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Research Report
**Neural circuits of hierarchical visuo-spatial
sequence processing**
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ABSTRACT

Sequence processing has been investigated in a number of studies using serial reaction time tasks or simple artificial grammar tasks. Little, however, is known about higher-order sequence processing entailing the hierarchical organization of events. Here, we manipulated the regularities within sequentially occurring, non-linguistic visual symbols by applying two types of prediction rules. In one rule (the adjacent dependency rule), the sequences consisted of alternating items from two different categories. In the second rule (the hierarchical dependency rule), a hierarchical structure was generated using the same set of item types. Thus, predictions about non-adjacent elements were required for the latter rule. Functional Magnetic Resonance Imaging (fMRI) was used to investigate the neural correlates of the application of the two prediction rules. We found that the hierarchical dependency rule correlated with activity in the pre-supplementary motor area, and the head of the caudate nucleus. In addition, in a hypothesis-driven ROI analysis in Broca's area (BA 44), we found a significantly higher hemodynamic response to the hierarchical dependency rule than to the adjacent dependency rule. These results suggest that this neural network supports hierarchical sequencing, possibly contributing to the integration of sequential elements into higher-order structural events. Importantly, the findings suggest that Broca's area is also engaged in hierarchical sequencing in domains other than language.

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1. Introduction

Sequence processing is inherent in several cognitive domains. In the field of language, sequencing is necessary, e.g. for concatenating letters to words, words to phrases, and phrases to sentences. Similarly, sequential organization is also required in non-language domains such as goal-directed

behavior or action perception and prediction. Accordingly, experiments dealing with the sequential structure of stimuli are rooted in different research domains (cf. [Fiebach and Schubotz, 2006](#); [Schubotz and Fiebach, 2006](#)). Recently, efforts have been directed to the investigation of hierarchical sequence processing not only in the field of language, but also in non-language domains.

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Some language processing studies used fMRI to determine which cognitive processes were involved in the processing of sentences with different hierarchical structures. In these linguistic structures, the syntactic complexity was manipulated, while the meaning (semantics) of the sentences was kept constant. For example, the syntactic structure was varied by manipulating the hierarchical order of phrases within sentences, and this manipulation resulted in increased brain activity in Broca's area. It was suggested that Broca's area, among other brain regions, played an important role in parsing syntactically complex natural sentences (e.g. Roder et al., 2002; Santi and Grodzinsky, 2007).

Hierarchical sequencing was also examined in studies that used the serial reaction time task (Koch and Hoffmann, 2000), the serial prediction task (Schubotz and von Cramon, 2002) and in a study that applied abstract, hierarchically organized action plans (Koechlin and Jubault, 2006). Schubotz and von Cramon (2002) suggested that a premotor-parietal network plays a crucial role in predicting and planning in the motor domain. Importantly, this study did not differentiate between hierarchical and non-hierarchical sequences. In contrast, Koechlin and Jubault (2006) found Broca's area and its right hemispheric homologue to be sensitive to the start and the end states of hierarchically organized event sequences.

Recent PET and fMRI studies that investigated the learning and processing of artificial grammar (AG) rules addressed the question of explicit versus implicit rule learning (Fletcher et al., 1999; Forkstam et al., 2006; Peigneux et al., 2000; Seger et al., 2000; Skosnik et al., 2002) or chunk versus pattern learning (Lieberman et al., 2004). Most of these imaging studies, applied Reber's artificial grammar (Reber, 1967). Brain activations reported for the processing of Reber's artificial grammar varied as a function of the type of the design used, and of the type of the comparison conducted. Several different regions were reported to be involved in the learning of a Reber grammar. Most common activations were found in different parts of the left prefrontal cortex (Forkstam et al., 2006; Lieberman et al., 2004; Peigneux et al., 2000; Seger et al., 2000). Moreover, the basal ganglia were also engaged in artificial grammar learning. Activation in the caudate nucleus was reported by Peigneux et al. (2000), Lieberman et al. (2004), and Forkstam et al. (2006). Another group of studies working with artificial grammars used rules containing a more language-like structure than the Reber grammar (Bahlmann et al., 2008; Opitz and Friederici, 2003; Opitz and Friederici, 2004). Opitz and Friederici (2003) found that learning of the artificial language came along with activation in the hippocampal formation in the early learning stage; the left inferior frontal gyrus was engaged during the later part of the task.

A growing number of behavioral and fMRI studies have been published which have focused on the processing of hierarchical structure (Bahlmann et al., 2008; de Vries et al., 2008; Friederici et al., 2006; Gentner et al., 2006; Opitz and Friederici, 2007; Perruchet and Rey, 2005). The hierarchical stimulus structure employed in these experiments comprised center-embedded stimuli also present in natural sentences (e.g. [the football player [who scored the goal] was praised]). Studies on hierarchical sequence processing suggest that Broca's area plays an important role whenever language-related stimuli (e.g. letters, syllables, words, or sentences) are

to be processed. Beyond this, in a theoretical article, Fiebach and Schubotz (2006) recently proposed that Broca's area plays a crucial role in hierarchical sequence processing, independent of the cognitive domain. In a similar vein, based on their fMRI data, Koechlin and Jubault (2006) suggested that Broca's area is involved in hierarchical sequence processing in non-language domains, for example, the executive control of the hierarchical organization of goal-directed behavior.

