6
Investigation with fMRI

The behavioral studies reported in the previous two chapters indicate the possibility that stimuli are processed in a different manner when presented simultaneously and sequentially. Specifically, these studies show that location information differentially affects performance in the simultaneous and sequential presentation conditions. To observe the underlying differences and the interaction pattern in brain areas related to feature binding, the experiment reported in this chapter was designed to obtain and compare fMRI BOLD data under all the four experimental conditions studied in the behavioral experiments, comprising simultaneous and sequential presentation, each with unchanged and random locations.

Although previous researches at the brain level directly comparing simultaneous and sequential presentation testing feature binding are relatively rare, they do show that the processing of multiple objects differs in the brain, when they are presented simultaneously and sequentially. Shafritz *et al.* [2002] specifically compared memory for bindings and uni-feature objects, when the stimuli were presented sequentially and simultaneously. They found greater activation in the right superior parietal lobule and intra parietal cortices in the binding condition than in the single features condition. This was true, however, for only simultaneous presentation, with no clear results for sequential presentation. Shafritz *et al.* [2002] concluded that simultaneous presentation of several stimuli at different locations was essential for feature binding.

Coull *et al.* [2003] specifically aimed to explore whether the superior parietal cortex is strictly spatial in nature. They compared detection of single features and conjunctions [bindings] using a search paradigm with the stimuli presented simultaneously or sequentially. They found that the medial parietal cortex was bilaterally more activated for simultaneous than sequential presentation in conjunction search. With sequential presentation, the right superior parietal cortex and the bilateral intra-parietal sulci were activated in conjunction search [besides the frontal operculum and putamen]. So, different regions emerged to be important for simultaneous and sequential presentation within the parietal cortex. The right lateral parietal cortex [BA7] was however, equally activated in simultaneous as well as sequential presentation, confirming a similar result by Wojciulik and Kanwisher [1999].

Xu and Chun [2006] studied unicolor shapes in three conditions – stimuli presented simultaneously but off-center, stimuli presented sequentially off-centre, and stimuli presented sequentially at a central location. They reported an increment in activation in the superior and inferior intra parietal sulci with off-center presentations, sequential as well as simultaneous, as compared to the central location presentation. They concluded that intra parietal activity is caused by attention to multiple locations. Xu and Chun [2009] theorized that the inferior IPS is linked to spatial attention and leads to "object individuation", but the superior IPS and the lateral occipital complex is linked to maintenance of a subset of attended objects, depending on their complexity, and are therefore linked to "object identification". More recently, Bettencourt

and Xu [2016] also distinguished between location based and feature based processing in the intra parietal sulcus. They concluded that there is a need to understand the multiplex nature of this brain region not in terms of 'where' activity is seen, but how the same region participates in different cognitive tasks. Perhaps this is true, not only for the intra parietal sulcus, but also for all the brain regions.

None of the aforementioned neuroimaging studies of simultaneous vs. sequential presentation, conceptualized locations as a distinct factor. Indeed, these researchers [Coull *et al.*, 2003; Shafritz *et al.*, 2002; Xu and Chun, 2006] primarily designed their studies to compare simultaneous and sequential presentation, but ended up drawing conclusions regarding locations. Thus, the present study, which orthogonally manipulates mode of presentation and locations to study their independent as well as interactive effect, will augment this literature, as it will precisely delineate the role of location information in the effects of simultaneous as well as sequential presentation.

As far as other factors affecting performance in the sequential presentation condition are concerned, as explained in Section 2.3 [in the chapter on review of literature], several behavioral studies have shown the greater involvement of working memory resources, particularly related to executive attention, when stimuli are sequentially presented [e.g., Ihssen *et al.*, 2010; Rudkin *et al.*, 2007]. One may also speculate that with sequential presentation, participants need to store the previous stimulus and at the same time process the next. Alternatively, they may be still processing a stimulus, when the next one arrives. In both cases, there is a greater utilization of working memory resources [for storage and/or processing at the same time]. Therefore, in fMRI, greater activation may be reported from the working memory related areas with sequential presentation.

Working memory was initially associated with single cell stimulation in the frontal cortex in monkeys [Goldman-Rakic, 1988]. Several studies subsequently used fMRI to establish activation of the frontal cortex particularly the dorsolateral prefrontal cortex in working memory [Prabhakaran *et al.*, 2000; Rowe *et al.*, 2000; Smith and Jonides, 1997]. Since then, several other areas of the brain have been associated with working memory, particularly the parietal cortex [Linden *et al.*, 2003; Todd and Marois, 2004]. Combining magneto encephalography and electro encephalography, Palva *et al.* [2010] reported increased synchrony in the fronto-parietal regions with increased memory load, whereas individual working memory capacity was predicted by synchrony in a network with the intra-parietal sulcus as its hub. Salazar *et al.* [2012] reported widespread, task-dependent, and content-specific synchronization of activity across the fronto-parietal network during visual working memory. The stimulus-selective neurons were governed by signals arising in the parietal cortex.

Recent studies have focused on finding dissociations between causal influences of the frontal and parietal areas. Mackey and Curtis [2017] used TMS to establish that the parietal cortex mainly codes for retrospective sensory information, whereas the frontal cortex codes for prospective action. More pertinent to the current work is the study by Li *et al.* [2017]. Using trans cranial direct current stimulation, they found that stimulation of the right PPC specifically increased the visual working memory capacity under the no-distractor condition, whereas stimulation of the right PFC specifically increased visual working memory capacity under a distractor present condition. They also showed that compared to central presentation of the stimuli, bilateral presentation of the stimuli led to a greater demand for attention control.

Whilst noting the areas activated in sequential performance, it is also important to consider the work by researchers who have demonstrated a role for the parietal cortex [Arend, *et al.*, 2011] and the MTL [Pertzov *et al.*, 2013] in sequential presentation. However, these studies do not involve a direct comparison with simultaneous presentation. The details of these studies are in the review of literature [Section 2.6.2].

A direct comparison of simultaneous and sequential presentation exists in the work of Ihssen *et al.* [2014] but for single feature objects. They found activation in the primary visual area V1 and extra striate areas in their experiment, when the sequential to simultaneous contrast was made. They had presented an array of 8 stimuli in three ways: simultaneously for 700 ms, repeated twice for 350 ms, and divided into two 4-objects arrays presented for 350 ms each, and had found enhanced memory for their single feature objects in the repeated and half sequence modes as compared to simultaneous presentation.

As far as the independent variable of locations is concerned, in the present research, it is unchanged in one condition and random in the other condition, from the study to the test display. The test display always comprises multiple stimuli presented together. As such, when the stimuli remain unchanged, the participant can rely on relatively automatic encoding of the stimuli together as a pattern, and the subsequent iconic memory; particularly in the simultaneous presentation condition. However, when stimuli are random from study to test display, configural encoding and iconic memory can actually work against good performance on the binding task. Random locations require the participants to move the focus of attention from one item to the other, in search of the item that has changed. This shifting focus of attention, with or without eye movements, recruits a core network of fronto-parietal and temporal brain regions as concluded by Grosbras *et al.* [2005] after a meta-analysis of 59 brain imaging experiments. For the present research, this suggests the recruitment of areas in the frontal, parietal, and temporal cortices, to a greater extent in the random locations condition.

Two types of analyses of fMRI data were envisaged. First, conjunction null analyses were to be carried out to reveal the areas commonly activated in the different levels of each independent variable. Conjunction analysis is a method that gathers evidence of activation in brain areas, which would *definitely* be involved in all the experimental conditions considered in that analysis, ignoring the interactions if any. Second, specific ROI delineated on the basis of previous studies, were to be explored for the main effects of locations and mode of presentation, and their interaction. Focusing on specific ROI rather than whole brain ensured that the specific expectation regarding the results of the two independent variables and their interaction would be tested in all the binding related areas.

The most well-known ROI in binding studies is the parietal cortex. The parietal lobe was among the first brain regions associated with feature binding through clinical studies [Friedman-Hill *et al.*, 1995] as well as fMRI evidence and is consequently an ROI in the present research. Shafritz *et al.* [2002] reported that the regions from right parietal areas were significantly more activated with simultaneous presentation at multiple locations than sequential presentation in a single location. Parietal activation in their experiment could be because of simultaneous presentation or multiple locations. The present research endeavors to unravel this confound and study whether parietal activation is associated with attention to multiple locations or to multiple stimuli.

Todd and Marois [2004] found a major role of the intra parietal sulcus [IPS] in binding, when they studied the binding of color and location using simultaneous presentation. Song and Jiang [2006] tested the capacity for working memory in single feature and binding conditions. They found significantly increased activity in bilateral superior parietal lobules for binding and shape only conditions as compared to the color only condition. Xu [2007] investigated the role of visual short-term memory in the process of binding of color and shape, and found that bilateral intra-parietal sulci are involved in the storage of bindings. Recently, Bettencourt and Xu [2016] confirmed that the inferior IPS is involved in location-based object processing whereas the superior IPS primarily encodes and stores surface features of objects. This indicates that one may obtain a differential pattern of activation in the superior and inferior parietal sulci depending on which features are processed. More precisely, in the present study, different

levels of activation may be observed in various regions of IPS depending on whether or not processing of locations is endemic to the task.

The precentral gyrus is another important ROI as per previous studies. Mitchell *et al.* [2000] studied the binding of object identity and location in younger and older samples using sequentially presented objects in multiple locations. They found the left pre-central gyrus activated for binding in both the participant groups, whereas the anterior hippocampus activated only among the younger adults. Donner *et al.* [2002] found activation in both, the pre and the post central sulci, when participants searched for conjunctions as well as for orientations [assumed to be the hard features] in comparison to the search for color [termed the easy feature by the authors]. Raabe *et al.* [2013], using a delayed matching-to-sample task involving binding of color and orientation in spatial working memory, found a major contribution from the post-central gyrus [in addition to the inferior parietal lobe and precuneus]. These studies show the involvement of the precentral gyrus whenever participants have to search an object in visual space. As far as the experimental condition in the present experiment requires search in visual space, activation in the pre-central gyrus is expected. Search for an object may be crucial to performance in the condition involving simultaneous presentation with random locations and with sequential presentation in the unchanged as well as random location conditions.

Another ROI, the hippocampus, is suggested by studies comparing bindings with uni-feature stimuli. Mitchell *et al.* [2000] reported that the left anterior hippocampus was more activated in young adults for binding of objects and their location as compared to memory for objects alone or locations alone. Memel and Ryan [2017] reported higher activity in the left hippocampal region for older as well as younger population in a visual integration task. In the present research, the hippocampus is expected to be active, but no specific prediction is made for differential activation in the four experimental conditions.

