
PREPROCESSING

1. Setting the Working Directory

Utilities → CD → *Select directory* [the selected directory is always shown at the bottom of the window; you can unselect it by simply clicking on its name] → Done

Done

2. Choosing the VBM5 Toolbox

Toolboxes → vbm5 [VBM5.1 appears in 2nd window]

3. Normalizing, Bias-Correcting, Segmenting

VBM 5.1 → Estimate and write

- Data <-X → Specify Files → *Select raw data* [the selected files are always shown at the bottom of the window; you can unselect them by simply clicking on their names] → Done
- Estimation options [double-click on the “+” to adjust options]
 - Tissue probability maps → Specify Files → *Choose the files “grey.nii”, “white.nii”, “csf.nii”* in /Spm/spm5/tpm
 - Gaussians per class → Specify Text → *Type “2 2 2 4”*
 - Affine Regularisation → Specify Menu Item → *Select “ICBM space template – European”*
 - Warping Regularisation → Specify Text → *Type “1”*
 - Warp Frequency Cutoff → Specify Text → *Type “25”*
 - Bias regularization → Specify Menu Item → *Select “very light regularization (0.0001)”*
 - Bias FWHM → Specify Menu Item → *Select “70mm cutoff”*
 - Sampling distance → Specify Text → *Type “3”*
 - Masking image → Specify Files → *Select none*
 - Set origin → Specify Menu Item → *Select “use center of mass to set origin”*
- Writing options
 - Gray matter
 - Native space → Specify Menu Item → *Select “no”*
 - Unmodulated normalized → Specify Menu Item → *Select “yes”*
 - Modulated normalized → Specify Menu Item → *Select “non-linear only”*
 - White matter
 - Native space → Specify Menu Item → *Select “no”*
 - Unmodulated normalized → Specify Menu Item → *Select “yes”*
 - Modulated normalized → Specify Menu Item → *Select “non-linear only”*
 - Cerebro-Spinal Fluid (CSF)
 - Native space → Specify Menu Item → *Select “no”*
 - Unmodulated normalized → Specify Menu Item → *Select “yes”*
 - Modulated normalized → Specify Menu Item → *Select “non-linear only”*
 - Bias Corrected

- Native space → Specify Menu Item → *Select “no”*
- Normalized Space → Specify Menu Item → *Select “yes”*
- Additional scalp editing → Specify Menu Item → *Select “no”*
- Extended options
 - Use tissue priors (experimental) → Specify Menu Item → *Select “use priors (old SPM5 approach)”*
 - HMRF weighting → Specify Menu Item → *Select “iterative HMRF weighting”*
 - Clean up any partitions → Specify Menu Item → *Select “Light Clean”*
 - Voxel size for normalization → Specify Text → *Type “1 1 1”*
 - Bounding box → Specify Text → *Type “-78 -112 -70; 78 76 85;”*
 - Write affine only → Specify Menu Item → *Select “use warps and affine for writing data”*
 - Display and print results → Specify Menu Item → *Select “yes”*
- Save [optionally save your selections as a *.mat file; the next time you can simply load this *.mat file with “Load” and only modify the respective sections]
- Run [as per your specifications above, this will calculate the bias-corrected whole-brain image in normalized space (wm*), as well as the modulated (m0*) and unmodulated (w) tissue segments].

4. Checking Segmentation Outcomes

Toolboxes → vbm5

VBM 5.1 → Tools → Check sample homogeneity using standard deviation across...

- Data <-X → Specify Files → *Select the newly written data [e.g., select the “m0wc1*” files, which are the normalized (w) GM segments (c1), modulated for non-linear components (m0)]: Specify filter: ^m0wc]*
- Proportional scaling? → Specify Menu Item → *Select “no”*
- Output mean file → Specify Text → *Type “Mean.img”*
- Output standard deviation file → Specify Text → *Type “SD.img”*
- Run [this will provide a boxplot]
 - Show Files with Check Reg → *Select “yes” and the respective files if you just found partitions outside the common range (in the boxplot); if necessary, repeat preprocessing with modified parameters in order to improve segmentation outcomes (or replace / exclude subjects)*

5. Smoothing

Smooth

- Images to Smooth <-X → Specify Files → *Select M0wc1* files (i.e., the normalized and modulated GM segments)*
- FWHM → Specify Text → *Type “12 12 12”*
- Data Type → Specify Menu Item → *Select “SAME”*

BUILDING THE STATISTICAL MODEL: TWO-SAMPLE T-TEST

Specify 2nd-level

- Factorial design specification
 - o Design → Two-sample t-test
 - * Group 1 scans → Specific Files (...)
 - * Group 2 scans → Specify Files (...)
 - * Independence → Yes
 - * Variance → Equal
 - * Grand mean scaling → no
 - * ANCOVA → no
 - o Covariates¹
 - o Masking
 - * Threshold masking → Absolute
⇒ Absolute Threshold → 0.1
 - * Implicit Mask → Yes
 - * Explicit Mask → <None>
 - o Global calculation → Omit
 - o Global normalization
 - * Overall grand mean scaling → No
 - o Normalisation → None
 - o Directory → Specify Files (...)

