaskImage,
                      moving = aalImage, typeofTransform = "ElasticSyN",
                      outprefix = paste0( dataDirectory, "rsboldxaal" ) )

aalWarpedImage <- antsApplyTransforms( fixed = restingStateBoldMaskImage,
                      moving = aalImage, interpolator = 'genericLabel',
                      transformlist = aalRegistration$fwdtransforms )

invisible( plot.antsImage( restingStateBoldAverage, aalWarpedImage,
   alpha = 0.9, ncolumns = 5 ) )
```



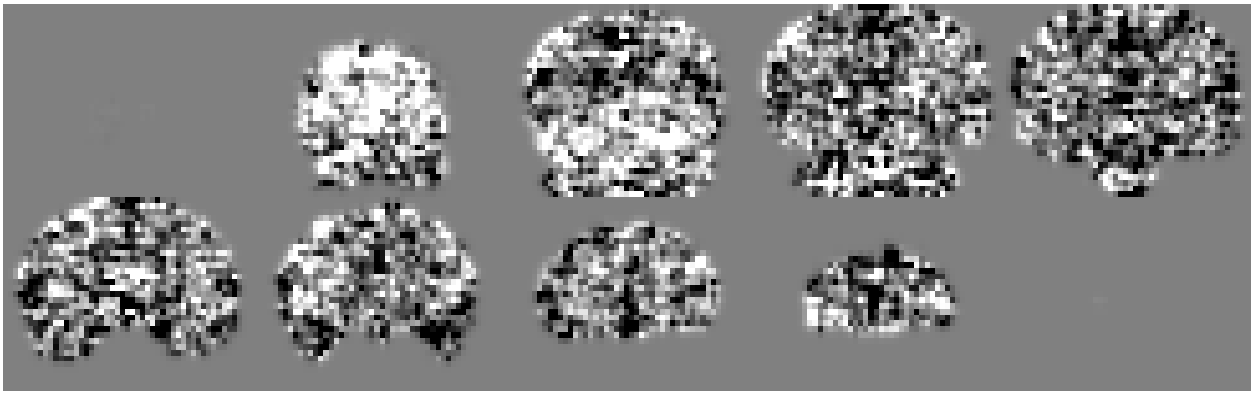
Preprocessing the resting state fMRI data

```
# The evolution of fMRI functionality in ANTsR is still ongoing. We process our
# current subject with ``preprocessRestingBOLD`` and plot the average of the
# resulting processed fMRI.  Note that we're doing motion correction on the lowest
# accuracy level for tutorial purposes.  For actual data, one would probably
# want to increase the accuracy level.

preprocessedRestingState <-
  preprocessRestingBOLD( restingStateBoldImage,
                         maskImage = restingStateBoldMaskImage,
                         denseFramewise = FALSE, numberOfCompCorComponents = 6,
                         doMotionCorrection = TRUE, motionCorrectionAccuracyLevel = 0,
                         motionCorrectionIterations = 1, frequencyLowThreshold = 0.01,
                         frequencyHighThreshold = 0.1,
                         spatialSmoothingType = "gaussian",
                         spatialSmoothingParameters = 2 )