The fusiform gyrus is another ROI as it has been found to be active in feature binding studies. Schoenfeld *et al.* [2003] found activation in the fusiform gyrus when relevant as well as irrelevant features were integrated in a binding task. Parra *et al.* [2014] also found that the fusiform gyrus is significantly more active in encoding and maintenance of color-shape bindings as compared to shape only and color only conditions. The fusiform gyrus is also associated with object recognition [Grill-Spector *et al.*, 2001] To the extent that object based processing is required in the conditions of the current fMRI experiment, the fusiform gyrus is expected to be differentially activated. Object based processing is less likely in the simultaneous presentation condition with unchanged locations.

Recent studies on spatial working memory indicated the supra marginal gyrus as another ROI for the present experimenter. Silk *et al.* [2010] used a dual task paradigm and studied the underlying brain areas, using a task of spatial working memory and visual search. They suggested that the supra marginal gyrus as well as the intra-parietal sulcus are critical for spatial working memory as well as shifts in spatial attention. Yang *et al.* [2017] observed the change in BOLD activity during object identity and location binding task whilst manipulating memory load. They reported less suppressed activity in left supra marginal gyrus in the changed locations condition than the same locations condition. In keeping with these results, in the present experiment, the left supra marginal gyrus is expected to show higher activation in the random locations condition as compared to the unchanged locations condition.

All these binding related ROIs were investigated to study the differential pattern of activity associated with the main effects of mode of presentation and locations, and particularly the interaction effect. The available literature shows no fMRI study focusing on the interaction of these two variables.

The exact coordinates defining the center of each ROI and the studies on the basis of which they were defined are given in Table 6.1.

Table 6.1: Brain regions of interest associated with feature binding

Region	*Side*	*MNI*			*As suggested by*
		x	y	z	
Intra-parietal cortex	R	39	-43	51	Shafritz *et al.* [2002]
Superior parietal cortex	R	15	-62	63	
		30	-60	48	Song and Jiang [2006]
Superior parietal lobule	L	-24	-60	48	
Intra parietal sulcus	L	-22	-69	42	Todd and Marois [2004]; Xu [2007]
Intra parietal sulcus	R	23	-63	50	
Pre-central gyrus	L	-54	-6	36	Mitchell *et al.* [2000]
Anterior hippocampus	L	-35	-14	-17	
Fusiform gyrus	R	45	-63	-15	Parra *et al.* [2014]
Supra marginal gyrus	R	57	-48	33	Yang *et al.* [2017]

Based on the foregoing review of research, the expectations from the fMRI study are summarized in the following paragraphs.

Conjunction null analyses were carried out to study the common areas recruited in all conditions, and in two levels of each independent variable, i.e., the two modes of presentation and unchanged and random locations.. The task being visual binding, activation in the parietal cortex was definitely expected to be common across all conditions as it is almost universally accepted as essential to binding.

Greater overall activation was expected with random locations, particularly in the fronto parietal and temporal brain areas [based on Grosbras *et al.*, 2005] and supra marginal gyrus [as found by Yang *et al.*, 2017]. Greater activation was also expected with sequential presentation than with simultaneous presentation in visual areas [in consonance with Ihssen *et al.*, 2014]. If sequential presentation recruits extra attentional and/or working memory resources, as per evidence regarding working memory being based on activation of the fronto parietal network [e.g., Li *et al.*, 2017, Palva *et al.*, 2010, Salazar *et al.*, 2012], greater activation in the frontal and parietal regions was also expected with sequential than simultaneous presentation.

As far as the analyses of specific ROIs are concerned, based on the behavioral experiments reported earlier in this thesis, a larger difference in the amount and pattern of activation due to unchanged and random locations was expected with simultaneous presentation than with sequential presentation.

Specifically, the least activation was expected in the attention related or object focused ROIs, such as the parietal regions and the fusiform area, in the simultaneous presentation unchanged locations condition. The precentral gyrus, closely associated with oculomotor responses, was also expected to be less involved in the simultaneous presentation unchanged locations condition. This is because, it was speculated that the simultaneous presentation condition with unchanged locations yields superior behavioral performance due to relatively

automatic configural encoding which uses lesser attentional resources. In contrast, all other conditions of the experiment require a shifting focus of attention on all stimulus objects, recruiting greater resources in terms of shifting focus of attention and working memory for storage and/or processing.

6.1 DESIGN AND PROCEDURE

6.1.1 Experimental Design

The fMRI experiment was similar to Experiment 3, with the following changes necessitated by the fMRI environment:

1. A block design was used, such that random and unchanged locations were also presented in separate blocks, besides simultaneous and sequential presentation.
2. The duration of fixation display was constant at 500 ms, and the response time window was constant at 1000 ms, so that a fixed sequence protocol could be defined for each participant.
3. The total number of experimental trials was reduced to half in every experimental condition yielding a total of 192 experimental trials. This was done to shorten the session and avoid fatigue inside the scanner. All participants also completed a practice session of 48 trials outside the scanner.
4. Articulatory suppression was not used to avoid language and memory related activity.

The experiment was a 2 × 2 factorial design with repeated measures on both factors – mode of presentation [simultaneous vs. sequential] and locations [unchanged vs. random].

The whole experiment was conducted in a single session using a block design. Mode of presentation was counterbalanced in the sequence ABBABAAB, where A and B respectively imply simultaneous and sequential presentation. Unchanged [U] and random [R] locations alternated as URURURUR in a separate sequence. Baseline blocks, which were simply a fixation display, alternated with every experimental block. The sequence started and ended with a baseline block. Cumulatively, there were 17 blocks comprising eight experimental blocks and nine baseline blocks. Trials requiring 'same' or 'different' responses were equal in number and random in order within each block.

The task was presented using the NordicNeuroLab presentation hardware with 800×600 screen resolution. Each trial began with a fixation display of 500 ms, and then presented four stimuli with the participant required to remember the color and shape binding. The presentation time was 250 ms for all stimuli in the simultaneous presentation condition and for each stimulus in the sequential presentation condition, with each stimulus offset with the onset of the next stimulus.

The test display presented all the stimuli together for change detection and remained until the response of the participant, although it was also set to jump after 1000 ms if the participant did not respond, to the fixation screen of the next trial. Behavioral experiments had shown that this was a sufficient time window for the response of the participant.

The sequence of events in each trial is shown in Figure 6.1.

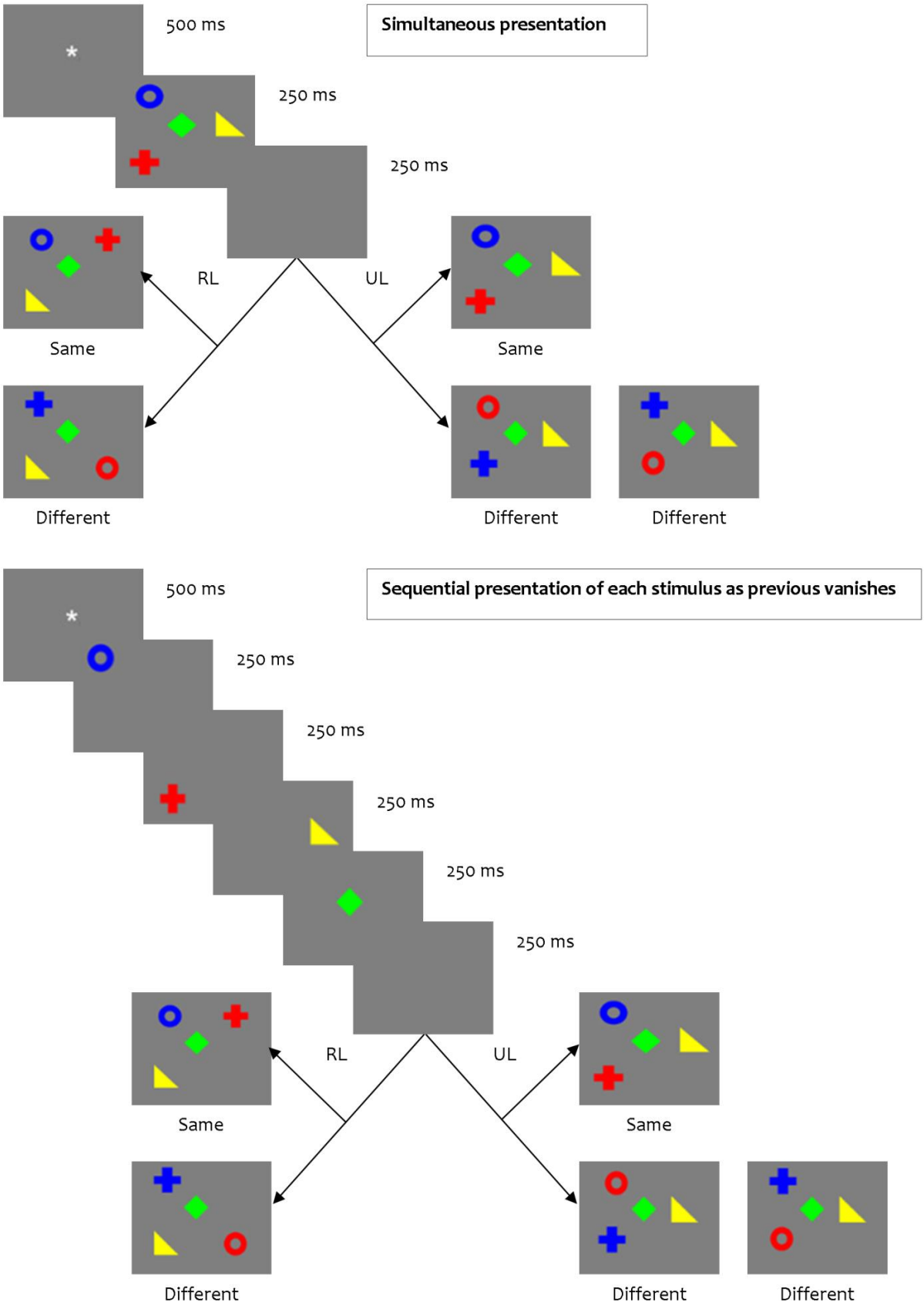


Figure 6.1: Sequence of events in the fMRI experiment

6.1.2 Acquisition of fMRI Data

All participants were scanned in a single session to obtain an fMRI time-series. The stimuli were presented through the NordicNeuroLab visual system. The experiment was carried out using the blocks described in the previous section. Participants detected changes in the binding of color and shape by pressing the thumb button [left for 'different' and right for 'same' response] on the Nordic response grip. The neuroimaging data was obtained by 3T Siemens MAGNETOM Skyra Whole-body MRI system, equipped with a 20-channel head and neck coil. The head was supported and immobilized [using foam pads] within the head coil to minimize head movement and gradient noise. Anatomical T1-weighted images were collected using a three-dimensional magnetization-prepared rapid gradient echo [MPRAGE] sequence, with 160 contiguous 1 mm thick sagittal slices [echo time [TE] = 2.07 ms; repetition time [TR] = 1900 ms; field of view [FOV] = 256 mm; flip angle = 9°; voxel size = 1×1×1 mm]. A total of 242 functional brain volumes were acquired using echo-planar T2*weighted sequence [TE = 30 ms, TR = 3000 ms, 64×64×30 matrix size, flip angle = 90°, 3.75×3.75×5 mm voxels].

