

## Chapter 16

# Psychophysiological Interactions (PPI)

### 16.1 Theoretical background

Psychophysiologic interactions (PPI) model the response in one cortical area as the influence of another region and its interaction with an experimental treatment (sensory stimulus, task, etc.) [12]. This technique is one way of examining effective connectivity, or the influence that one neural system exerts on another [11]. Mechanistically, a PPI analysis involves the following steps.

1. Performing a standard GLM analysis.
2. Extracting BOLD signal from a source region identified in the GLM analysis.
3. Forming the interaction term (source signal x experimental treatment)
4. Performing a second GLM analysis that includes the interaction term, the source region's extracted signal and the experimental vector in the design. The inclusion of the source region's signal and the experimental vector is analogous to including the main effects in an ANOVA in order to make an inference on the interaction.

Forming the proper interaction term turns out to be a challenge because of the unique characteristics of fMRI (BOLD) data in which the underlying neural signal is convolved with a hemodynamic response function. However, interactions in the brain take place at the neural and not the hemodynamic level. Therefore, appropriate models of the interactions require the neural signal, which is not measured directly, but instead must be derived by deconvolving the HRF. The PPI software (`spm_peb_ppi.m`) was developed in order to provide robust deconvolution of the HRF and the proper derivation of the interaction term [20].

### 16.2 Practical example

The dataset in this exercise is from one subject who was studied in the [7] report and refers to the “attention to motion” dataset available from the SPM website<sup>1</sup>. It has already been described in the previous chapter for DCM.

The goal is to use PPI to examine the change effective connectivity between V2 and V5 while the subject observes visual motion (radially moving dots) under the experimental treatments of attending vs. not attending to the speed of the dots. The psychophysiologic interaction can be conceived of as looking for a significant difference in the regression slopes of V1 vs. V5 activity under the influence of the different attentional states [12].

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<sup>1</sup><http://www.fil.ion.ucl.ac.uk/spm/data/attention/>

### 16.2.1 GLM analysis - Design setup and estimation

This dataset has already been preprocessed (coregistered, normalised and smoothed) using an earlier version of SPM.

1. The analysis directory should include
  - (a) A directory named `functional`, which includes the preprocessed fMRI volumes.
  - (b) A directory named `structural`, which includes a T1 structural volume
  - (c) Files: `factors.mat`, `block_regressors.mat`, `multi_condition.mat` and `multi_block_regressors.mat`.
  - (d) You will also need to make 2 empty directories called GLM and PPI for performing the analyses.
2. In MATLAB type
 

```
>> cd 'path-to-analysis-directory'
```

```
>> spm_fmri
```
3. From the Tasks menu at the top of the Graphics window select BATCH
4. Select NEW "STATS" from the Options box

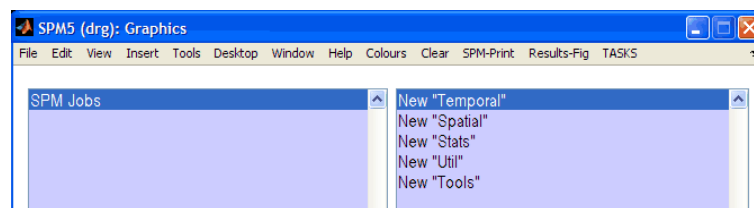


Figure 16.1: Select NEW "STATS" from the Options box

5. Click STATS in the Batch box and then select NEW "FMRI MODEL SPECIFICATION", NEW "MODEL ESTIMATION" and "NEW CONTRAST MANAGER" from the Options box

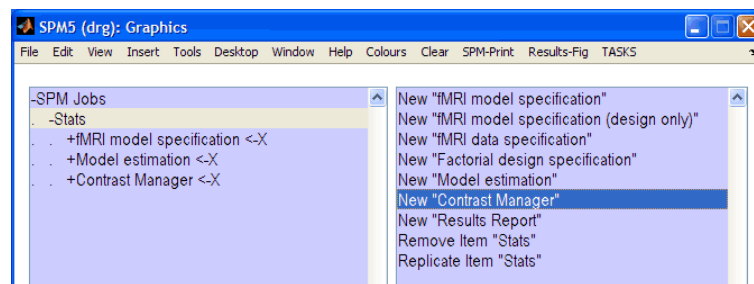


Figure 16.2: Click STATS in the Batch box and then select NEW "FMRI MODEL SPECIFICATION", NEW "MODEL ESTIMATION" and "NEW CONTRAST MANAGER" from the Options box

6. Right click FMRI MODEL SPECIFICATION > EXP/CON ALL > EXPAND ALL
7. Fill in the FMRI MODEL SPECIFICATION
8. Click DIRECTORY and choose the GLM directory that you made above.
9. UNITS FOR DESIGN [SCANS]
10. INTERSCAN INTERVAL [3.22]
11. MICROTME RESOLUTION [16]

