Voxel-Based Morphometry
PART I
A. Introduction to structural brain analysis
B. What is VBM?

PART II
A. Pre-processing fundamentals
B. Step-by-step guide
C. Pitfalls and hints

PART III
A. Longitudinal analyses
B. Quantitative MRI & voxel base quantification
INTRODUCTION
ANATOMICAL ANALYSIS

Structural properties across homologous (i.e. like-for-like) regions

WHOLE BRAIN PROPERTIES

Manually defined region of interest (ROI) volumes

REGIONAL SHAPE PROPERTIES

Voxel-wise tissue “density”
Voxel based morphometry

TISSUE MICROSTRUCTURE

Average signal in manually defined ROIs

MRI PROPERTIES

Voxel-wise tissue maps:
Voxel based quantification

HISTOLOGY
What is VBM?

- **Voxel-Based Morphometry:**
  - **Size** and shape of the brain and its structures ("morphometry")
  - Compared at a voxel wise level across a population
Examples applications of VBM

• Many scientifically or clinically interesting questions might relate to changes in local volume of anatomical regions of the brain

• For example, whether (and where) patterns of brain morphometry help to:

  1. Distinguish between groups (e.g. Alzheimer's vs. healthy controls)
  2. Explain changes seen in development and aging
  3. Identify plasticity, e.g. when learning new skills
  4. Find structural correlates (i.e. regions where the size correlates with scores, traits, genotype etc.,)
1. Phenotypic patterns of disease

2. Ageing: GM atrophy

3. Plasticity: Computer game

4. Correlates: Political orientation

Atrophy - AD vs. healthy Controls (ADNI2 Dataset)

Callaghan et al., 2014

Kühn et al., 2014

Conservative - Decrease

Conservative - Increase

Kanai et al., 2011
Overview of SPM

- Realignment
- Smoothing
- Normalisation
- Design matrix
- Image time-series
- Kernel
- General linear model
- Template
- Parameter estimates
- Statistical parametric map (SPM)
- Gaussian field theory
- Statistical inference
- p < 0.05
TISSUE SEGMENTATION
Tissue segmentation for VBM

• High-resolution MRI reveals fine structural detail in the brain, but not all of it reliable or interesting
  • Noise, intensity-inhomogeneity, vessels
  • MR Intensity is usually not quantitatively meaningful
  • Quantitative MRI is possible though, and promising, see Voxel Based Quantification (VBQ) e.g. Draganski et al. (2011) PMID:21277375

• Regional volumes of the three main tissue types: gray matter, white matter and CSF, are well-defined and potentially have interesting properties
Tissue segmentation in SPM12

- Main Segmentation button on the SPM GUI
**Structural MRI** (e.g. T1 weighted MRI)

Output T1 with bias field inhomogeneities removed

Tissue segmentations (c1->c6)

Can output "native" or "dartel imported", make sure to select what you want.

Can use either, but if native make sure the data is all well aligned across subjects (see later example)
Tissue segmentations
Total intracranial volume (TIV)

- TIV is a useful measure, as the regional size of brain structures correlate with overall brain size.
- Used as a covariate in VBM analysis
- Once you have run SPM SEGMENT, each subject will have a “_seg8.mat” file
- Can use this in “Tissue Volumes” function
- Output – Volume (mm\(^3\)) of each tissue class
VOXEL BASED MORPHOMETRY
Voxel-Based Morphometry

• In essence VBM is Statistical Parametric Mapping of regional segmented tissue density or volume

• The exact interpretation of gray matter density or volume is complicated, and depends on the preprocessing steps used:
  • It is not interpretable as neuronal packing density or other cytoarchitectonic tissue properties
  • The hope is that changes in these microscopic properties may lead to macro- or mesoscopic VBM-detectable differences
  • One technique is to use VBM in combination with other quantitative structural measures (diffusion, MT, R2*, SWI) to make biophysical inferences (see later)
Overview of VBM

- Anatomical analyses in SPM (VBM, VBQ) use the same standard SPM pipeline, but with a few adaptations to allow us to derive and use morphometric properties
VBM
Step-by-step overview
STEP 1!

** ALWAYS VISUALLY CHECK YOUR DATA**

Examples: Poor scan quality, artefacts, abnormal tissue (ischaemia, dural thickening, lesions), abnormal brains (hydrocephalus), alignment (need to be relatively close), header issues (flipped brains), missing data
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1. Unified segmentation and spatial normalisation
   i. More flexible groupwise normalisation using Shoot (supersedes DARTEL)

2. Modulation to preserve tissue volume
   i. Otherwise, tissue “density” (harder to interpret, registration errors)
   ii. See also Radua et al. (2014) [PMID:23933042]