6.1.3 Processing and analyses of fMRI Data

Data preprocessing

The data were analyzed with SPM 12 [Ashburner *et al.*, 2020; Wellcome Department of Cognitive Neurology, http://www.fil.ion.ucl.ac.uk/] running on MATLAB R2014a [MathWorks, 2014]. In brief, fMRI images were slice time corrected and then they were realigned using the first image as the reference image. The translational and rotational movement for all the participants was within ± 1.5 mm and ± 1.5° respectively, hence all the 18 participants were included in the analysis. The realignment parameters were used as regressors during participant level analysis to remove the motion artifacts. The anatomical image for each participant was co-registered with the mean functional image generated from realignment. The registered anatomical images were then segmented to obtain deformation fields, which were subsequently used to normalize the realigned functional images and registered anatomical images to the MNI space. The spatially normalized images were then smoothed with a Gaussian kernel of 8 mm FWHM [full width half maximum]. The first baseline block was removed to avoid initial transit signal fluctuation. Low frequency drifts were removed using a high pass filter with cut-off frequency of 128 seconds [~.0078 Hz] during analysis. The anatomical details of the activation clusters were obtained using SPM anatomy toolbox [Eickhoff *et al.*, 2005].

Conjunction null analyses

Conjunction null analyses were carried out to find the common areas activated in the different levels of each independent variable. Participant-specific contrasts were obtained by subtracting activation in the baseline from each experimental condition. At the second level, random effect analysis was performed to find significant activation across participants in each of the four experimental conditions by carrying out a one way *ANOVA*. Thereafter, five conjunctions were explored. The first revealed common areas associated with simultaneous presentation for unchanged and random locations [[SIMU-Base] ∩ [SIMR-Base]]. The second revealed common areas associated with sequential presentation for unchanged and random locations [[SEQU-Base] ∩ [SEQR-Base]]. The third was for common areas recruited with unchanged locations across simultaneous and sequential presentation [[SIMU-Base] ∩ [SEQU-Base]]. The fourth was for common areas recruited with random locations across simultaneous and sequential presentation [[SIMR-Base] ∩ [SEQR-Base]]. The last conjunction revealed common areas recruited in all four conditions [[SIMU-Base] ∩ [SIMR-Base] ∩ [SEQU-Base] ∩ [SEQR-Base]]. As conjunction null analysis is a conservative method used to find commonly activated areas across conditions and limits the risk of false positives, the thresholds used for activation maps were $p<.01$ [FWE corrected] and $k>20$ voxels.

ROI analyses

The ROIs were explored using Marsbar Toolbox designed for SPM [Brett *et al.*, 2002]. For each participant, contrast files were obtained by subtracting the baseline from the experimental condition. All the ROIs were built as a sphere with 8 mm radius around the defined coordinates as given in Table 6.1. The mean signal [parameter estimate] across all voxels in each experimental condition was then extracted, resulting in four values per ROI [one for each experimental condition] for each participant. For each ROI, a 2×2 repeated measures *ANOVA* was carried out on the mean parameter estimates obtained for the eighteen participants, testing the main effects and the interaction of modes of presentation and locations.

6.2 RESULTS

6.2.1 Behavioral results

Primary Analyses

Mean change detection performance calculated from d primes is shown in Figure 6.2. A repeated measures *ANOVA* revealed the main effect of unchanged and random locations, $F[1,17]=7.855$, $MSE=.797$, $p<.012$, *partial* $\eta^2=.316$, $BF_{10}=6.727$. Overall performance was reduced when locations were randomly changed from study to test display than when locations were unchanged. The main effect of simultaneous and sequential presentation was not significant. The interaction effect of mode of presentation and locations was also not significant. In Bayesian *ANOVA*, the model comprising both main effects and the interaction effect [$BF_{10}= .051$] was compared with the model comprising only the main effects [$BF_{10}= 1.651$]. The model comprising only the main effects better fit the data by a factor of 3.295:1.

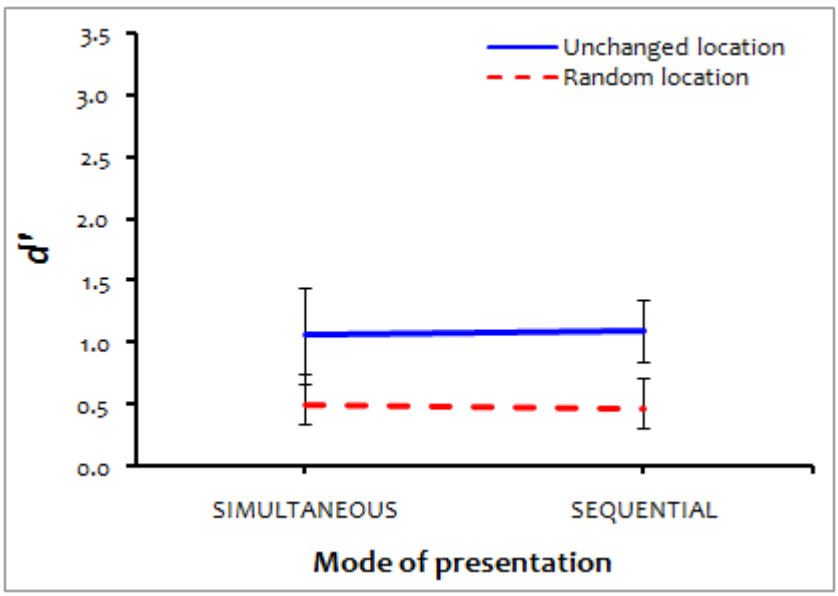


Figure 6.2: Mean d prime scores of behavioral responses in the fMRI experiment

Serial position effects in sequential presentation

The serial position effects in the sequential presentation condition were explored using a 2×4 repeated measures *ANOVA* [location × swaps]. The swaps selected for this analysis were between stimuli shown at serial positions 1 and 4 [showing the joint effect of primacy as well as recency], 1 and 2 [showing only primacy effect], 2 and 3 [items in the middle positions], and 3

and 4 [showing only the recency effect]. Neither the main effects nor the interaction effect was significant. To explore the interaction using Bayesian *ANOVA*, the model comprising both main effects and the interaction effect [BF_{10}= .273] was compared with the model comprising only the main effects [BF_{10}= 2.937]. The model comprising only the main effects better fit the data by a factor of 10.758:1. Figure 6.3 shows these results.

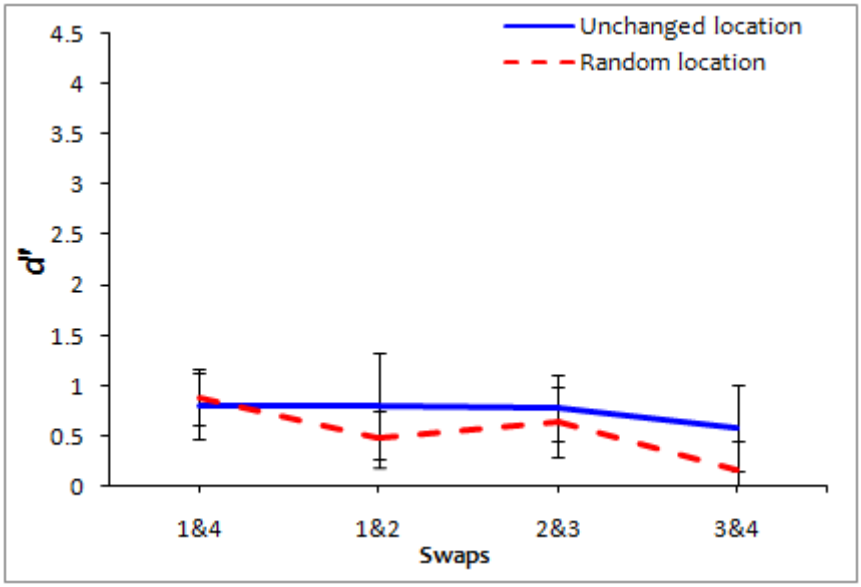


Figure 6.3: Serial position effects in behavioral responses in the fMRI experiment

Comparison with Experiment 3

To compare the behavioral data obtained in this experiment with Experiment 3 [with which the design was most closely aligned], a three way analysis was carried out with experiments as the between participants variable, and mode of presentations and locations as the repeated measures variables. A significant main effect of experiments was found, F [1, 34]= 9.326, MSE = 2.058, p< .004, *partial* η^2= .215, BF_{10}=8.513, as overall performance in the fMRI experiment [M=.782, SD = .169] reduced in comparison with Experiment 3 [M=1.512, SD =.169].

The three way interaction was also significant, F[1,34]= 10.734, MSE = .450, p< .002, *partial* η^2= .240, BF_{10}= 11.525. The model comprising all the main effects, all two way interaction effects, and the three way interaction effect [$BF_{10}=2.543\times10^6$] was compared with the model comprising all main effects and all two way interaction effects [$BF_{10}=2.206\times10^5$]. The model with the three-way interaction effect better fit the data by a factor of 11.527:1.

The pattern of interaction of mode of presentation and locations is different in the two experiments. A comparison of Figure 6.2 with Figure 5.2 suggests that this was likely due to reduced performance in the condition with simultaneous presentation and unchanged locations in the fMRI experiment, the reduction being much more, than in the other three conditions. This could be because the fMRI environment possibly disrupts spatiotopic/iconic memory [Coltheart, 1980], which, as is argued later in the discussion, is primarily responsible for better performance in this condition. Iconic memory is disrupted even by small distractions such as voluntary eye blinks [Thomas and Irwin, 2006], what to speak of the noise, motion restriction, and claustrophobic conditions in the scanner. Also, when disrupted, the memory for locations is more affected than memory for objects [Dick, 1969, Irwin and Yeomans, 1986; Townsend 1973].