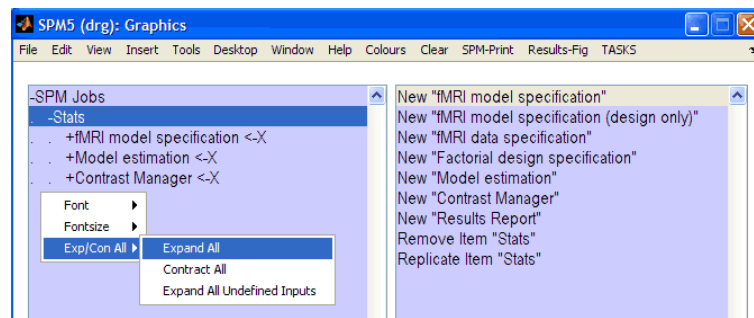


Figure 16.3: *Right click* FMRI MODEL SPECIFICATION > EXP/CON ALL > EXPAND ALL

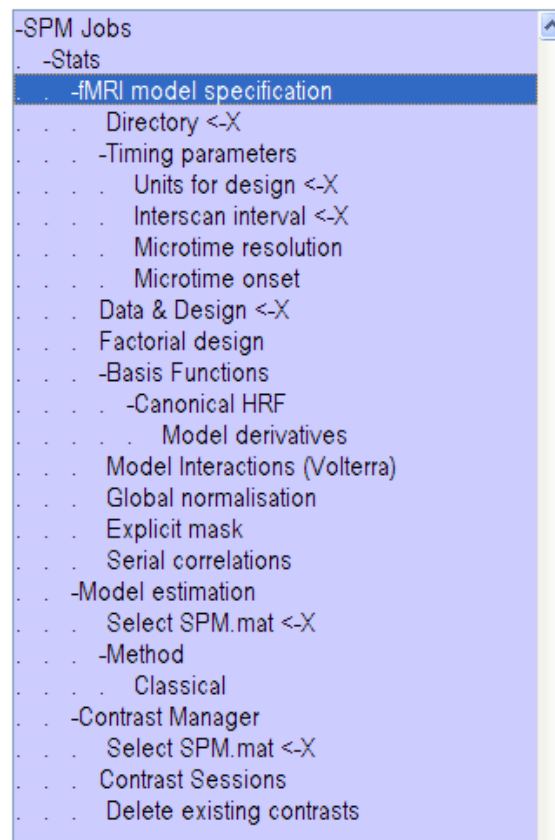


Figure 16.4: *Fill in the* FMRI MODEL SPECIFICATION

12. MICROTOME ONSET [1]
13. Click DATA & DESIGN, Choose NEW "SUBJECT/SESSION" from the options box then right click the Batch box and expand all.
14. Click SCANS and choose all the functional scans `snffM00587_00xx.img`. There should be 360 \*.img files.
15. The experimental conditions can be defined either individually or using a multiple condition mat-file. This exercise shows both methods for educational purposes, but you just need to follow one of the two approaches below.

### Define conditions individually

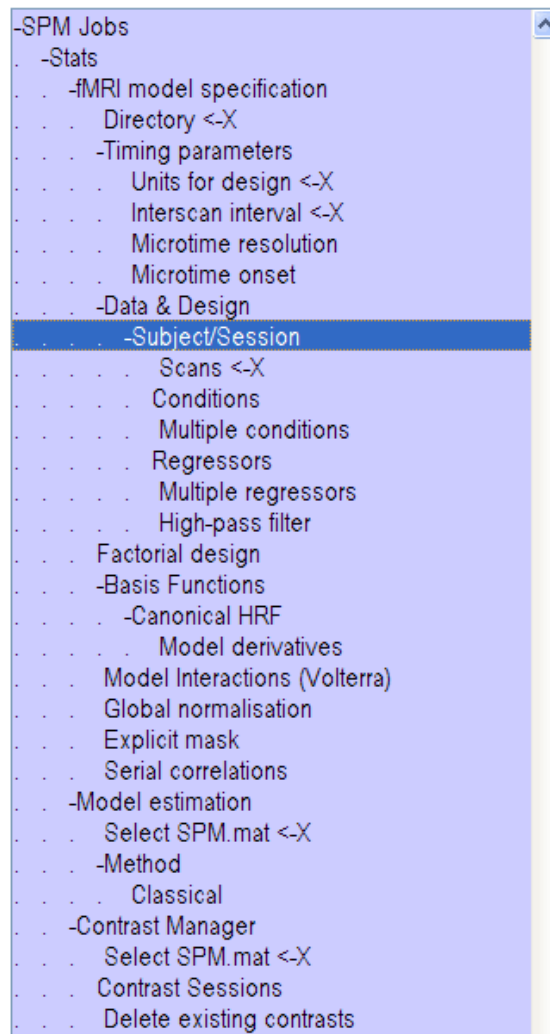


Figure 16.5: Click DATA & DESIGN and Choose NEW "SUBJECT/SESSION"