In a recent fMRI study, we used two AG rules that varied in complexity in an fMRI setting (Bahlmann et al., 2008). The 'lexicon' of this language consisted of consonant-vowel syllables. The less complex structure followed the rule $(AB)^n$ and employed an adjacent dependency rule. It involved the generation of sequences of alternating, adjacent category pairs. The more complex structure was coded as A^nB^n . That is, sequence structuring was governed by a hierarchical dependency rule, since individual items related to each other according to non-adjacent, hierarchically organized sequences of stimuli. The direct comparison of hierarchical versus adjacent dependency syllable sequences revealed activity in a network comprising Broca's area, the ventral premotor cortex (BA 44/6) and other cortical and sub-cortical areas.

In the present study we aimed to identify brain regions involved in hierarchical sequence processing in the visuo-spatial domain. We applied a hierarchical and a non-hierarchical AG to non-linguistic stimuli (see Fig. 1). These materials consisted of abstract visual stimuli that were characterized by their spatial orientation and surface pattern. Importantly, these stimuli were considered to be novel because they were not associated with a linguistic label or any conceptual knowledge (cf. Koester and Prinz, 2007). If Broca's area is engaged in domain-general hierarchical sequencing, we expect this region to also be activated for hierarchical structures in an AG using non-linguistic stimuli.

2. Results

2.1. Behavioral results

In the present study two different sequencing rules (factor RULE) were applied to abstract visual stimuli (see Fig. 1). The sequences consisted of four or six stimuli (factor LENGTH) and were grammatical or ungrammatical (factor GRAMM). Participants entered the scanner after they successfully learned the two rule types. Each participant learned and processed both rules in two separate fMRI sessions, with one week delay between the sessions. A grammaticality judgment task was applied.

An ANOVA on errors rates was conducted with the factors RULE (adjacent dependency, hierarchical dependency rule), LENGTH (short sequences, long sequences), and GRAMM (grammatical sequences, ungrammatical sequences; see the Experimental procedures section for a detailed description of the factors). A significant main effect of RULE was found [$F(1,14)=12.7, p<.01$], indicating that the hierarchical dependency rule induced more errors (5.9%, $SD=6.1$) than the adjacent dependency rule (1.4%, $SD=2.5$; see Fig. 2). The interaction between RULE and GRAMM also reached significance [$F(1,14)=12.0, p<.01$].

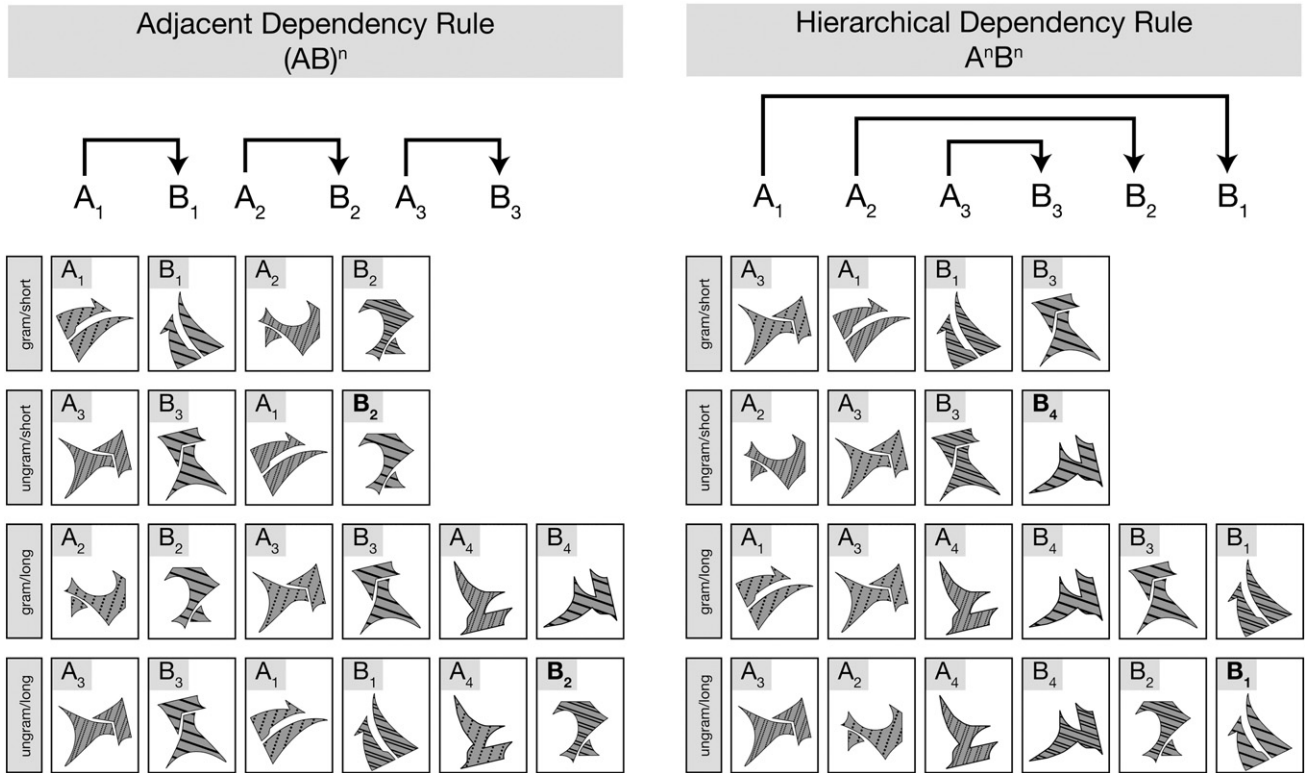


Fig. 1 – Description of the rules used. Adjacent dependency rule and hierarchical dependency rule with examples of abstract symbol sequences. The adjacent dependency rule involves the processing of local transitions between single elements. By contrast, in the hierarchical dependency rule, long-distance dependencies between non-adjacent elements must be processed. Note that during the experiment, each element was presented separately on the screen. Examples are provided for grammatical short (gram/short), ungrammatical short (ungr/short), grammatical long (gram/long), and ungrammatical long (ungr/long) sequences. Violated sequences in the example are marked with bold letters.