6.3.2 Neuroimaging results

Conjunction null analyses

Conjunction null analyses revealed the areas of the brain, which were recruited in common by the different levels of the two independent variables, and by all experimental conditions as well. The results of the five conjunctions tested in the present work are as follows.

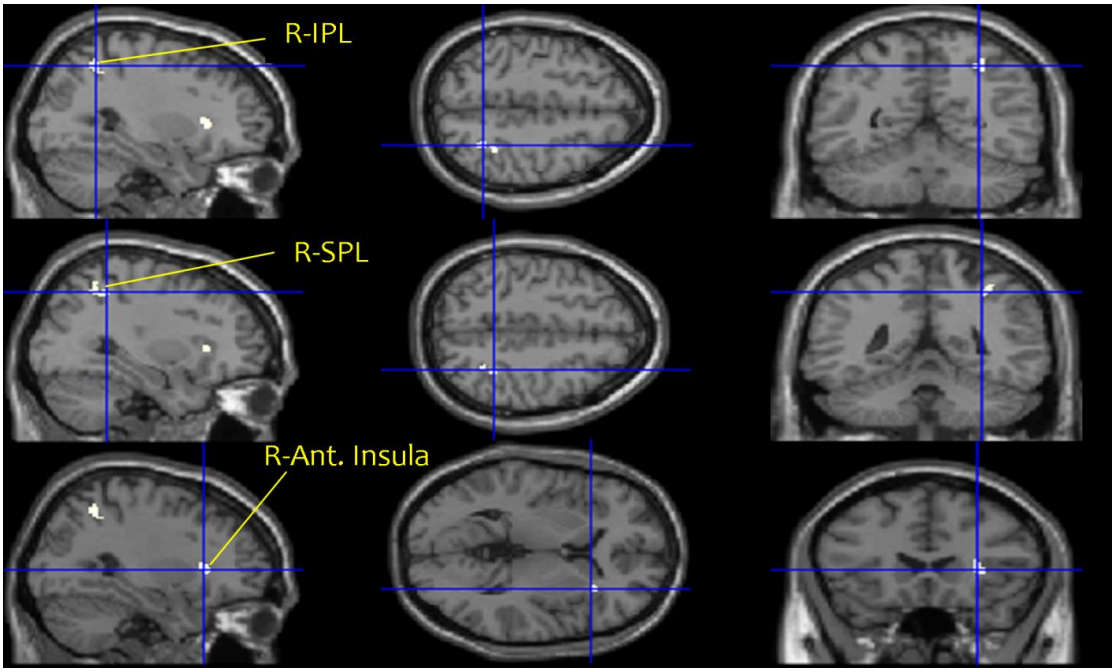
Brain areas activated with simultaneous presentation [[SIMU-Base] ∩ [SIMR-Base]]

To observe the brain regions recruited by simultaneous presentation, conjunction analysis was carried out for unchanged and random locations within this level. Major areas of activation were the inferior parietal lobule, superior parietal lobule, and anterior insula, in the right hemisphere as shown in Table 6.2 and Figure 6.4.

Table 6.2: Activation with simultaneous presentation [[SIMU-Base] ∩ [SIMR-Base]]

Cluster	L/R	BA	Anatomical region	SPM{Z}	x	Y	z	No. of Voxels
1	R	7	Inferior Parietal Lobule	5.94	+30	-54	+50	59
		7	Inferior Parietal Lobule	5.82	+36	-44	+52	
		7	Superior Parietal Lobule	5.74	+32	-46	+48	
2	R	13	Anterior Insula	5.36	+30	+22	+4	36

All regions are significant at p<.01 [FWE corrected] and k>20



[Activated areas are shown in sagittal, axial, and coronal planes from left to right. R-IPL = Right Inferior Parietal Lobule; R-SPL = Right Superior Parietal Lobule; R-Ant. Insula = Right Anterior Insula]

Figure 6.4: Activation with simultaneous presentation

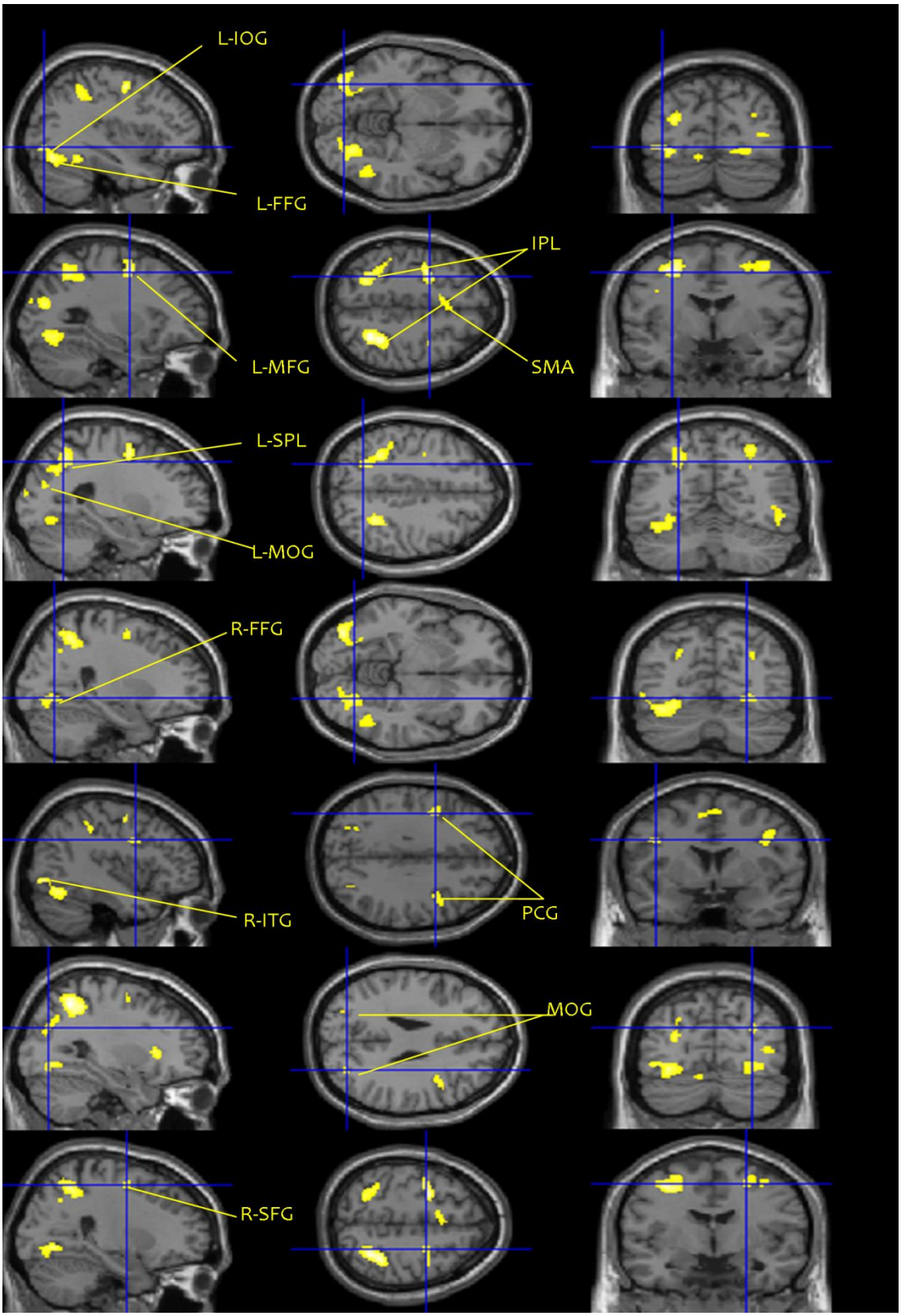
Brain areas activated with sequential presentation [[SEQU-Base] ∩ [SEQR-Base]]

To obtain the areas activated with sequential presentation, conjunction analysis was carried out on unchanged and random locations conditions within sequential presentation. Significant activation was in bilateral occipital and parietal regions, bilateral frontal cortex including middle and inferior frontal gyri, precentral gyri, supplementary motor areas, and left cerebellum. These areas are shown in Table 6.3 and Figure 6.5.

Table 6.3: Activation with sequential presentation [[SEQU-Base] ∩ [SEQR-Base]]

Cluster	L/R	BA	Anatomical region	SPM[Z]	x	y	z	No. of Voxels
1	L	19	Inferior Occipital Gyrus	7.51	-36	-78	-6	728
		37	Fusiform Gyrus	7.27	-36	-70	-16	
		37	Fusiform Gyrus	7.17	-34	-72	-14	
		37	Fusiform Gyrus	7.09	-42	-64	-16	
		37	Fusiform Gyrus	6.74	-36	-48	-16	
2	R	7	Inferior Parietal Lobule	8.48	+30	-54	+50	564
		7	Inferior Parietal Lobule	7.88	+34	-44	+50	
		7	Inferior Parietal Lobule	7.83	+34	-46	+54	
3	L	7	Superior Parietal Lobule	7.31	-24	-60	+44	547
		7	Inferior Parietal Lobule	7.03	-32	-46	+44	
		7	Inferior Parietal Lobule	6.71	-30	-54	+50	
		39	Middle Occipital Gyrus	6.56	-24	-66	+36	
4	L	6	Middle Frontal Gyrus	8.17	-28	-2	+50	243
		6	Middle Frontal Gyrus	7.70	-34	-4	+52	
5	R	37	Inferior Temporal Gyrus	6.66	+50	-60	-10	218
		37	Inferior Temporal Gyrus	6.18	+42	-54	-10	
6	R	19	Fusiform Gyrus	6.85	+26	-68	-6	214
		19	Fusiform Gyrus	6.30	+34	-74	-9	
7	R	44	Inferior Frontal Gyrus	6.39	+42	+10	+28	182
		8	Precentral Gyrus	6.33	+46	+8	+38	
		44	Inferior Frontal Gyrus	6.24	+40	+6	+30	
		8	Inferior Frontal Gyrus	6.23	+40	+4	+36	
8	L	6	Supplementary Motor area	6.26	-6	+8	+54	148
	R	6	Supplementary Motor area	6.15	+4	+6	+58	
	L	6	Supplementary Motor area	5.87	-0	+14	+50	
	R	6	Supplementary Motor area	5.64	+4	+20	+46	
9	L	19	Middle Occipital Gyrus	6.78	-28	-76	+20	131
		19	Middle Occipital Gyrus	6.07	-30	-86	+22	
10	R	6	Superior Frontal Gyrus	6.58	+26	-4	+54	97
		6	Superior Frontal Gyrus	6.40	+38	-2	+56	
11	R	39	Middle Occipital Gyrus	5.97	+30	-74	+28	64
		7	Middle Occipital Gyrus	5.88	+28	-64	+36	
		7	Middle Occipital Gyrus	5.83	+30	-66	+36	
12	L	6	Precentral Gyrus	6.37	-40	+4	+32	49
13	L	18	Superior Occipital Gyrus	6.62	-16	-96	+14	39
		18	Middle Occipital Gyrus	6.44	-22	-94	+14	
14	R	19	Middle Occipital Gyrus	5.92	+38	-76	+8	38
15	L		Cerebellum	5.86	-10	-76	-16	25

All regions are significant at p<.01 [FWE corrected] and k>20



[Activated areas are shown in sagittal, axial, and coronal planes from left to right. L-IOG = Left Inferior Occipital Gyrus; L-FFG = Left Fusiform Gyrus; L-MFG= Left Middle Frontal Gyrus; IPL= Intra Parietal Lobule; SMA= Supplementary Motor Area; L-SPL= Left Superior Parietal Lobule; L-MOG= Left Middle Occipital Gyrus; R-FFG= Right Fusiform Gyrus; R-ITG= Right Inferior Temporal Gyrus; PCG= Precentral Gyrus; MOG= Middle Occipital Gyrus; R-SFG= Right Superior Frontal Gyrus]

Figure 6.5: Activation with sequential presentation

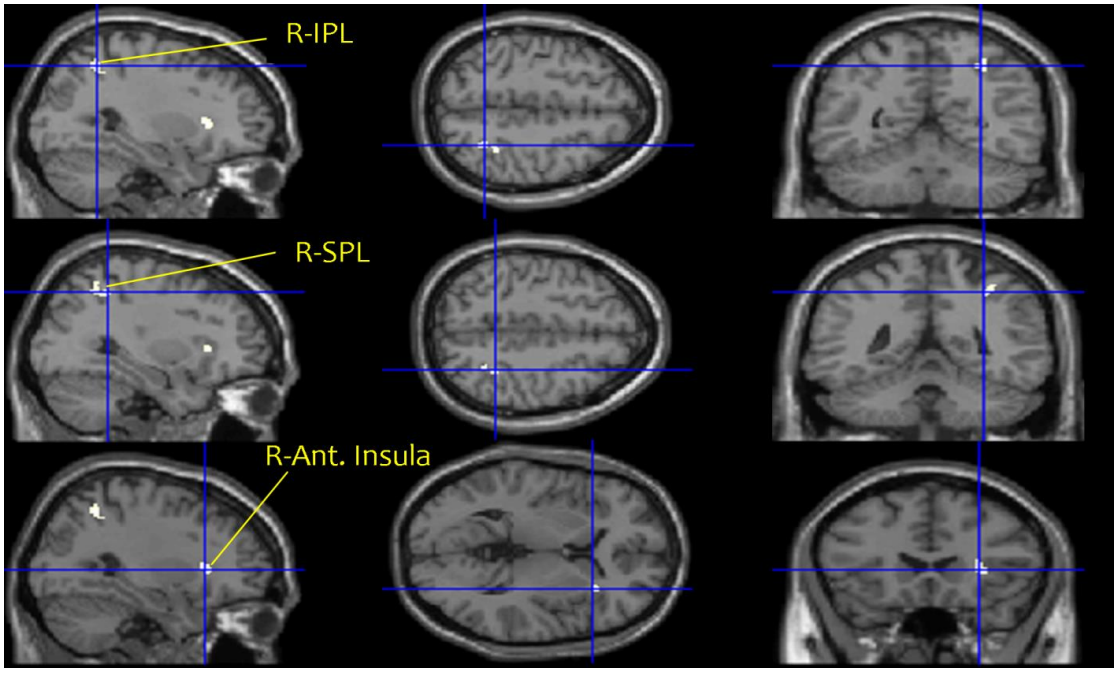
Brain areas activated with unchanged locations [[SIMU-Base] ∩ [SEQU-Base]]

To obtain the common areas recruited with unchanged locations, conjunction analysis was carried out on simultaneous and sequential presentation conditions with unchanged locations. Major contributions from the right inferior parietal lobule, right superior parietal lobule, and the right anterior insula were observed. The coordinates of these significant areas of activation are given in Table 6.4 and depicted in Figure 6.6.

Table 6.4: Activation with unchanged locations [[SIMU-Base] ∩ [SEQU-Base]]

Cluster	L/R	BA	Anatomical area	SPM{Z}	X	y	z	No. of Voxels
1	R	7	Inferior Parietal Lobule	5.94	+30	-54	+50	59
		7	Inferior Parietal Lobule	5.82	+36	-44	+52	
		7	Superior Parietal Lobule	5.74	+32	-46	+48	
2	R	13	Anterior Insula	5.91	+30	+22	+4	30

All regions are significant at p<.01 [FWE corrected] and k>20



[Activated areas are shown in sagittal, axial, and coronal planes from left to right. R-IPL = Right Inferior Parietal Lobule; R-SPL = Right Superior Parietal Lobule; R-Ant. Insula = Right Anterior Insula]

Figure 6.6: Activation with unchanged locations

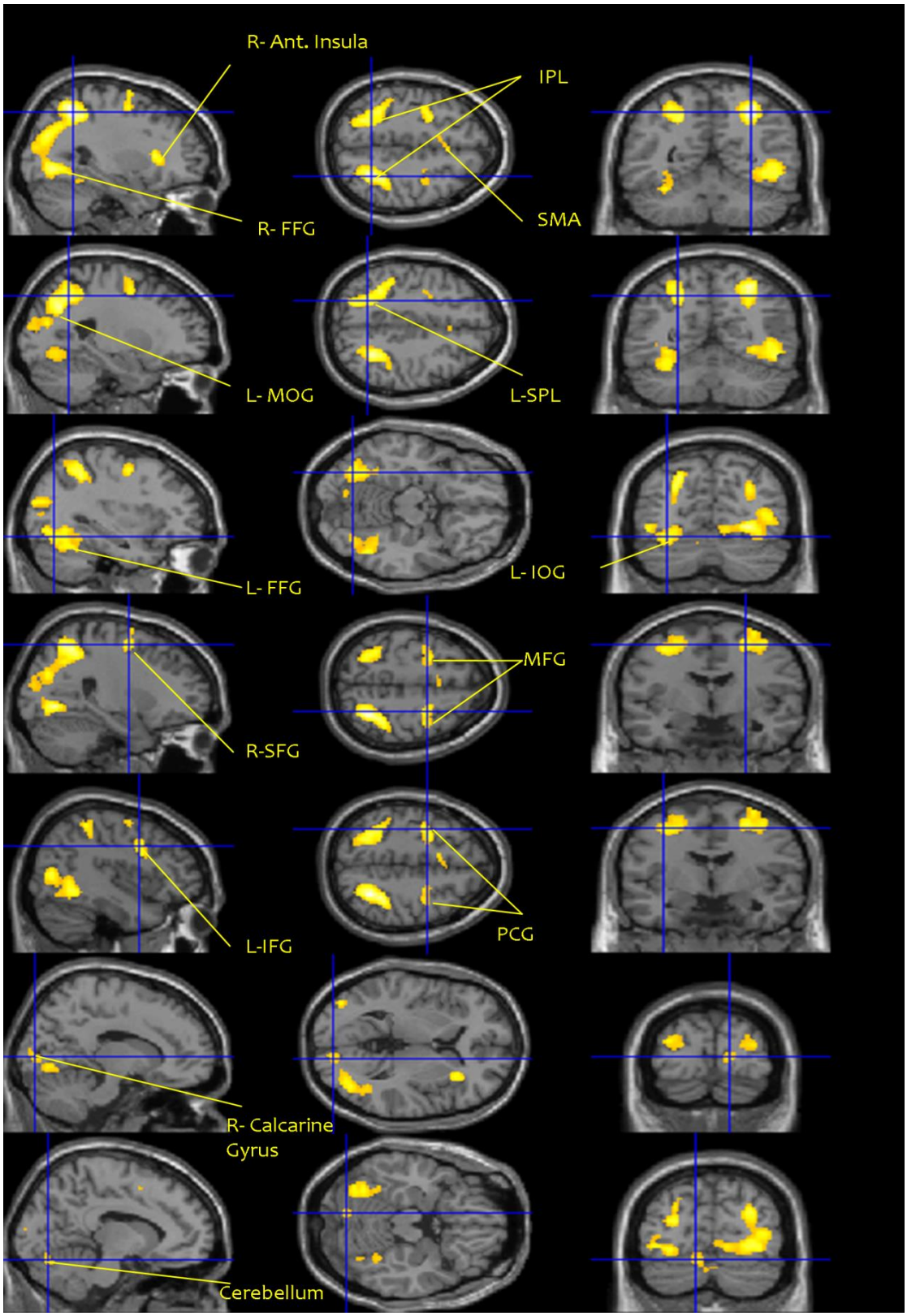
Brain areas activated with random locations [[SIMR-Base] ∩ [SEQR-Base]]

To obtain the common areas recruited with random locations, conjunction analysis was carried out on simultaneous and sequential presentation conditions with random locations. Significant activation was seen in bilateral occipital and parietal regions. In addition, almost the whole frontal cortex in the right hemisphere as well as the middle frontal cortex in the left hemisphere were activated. Relatively smaller clusters in right anterior insula, right calcarine gyrus, left supplementary motor area, and left cerebellum were also observed. The coordinates of these areas of activation are shown in Table 6.5 and depicted in Figure 6.7.

Table 6.5: Activation with random locations [[SIMR-Base] ∩ [SEQR-Base]]

Cluster	L/R	BA	Anatomical area	SPM{z}	x	y	z	No. of Voxels
	R	7	Inferior parietal lobule	10.07	30	-54	50	
	R	7	Inferior parietal lobule	8.74	32	-46	50	
1	R	19	Fusiform gyrus	8.60	26	-70	-4	3576
	L	7	Middle occipital gyrus	8.87	-24	-66	36	
	L	7	Superior parietal lobule	8.50	-24	-58	46	
2	L	7	Inferior parietal lobule	8.94	-34	-46	46	1624
	L	19	Inferior occipital Gyrus	7.23	-36	-78	-6	
	L	19	Fusiform gyrus	7.17	-32	-70	-14	
	L	19	Inferior occipital Gyrus	7.11	-46	-72	-6	
3	L	37	Fusiform gyrus	7.09	-32	-60	-16	1116
	R	6	Superior frontal gyrus	7.56	26	-4	56	
	R	6	Superior frontal gyrus	6.99	32	-2	70	
4	R	6	Middle frontal gyrus	6.72	38	-4	58	327
	L	6	Middle frontal gyrus	7.49	-28	-2	50	
5	L	6	Precentral gyrus	7.31	-34	-4	52	284
	R	44	Inferior frontal gyrus	7.01	42	8	28	
	R	8	Precentral gyrus	6.91	42	6	34	
6	R	8	Middle frontal gyrus	6.74	42	6	38	220
7	R	8	Anterior Insula	7.47	30	20	8	157
8	R	17	Calcarine gyrus	6.36	14	-88	4	95
9	L		Cerebellum	6.41	-10	-76	-18	92
10	L	6	Supplementary Motor Area	6.23	-8	6	52	78

All regions are significant at p<.01 [FWE corrected] and k>20



[Activated areas are shown in sagittal, axial, and coronal planes from left to right. R-Ant. Insula = Right Anterior Insula; R-FFG= Right Fusiform Gyrus; IPL= Intra Parietal Lobule; SMA= Supplementary Motor Area; L-MOG= Left Middle Occipital Gyrus; L-SPL= Left Superior Parietal Lobule; L-FFG= Left Fusiform Gyrus; L-IOG= Left Inferior Occipital Gyrus; R-SFG= Right Superior Frontal Gyrus; MFG= Middle Frontal Gyrus; L-IFG= Left Inferior Frontal Gyrus; PCG= Precentral Gyrus; R-Calcarine Gyrus= Right Calcarine Gyrus]

Figure 6.7: Activation with random locations

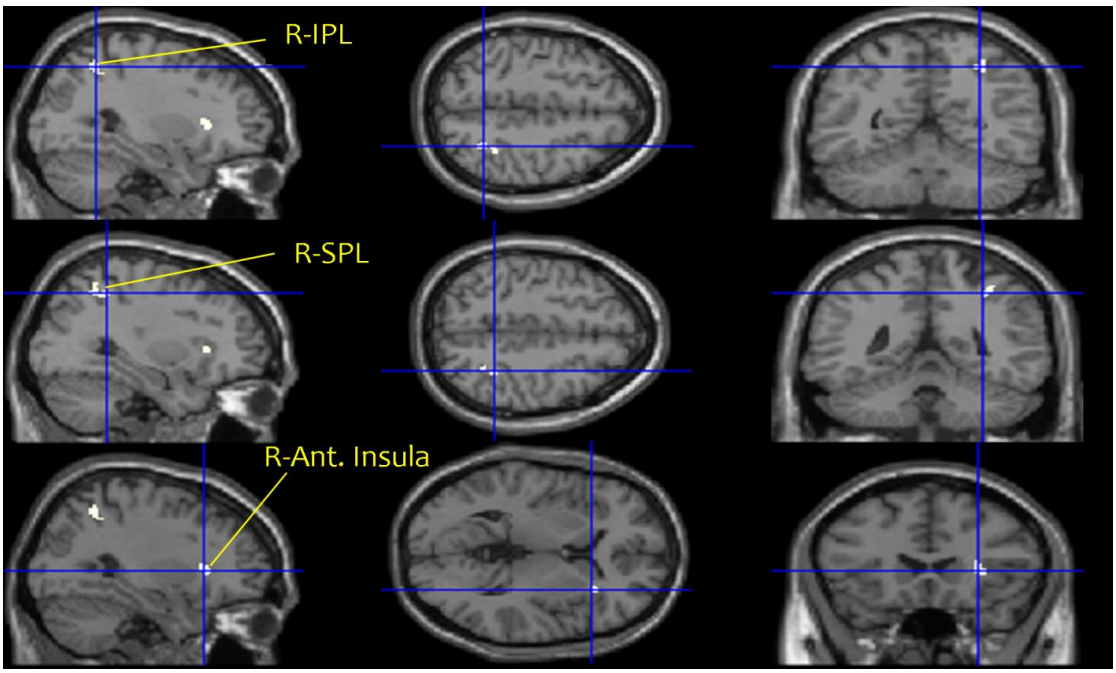
Brain areas activated in all four conditions [[SIMU-Base] ∩ [SIMR-Base] ∩ [SEQU-Base] ∩ [SEQR-Base]]

To obtain the brain regions active commonly in all four conditions, conjunction analysis was carried out on the contrasts of all four conditions to the baseline. The right inferior parietal lobule, right superior parietal lobule, and right anterior insula showed significant brain activity. The coordinates are shown in Table 6.6, and depicted in Figure 6.8. Notice that these are the same areas that show activation with simultaneous presentation, and with unchanged locations.

Table 6.6: Activation in all four conditions [[SIMU-Base] ∩ [SIMR-Base] ∩ [SEQU-Base] ∩ [SEQR-Base]]

Cluster	L/R	BA	Anatomical region	SPM{Z}	X	y	z	No. of Voxels
1	R	7	Inferior Parietal Lobule	5.94	+30	-54	+50	59
		7	Inferior Parietal Lobule	5.82	+36	-44	+52	
		7	Superior Parietal Lobule	5.74	+32	-46	+48	
2	R	13	Anterior Insula	5.91	+30	+22	+4	30

All regions are significant at p<.01 [FWE corrected] and k>20



[Activated areas are shown in sagittal, axial, and coronal planes from left to right. R-IPL = Right Inferior Parietal Lobule; R-SPL = Right Superior Parietal Lobule; R-Ant. Insula = Right Anterior Insula]

Figure 6.8: Activation common to all four conditions

In a nutshell, the results of conjunction null analyses revealed that exactly the same areas were commonly activated with simultaneous presentation and with unchanged locations [and in all four experimental conditions]. Many more [and different] areas show activation with random locations and with sequential presentation.

ROI analysis

Activation was searched in ten ROIs defined on the basis of previous studies of feature binding as mentioned in Table 6.1. Several studies of feature binding have confirmed the important role of the parietal regions in feature binding [e.g., Coull *et al.*, 2003; Shafritz *et al.*, 2002; Song and Jiang, 2006; Todd and Marois [2004]; Xu [2007]. However, these different studies had reported slightly different coordinates within this region, and so the bilateral parietal cortex was searched at several different coordinates. Thus, although the main effects and interactions were tested only in the bilateral parietal regions, left precentral gyrus, right fusiform gyrus, left anterior hippocampus, and the left supra marginal gyrus, actually ten different coordinates were searched as listed in Table 6.1. A 2×2 repeated measures *ANOVA* [mode of presentation × locations] was conducted on mean parameter estimates for each ROI. All significant results in the data for the different regions of interest are reported in Figure 6.9.

R-intra parietal cortex

Repeated measures *ANOVA* on mean parameter estimates from the R-intra parietal cortex, defined on the basis of Shafritz *et al.* [2002], showed a significant main effect of locations, F[1,17]=4.979, MSE=.256, p<.039, *partial* η^2=.227, BF_{10}=1.672, with higher activation in the random locations condition [M=1.181, SD=.593] than the unchanged locations condition [M=.915, SD= .763]. The main effect of mode of presentation was not significant. However, there was a significant interaction, F[1,17]=5.412, MSE=.268, p<.033, *partial* η^2=.241, BF_{10}=2.637 with the difference between unchanged and random location being larger with simultaneous presentation [t[17]= 2.840, p<.01, d=.670, BF_{10}= 4.782] as compared to sequential presentation [t[17]=.122, p<.904 ns, d= .029, BF_{01}=4.081]. The model comprising both main effects and the interaction effect [BF_{10}= 1.063] was compared with a model comprising only the main effects [BF_{10}=.403]. The model comprising main effects and interaction effect better fit the data by a factor of 2.637:1. In fact, maximum activity is shown in the condition with simultaneous presentation and random locations, which is significantly different from all other conditions, where activation levels are not significantly different from each other.

R-superior parietal cortex

Repeated measures *ANOVA* on mean parameter estimates in the ROI defined on the basis of Shafritz *et al.* [2002] showed a significant main effect of locations, F[1,17]=23.989, MSE=.466, p<.001, *partial* η^2=.585, BF_{10}=353.177, with higher activation in the random locations condition [M=1.505, SD= 1.224] than the unchanged locations condition [M=.717, SD= 1.123]. Neither the main effect of mode of presentation and nor its interaction with locations was significant. The model comprising both main effects and the interaction effect [BF_{10}=107.121] was compared with a model comprising only the main effects [BF_{10}= 80.496]. The model comprising main effects and interaction effect better fit the data by a factor of 1.33:1.

R-superior parietal lobule

Repeated measures *ANOVA* was carried out on mean parameter estimates in the right superior parietal lobule at the coordinates defined on the basis of Song and Jiang [2006]. Results showed a main effect of locations, F[1,17]=15.637, MSE=.415, p<.001, *partial* η^2=.479, BF_{10}=204.75, with higher activation in the random locations condition [M=1.628, SD= 1.014], than the unchanged locations condition [M=1.028, SD= .811]. The main effect of mode of presentation was not significant. However, there was a significant interaction, F[1,17]=7.748, MSE=.321, p<.013, *partial* η^2=.313, BF_{10}= 5.815, with the difference between unchanged and random locations being larger with simultaneous presentation [t[17]= 4.536, p<.001, d=1.069, BF_{10}= 109.374] as compared to sequential presentation [t[17]=1.207, p<.244 ns, d=.285, BF_{01}=2.192]. The model comprising both main effects and the interaction effect [BF_{10}= 326.101] was compared with a model comprising only the main effects [BF_{10}=56.071]. The model comprising the main effects and interaction effect better fit the data by a factor of 5.815:1.

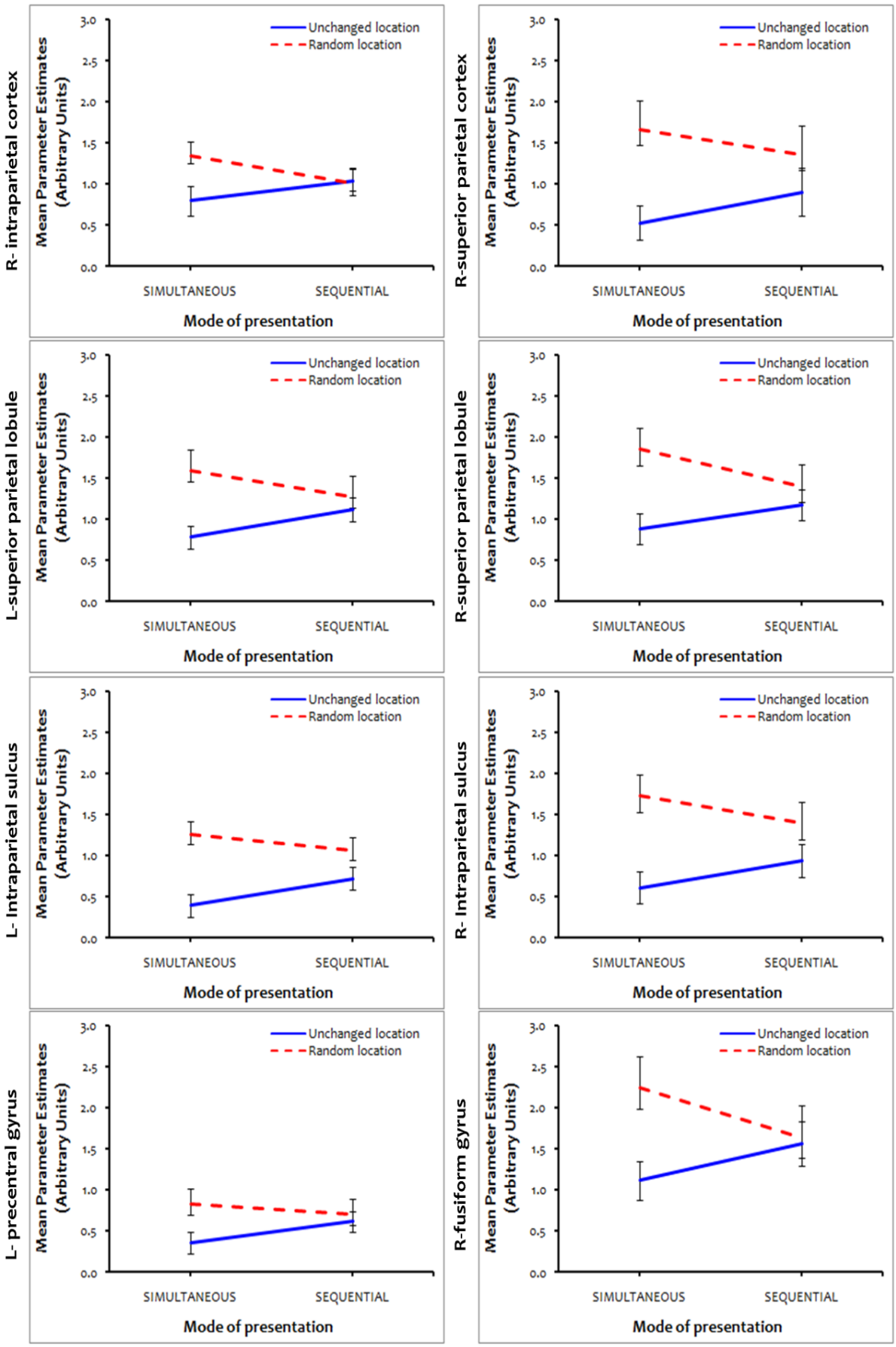


Figure 6.9: Mean parameter estimates in ROIs showing significant effects

L-superior parietal lobule

Repeated measures *ANOVA* on mean parameter estimates showed a significant main effect of locations, F[1,17]=9.671, MSE=.438, p<.006, *partial* η^2=.363, BF_{10}= 34.226 with higher activation in the random locations condition [M=1.435, SD= .880] than the unchanged locations condition [M=.949, SD= .650]. The main effect of mode of presentation was not significant. However, there was a significant interaction, F[1,17]=5.876, MSE=.332, p<.027, *partial* η^2=.257, BF_{10}=3.707 with the difference between unchanged and random locations being larger with simultaneous presentation [t[17]= 3.049, p<.007, d= .719, BF_{10}= 6.910] as compared to sequential presentation [t[17]=1.304, p<.209 ns, d= .308, BF_{01}=1.984]. The model comprising both main effects and the interaction effect [BF_{10}= 30.820] was compared with a model comprising only the main effects [BF_{10}=8.313]. The model comprising main effects and interaction effect better fit the data by a factor of 3.707:1.

L-intra parietal sulcus

Repeated measures *ANOVA* on mean parameter estimates showed a significant main effect of locations, F[1,17]=17.076, MSE=.382, p<.001, *partial* η^2=.501, BF_{10}= 1498.968 with higher activation in the random locations condition [M=1.161, SD= .612] than the unchanged locations condition [M=.559, SD= .613]. The main effect of mode of presentation was not significant. However, there was a significant interaction, F[1,17]=4.433, MSE=.282, p<.050, *partial* η^2=.207, BF_{10}=1.763, with the difference between unchanged and random locations being larger with simultaneous presentation [t[17]= 4.131, p<.006, d=.974, BF_{10}= 51.146] as compared to sequential presentation [t[17]=1.957, p< .067 ns, d=.461, BF_{10}=1.151]. The model comprising both main effects and the interaction effect [BF_{10}= 709.904] was compared with the model comprising only the main effects [BF_{10}=402.544]. The model comprising the main effects and interaction effect better fit the data by a factor of 1.763:1.

R-intra parietal sulcus

Repeated measures *ANOVA* on mean parameter estimates showed a significant main effect of locations, F[1,17]=18.662, MSE=.589, p<.001, *partial* η^2=.523, BF_{10}=1723.461, with higher activation in the random locations condition [M=1.557, SD= 1.026] than the unchanged locations condition [M=.776, SD= .862]. The main effect of mode of presentation was not significant. However, there was a significant interaction, F[1,17]=5.686, MSE=.346, p<.029, *partial* η^2=.251, BF_{10}=1.737 with the difference between unchanged and random locations being larger with simultaneous presentation [t[17]=4.605, p<.001, d=1.086, BF_{10}=124.635] as compared to sequential presentation [t[17]=2.109, p< .049, d=.497, BF_{10}=1.443]. The model comprising both main effects and the interaction effect [BF_{10}=725.024] was compared with a model comprising only the main effects [BF_{10}=417.237]. The model comprising the main effects and interaction effect better fit the data by a factor of 1.737:1.

L-precentral gyrus

Repeated measures *ANOVA* on mean parameter estimates at the coordinates given by Mitchell *et al.* [2000] which were MNI [-54, -6, 36] did not yield any significant result. However, when the L-precentral gyrus was searched using the coordinates that emerged significant in the conjunction null analyses, the results were fruitful. None of the main effects emerged significant. But there was a significant interaction effect, F[1,17]=7.127, MSE=.489, p<.016, *partial* η^2=.295, BF_{10}=2.257 with a larger difference between unchanged and random locations observed with simultaneous presentation [t[17]= 2.480, p<.02, d=585, BF_{10}=2.593] than with sequential presentation [t[17]=.375, p<.712 ns, d=.088, BF_{01}=3.861]. The model comprising both main effects and the interaction effect [BF_{10}= .745] was compared with the model comprising only the main effects [BF_{10}=.330]. The model comprising main effects and interaction effect better fit the data by a factor of 2.257:1. The least activation was observed in the simultaneous presentation

condition with unchanged locations, which is significantly different from the other three conditions, which do not differ among themselves in activation levels.

R-fusiform gyrus

Repeated measures *ANOVA* on mean parameter estimates showed a significant main effect of locations, $F[1,17]=5.895$, $MSE=1.088$, $p<.027$, *partial* $\eta^2=.257$, $BF_{10}= 5.799$ with higher activation in the random locations condition [M=1.937, SD= 1.420] than the unchanged locations condition [M=1.340, SD= 1.101]. The main effect of mode of presentation was not significant. However, there was a significant interaction, $F[1,17]=9.660$, $MSE=.515$, $p<.006$, *partial* $\eta^2=.362$, $BF_{10}= 3.768$, with the difference between unchanged and random locations being larger with simultaneous presentation [$t[17]= 3.480$, $p<.002$, $d=.822$, $BF_{10}= 15.340$] as compared to sequential presentation [$t[17]=.122$, $p<.796$ ns, $d=.062$, $BF_{01}=3.984$]. The model comprising both main effects and the interaction effect [$BF_{10}= 5.815$] was compared with a model comprising only the main effects [$BF_{10}=1.543$]. The model comprising the main effects and interaction effect better fit the data by a factor of 3.768:1.

L-anterior hippocampus

Repeated measures *ANOVA* on mean parameter estimates showed no significant effects in the L-anterior hippocampus. However the main effect of locations showed a trend toward significance, $F[1,17]=3.906$, $MSE=.075$, $p<.065$, *partial* $\eta^2=.187$, $BF_{10}= 3.93$. The model comprising both main effects and the interaction effect [$BF_{10}= .312$] was compared with a model comprising only the main effects [$BF_{10}=1.006$]. The model comprising only the main effects better fit the data by a factor of 3.224:1.

L-supra marginal gyrus

Repeated measures *ANOVA* on mean parameter estimates showed no significant effects in the L-supra marginal gyrus. Bayesian analysis also supported the null hypothesis. For the interaction, the model comprising both main effects and the interaction effect [$BF_{10}= .065$] was compared with a model comprising only the main effects [$BF_{10}=.187$]. The model comprising only the main effects better fit the data by a factor of 2.876:1.

To recapitulate the results of the ROI analyses, the main effect of locations as well as its interaction with mode of presentation was significant in the bilateral intra-parietal sulci, bilateral superior parietal regions, and the right fusiform area. In addition, the main effect of location was also significant in the right superior parietal cortex around the coordinates [15, -62, 63] defined on the basis of Shafritz *et al.* [2002]. However, the interaction effect was not significant here. In contrast, the left precentral gyrus, showed only a significant interaction effect, despite none of the main effects being significant.

6.4 DISCUSSION

The fMRI experiment was carried out to study the underlying activation in the brain as the participant performed the binding change detection task in the four different experimental conditions resulting from orthogonally manipulating locations and mode of presentation.

Conjunction null analyses revealed the areas commonly active in all the conditions under study. It showed the contribution of right inferior and superior parietal lobules, and the anterior insula. This accords well with several previous studies which have zeroed in on the parietal cortex as being crucial for feature binding [e.g., Braet and Humphreys, 2009; Friedman-Hill *et al.*, 1995; Shafritz *et al.*, 2002;]. The insula is also associated with binding, awareness, and consciousness [Bushara et al, 2003; Craig, 2009; Sterzer and Kleinschmidt, 2010]

When activations associated with simultaneous presentation and with sequential presentation conditions were assessed, the relatively larger clusters of activation with sequential

presentation are strikingly apparent. With simultaneous presentation, activation is observed only in the right inferior and superior parietal lobules and the anterior insula. However, activation is larger with sequentially presented stimuli in intensity as well as spread, the areas recruited in this condition being the bilateral parietal lobules, bilateral fusiform gyri, bilateral middle occipital gyri, bilateral supplementary motor areas, right inferior and superior frontal gyri, right inferior temporal gyrus, left precentral gyrus, and left cerebellum.

The conjunction null analysis for random locations also caused very high BOLD responses across the brain. The largest clusters spread from the parietal to the occipital areas, in both hemispheres. Random locations also recruited areas from the frontal cortex [i.e. bilateral middle frontal gyrus, precentral gyrus, right superior and inferior frontal gyri]. Activation was also observed in the right insula and calcarine gyrus, and the left cerebellum and supplementary motor area. Such large clusters of activation may indicate that the random locations condition was highly resource demanding in both the presentation modes. In contrast, the conjunction null analyses for unchanged locations caused comparatively lower BOLD responses, the largest cluster being only 59 voxels. The activation in unchanged locations condition is in the right inferior and superior parietal lobules and the right anterior insula.

It is noteworthy that activation was noted in exactly the same areas in unchanged locations as well as the simultaneous presentation condition, substantiating the confound and/or convergence between simultaneous presentation and location information. This suggests that whenever stimuli are presented simultaneously, the participants process stimuli in terms of information regarding relative locations.

In contrast, it is interesting to note the difference in the areas activated with random locations and sequential presentation. Although, both show large areas of activation, the specific regions involved are quite different. Most notably, the right inferior temporal gyrus is activated only with sequential presentation. The temporal gyrus is related to memory for visual objects, more precisely the recognition/ recall of object identity. It is the final location of the ventral cortical visual system. It perceives and processes visual stimuli amplified in the V1, V2, V3, and V4 regions of the occipital lobes, identifying the object on the basis of color and form and comparing that incoming information to stored memories of objects to identify that object. This allows the inference that stimuli are primarily processed as objects when sequentially presented.

Collectively, the conjunction null analyses show that random locations recruit the largest number of areas with high intensity. The activated areas are exactly the same [and the least in number and extent] with simultaneous presentation and with unchanged locations, with only the parietal cortex and anterior insula being involved. It is clear that simultaneous mode of presentation, and unchanged locations, recruit very few brain areas and are less demanding of brain resources. The right inferior temporal gyrus related to memory for objects is involved only in the sequential presentation condition. The next chapter will discuss these results in the context of other results and theoretical considerations.

The main aim of conducting the experiment using fMRI was to find the differences in brain activation for simultaneous and sequentially presented stimuli when locations were unchanged and random from study to test in the ROIs associated with binding. One expected some overlap, but also differences in the areas recruited by the different experimental conditions. More importantly, the research tested for the differential intensity of activation in predefined ROIs.

Repeated measures *ANOVA* revealed a significant main effect of locations in the bilateral parietal regions and right fusiform gyrus, with higher activation in all these areas in the random locations condition. Activation in the bilateral parietal areas probably denotes spatial as well as the top down object based attention [Bettencourt and Xu, 2016; Bressler and Silver, 2010; Haxby

et al., 1991, Kastner and Ungerleider, 2000], both of which are clearly required more in the random locations condition.

Another point to note is that all parietal regions show a significant interaction in this experiment, except the right superior parietal cortex, which shows only the main effect of locations, not an interaction. The coordinates for this area were taken from Shafritz *et al.* [2002], who found the area to be activated during simultaneous presentation, but not sequential presentation. It may be recalled that Shafritz *et al.* [2002] presented their stimuli in a single central location in the sequential presentation condition, whereas stimuli were presented in several different locations in the simultaneous presentation condition. Thus, their comparison of mode of presentation was confounded by a comparison of single vs. multiple locations. In the present experiment an attempt was made to unravel this confound by orthogonally manipulating mode of presentation and locations. The present results clearly show that the right superior parietal cortex shows a difference in activation due to manipulation of locations but not mode of presentation. One may surmise that the results of Shafritz *et al.* [2002] manifest the effect of locations rather than mode of presentation.

The activation in the fusiform gyrus resonates with earlier studies finding activation in this area using color-shape binding [Parra *et al.*, 2014; Song and Jiang, 2006]. The fusiform gyrus is associated with object processing [review by Grill-Spector *et al.*, 2001]. The greater activation of the fusiform gyrus in the random locations condition suggests that the participants might be focusing on a few [or single] object in this condition as they cannot rely on the spatiotopic representation of the study display to find changes in binding. With unchanged locations, participants can just match the pattern of stimuli at study and test to figure out if any stimulus has changed color-shape binding. They do not need to focus on every single stimulus object, hence the fusiform gyrus is less activated in this condition.

The main effect of mode of presentation did not emerge significant in any of the ROIs. However, a significant interaction effect was found in seven out of the ten ROIs investigated in this research. [All these areas also showed a significant main effect of locations, except the left precentral gyrus, which showed only a significant interaction].

The right intra parietal cortex shows maximum activation in the condition with simultaneous presentation and random locations, which in turn, is significantly higher than the other three experimental conditions, which do not differ among themselves. It is notable that this area is associated with decisions about conjunctions rather than single feature stimuli, but only when many stimuli are presented together [Shafritz *et al.*, 2002]. In general, the parietal cortex is associated with allocation of attention to visual locations [Corbetta, 1998] and is therefore perhaps maximally active in the simultaneous presentation random locations condition because it is this condition, which requires search for an object in various locations in the visual space, an object which is different from the ones in the mental representation.

The bilateral superior parietal lobules, associated with binding in several studies. [e.g., Coull *et al.*, 2003; Shafritz *et al.*, 2002; Song and Jiang, 2006] also show a significant interaction with the difference between random and unchanged locations being larger with simultaneous presentation [favouring random locations], but not being significant with sequential presentation. Perhaps this is because the competition between various objects and resultant confusion is much more in the random location condition with simultaneous presentation. Song and Jiang [2006] reported increased activity in bilateral superior parietal lobules with increasing set size for both binding and single feature conditions, presumably because increasing set size also increases competition and confusion.

The bilateral intra parietal sulci also show significant interaction effects. Some researchers have related the intra parietal sulcus to distracter suppression [Chun and Marois, 2002; Marois *et al.*, 2000] while still others have linked it to attentional control and enhancement

[Hahn *et al.*, 2006; Painter *et al.*, 2015]. In the present experiment, distracter suppression as well as focus on the target is required to the maximum possible extent in the condition with random locations and simultaneous presentation and hence significantly higher activity in the right intra parietal cortex is obtained in this condition. In contrast, the stimuli in the condition with unchanged locations and simultaneous presentation can easily be encoded as a pattern or configuration and neither require a focus on the target nor suppression of distracters. Indeed, the presence of distracters allows the relational encoding. Hence, the least activity in the bilateral intra-parietal sulci is found in this condition.

The left precentral gyrus shows the lowest activity with unchanged locations in the simultaneous presentation condition, with similar activation in the rest of the three conditions. To reiterate, the simultaneous presentation with unchanged locations is the only condition in which the participants could do the task without shifting the focus of attention. Indeed, they can rely on a spatiotopic representation of all the stimuli. In the random locations condition, the participants have to search for the object, which has changed its binding, in an array of objects. This entails shifting the focus of attention from one place or object to another. The left precentral gyrus has been associated with a shifting focus of attention in several studies [e.g., Fan *et al.*, 2005; Hopfinger *et al.*, 2000]. The sequential condition also requires shifting the focus of attention during encoding, as stimuli are presented one by one in multiple locations. This is true for unchanged locations as well as random locations in this mode of presentation. Mitchell *et al.* [2000] found activity in the precentral gyrus in their sequential binding task, which used multiple locations. Beauchamp *et al.* [2001] found that the precentral gyrus activated with both, covert and overt shifts of attention.

The right fusiform gyrus also shows a significant interaction. The fusiform gyrus is least active in the unchanged locations condition with simultaneous presentation, and maximally active in the random locations condition, with activity in both the sequential presentation conditions being in-between and similar to each other. Schoenfeld *et al.* [2003] found increased activation in the color-selective region of the fusiform gyrus even when color was irrelevant to the task. It seems an inferential/ conceptual leap, but perhaps the fusiform gyrus shows increased activation if a feature is irrelevant, as is the case with the random locations condition in the present experiment.

In addition, the fusiform gyrus shows reduced activation to sequential stimuli [both unchanged and random] as compared to stimuli in the condition with simultaneous presentation and random locations. Reber *et al.* [2005] found direct priming effects in the fusiform gyrus and noted a reduction in the neural activity beyond the second presentation of the stimulus. Jiang, Haxby, *et al.* [2000] used a sequential face recognition task to find that reduced responses in the extra-striate visual cortex [fusiform gyrus] were associated with stimulus repetition. Henson *et al.* [2000] found that the right fusiform region showed reduced responses to repetition of familiar faces as well as symbols, but the left fusiform gyrus showed increased responses. When the stimuli were not familiar, there was increased activation in bilateral fusiform gyrus. Together, these studies suggest that repetition of stimuli leads to reduction in the activity of the fusiform gyrus. Stimuli in the sequential mode of presentation in the present experiment can be considered similar to repetitions as each one is sampled from the same limited pool, are quite similar to each other, and each has no particular meaningfulness for the participant. This may be the reason for the relatively reduced performance in this condition, as compared with the condition with simultaneous presentation and random locations. The fusiform gyrus shows the least activity in the condition with simultaneous presentation and unchanged locations, indicative of a different process operating in this condition, which probably does not recruit object-focused areas. As suggested before, performance in this condition probably relies on a spatiotopic representation which involves relatively automatic encoding of the stimuli as a pattern.

Thus, significant interaction effects in the bilateral parietal areas, left precentral gyrus, and the right fusiform, substantiate the assertion that location is a factor in processing simultaneously presented stimuli, but not sequentially presented ones. Overall, the orthogonal manipulation of locations and mode of presentation was successful in showing the different role of locations in simultaneous and sequential presentation at the behavioral as well as brain levels. The next chapter explains the importance of these results together with the behavioral outcomes noted in Experiments 1-5.