16. Load the mat file containing the individual conditions:

```
>> load factors.mat
```

You can look at the loaded variables by typing the variable names. (**att** = attention, **natt** = no attention, **fix** = fixation, **stat** = stationary)

```
>> att
>> natt
>> fix
>> stat
```

17. Click CONDITIONS under SUBJECT/SESSION and from the Options box add 4 "NEW CONDITIONS", then expand all.

18. Condition 1:

- NAME [**Fix**]
- ONSETS [**fix**] (this will enter the variable "fix")
- DURATIONS [10]

19. Condition 2: Name = `Stationary`, `ONSETS = stat`, `DURATIONS = 10`
20. Condition 3: Name = `No-attention`, `ONSETS = natt`, `DURATIONS = 10`
21. Condition 4: Name = `Attention`, `ONSETS = att`, `DURATIONS = 10`
22. Next you will enter 4 regressors to model block effects. This will account for the fact that the experiment took place over 4 runs that have been concatenated into a single session to make the PPI analysis easier. First load the regressors:

```
>> load block_regressor.mat
```

23. Click `REGRESSORS` and add 4 `NEW REGRESSORS`, then expand all.
24. Regressor 1: NAME = `Block 1`, VALUE = `block1`
25. Regressor 2: NAME = `Block 2`, VALUE = `block2`
26. Regressor 3: NAME = `Block 3`, VALUE = `block3`
27. Regressor 4: NAME = `Block 4`, VALUE = `block4`

#### Define conditions using multiple condition and multiple regressor files

28. To look at the organization of the variables in the multiple condition file, first load it. (Note: You only need to do this step if you want to look at the organization of the variables. You do not have to load the file in order to enter it into the design.)

```
>> load multi_condition.mat
```

If you want to look at the loaded variables type:

```
>> names
>> onsets
>> durations
```

29. To use the multiple conditions file in the design, click `MULTIPLE CONDITIONS`, then `SPECIFY FILES` in the Options box and choose the `multi_condition.mat` file.
30. Next you will enter 4 regressors to model block effects. This will account for the fact that the experiment took place over 4 runs that have been concatenated into a single session to make the PPI analysis easier. The regressors will be entered as a multiple regressor file. To look at the organization of the multiple regressor variable, first load it. (Note: Again you do not have to load the multiple regressor file in order to use it. This step is just to allow you to examine the variable.)

```
>> load multi_block_regressor.mat
>> R
```

Notice that there is single variable, `R`, which contains each regressor in a separate column.

31. To use the multiple regressor file, click `MULTIPLE REGRESSOR` then select the `multi_block_regressor.mat` file

#### Complete the design setup

32. `HIGH-PASS FILTER [192]` (Note: most designs will use a high-pass filter value of 128. However, this dataset requires a longer high-pass filter in order not to lose the low frequency components of the design.)