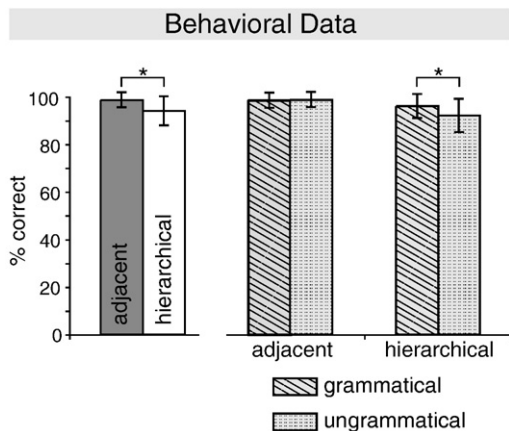


Fig. 2 – Behavioral data. Response accuracy for the hierarchical and adjacent dependency rule sequences. Left bars: Slightly more errors (5.9%) were observed during the processing of items governed by the hierarchical dependency (white) compared with the processing of the items governed by the adjacent dependency rule (gray, 1.4%). Middle and right bars: Slightly more errors were observed when rejecting ungrammatical sequences than when confirming correct grammatical sequences (but only for items governed by the hierarchical dependency rule).

The step down analysis revealed no difference between grammatical and ungrammatical items for the adjacent dependency rule (grammatical: 1.5%, SD=2.3; ungrammatical: 1.4%, SD=2.6). In contrast, in the hierarchical dependency rule, more errors were made when rejecting ungrammatical items (7.9%, SD=6.7) than when accepting grammatical items (4.0%, SD=4.7). An additional analysis for the violation type (Category-violation: e.g. A₂A₃B₃A₄; Index-violation: e.g. A₁A₂B₂B₃) was performed to test for possible performance differences between the two types of classification. An ANOVA with the factors RULE (adjacent dependency, hierarchical dependency rule), LENGTH (short sequences, long sequences), and VIOLATION TYPE (Category-violation, Index-violation) was performed. As a result, no main effect or any interactions were found, suggesting no differences in performance between the two types of classifications. Due to the delayed response mode used in the present study, reaction times were not analyzed.

2.2. fMRI results

The resting contrast in the adjacent dependency rule condition (collapsed across subconditions) revealed a network of prefrontal, parietal, and occipital brain regions. Significantly increased activity was found in the right ventrolateral premotor cortex and the adjacent inferior frontal junction,

the bilateral cuneus, the bilateral fusiform gyrus, and the right anterior insula. The resting contrast in the hierarchical dependency rule (collapsed across subconditions) also showed significant activation in the right ventrolateral premotor cortex, the bilateral precuneus, and the bilateral fusiform gyrus. However, additional activation was found in the pre-supplementary motor area (pre-SMA), the anterior cingulate cortex (ACC), the left premotor cortex, and the postcentral gyrus (see Table 1 and Fig. 3).

The main effect of RULE (hierarchical>adjacent dependency) was significant with an increased hemodynamic response in the right pre-SMA, the left middle pre-central sulcus (M1) and the head of the right caudate nucleus (see Fig. 4). In a meta-analysis, Fox et al. (2001) classified the mouth region to be located at the coordinates: $-47 (\pm 5.2) -13 (\pm 5.2) 36 (\pm 5.5)$. Thus, the activity in the left middle pre-central sulcus ($-50 -8 33$) corresponds to the human M1-mouth representation. No significant activity was found for the opposite contrast (adjacent>hierarchical dependency). For the main effect of GRAMM (ungrammatical>grammatical and vice versa) and the main effect of LENGTH (long>short and vice versa), no activity was found.

Table 1 – Anatomical areas, approximate Brodmann's Area (BA), mean Talairach coordinates (x, y, z), maximal Z values of the significant activations.

	Brain region	BA	x	y	z	Z _{max}
(A)	Adjacent dependency rule>baseline					
	R ventrolateral premotor C	6	43	1	27	4.38
	R precuneus	7	22	-65	45	3.71
	L precuneus	7	-26	-56	54	4.28
	R fusiform G	19/37	31	-74	-6	4.94
	L fusiform G	19/37	-35	-56	-12	4.73
	R middle occipital G	19	-26	-87	9	4.19
	L middle occipital G	18	25	-84	3	4.63
	R anterior insula		31	19	0	4.34
(B)	Hierarchical dependency rule>baseline					
	R pre-SMA	6	4	3	63	4.08
	R ventrolateral premotor C	6	43	6	27	5.01
	L premotor C	6	-50	6	30	4.40
	R medial frontal G	46	43	27	16	5.28
	L postcentral G	2	-47	-24	39	4.55
	R anterior cingulate C	32	7	22	36	6.17
	R precuneus	7	25	-48	45	4.70
	L precuneus	7	-26	-48	45	4.93
	R fusiform G	19/37	31	-57	-6	5.29
	L fusiform G	19/37	-35	-57	-9	5.77
	R middle occipital G	19	-29	-87	9	5.21
	L cuneus	17	22	-84	6	4.86
	R anterior insula		31	24	0	6.17
L anterior insula		-29	24	3	5.47	
(C)	Hierarchical>adjacent dependency rule					
	R pre-SMA	6	1	4	57	4.77
	L pre-central G/S	4/6	-50	-8	33	4.30
(D)	gramm: hierarchical > adjacent dependency rule					
	R pre-SMA/SMA	6	7	0	60	4.75
(E)	ungr: hierarchical > adjacent dependency					
	L pre-central G/M1	6	-47	-11	42	4.42

L, left hemisphere; R, right hemisphere; G, gyrus; S, sulcus; C cortex; IFJ, inferior frontal junction.