→ Save

→ Run

¹You could specify a covariate (i.e., partializing out the variance of a particular factor when looking at group differences)

- * Covariates → New Covariate
- * Vector <-X → Type
“spm_load”(you can upload a *.txt file with the covariates in the same order like the filenames)
- * Name <-X → Specify text (e.g., “age”)
- * Interactions → None
- * Centering → No centering

BUILDING THE STATISTICAL MODEL: USING THE FULL FACTORIAL MODEL (FOR A 2x2 ANOVA)

Specify 2nd-level

- Factorial design specification
 - o Design → Full factorial
 - * Factors → New Factor; New Factor
- Factor
- ~ Name → Specify Text (e.g., group)
 - ~ Levels → 2
 - ~ Independence → Yes
 - ~ Variance → Equal
 - ~ Grand mean scaling → No
 - ~ ANCOVA → No
- Factor

- ~ Name → Specify Text (*e.g., sex*)
 - ~ Levels → 2
 - ~ Independence → Yes
 - ~ Variance → Equal
 - ~ Grand mean scaling → No
 - ~ ANCOVA → No
 - * Specify Cells → New Cell; New Cell; New Cell; New Cell
 - Cell
 - ~ Levels → Specify Text (*e.g., [1 1]*)
 - ~ Scans → Specify Files (*e.g., groupA-males*)
 - Cell
 - ~ Levels → Specify Text (*e.g., [1 2]*)
 - ~ Scans → Specify Files (*e.g., groupA-females*)
 - Cell
 - ~ Levels → Specify Text (*e.g., [2 1]*)
 - ~ Scans → Specify Files (*e.g., groupB-males*)
 - Cell
 - ~ Levels → Specify Text (*e.g., [2 2]*)
 - ~ Scans → Specify Files (*e.g., groupB-females*)
 - Covariates
 - Masking
 - * Threshold masking → Absolute
 - ⇒ Absolute Threshold → 0.1
 - * Implicit Mask → Yes
 - * Explicit Mask → <None>
 - Global calculation → Omit
 - Global normalization
 - * Overall grand mean scaling → No
 - Normalisation → None
 - Directory → Specify Files (...)
- Save
→ Run

BUILDING THE STATISTICAL MODEL: MULTIPLE REGRESSION (CORRELATION)

Specify 2nd-level

- Factorial design specification
 - Design → Multiple Regression
 - * Scans <-X → Specify Files
 - * Covariates → New Covariate
 - Covariate

- ~ Vector <-X → Type “spm_load”(you can upload a *.txt file with the covariates in the same order like the filenames)
 - ~ Name <-X → Specify text (e.g., “age”)
 - ~ Centering → No centering
 - Intercept → Omit Intercept
 - Covariates
 - Masking
 - * Threshold masking → Absolute
 - ⇒ Absolute Threshold → 0.1
 - * Implicit Mask → Yes
 - * Explicit Mask → <None>
 - Global calculation → Omit
 - Global normalization
 - * Overall grand mean scaling → No
 - Normalisation → None
 - Directory → Specify Files (...)
- Save
→ Run

ESTIMATING THE STATISTICAL MODEL

Estimate

- Model estimation
 - Select SPM.mat <- → Specify Files
 - Method → Classical
- Run

DEFINING CONTRASTS & LOOKING AT THE RESULTS

Results

- Select SPM.mat
 - Define new contrast... [t-contrasts]
- (a) Simple group difference
⇒ use *.mat from model “2-sample T-test” (with / or without covariate):
- * for GroupA > GroupB: **1 -1**
 - * for GroupA < GroupB: **-1 1**
- (b) Correlation from model “Multiple Regression”
- * for positive Correlation: **1**
 - * for negative Correlation: **-1**

(b) 2x2 Anova

⇒ Use *.mat from model “Full factorial” (some contrasts are already defined)

- * For GroupA_males>GroupB_males: **1 0 -1 0**
- * For GroupA_males<GroupB_males: **-1 0 1 0**
- * For GroupA_females>GroupB_females: **0 1 0 -1**
- * For GroupA_females<GroupB_females: **0 -1 0 1**

→ Done

- Mask with other contrasts: no
- Titel for comparison:
- P value adjustment to...: none
- Threshold: 0.001
- Extent threshold: 0
- Correct for non-isotropic: yes
 - Right mouse button → Go to global maxima
 - Small volume correction → Search volume: Sphere
 - Radius of VOI (mm): 30

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