33. FACTORIAL DESIGN is not used
34. The BASIS FUNCTION is the CANONICAL HRF as shown and MODEL DERIVATIVES [NO DERIVATIVES]
35. MODEL INTERACTIONS (VOLTERRA): [DO NOT MODEL INTERACTIONS]
36. GLOBAL NORMALISATION [NONE]
37. EXPLICIT MASK [NONE]
38. SERIAL CORRELATIONS [AR(1)]
39. Under MODEL ESTIMATION click SELECT SPM.MAT then choose OUTPUT FROM “FMRI MODEL SPECIFICATION” from the Options box.
40. Under CONTRAST MANAGER click SELECT SPM.MAT then choose OUTPUT FROM “MODEL ESTIMATION” from the Options box.
41. Click CONTRAST SESSIONS then click NEW “F-CONTRAST” once, and NEW “T-CONTRAST” twice from the Options box, and Expand all.
42. Click F-CONTRAST → CONTRAST VECTORS and then choose NEW “F CONTRAST VECTOR” 4 times from the Options box
43. Under the F-contrast the name is: **Effects of Interest**
44. The F contrast vectors are as follows. Enter each one in turn into an F contrast vector.
 

```

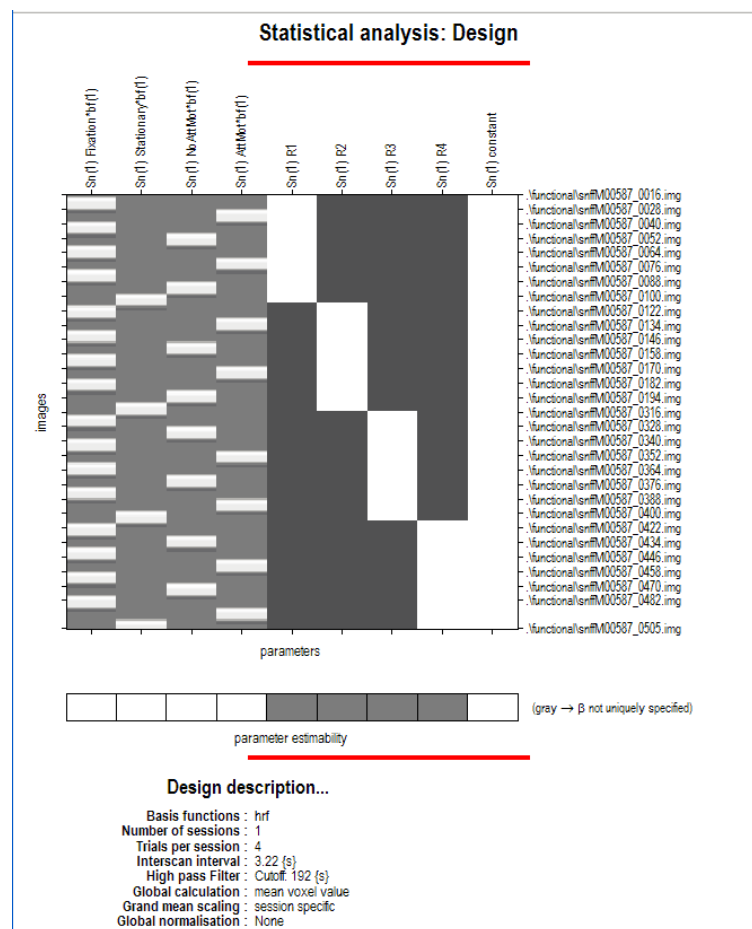
1 0 0 0 0 0 0 0 0
0 1 0 0 0 0 0 0 0
0 0 1 0 0 0 0 0 0
0 0 0 1 0 0 0 0 0
      
```
45. For the first T-contrast click NAME, then click SPECIFY TEXT in the Options box and enter: **Attention**.
46. Click T CONTRAST VECTOR, then SPECIFY TEXT in the Options box and enter: 0 0 -1 1 (Note the order of the conditions in the design matrix is: Fixation, Stationary, NoAttMot and AttMot).
47. For the second T-contrast the name is: **Motion**. The vector is: -2 0 1 1
48. Click the SAVE button and save the batch file.

### Design estimation

49. If everything has been entered correctly the RUN button is no longer grayed out. Click RUN to estimate the design.
50. The design matrix should look as follows.

## 16.2.2 GLM analysis - Results

1. Click RESULTS and select the SPM.mat file.
2. Choose the **Attention** contrast
3. Mask with other contrasts [No]
4. Title for comparison [Attention]
5. p value adjustment to control [None]

Figure 16.6: *Design matrix*

6. threshold T or p value [0.0001]
7. & extent threshold voxels [10]
8. You should see an SPM that looks like the one shown below Note the Superior Parietal and Dorso-Lateral Prefrontal activations, among others. By selecting OVERLAYS *rightarrow* SECTIONS, and selecting the normalised structural image you should be able to identify the anatomy more accurately.
9. To look at the Motion contrast where Attention is greater than No Attention, click RESULTS, choose the SPM.mat file and choose the Motion contrast.
10. Mask with other contrasts [Yes]
11. Select contrast for masking: Choose the Attention contrast
12. Uncorrected mask p-value [0.01]
13. Nature of Mask: [inclusive]
14. Title for comparison: leave as the defaults, which is [Motion (masked [incl.] by Attention at p=0.01)]
15. p value adjustment to control [FWE]
16. threshold T or p value [0.05]
17. & extent threshold voxels [3]
18. The masked motion contrast on the glass brain is shown below