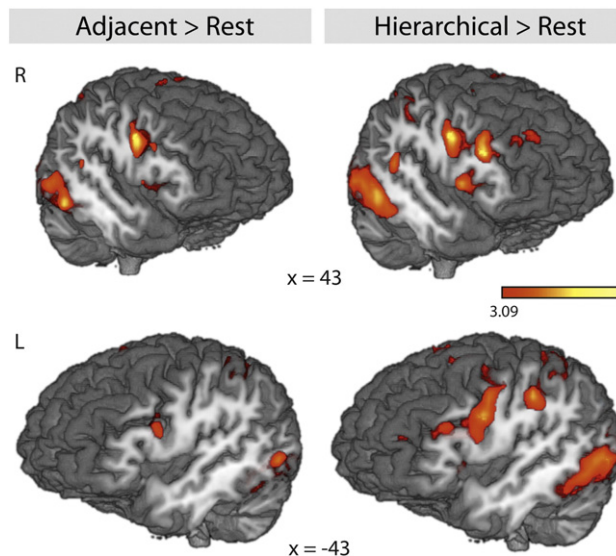


Fig. 3 – Baseline contrast for both rule types. Brain regions showing increased activity in the adjacent dependency rule versus baseline (left), and in the hierarchical dependency rule versus baseline (right). The processing of the hierarchical dependency rule caused activity in several temporal, parietal, and prefrontal areas. The adjacent dependency rule engaged fewer brain regions.

A region of interest (ROI) analysis was conducted to examine the influence of the other two factors (GRAMM and LENGTH) on the regions activated in the RULE contrast. An ANOVA in each ROI was conducted with the three factors applied in this experiment. In all three regions, only the RULE effect reached significance (pre-SMA: $F(1,14)=12.7, p<.005$; pre-central sulcus: $F(1,14)=7.3, p<.05$; caudate head: $F(1,14)=17.5, p<.005$). No other main effect or interaction was significant.

In order to test the specific hypothesis of whether Broca's area (BA 44) is engaged in hierarchical processing of non-linguistic stimuli, an additional ROI analysis with the factors RULE, GRAMM, and LENGTH was performed in this area. The ANOVA revealed a main effect of RULE ($F(1,14)=8.93, p<.01$): The mean beta-values for the hierarchical dependency rule were significantly higher in comparison to the adjacent dependency rule. No other main effect or interaction reached significance.

With the purpose to test whether the rule type difference is specific to Broca's area or whether there is a bias for a more general higher activation level in the brain during the processing of the hierarchical dependency rule, we additionally conducted several ROI analyses in different parts of the brain. In four different fMRI experiments we compared the processing of hierarchical structures with the processing of non-hierarchical structures. The stimuli in these experiments comprised of natural language sentences (Makuuchi et al., 2009), language-like artificial grammar sequences (Bahmann et al., 2008), mathematical formulae (Friedrich and Friederici, 2009), and non-linguistic artificial grammar sequences (present study). The ROIs were defined by the main effect of hierarchy (RULE in the present study) from all four studies. As a result we found no significant differences between the two rule types in the present study in the following regions: left BA6, 10, and 47, left superior

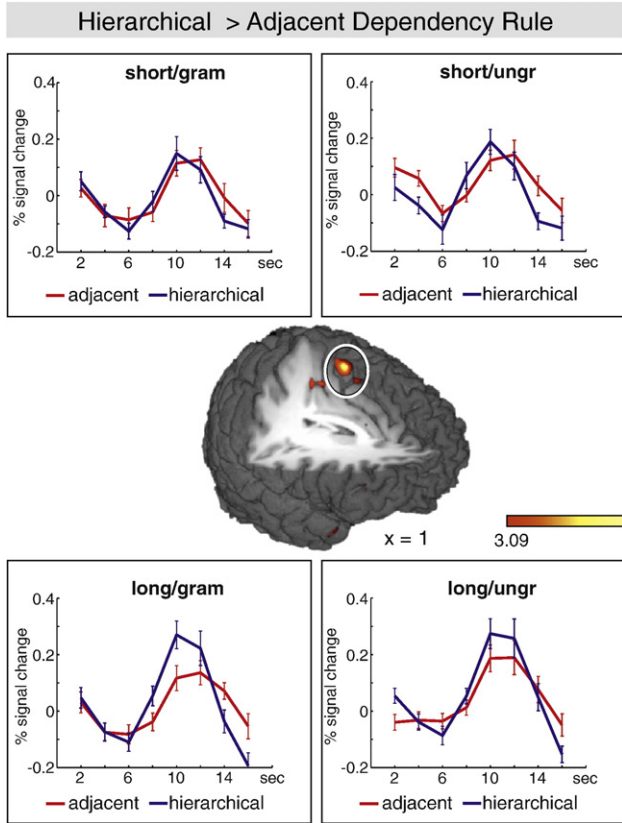


Fig. 4 – Direct contrast of activations from processing hierarchical versus adjacent dependency rules. This contrast resulted in activity in the pre-SMA, the left M1, and the right caudate head. The plots show the time course of the activations in the pre-SMA for adjacent (red) and hierarchical dependency rule (blue) for short grammatical (short/gram), short ungrammatical (short/ungr), long grammatical (long/gram), and long ungrammatical (long/ungr) sequences.

temporal gyrus and sulcus, left intra parietal and superior parietal lobe, left lingual gyrus. These results indicate that the main effect of RULE in the left BA44 in the present study was not caused by a general higher activation level for the hierarchical dependency rule.