pander( summary( preprocessedRestingState ), style = "rmarkdown",
  caption = "Returned values from the function preprocessRestingBOLD." )
```

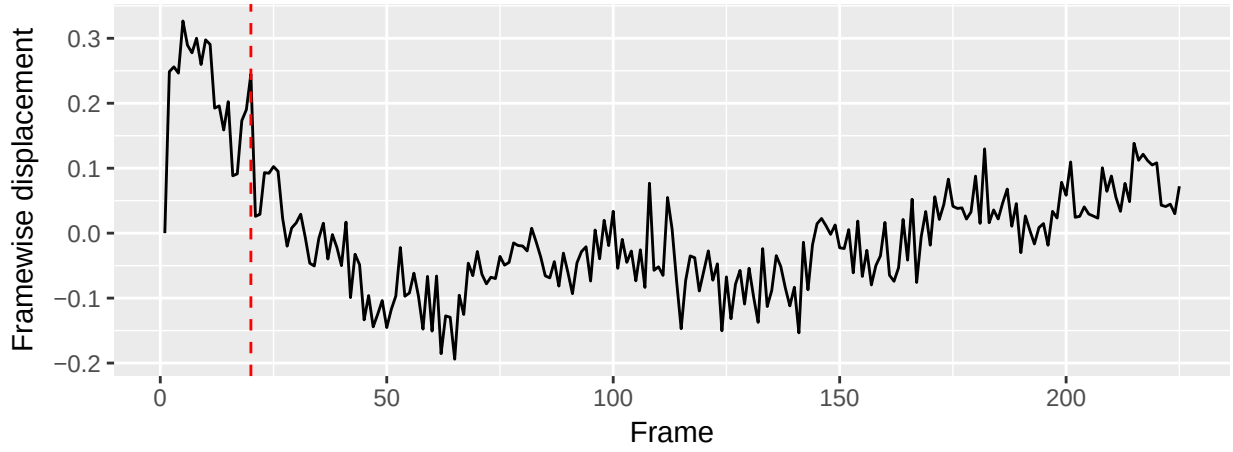
Table 1: Returned values from the function preprocessRestingBOLD.

	Length	Class	Mode
cleanBoldImage	1	antsImage	S4
maskImage	1	antsImage	S4
DVARS	225	-none-	numeric
DVARSpostCleaning	225	-none-	numeric
FD	225	-none-	numeric
globalSignal	225	-none-	numeric
nuisanceVariables	1350	-none-	numeric

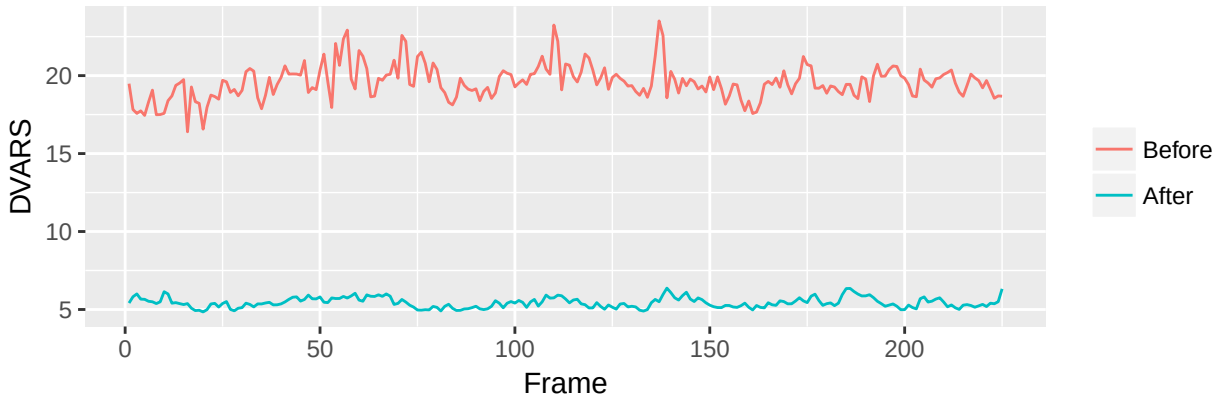
```
invisible( plot.antsImage(
   getAverageOfTimeSeries( preprocessedRestingState$cleanBoldImage ), ncolumns = 5 ) )
```



```
# We continue to check the preprocessing by plotting:
#   1. the framewise displacement (FD)
#   2. the global signal before and after regression (globalSignal)
#   3. comparing the DVARS of the original data (DVARS) and the processed
#        data (DVARSpostCleaning)
#   4. Plot the CompCor nuisance variables

numberOfTimeFrames <- dim( restingStateBoldImage )[4]

# Plot the framewise displacement.  Note the extreme displacements during the
# initial acquisition.  This is common and preprocessing often involves discarding
# the first N frames.