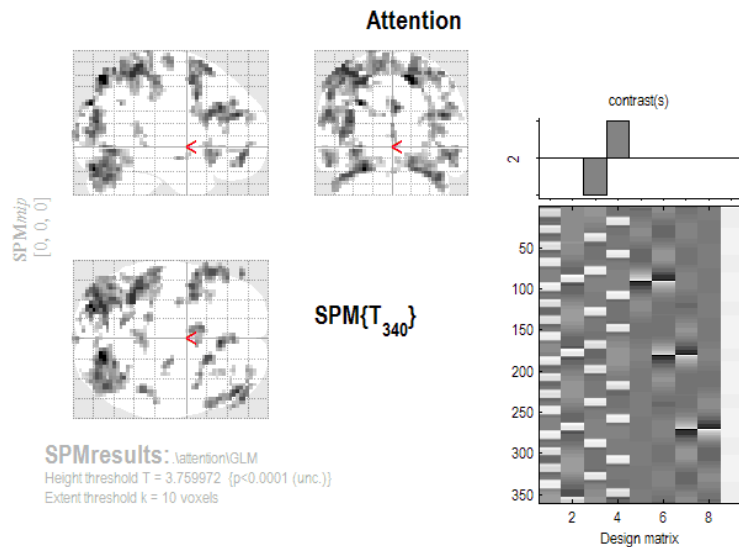


Figure 16.7: *Statistical parametric map for the Attention contrast*

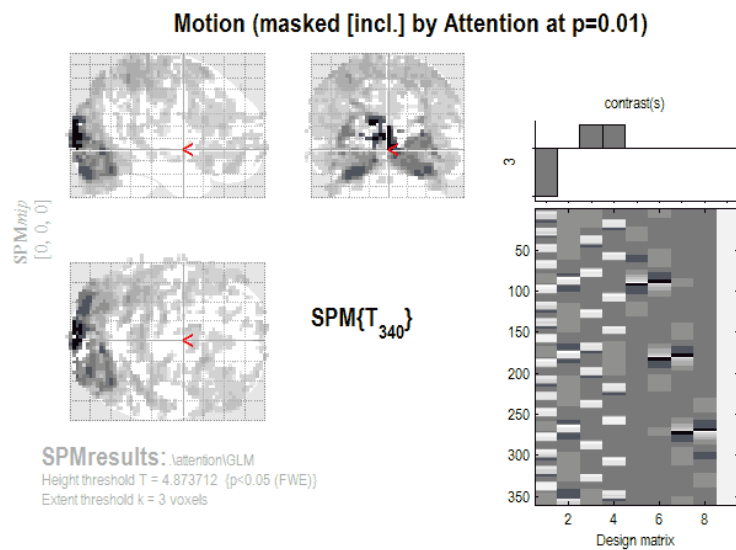
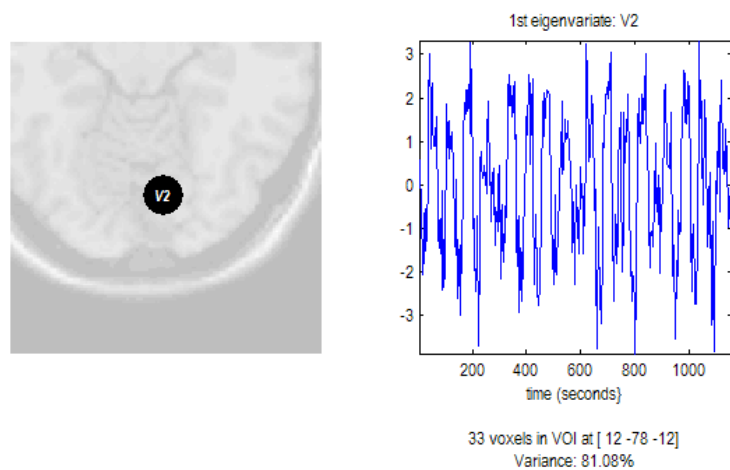
### 16.3 GLM analysis - Extracting VOIs

1. First select the **Motion** contrast, but do not include masking. Use a p-value adjustment of FWE with height threshold of 0.05 and a cluster threshold of 3.
2. Go to point [12 -78 -12]
3. Press **eigenvariate**
4. Name of region [V2]
5. Adjust data for [effects of interest]
6. VOI definition [sphere]
7. VOI radius(mm) [6]

This saves the extracted VOI data in the file `VOI_V2.1.mat` in the working directory, and displays the figure below. The left side shows the location on a standard brain. The right side shows the first eigenvariate of the extracted BOLD signal.

### 16.4 PPI analysis - Create PPI variable

1. Press the PPIs button, and select the previous `SPM.mat` file in the GLM directory.
2. Analysis type ? [psychophysiologic interaction]
3. Select `VOI_V2.1.mat`
4. Include Fixation [No]
5. Include Stationary [No]
6. Include No-Attention [Yes], contrast weight [-1]
7. Include Attention [Yes], contrast weight [1]
8. Name of PPI [ V2x(Att-NoAtt) ]

Figure 16.8: *Statistical parametric map for the Motion contrast*Figure 16.9: *First eigenvariate of the extracted BOLD signal in V2*

After a few seconds the PPI will be calculated and a graphical window will appear, see below. A file `PPI_V2x(Att-NoAtt).mat` will be created in the current working directory. It contains the variable `PPI.ppi` (the interaction term), `PPI.Y` (the original ROI eigenvariate) and `PPI.P` (the **Attention - No Attention** task vector). You will use these vectors in setting up your psychophysiological interaction GLM analysis. See `spm_peb_ppi` for a full description of the PPI data structure.