To test how much variance in the main effect of RULE was explained by grammatical versus ungrammatical items, additional contrasts were calculated. For grammatical items (i.e. grammatical hierarchical > grammatical adjacent dependency), the pre-SMA and the head of the right caudate nucleus showed a significant BOLD response. For ungrammatical items (i.e. ungrammatical hierarchical > ungrammatical adjacent dependency), only the left middle pre-central sulcus (M1) remained significant.

3. Discussion

In the present study, we investigated which brain regions were involved during the processing of abstract, non-linguistic, hierarchical structures in the visuo-spatial domain. In a direct

comparison of hierarchically organized abstract stimuli with those that followed an adjacent dependency rule, we observed activity in the right pre-SMA, the left pre-central gyrus/sulcus, and the right caudate head. An additional hypothesis-driven ROI analysis confined to Broca’s area revealed a higher hemodynamic response for the hierarchical dependency rule in comparison to the adjacent dependency rule.

3.1. Premotor cortex and pre-SMA

The left prefrontal cortex showed the major activation difference for the contrast of the adjacent dependency rule versus rest condition and the contrast of the hierarchical dependency rule versus rest in the present study (see Fig. 3). The overall stronger BOLD response to the hierarchical dependency rule in this and additional regions suggests general higher cognitive demands during the processing of this rule type. The activity in the left premotor cortex observed for the hierarchical dependency rule was broadly distributed along the whole pre-central gyrus/sulcus. In contrast, the right-hemisphere activity was restricted to the ventral portion of the premotor cortex and was present for both rule types. The premotor cortex has frequently been reported to be co-activated during the observation and imagery of movement sequences (e.g. Dechent et al., 2004; Ehrsson et al., 2003; Gerardin et al., 2000; Lamm et al., 2001) and with increased response competition (Badre and D’Esposito, 2007). It has been suggested that this region is particularly involved in the prediction of dynamics in abstract and concrete event sequences (Schubotz, 2007). The present right ventrolateral premotor cortex activation for both rule types is in line with this latter idea. To successfully

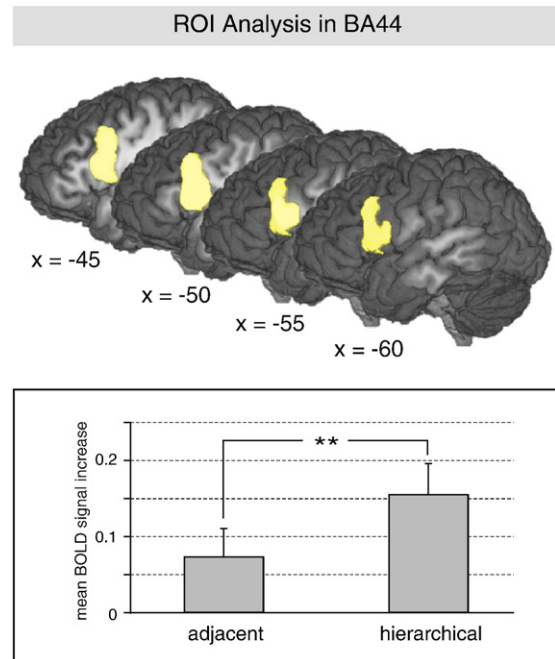


Fig. 5 – ROI analysis in BA44. A ROI in BA44 was defined that comprised of the voxels with a value of at least 30% overlap in the cytoarchitectonical probability map. The BOLD response in BA44 on the hierarchical dependency rule (right bar) was significantly higher than on the adjacent dependency rule (left bar).

process either of the two rules, a prediction regarding the forthcoming elements is required. We suggest that the right ventrolateral premotor cortex plays an important role in the prediction of abstract, sequentially presented elements.

In the present study, we found specific activation in the pre-SMA revealed by a main effect of RULE (hierarchical > adjacent dependency rule). Importantly, the activity in the pre-SMA was specific for grammatical sequences and not present for ungrammatical sequences, thus making an interpretation of an error detection mechanism highly unlikely.

There is strong evidence from a number of tracer studies for a reciprocal connection between the pre-SMA and various prefrontal areas in the monkey (e.g. Bates and Goldman-Rakic, 1993; Luppino et al., 1993). The pre-SMA also receives projections from the basal ganglia and the cerebellum (see for instance Akkal et al., 2007 for a recent study). This connectivity pattern supports the hypothesis that the pre-SMA is more involved in cognitive processing compared with the SMA proper which is, rather, engaged in motor functions (Picard and Strick, 2001). Using electrophysiological single-cell recording, Clower and Alexander (1998) analyzed cell populations of the SMA and the pre-SMA while monkeys performed different movement sequences. Interestingly, the authors found neurons that fired in response to items with a certain rank (i.e. the position of the movement in a sequence), independent of the start point, the direction, or the endpoint of a movement. Neurons tuned specifically to rank-order were predominantly recorded in the pre-SMA. In line with this finding, Shima and Tanji (2000) also reported order-selective activity in pre-SMA neurons. These neurons were specifically activated for the rank-order of movements, independent of the type of movement tested. A similar rank-specific activity pattern in the pre-SMA was also found for saccadic eye movement sequences (Isoda and Tanji, 2004), suggesting an involvement of this region in higher-order sequential processing not only during limb movements but also during other movement modalities. Imaging studies in humans investigating the acquisition and the processing of abstract rules reported activity in the pre-SMA (Bunge et al., 2003; Hanakawa et al., 2002; Sakai et al., 1999). The connectivity between the pre-SMA and the prefrontal cortex, the rank-order selectivity, and the activity during abstract rule processing, suggest a strong involvement of this area during the processing of abstract sequences. The results of the present study are in line with this notion. We found activity in the pre-SMA during the processing of abstract visuo-spatial event sequences. Moreover, this region together with Broca's area seems particularly engaged during hierarchical processing of sequentially presented events, and thus, with the on-line building of a hierarchical representation of abstract items in the visuo-spatial domain.