fdDataFrame <- data.frame( Frame = 1:numberOfTimeFrames,
   FD = preprocessedRestingState$FD - mean( preprocessedRestingState$FD ) )

ggplot( fdDataFrame ) +
   geom_line( aes( x = Frame, y = FD ), size = 0.5 ) +
   geom_vline( xintercept = 20, linetype = "dashed", size = 0.5, color = "red" ) +
   xlab( "Frame" ) + ylab( "Framewise displacement" ) +
   theme( legend.title = element_blank() ) + theme( aspect.ratio=1/3 )
```

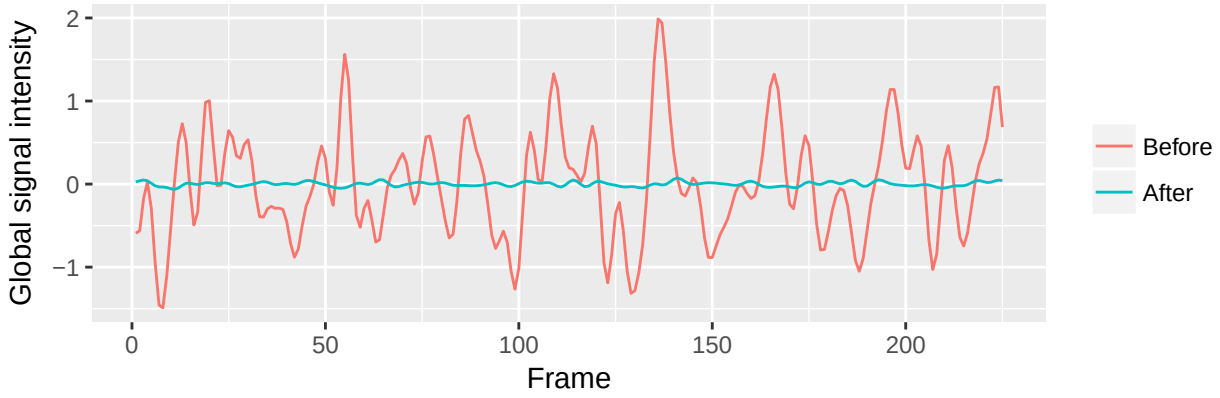



```
# Plot the DVARS.  Defined as the framewise backwards RMS voxelwise difference averaged
# over each time frame.

dvarsDataFrame <- data.frame( Frame = rep( 1:numberOfTimeFrames, 2 ),
       DVARS = c( preprocessedRestingState$DVARS,
                  preprocessedRestingState$DVARSpostCleaning ),
       Type = factor( c( rep( "Before", numberOfTimeFrames ),
              rep( "After", numberOfTimeFrames ) ), levels = c( "Before", "After" ) ) )

ggplot( dvarsDataFrame ) +
   geom_line( aes( x = Frame, y = DVARS, colour = Type ), size = 0.5 ) +
     xlab( "Frame" ) + ylab( "DVARS" ) +
     theme( legend.title = element_blank() ) + theme( aspect.ratio = 1/3 )
```

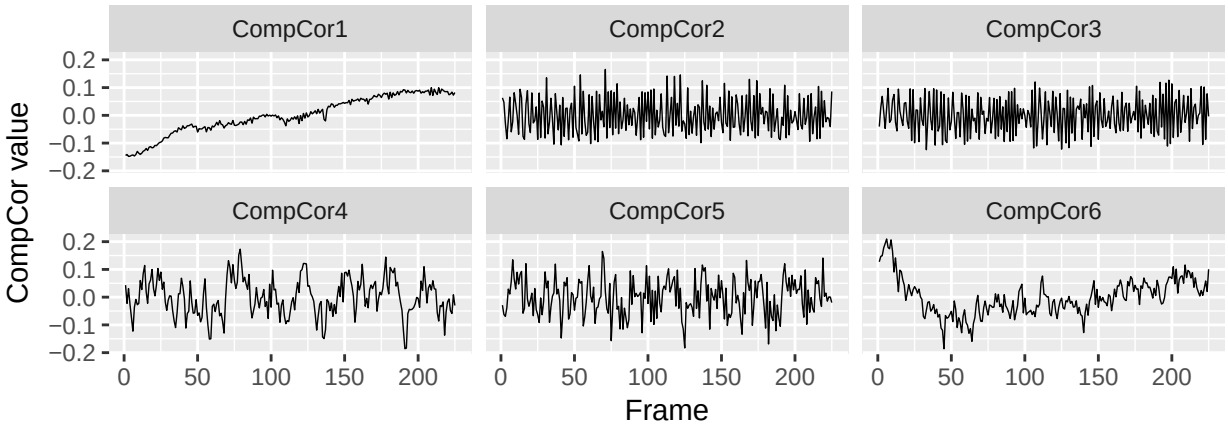



```
# Plot the global signal.  Do we regress out the global signal? Still an open issue.
# Let's just explore the approach to regressing it out afterwards.  A better way would
# be to include it as an ``initialNuisanceVariable`` in ``preprocessRestingBOLD()``.

boldMatrix <- timeseries2matrix(
  preprocessedRestingState$cleanBoldImage, restingStateBoldMaskImage )
boldMatrixGlobalSignalRegressedOut <-
  residuals( lm( boldMatrix ~ scale( preprocessedRestingState$globalSignal ) ) )

globalSignalDataFrame <- data.frame( Frame = rep( 1:numberOfTimeFrames, 2 ),
       GlobalSignal = c( preprocessedRestingState$globalSignal,
```

```
          apply( boldMatrixGlobalSignalRegressedOut, mean, MARGIN = 1 ) ),
       Type = factor( c( rep( "Before", numberOfTimeFrames ),
              rep( "After", numberOfTimeFrames ) ), levels = c( "Before", "After" ) ) )

ggplot( globalSignalDataFrame ) +
   geom_line( aes( x = Frame, y = GlobalSignal, colour = Type ), size = 0.5 ) +
     xlab( "Frame" ) + ylab( "Global signal intensity" ) +
     theme( legend.title = element_blank() ) + theme( aspect.ratio = 1/3 )
```

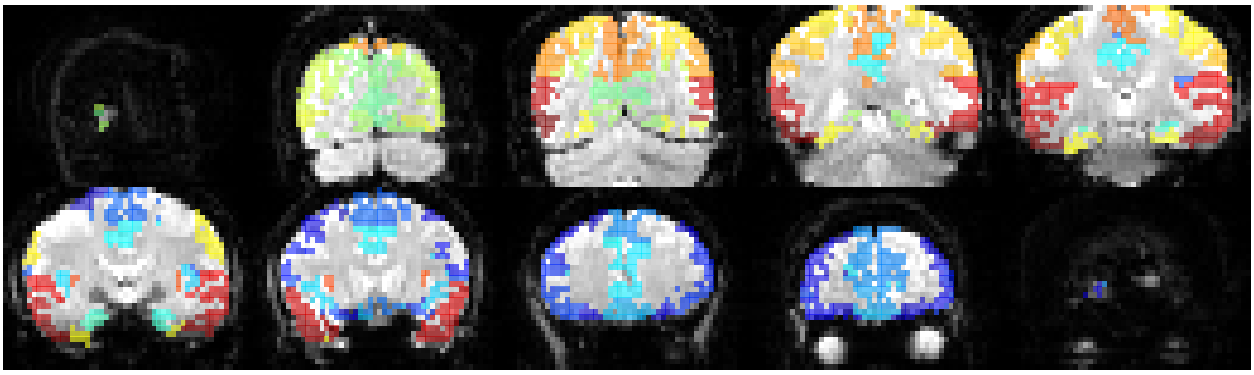



```
# Plot the CompCor nuisance signals.  Defined in terms of the PCA decomposition of
# the high frequency components of the BOLD signal.

numberOfCompCorComponents <- ncol( preprocessedRestingState$nuisanceVariables )
whichComponentLevels <- paste0( "CompCor", 1:numberOfCompCorComponents )
whichComponent <- factor( as.vector(
  matrix( rep( whichComponentLevels, numberOfTimeFrames ),
    nrow = numberOfTimeFrames, byrow = TRUE ) ), levels = whichComponentLevels )

compCorDataFrame <- data.frame(
  Frame = rep( 1:numberOfTimeFrames, numberOfCompCorComponents ),
  WhichComponent = whichComponent,
  Values = as.vector( preprocessedRestingState$nuisanceVariables ) )

ggplot( compCorDataFrame ) +
   geom_line( aes( x = Frame, y = Values ), size = 0.25 ) +
     facet_wrap( ~ WhichComponent, ncol = 3 ) +
     xlab( "Frame" ) + ylab( "CompCor value" ) +
     theme( legend.title = element_blank() ) + theme( aspect.ratio = 1/3 )
```



Calculate functional connectivity measures

```
# We use the AAL labels to calculate various functional connectivity measures for
# this subject.

# Mask out non-gray matter pixels from aalImage and plot image to ensure things look
# correct.

aalWarpedImage[restingStateBoldSegImage != 2] <- 0

invisible( plot.antsImage( restingStateBoldAverage, aalWarpedImage,
   alpha = 0.9, ncolumns = 5 ) )
```



```
# Determine the unique AAL labels and construct the correlation matrix

aalRoiLabelVector <- as.vector( as.array( aalWarpedImage[aalWarpedImage > 0] ) )
aalUniqueLabels <- sort( unique( aalRoiLabelVector ) )

boldMatrix <- timeseries2matrix(
  preprocessedRestingState$cleanBoldImage, restingStateBoldMaskImage )

boldLabelMatrix <- matrix( NA, nrow = nrow( boldMatrix ), ncol = length( aalUniqueLabels ) )
for( j in 1:length( aalUniqueLabels ) )
  {
  currentLabelIndices <- which( aalRoiLabelVector == aalUniqueLabels[j] )
```

```
  if( length( currentLabelIndices ) > 1 )
    {
    boldLabelMatrix[, j] <- rowMeans( boldMatrix[, currentLabelIndices] )
    }
  else
    {
    boldLabelMatrix[, j] <- mean( boldMatrix[, currentLabelIndices] )
    }
  }
correlationMatrix <- cor( boldLabelMatrix, boldLabelMatrix )
correlationMatrix[which( is.na( correlationMatrix ) )] <- 0
rownames( correlationMatrix ) <- colnames( correlationMatrix ) <-
  aalLabelTable$label_name[aalUniqueLabels]

# We calculate the significance for each entry.

cor.mtest <- function( mat, ... )
  {
  mat <- as.matrix( mat )
  n <- ncol( mat )
  p.mat <- matrix( NA, n, n )
  diag( p.mat ) <- 0

  for( i in 1:( n - 1 ) )
    {
    for( j in ( i + 1 ):n )
      {
      tmp <- cor.test( mat[, i], mat[, j], ... )
      p.mat[i, j] <- p.mat[j, i] <- tmp$p.value
      }
    }
  colnames( p.mat ) <- rownames( p.mat ) <- colnames( mat )
  p.mat
  }

p.mat <- cor.mtest( correlationMatrix )

uvaColors <- colorRampPalette( c( "#F59A2C", "#F1E5C7", "#E6E7E8", "#46A8C2", "#0D3268" ) )
correlationPlotFile <- paste0( figuresDirectory, '/CorrelationMatrix.pdf' );
pdf( height = 10, width = 10, file = correlationPlotFile )
corrplot( correlationMatrix, method = "circle", diag = FALSE, type = "upper",
  tl.col = "black", tl.cex = 0.48, tl.srt = 45, col = uvaColors( 200 ),
  p.mat = p.mat, sig.level = 0.01, insig = "blank" )
invisible( dev.off() )
```

```
# Instead of exploring the connectivity of the entire set of AAL labels, let's just
# look at specific networks (default mode and salience networks).  This information is in
# ``aalLabelTable``.

# Default mode network

aalDmnLabels <- aalLabelTable$label_num[which( aalLabelTable$isdmn > 0 )]
```

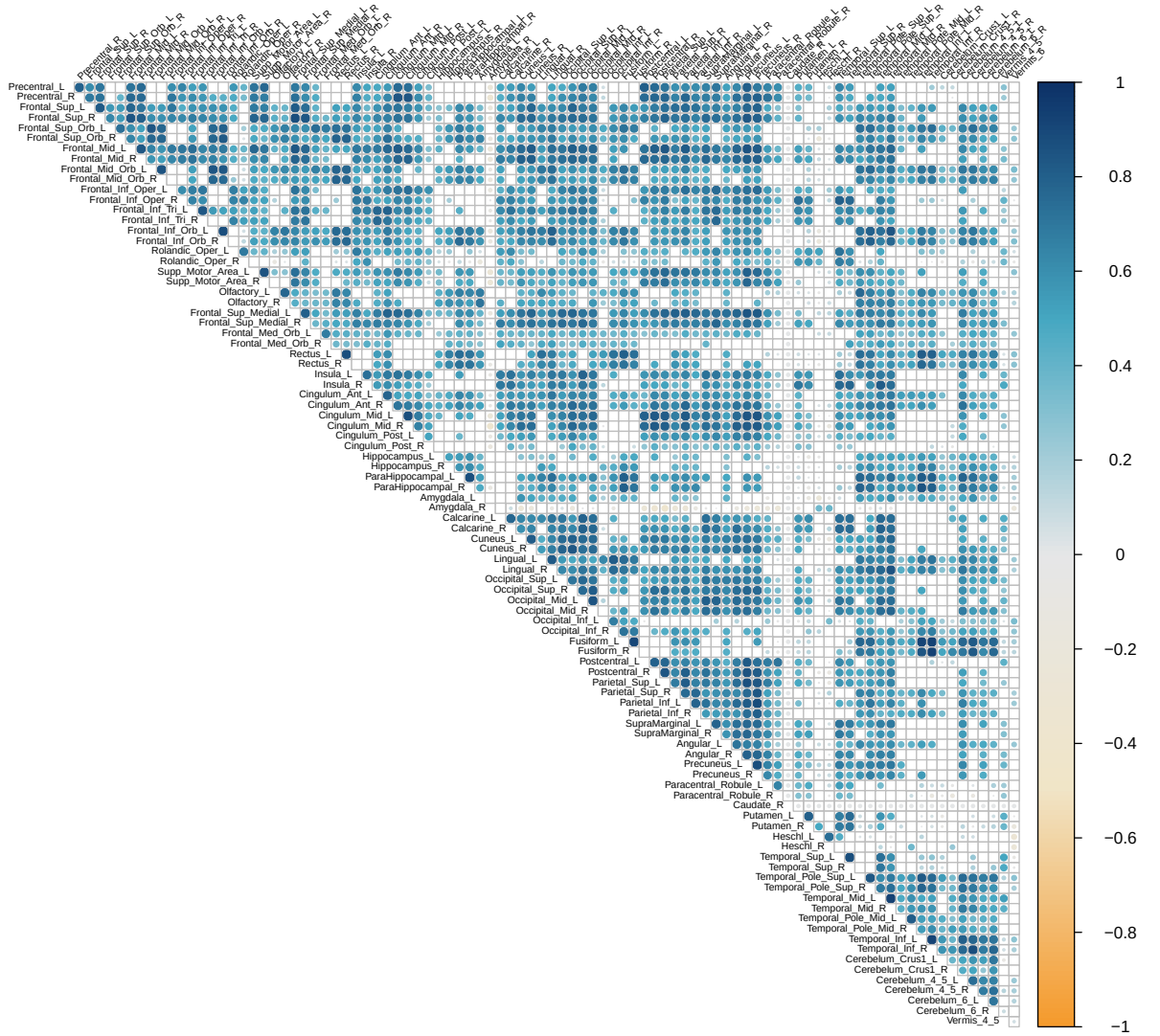


Figure 1: Correlation plot illustrating the functional connectivity relationships between AAL-defined regions.

```
dmnBoldLabelMatrix <- matrix( NA, nrow = nrow( boldMatrix ), ncol = length( aalDmnLabels ) )
for( j in 1:length( aalDmnLabels ) )
  {
  currentLabelIndices <- which( aalRoiLabelVector == aalDmnLabels[j] )
  if( length( currentLabelIndices ) > 1 )
    {
    dmnBoldLabelMatrix[, j] <- rowMeans( boldMatrix[, currentLabelIndices] )
    }
  else
    {
    dmnBoldLabelMatrix[, j] <- mean( boldMatrix[, currentLabelIndices] )
    }
  }
dmnCorrelationMatrix <- cor( dmnBoldLabelMatrix, dmnBoldLabelMatrix )
dmnCorrelationMatrix[which( is.na( dmnCorrelationMatrix ) )] <- 0
rownames( dmnCorrelationMatrix ) <- colnames( dmnCorrelationMatrix ) <-
  aalLabelTable$label_name[aalDmnLabels]

p.mat <- cor.mtest( dmnCorrelationMatrix )

dmnCorrelationPlotFile <- paste0( figuresDirectory, '/dmnCorrelationMatrix.pdf' )
pdf( height=5, width=5, file = dmnCorrelationPlotFile )
corrplot( dmnCorrelationMatrix, method = "circle", diag = FALSE, type = "upper",
  tl.col = "black", tl.cex = 0.75, tl.srt = 45, col = uvaColors( 200 ),
  p.mat = p.mat, sig.level = 0.01, insig = "blank" )
invisible( dev.off() )
```

```
# Now we calculate various graph-based measures from the connectivity relationships
# in the default mode network and plot the resulting graph.

networkGraph <- makeGraph( dmnCorrelationMatrix, graphdensity = 0.25, getEfficiency = T )

V( networkGraph$mygraph )$color <- "#F59A2C"
V( networkGraph$mygraph )$label.cex <- 1.0
E( networkGraph$mygraph )$width <- 1.5

dmnGraphPlotFile <- paste0( figuresDirectory, '/dmnGraph.pdf' )
pdf( height = 10, width = 10, file = dmnGraphPlotFile )
plot.igraph( networkGraph$mygraph, layout = layout.fruchterman.reingold )
invisible( dev.off() )
```

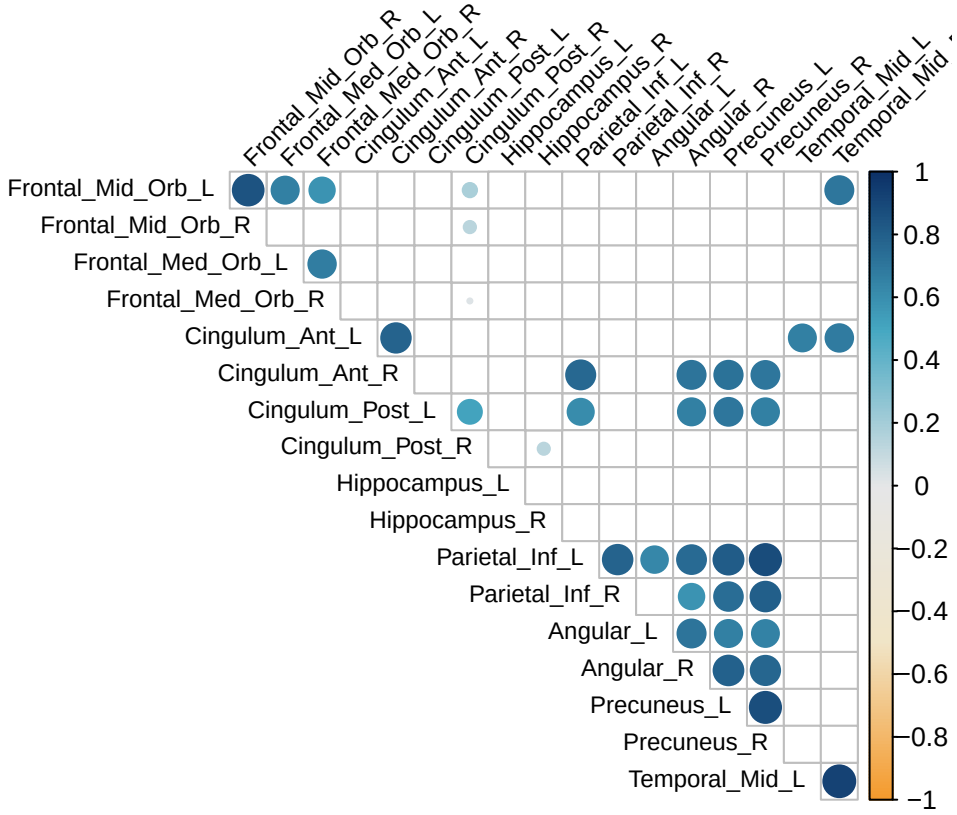


Figure 2: Correlation plot illustrating the functional connectivity relationships in the default mode network.

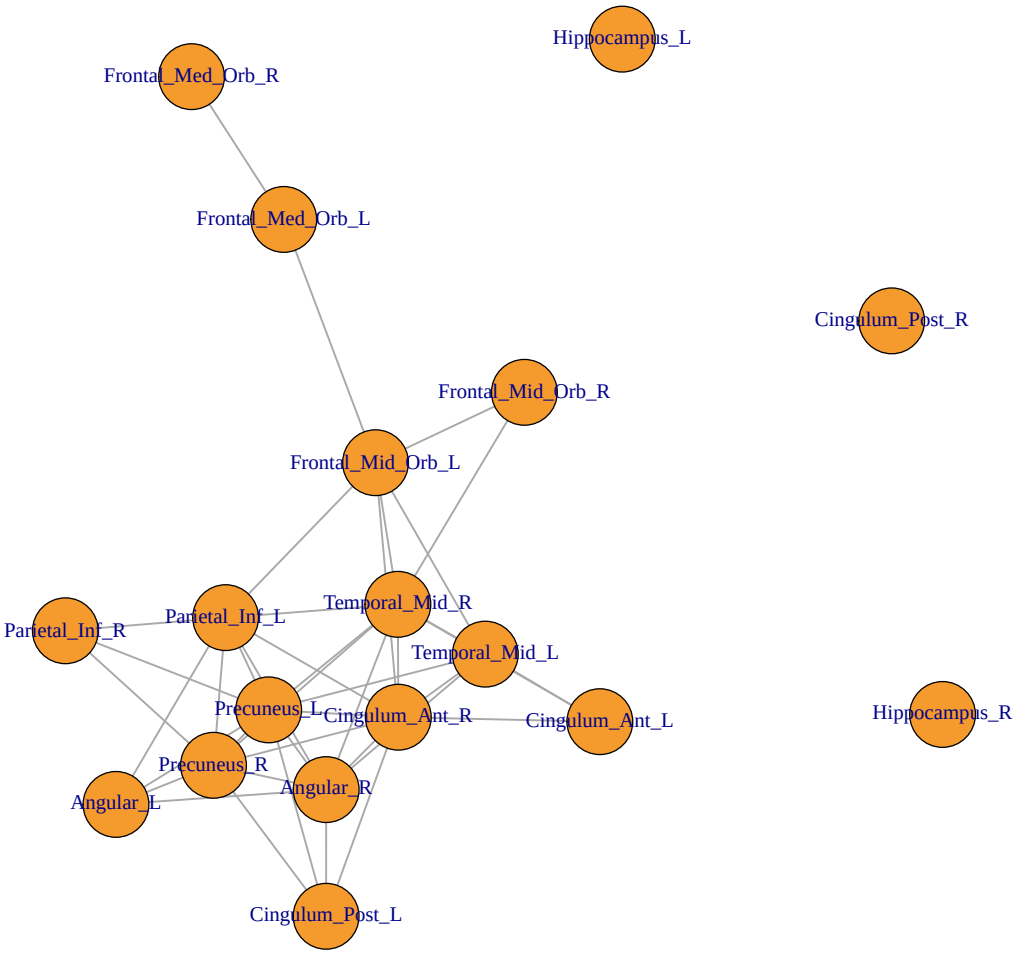


Figure 3: Graph of the functional connectivity of the default mode network.

References

1. Behzadi, Y., Restom, K., Liau, J., and Liu, T. T. "**A Component Based Noise Correction Method (CompCor) for BOLD and Perfusion Based FMRI**" *Neuroimage* 37, no. 1 (2007): 90–101. doi:10.1016/j.neuroimage.2007.04.042

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