### 16.4.1 PPI GLM analysis - Design setup and estimation

1. Copy the file `PPI_V2x(Att-NoAtt).mat` to the PPI directory that you created at the start of this exercise.
2. Create a new directory and copy the file `PPI_V2x(Att-NoAtt).mat` to this directory.
3. `cd` (change directory) to the new directory.
4. At the MATLAB prompt type

```
>> load PPI_V2x(Att-NoAtt)
```

Psychophysiological Interaction: V2x(Att-NoAtt)  
 VOI File: V2  
 Factors: No-Attention [-1], Attention [1]

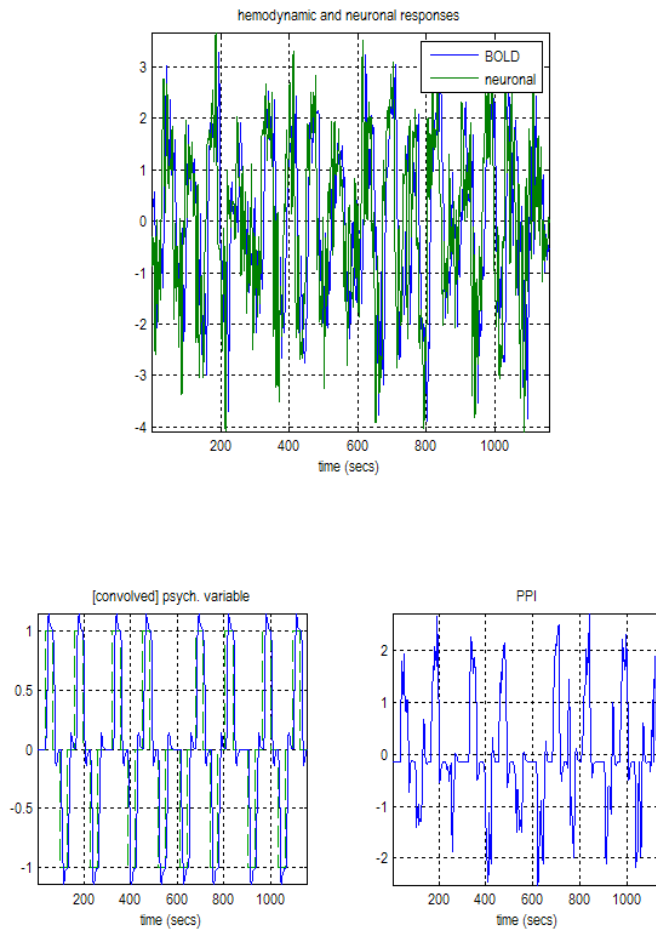


Figure 16.10: *PPI output graphics*

```
>> whos
```

5. From the Tasks menu at the top of the Graphics window select Batch
6. Select “NEW STATS” from the Options box, then add jobs NEW “fMRI MODEL SPECIFICATION”, NEW “MODEL ESTIMATION”, and NEW “CONTRAST MANAGER”, and Expand All.
7. Directory: Choose the PPI directory
8. Units for design [scans]
9. Interscan interval [3.22]
10. Add a NEW “SUBJECT/SESSION” under DATA & DESIGN
11. Click SCANS and choose all the functional scans `snffm00587_00xx.img`. There should be 360 `*.img` files.
12. Click REGRESSORS, add 3 regressors and Expand All
13. Regressor 1: Name: PPI-interaction, Value: PPI.ppi
14. Regressor 2: Name: V2-BOLD, Value: PPI.Y

15. Regressor 3: Name: Psych.Att-NoAtt, Value: PPI.P
16. High Pass Filter [192]
17. MODEL ESTIMATION → SELECT SPM.MAT → OUTPUT FROM “fMRI MODEL SPECIFICATION”
18. CONTRAST MANAGER → SELECT SPM.MAT → OUTPUT FROM “MODEL ESTIMATION”
19. CONTRAST MANAGER → CONTRAST SESSIONS → NEW “T-CONTRAST”. Double click T-CONTRAST to expand.
20. T-contrast: Name: PPI-Interaction
21. T-contrast vector: 1 0 0 0
22. Save the batch file.
23. Run

The design matrix is shown below.