3.2. Broca's area

Under the hypothesis that Broca's area is engaged in a domain-general processing of hierarchical sequences, it is expected that Broca's area is involved not only for the processing of hierarchically structured language materials but also for non-linguistic materials. The present data support this hypothesis by indicating a higher BOLD response in

Broca's area for hierarchically structured event sequences than for non-hierarchically structured event sequences.

In the language domain, a number of fMRI studies reported activity in Broca's area for the processing of syntactic complexity. A syntactically more complex sentence comprises of a different hierarchical structure than a syntactically less complex sentence. An object-before-subject structure (e.g. "It was Tom who Mary liked.") is syntactically more complex than a subject-before-object structure (e.g. "Mary liked Tom."), irrespective of the content of the sentence (semantics), which is the same in both sentences. Broca's area activity correlated with syntactic complexity (hierarchy) was reported for English (Peelle et al., 2004; Stromswold et al., 1996), German (Bahlmann et al., 2007; Fiebach et al., 2005; Roder et al., 2002), Hebrew (Ben-Shachar et al., 2003), and Japanese (Musso et al., 2003). Interestingly, studies that manipulated the hierarchical dependencies of elements in an artificial grammar design also reported activity in Broca's area (Bahlmann et al., 2008; Friederici et al., 2006; Opitz and Friederici, 2007). Broca's area was also found to be engaged in the hierarchical processing of action plans Koechlin and Jubault (2006). Interestingly, in a study with agrammatic Broca's aphasic patients, Hoen et al. (2003), reported a transfer effect of the training with non-linguistic transformational sequences to a language comprehension task. During training, patients did not only improve the performance of a non-linguistic transformation rule, but they also improved in understanding sentences with relative clauses after training. These results may speak in favor for a cognitive domain-independent involvement of left perisylvian brain region in higher-order sequencing.

The findings from the present study can be compared directly to a previous study investigating the processing of syllable sequences that followed the rule A^nB^n (hierarchical dependency rule) and syllable sequences structured by the rule $(AB)^n$ (adjacent dependency rule). Hierarchically structured syllable sequences correlated with higher activity in Broca's area. In the present study, the same rules were applied using non-verbal stimuli. We employed the same learning procedure and the same number of items per condition, and switched from the verbal modality to the visual-spatial modality. We found Broca's area to be more activated for the hierarchical dependency rule compared to the adjacent dependency rule in both studies — at least when applying a ROI analysis.

The processing of hierarchical visuo-spatial sequences in comparison to non-hierarchically organized sequences resulted in an involvement of the pre-SMA, M1, and the right caudate head. The observed difference in the activation patterns in the two studies would suggest two different neural networks responsible for hierarchical processing in the language domain (previous study) and the visuo-spatial domain (present study). The only commonly activated region being Broca's area.

In recent years it has been attempted to specify neural subregions within the prefrontal cortex for hierarchical structures in different behavioral/cognitive domains (see Botvinick, *in press* for an overview). Tettamanti and Weniger (2006) put forward the notion that other non-linguistic, but hierarchically organized cognitive functions, such as visuo-spatial processing or action-related cognition, are processed in Broca's area. A cognitive domain-independent involvement of this region

during higher-order sequence processing was also postulated by [Fiebach and Schubotz \(2006\)](#). [Friederici \(2006\)](#) has pointed out that Broca's area receives its specificity for linguistic syntax only in a neural network specified for language, and, once part of another network, can serve the function of processing structured sequences in non-language domains such as music or action. The data to hand support this theoretical assumption. To our knowledge we showed for the first time that Broca's area is engaged in hierarchical processing in the visuo-spatial domain. Based on our results, we suggest that Broca's area might reflect the least common denominator for hierarchical processing in the different cognitive domains.

4. Conclusion

The present data indicate that Broca's area together with the pre-SMA subserves the processing of hierarchical structures in visual-spatial sequences thus supporting the view that Broca's area is engaged in hierarchical sequencing in other domains than language.

5. Experimental procedures

5.1. Participants

Fifteen right-handed participants took part in the fMRI study (seven female, mean age=25.6 years, SD=2.4 years). They were all native speakers of German and had normal, or corrected to normal vision. None of the participants had a history of neurological, major medical, or psychiatric disorder.

5.2. Stimuli

Two rule types were applied that were coded by different object categories (A and B). Category A objects were filled with thin or thick dotted lines. Category B objects were filled with thin or thick solid lines. This type of grouping was labeled *Category*. In a behavioral pilot study, a discrimination experiment was performed to select objects for the categories A and B with surface patterns that differed minimally in their perceived physical properties. Additionally, pairs of As and Bs were combined using different spatial orientations of the objects. This type of grouping was labeled *Index*, since A and B were concatenated by their indices. Category B_x objects were generated by rotating category A_x items. The index x indicates the conjunction between single elements of A and B categories. Hence, the concatenation of A and B objects was achieved by their spatial properties (see [Fig. 1](#)).

Two different artificial grammar rules were applied. The rule A^nB^n (e.g. $A_3A_2A_1B_1B_2B_3$) generated center-embeddings and long-distance dependencies between the two different categories (A and B). In this case, the first element in a sequence (A_3) predicted the last element (B_3), the second element (A_2) predicted the fifth (B_2), and the third element (A_1) was concatenated with the fourth (B_1). The three different predictive dependencies generated a hierarchical order within a sequence: both the match between A_x and the corresponding

B_x , and the correct order of the elements (3–2–1 1–2–3) was important. Thus, sequences following this rule were labeled hierarchical. In the present study, four different object categories were applied ($x=4$). To prevent mere pattern learning, the positions of the categories in the sequences were varied (e.g., $A_4A_2A_1B_1B_2B_4$; $A_2A_1A_3B_3B_1B_2$; $A_3A_4A_2B_2B_4B_3$; etc.). The adjacent dependency rule followed the formula $(AB)^n$. This rule generated simple transitions between the two categories (e.g., $A_1B_1A_2B_2A_3B_3$), resulting in the establishment of dependencies between adjacent elements. No processing of long-distance dependencies, center-embeddings, and hierarchy was necessary to process this sequence. As with the hierarchical sequences, the positions of the categories were systematically changed ($A_2B_2A_1B_1A_3B_3$; $A_1B_1A_3B_3A_2B_2$; $A_4B_4A_3B_3A_2B_2$; etc.).

We varied the positions of the dependent elements in a sequence (six combinations for long sequences with 3 elements each: $A_3A_2A_1B_1B_2B_3$; $A_2A_1A_3B_3B_1B_2$; $A_3A_1A_2B_2B_1B_3$; $A_2A_3A_1B_1B_3B_2$; $A_1A_2A_3B_3B_2B_1$; $A_1A_3A_2B_2B_3B_1$; six combinations for long sequences with 3 elements each (including the fourth category): $A_4A_2A_1B_1B_2B_4$; $A_4A_1A_3B_3B_1B_4$; $A_4A_1A_2B_2B_1B_4$; $A_4A_3A_1B_1B_3B_4$; $A_4A_2A_3B_3B_2B_4$; $A_4A_3A_2B_2B_3B_4$; etc. = 6 combinations \times 4 elements per position = 24 combinations). Furthermore, $2^6=64$ combinations of the permutations of the categories A (one dotted line, two dotted lines) and B (one solid line, two solid lines) were generated for long sequences and $2^4=16$ category combinations for short sequences. Hence, $64 \times 24=1536$ items for long sequences and $16 \times 12=192$ items for short sequences were computed.

In both rule types, short (four elements) and long (six elements) sequences were employed, as were grammatical and ungrammatical sequences (50% each). This manipulation resulted in a $2 \times 2 \times 2$ design, with the within-subject factors RULE (adjacent dependency, hierarchical dependency rule), LENGTH (short sequences, long sequences) and GRAMM (grammatical sequences, ungrammatical sequences). In ungrammatical sequences, the violating object was always placed at one of the last two positions in the short (AABB) sequences and at one of the last three positions in the long sequences (AAABBB).

At least two studies exist claiming that the rule A^nB^n can be processed by means of simpler strategies rather than using the processing of long-distance dependencies ([de Vries et al., 2008](#); [Perruchet and Rey, 2005](#)). In order to exclude this possible confound, we introduced Index grouping. To ensure and measure the processing of the rule in a center-embedded manner, violations of both types of grouping (Index and Category) were included in the sequences. In a *Category-violation*, an element of category B was exchanged with an element of category A (e.g. linear rule, short sequence: $A_1B_1A_2A_3$; hierarchical rule, long sequence: $A_1A_2A_3B_3A_4B_1$). *Index-violations* comprised incorrect indices, i.e. the category B element did not match the preceding A element (e.g. linear rule, long sequence: $A_1B_1A_2B_2A_3B_4$; hierarchical rule, short sequence: $A_1A_2B_2B_3$; see [Fig. 1](#)). As mentioned above, in the present study, four different object categories were applied, instead of three categories as in the previous experiment. This was done for the following reason: an *Index-violation* in long sequences could be identified by detecting the repetition of a category when only three categories are selected (e.g.

$A_1A_2A_3B_3B_2B_3$; see de Vries et al., 2008 for a similar argumentation). However, as sequences were presented in random order, participants could not know when to apply an embedded-processing strategy or a simple repetition detection tactic. Nonetheless, to prevent repetition detection strategies for *Index-violation*, in the present study a fourth type of category was included (e.g., $A_1A_2A_3B_3B_2B_4$). The number of violations was counterbalanced across positions, sequence length, and rule type.

5.3. Procedure

The structure and task of the experiment were identical to the previous study (Bahlmann et al., 2008). A learning session and a testing session were applied for each rule type. The learning session was scheduled for two days prior to testing. After a week's delay, the second rule was trained and (two days later) learning was assessed in the scanner.

Before training, participants were instructed to extract the rule behind the object sequences. During the learning session, first, a block of ten correct probe sequences was presented, followed by a block with five grammatical and five ungrammatical object sequences. Participants were required to indicate whether the sequences were structured according to a rule or not by pressing a button. Feedback was given. Hence, learning was explicit and guided by trial and error. Two object items (e.g. A_1B_1) were first employed in the learning phase of the experiment. When participants reached a criterion of 90% correctly answered items in two successive blocks, they entered the next learning step where they were presented with four object items (e.g. $A_2A_3B_3B_2$). Again, after correctly responding to 9 out of 10 test sequences in two successive blocks, the third learning step was reached, which involved the processing of six object sequences. Again, this was completed once 90% of the sequences in two successive blocks were judged correctly. On average, the learning procedure took approximately 32min. Note, that this learning procedure is different from that used by de Vries et al. (2008).