3. Compute tissue totals/globals

4. Gaussian smoothing

5. Voxel-wise statistical analysis
VBM in pictures

Segment

Normalise
VBM in pictures

Segment

Normalise

Modulate

Smooth
VBM in pictures

Segment
Normalise
Modulate
Smooth
Voxel-wise statistics

\[
\begin{bmatrix}
  a1_{xyz} \\
  a2_{xyz} \\
  \vdots \\
  aN_{xyz}
\end{bmatrix} = Y = X\beta_{xyz} + e_{xyz}
\]

\[e_{xyz} \sim N(0, \sigma^2_{xyz} V)\]

\[
X = \begin{bmatrix}
1 & 0 \\
1 & 0 \\
\vdots & \vdots \\
0 & 1 \\
0 & 1
\end{bmatrix}
\]
VBM in pictures

Segment

Normalise

Modulate

Smooth

Voxel-wise statistics

beta_0001

con_0001

ResMS

spmT_0001

FWE < 0.05
VBM SUBTLETIES

Modulation

How much to smooth

Interpreting results

Adjusting for total GM or Intracranial Volume

A priori regions of interest
Modulation ("preserve amounts")

- Multiplication of warped (normalised) tissue intensities so that their regional total is preserved
  - Can detect differences in completely registered areas
  - E.g. Focal atrophy

- Otherwise, we preserve concentrations, and are detecting mesoscopic effects that remain after approximate registration has removed the macroscopic effects
  - For example, gross folding differences in sulci but not focal atrophy

Native intensity = tissue probability

Unmodulated

Modulated

Native

Unmodulated

Modulated
JACOBIAN DETERMINANT IMAGE
(j_<image>.nii)
Modulation ("preserve amounts")

- Top shows “unmodulated” data (wc1), with intensity or concentration preserved
  - Intensities are constant

- Below is “modulated” data (mwc1) with amounts or totals preserved
  - The voxel at the cross-hairs brightens as more tissue is compressed at this point
Smoothing

• Analysis will be most sensitive to effects matching shape and size of the kernel

• The data will be more Gaussian and closer to a continuous random field for larger kernels
  • Usually recommend $\geq 6$mm

• Results will be rough and noise-like if too little smoothing is used

• Too much will lead to distributed, indistinct blobs (i.e. loss of spatial sensitivity)
  • Usually recommend $\leq 12$mm

• Small subcortical nuclei (e.g. STN/SN) represent a special case where $<<4$mm may be warranted (see de Hollander et al., 2015)
Smoothing

• The results below show two fairly extreme choices on the same data
  • 5mm on the left, and 16mm on the right
Smoothing is a *locally weighted ROI*

- VBM > ROI: no subjective (or arbitrary) boundaries
- VBM < ROI: harder to interpret blobs & characterise error
Interpreting findings

Thickening
Thinning
Mis-register
Mis-classify
Contrast
Thickening
Folding
Mis-register
Adjustment for “nuisance” variables

• Anything which might explain some variability in regional volumes of interest should be considered:

  • Total intracranial volume (TIV/ICV)

  • Age and biological sex are commonly used (i.e. brain size biases that may confound)
    • Consider age & age² to allow quadratic effects

  • Site or scanner if more than one
    (Note: model as factor, not covariate; multiple binary columns)
HINT: Define *a priori* regions of interest before analysis

- Before starting any analyses, you should try to identify brain regions where you expect to see structural changes:
  - Formal review of the literature (e.g., fMRI, VBM, functional networks, histopathology etc.,)
  - MRI-based meta-analyses tools

- Clearly defining *a priori* regions in advance allows you to accept $P < 0.001$ uncorrected in those regions, if peak voxel FWE $< 0.05$ fails

- You can also conduct small volume analysis in these regions to try and boost statistical sensitivity.

- Consider pre-registering ROIs in advance to clearly demarcate hypothesis testing from exploratory analyses
NORMALISATION
(warping)
NORMALISATION (warping)

• To analyse our data, we need to accurately align identical structures voxel wise

• To do this, we calculate the transformation to a “Template space”

• Calculating these transformations (warps) also encodes regional information about the amount of compression or expansion required compared to the average – Which are used for VBM
Spatial normalisation with Shoot

• VBM is crucially dependent on registration performance
  • The limited flexibility of DCT normalisation has been criticised
  • Inverse transformations are useful, but not always well-defined
  • MNI/ICBM templates/priors are not universally representative

• The **Shoot** (& DARTEL) toolbox combines several methodological advances to address these limitations
  • Voxel-wise DF, integrated flows, group-wise registration of GM & WM tissue segments to their (iteratively evolving) average
Two diffeomorphic approaches in SPM

“Dartel Tools” (SPM 8-12)

• Uses the same small deformation composed multiple times.
• Faster than Geodesic Shooting.
• Gives similar deformations to Geodesic Shooting.
• Currently more additional utilities.

Geodesic Shooting (SPM 12 onwards)

• Uses the optimal series of small deformations, which are composed together.
• More mathematically correct than Dartel.
• Gives nicer maps of volume change than Dartel.
• Growing number of utilities
• Recommended to use this instead of Dartel
Group-wise alignment in shoot

- Template implicitly generated from data in study.

- Findings less biased by choice of template.

START: Initial Average

Template 1 (few iterations)

Template 2 (more iterations)

Template 3 (.. even more iterations)

Template 4 (final)
Jacobian determinant
$j[filename].nii$
voxel wise volumetric information

Velocity field
$v[filename].nii$
4D file

Deformation field
$y[filename].nii$
4D file
Shoot Group Average Template
N = 5632
There are lots of examples of "Template" or "Group average" spaces e.g. MNI. However, each space represents the population from which it was created, which may bias your results (e.g. MNI tends to be larger than most). Creating specific "Group average space" for your study population will avoid this. You can always transform your coordinates (i.e. peak results) to MNI for reporting if needed (e.g. for meta-analysis).
Summary

• VBM performs voxel-wise statistical analysis on smoothed (modulated) normalised tissue segments
• SPM performs segmentation and spatial normalisation in a unified generative model
• The SHOOT (or DARTEL) toolboxes improves spatial normalisation for VBM (and probably also fMRI...)
• More specific details in the demo and practical sessions
LONGITUDINAL ANALYSIS
Longitudinal VBM – motivation

• Development, growth, plasticity, aging, degeneration, and treatment-response are inherently longitudinal

• Serial data have major advantages over multiple cross-sectional samples at different stages

• Increasing power
  • Subtlety of change over time vs. inter-individual variation

• Reducing confounds
  • Separating within-subject changes from cohort effects
  • Demonstrating causality with interventions
Longitudinal VBM – asymmetry & bias

• Within-subject image processing often treats one time-point differently from the others
  • Later scans registered (rigidly or non-rigidly) to baseline

• Asymmetry can introduce methodological biases
  • E.g. only baseline has no registration interpolation error
  • Baseline seg. more accurate than propagated segs.
  • Change in later intervals more regularised/constrained
Longitudinal VBM – registration in SPM12

No Longitudinal button, but found in Batch menu, like Dartel, etc.

Choice of paired or general serial. No difference in model, but easier specification and results for pairs.
Longitudinal Analysis Model

• Each individual is warped to their average
• Each average template will be warped to a total group average
• Each individual time-point will produce divergence & jacobian image
• These can be used to calculate single “rate” maps
• By repeating the segment-warp steps on the average images, VBQ-type analysis can be performed on the warped rate maps
• **Output/results**
  - Average image
  - Jacobians or divergences
  - Deformations

• **Next steps**
  - Segment avg
  - Run Shoot
  - Warp e.g. dv to standard space
  - SPM stats on dv (TBM)
  - Or combine with seg of avg (VBM)
Oasis Data

66 year old male, with MCI (MMSE=19, CDR=1).
Oasis Data

Data from first 82 subjects (OAS2 0001 to OAS2 0099).

Computed average expansion/contraction rates for each subject.

Warped all data to common anatomical space.

Generated averages.

Mean image intensity

Control subjects

Dementia subjects
RATE OF BRAIN TISSUE LOSS

NORMAL AGEING

PARKINSON’S DISEASE
AUGMENTING VBM:
Quantitative MRI & Voxel Based Quantification
What is quantitative MRI?

• Standard MRI acquisitions have signal intensities that depend on many factors (both biological and scanner related) – Provide pictures of the brain rather than accurate voxel wise measurements

• There are a number of dedicated MRI acquisitions that allow specific tissue properties to be quantified aka. quantitative MRI (qMRI)

• In addition to these, there are derived parameter maps that can also be treated as quantitative maps e.g. Cortical thickness, longitudinal rate maps/divergence maps
QUANTITATIVE MRI

Greater interpretability at the biophysical level

More sensitive measures to pathological processes

More precise measures across scanners and centres

Improved ability to map brain anatomy
Voxel based Quantification

This can be considered an extension of VBM, designed to allow quantitative MRI to be analysed in a voxel-wise manner using the SPM framework.

All of the VBM pre-processing steps are identical, the only difference comes in creating “weighted averages” when applying the spatial smoothing (to limit partial volume effects).

Can be implemented directly or using an SPM extension in the “The hMRI toolbox”.

Analysis of these quantitative weighted averages then uses standard SPM approach.
Combining the advances in anatomical mapping with the biophysical insights provided by qMRI measures allows us to go beyond VBM and understand the basis for structural brain changes.

**BRAINSTEM AGEING**

- **Focal regions of volume loss**
- **Focal regions of reduced myelin density**
  
  *(also associated with increased proton density)*

Location: \(\downarrow\)Volume + \(\downarrow\)Myelin + \(\uparrow\)Free water

= Cerebellothalamic tract degeneration

**TBM**

**MT**
CONCLUSION

Introduced VBM & Potential uses
Tissue Segmentation
Statistics
VBM Subtleties
Normalisation via SHOOT
Templates (e.g., MNI, population average etc.,)
Longitudinal Toolbox
Quantitative MRI and voxel based quantification

There is a lot more(!): In vivo histology, Cortical thickness, lesion analysis, structural covariance, combining with multivariate machine learning techniques.. etc.,

BASIC VBM HOW TO GUIDE: http://qmaplab.com/resources/how-to-guides