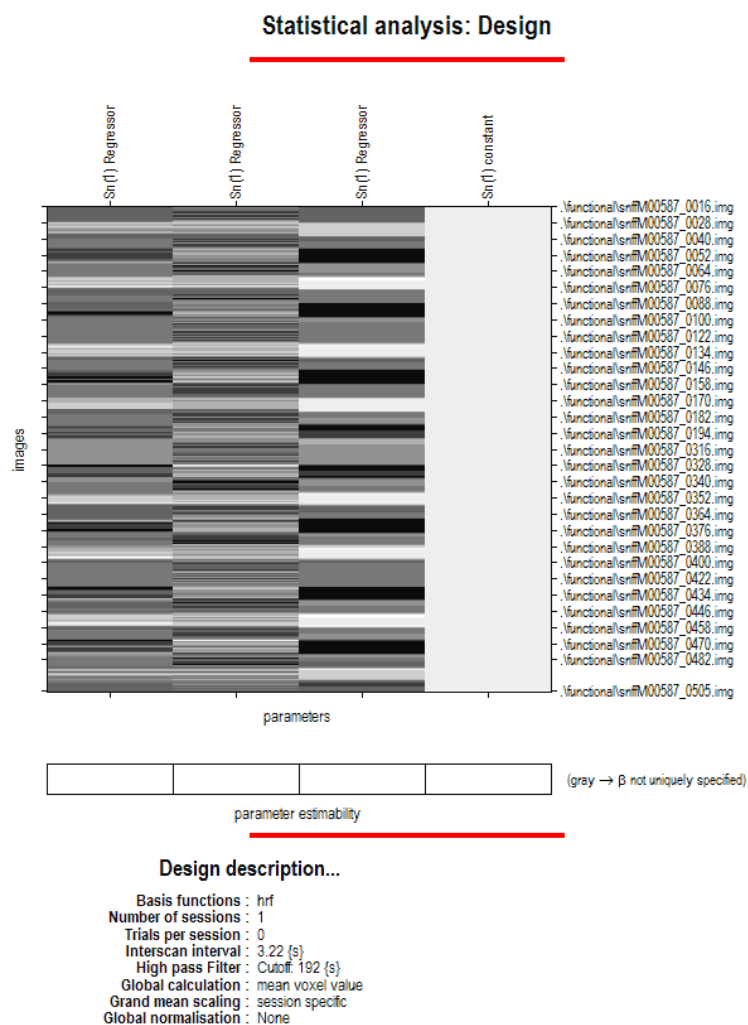


Figure 16.11: *Design matrix of the PPI analysis*

### 16.4.2 PPI analysis - Results

1. Press the RESULTS button and select the SPM.mat file in the PPI directory.
2. Choose the PPI-Interaction contrast
3. Mask with other contrasts [No]
4. Title for comparison [PPI-Interaction]
5. p value adjustment to control [None]
6. threshold T or p value [0.01]
7. & extent threshold voxels [10]
8. You should see an SPM that looks the same as the one shown below. The resulting SPM shows areas showing differential connectivity to V2 due to the effect of attention vs. no attention conditions. The effect in this subject is weak.

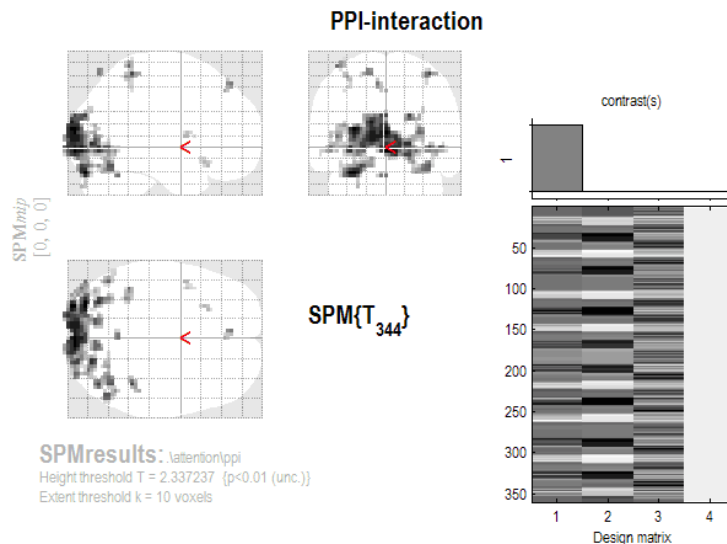


Figure 16.12: PPI results

### 16.4.3 PPI analysis - Plotting

1. One region showing the psychophysiologic interaction is the V5 region , which is located at [39 -72 0] in this subject. Move the cursor to this point to view the area of activation, as shown below.
2. In order to plot the PPI graph showing the effect of attention, you need to extract a VOI from the V5 region.
3. Select the GLM analysis SPM.mat file
4. Mask with other contrasts [No]
5. Title for comparison [Motion]
6. p value adjustment to control [None]
7. threshold T or p value [0.001]
8. & extent threshold voxels [3]