Two days after training, fMRI measurements were collected during testing. For this part of the experiment, 144 new items were presented (half short, half long sequences, of which half (36 for each length) were ungrammatical). The 36 ungrammatical sequences consisted of 18 *Category-violations* and 18 *Index-violations* for both long and short sequences.

The start of a sequence was indicated by a fixation cross (500 ms). Objects were presented separately in the middle of the screen for 600 ms each. After the presentation of a sequence, participants were asked to provide a judgment regarding the grammaticality of the sequence within 2000 ms, followed by feedback for 500 ms (i.e. the word "correct" or "wrong" was presented in green or red on the screen). Afterwards a fixation cross was shown for a further 2000 ms. Trials started with a jitter of 0, 500, 1000, or 1500 ms. Additionally, 36 null-events (presentation of a fixation cross) were included in random order.

Response-key assignment (right index finger vs. right middle finger) and order of rule type (adjacent dependency rule first vs. hierarchical dependency rule first) were counterbalanced across groups. Additionally, the number of male and

female participants was equal for response-key assignment and rule type order.

After each test session participants answered several questions about the experiment in a standardized questionnaire. In order to explore, how participants would describe the rule they used, we asked "How did you do the job?" After the session in which the hierarchical dependency rule was processed, 11 of the 15 participants provided indications that they had constructed a hierarchical structure during processing. In particular, they described the embedded nature of the sequences and they noticed the importance of the correct order of the items in a sequence. Thus, we take this as an indication that participants really applied the hierarchical rule.

5.4. fMRI Acquisition

Imaging was performed on a 3 T scanner (Medspec 30/100/ Bruker, Ettlingen). Stabilization cushions were used to reduce head motion. For registration purposes, two sets of two-dimensional anatomical images were acquired for each participant immediately prior to the functional imaging session. An MDEFT (data matrix 256×256 , TR=1.3 s, TE=7.4 ms) and an EPI-T1 (TE 14 ms, TR 3000 ms) sequence were used. Functional MRI scanning was carried out using a T2*-weighted BOLD sensitive gradient echo echo-planar imaging sequence (TR=2 s, TE=30 ms, FOV=19.2 cm, 64×64 matrix, resulting in an in-plane resolution of $3 \text{ mm} \times 3 \text{ mm}$). Twenty-four slices (thickness: 4 mm with an interslice gap of 1 mm) covering the whole brain were acquired. Anatomical and functional images were positioned parallel to AC-PC. Four functional runs with 220 volumes each were collected. The same imaging procedure was used for both experimental sessions.

5.5. fMRI analysis

Data processing was performed using the LIPSIA software package (Lohmann et al., 2001). Preprocessing of the time series involved: motion correction (rigid-body realignment); a slice-time correction using sinc interpolation; a spatial smoothing (Gaussian kernel with 5 mm FWHM); and, baseline correction using a temporal high-pass filter (cutoff frequency: 1/80 Hz). The time series were co-registered with high-resolution 3-D images that were acquired during previous scanning sessions. To achieve an optimal match between the 3-D image and the functional time series, co-registration was performed separately in each of the four functional runs. Functional images were then normalized to the Talairach stereotactic space (Talairach and Tournoux, 1988) using linear and non-linear normalization. The statistical evaluation was based on a least-squares estimation using the general linear model dealing with serially autocorrelated observations using pre-whitening of each voxel's time series (Worsley et al., 2002). Specifically, autocorrelation parameters were estimated from the least squares residuals using the Yule-Walker equations. These parameters were subsequently used to whiten both data and design matrix. Finally, the linear model was re-estimated using least squares on the whitened data to produce estimates of effects and their standard errors. Incorrectly answered trials were excluded from the analysis. Contrast images, i.e. estimates of the raw-score differences

between the conditions, were then generated for each participant. The single-participant contrast images were entered into a second-level random effects analysis for each of the contrasts. The group analysis consisted of a one-sample *t*-test across the contrast images for all participants to determine whether observed differences between conditions were significantly different from zero. Subsequently, *t*-values were transformed into *Z*-scores. The combination of voxel-based thresholds with a minimum cluster size has been argued to improve the statistical power (Forman et al., 1995). We applied this double-threshold approach to protect against false positive activations, considering an area to be activated only if it comprised a volume greater than or equal to 1215 mm³ (45 voxels) and had a *Z*-score of greater than 3.09 (*p* < .001, uncorrected). This non-arbitrary voxel cluster size was determined by using the program AlphaSim (afni.nimh.nih.gov/afni/doc/manual/AlphaSim by Ward, 2000). Figures show the resulting *Z*-map overlaid onto an anatomical reference image.

In order to test the specific hypothesis of whether Broca's area shows a significant difference in the BOLD response for the adjacent versus the hierarchical dependency rule, a region of interest (ROI) analysis was conducted. In order to examine the hemodynamic response to the different conditions, a ROI was defined according to the cytoarchitectonic probability map (Amunts et al., 1999). This map was used to provide an objective measurement for the definition of the ROI in BA44. We defined a ROI of BA44 that comprised of voxels with a value of at least 30% overlap (see Fig. 5). An ANOVA on the individual beta-values was calculated including the factors RULE (adjacent dependency, hierarchical dependency rule), LENGTH (short sequences, long sequences), and GRAMM (grammatical sequences, ungrammatical sequences).

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