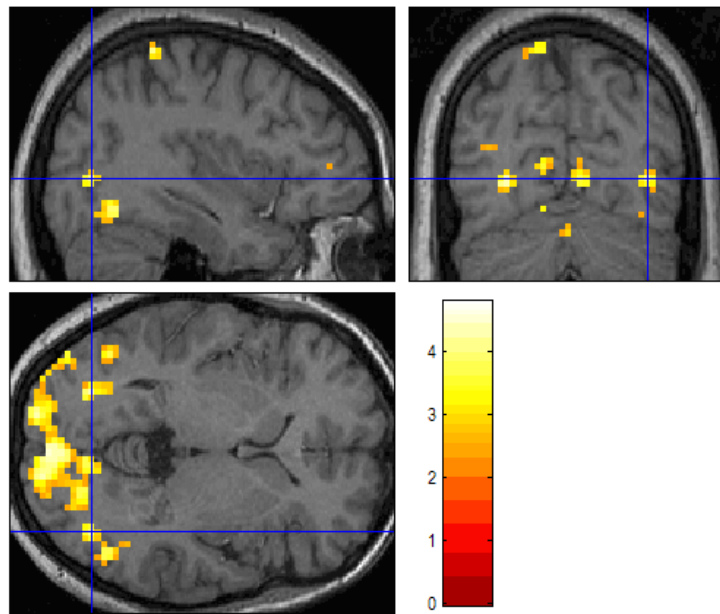


Figure 16.13: Time series extraction in V5

9. Go to point [39- 72 0]
10. Press eigenvariate
11. Name of region [V5]
12. Adjust data for [effects of interest]
13. VOI definition [sphere]
14. VOI radius(mm) [6]
15. Now you will create 4 PPIs (Follow the steps under Create PPI Variable, above). By using the PPI software machinery to create the interaction vectors, rather than just multiplying the extracted V2 and V5 eigenvariates by the behavioral vectors, the PPI vectors will be formed properly.
  - (a) **V2xNoAttention** (Use the V2 VOI and include **No-Attention** with a contrast weight of 1, do not include **Fixation, Stationary, Attention**)
  - (b) **V2xAttention** (Use the V2 VOI and include **Attention** with a contrast weight of 1, do not include **Fixation, Stationary, No-Attention**)
  - (c) **V5xNoAttention** (Use the V5 VOI and include **No-Attention** with a contrast weight of 1, do not include **Fixation, Stationary, Attention**)
  - (d) **V5xAttention** (Use the V5 VOI and include **Attention** with a contrast weight of 1, do not include **Fixation, Stationary, No-Attention**)
16. Load the PPIs you just created with the following commands at the MATLAB prompt:
 

```
>> v2att = load('PPI_V2xAttention.mat');
>> v5noatt = load('PPI_V5xNoAttention.mat');
>> v5att = load('PPI_V5xAttention.mat');
```
17. Plot the PPI datapoints with the following commands at the MATLAB prompt:

```
>> figure
>> plot(v2noatt.PPI.ppi,v5noatt.PPI.ppi,'k-');
>> hold on
>> plot(v2att.PPI.ppi,v5att.PPI.ppi,'r-');
```

18. To plot the best fit lines type the following first for NoAttention

```
>> x = v2noatt.PPI.ppi(:);
>> x = [x, ones(size(x))];
>> y = v5noatt.PPI.ppi(:);
>> B = pinv(x)*y;
>> y1 = (B(1)*x(:,1))+B(2);
>> plot(x(:,1),y1,'k-');
```

19. Then for Attention

```
>> x = v2att.PPI.ppi(:);
>> x = [x, ones(size(x))];
>> y = v5att.PPI.ppi(:);
>> B = pinv(x)*y;
>> y1 = (B(1)*x(:,1))+B(2);
>> plot(x(:,1),y1,'r-');
>> legend('No Attention','Attention')
>> xlabel('V2 activity')
>> ylabel('V5 response')
>> title('Psychophysilogic Interaction')
```

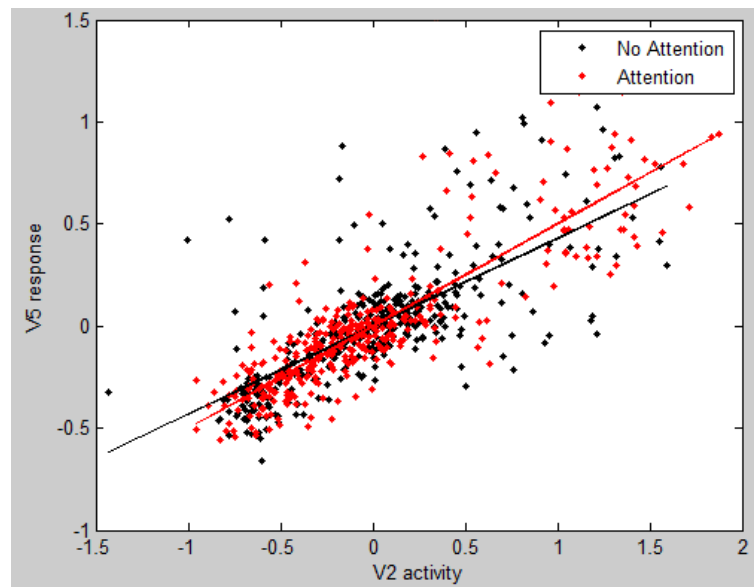


Figure 16.14: