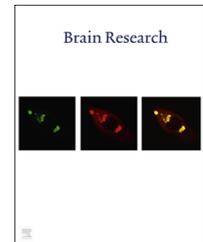
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Research Report

Inefficient stimulus processing at encoding affects formation of high-order general representation: A study on cross-modal word-stem completion task



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ABSTRACT

Priming is an implicit memory effect in which previous exposure to one stimulus influences the response to another stimulus. The main characteristic of priming is that it occurs without awareness. Priming takes place also when the physical attributes of previously studied and test stimuli do not match; in fact, it greatly refers to a general stimulus representation activated at encoding independently of the sensory modality engaged. Our aim was to evaluate whether, in a cross-modal word-stem completion task, negative priming scores could depend on inefficient word processing at study and therefore on an altered stimulus representation. Words were presented in the auditory modality, and word-stems to be completed in the visual modality. At study, we recorded auditory ERPs, and compared the P300 (attention/memory) and N400 (meaning processing) of individuals with positive and negative priming. Besides classical averaging-based ERPs analysis, we used an ICA-based method (ErpICASSO) to separate the potentials related to different processes contributing to ERPs. Classical analysis yielded significant difference between the two waves across the whole scalp. ErpICASSO allowed separating the novelty-related P3a and the top-down control-related P3b sub-components of P300. Specifically, in the component C3, the positive deflection identifiable as P3b, was significantly greater in the positive than in the negative priming group, while the late negative deflection corresponding to the parietal N400, was reduced in the positive priming group. In conclusion, inadequacy of specific processes at encoding, such as attention and/or meaning retrieval, could generate weak semantic representations, making words less accessible in subsequent implicit retrieval.

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1. Introduction

The defining characteristic of implicit memory is that it is used without awareness; thus, the content of implicit memory cannot be reported (Schacter, 1987; Schacter and Buckner, 1998). In contrast with explicit tests, in which participants consciously attempt to recollect information encountered in an earlier phase of the experiment (the study or encoding phase), in implicit tests participants perform tasks that are apparently unrelated to the previously presented information—i.e. identification of degraded words or word stems completion with the first word that comes to mind. In these tasks, subjects perform better at test when presented with information beforehand, even though they typically make no conscious attempt at remembering. This facilitation is termed priming. Priming has also been found when the physical attributes of the study stimuli do not match those of the test stimuli (Rajaram and Roediger, 1993; Jackson and Morton, 1984; Pilotti et al., 2000). The fact that mismatch does not eliminate priming supports the idea that stimulus processing forms representations spanning multiple modalities, even when information is conveyed by means of a specific one.

Linguistic processing is based on the integration of different modalities (Frost and Katz, 1989; Grainger and Ferrand, 1996; Plaut et al., 1996; Seidenberg and McClelland, 1989; Ziegler et al., 2003). One of the most accredited model of word recognition, based on the strong interactivity across modality-specific representations, is the bimodal interactive-activation mode (Grainger and Ferrand, 1994, 1996). According to the model, presentation of a word in the visual modality generates activation in orthographic codes, which rapidly activate the corresponding phonological codes thus improving the recognition process. The same applies for auditory word recognition, where phonological codes rapidly activate the corresponding orthographic representations (Kiyonaga et al., 2007). This interactive mechanism would be fundamental for a whole-word higher-level semantic representation.

Based on this assumption, implicit memory of linguistic information could refer to a general semantic representation of the word independent of the specific sensory modality engaged at encoding. According to this hypothesis, some studies have reported that the physical attributes of words play a role in priming even if these attributes are imagined rather than directly perceived (Pilotti et al., 2000; Stuart and Jones, 1996). In fact, imaging the sound of printed words at study (auditory imagery) (Pilotti et al., 2000) or forming images of spoken words as they would appear if printed (visual imagery) (Roediger and Blaxton, 1987) produces priming in the implicit tests of identification of words degraded by noise and in word fragment completion task, respectively.

Even though priming has been found when the physical attributes of studied words did not match those of the test words, smaller effects were generally found—i.e. spoken words produce less priming than printed words in the visual implicit tests of word stem and word fragment completion (Rajaram and Roediger, 1993), and printed words produce less priming than spoken words in the auditory version of these tests (Jackson and Morton, 1984; Pilotti et al., 2000).

In a pilot study of ours (Castellani, 2007, PhD Thesis) we found that about half of the individuals participating to a word stem completion task (WSCT) exhibited no implicit memory at all when the physical attributes of the study stimuli (spoken words) did not match those of the test stimuli (printed word stem). More specifically, their priming score that consists of the rate of correct completions on a studied list relative to the casual completions on a control list, was negative i.e. less correct completions on the studied than on control list. If cross-modal priming depends on the individuals' ability to form a general semantic representation, this effect could be related to factors at encoding such as attention and word processing that could have affected the formation and/or consolidation of the general high-order modality-independent representation of words.

In the present study we planned a cross-modal WSCT in which words were presented in the auditory modality at study, and word stems appeared in the visual modality in the test phase. Our aim was to evaluate whether negative priming scores at test could indicate a weak semantic representation likely owing to inefficient word processing at study. Specifically, we recorded auditory ERPs in the study phase and focused on the centro-parietal subcomponent of P300 (P3b) and the parietal N400 waves. P3b has been considered to reflect the activation of attentional networks and the integration of incoming information into memory representations of the stimulus (Polich, 2007; Azizian and Polich, 2007), and to be a possible electrophysiological marker of Locus Coeruleus (LC) noradrenergic function (Nieuwenhuis et al., 2005, 2011; Polich and Criado, 2006). On the other side, N400 is considered a good marker of the efficiency of word meaning processing (Kutas and Federmeier, 2011). Given that differences during word learning give rise to memory traces differing in strength and that word recognition is better with stronger traces than with weaker ones (Balass et al., 2010), we expected that, at study, individuals with negative implicit memory performance could be worse in terms of words processing than individuals with positive scores.

Besides a raw scalp-channels-based classical ERP analysis, we used a method based on independent component analysis (ICA) (Brown et al. 2001) in order to disentangle potentials related to the different processes contributing to the ERP dynamics (Pourtois et al. 2008). Our ICA approach named ErpICASSO, (Artoni et al., 2012b, 2014; Menicucci et al., 2013;) enabled us to derive templates of the independent ERP components that we chose to identify processing differences between subjects with different priming scores.

In particular, we used ErpICASSO with the aim to separate the two sub-components of P300 (P3a and P3b) that are easily discriminated with an oddball paradigm – P3a is evoked by unexpected novel stimuli and reflects stimulus-driven (bottom-up) attention capture, while P3b reflects top-down controlled detection of relevant/salient events – but appear greatly superimposed when task-relevant novel stimuli, as in our experimental protocol, are used (Debener et al., 2005; Heitland et al., 2013).

ErpICASSO could also help to discriminate between parietal N400 and FN400, a wave similar to N400 as for peak latency but with a more frontal localization than N400, which has been considered an index of familiarity rather than a general meaning retrieval one (Paller et al., 2007).

2. Results

2.1. Priming scores

According to the priming score ratings we selected a group of 10 individuals with positive PS values (PS-P, 0.18 ± 0.05 , mean \pm SE) and a group of 10 individuals with negative Priming Scores (PS-N, -0.10 ± 0.02) representing, respectively, individuals with and without implicit memory formation.

2.2. Classical ERPs

For each electrode site we performed the grand-average ERP across trials and subjects for PS-P and PS-N, separately. Fig. 1 shows the grand-average for midline (FZ, CZ), centro-parietal (CP3, CP4), temporal (T3, T4) and temporo-parietal (TP7, TP8) electrodes. Below each plot, the time course of statistical significance (unpaired t-test) relative to the comparison between the two groups performed at each time of sampling is shown. The threshold was set at $p < .005$.

It can be observed that in both groups word presentation elicited a small positive peak at about 50 ms followed by a negative deflection peaking at about 100 ms, likely corresponding to the auditory P50 and P100 waves. At later latencies a positive peak at about 300 ms was followed by a broad negative deflection. The presence of these waves at all the electrode sites did not allow to differentiate either the P3a from the P3b subcomponent of P300 or the N400 from the FN400 wave. In fact, their discrimination is mainly based on their different topographical distribution that is fronto-central for P3a and FN400 and centro-parietal for P3b and N400.

The comparison between the two groups did not yield any significant differences between peak latencies. In contrast, significant differences ($p < 0.01$) were found at P50 and in the 300–650 ms interval, at almost all the electrode sites. In particular, the amplitude of P50 and P300 was greater in the PS-P group than in the PS-N one, while the amplitude of the negative deflection (N400/FN400) was greater in the PS-N group than in the PS-P one.

2.3. Activity template components of ERPs

Fig. 2 shows, for each component, the topographical distribution on the scalp of PS-P and PS-N and their activity templates. In the plots, the sheaves of light lines show the variability of each component template whose estimate (the BAT) is highlighted with a thick line. Red and blue lines refer to PS-P and PS-N, respectively. Below each plot, the time course of the p values relative to the comparison between the two BATs performed at each time of sampling (unpaired t-test) are shown.

The component 1 (C1, $Q=0.64$) is lateralized on the temporal regions. It shows a small positive peak at about 150–200 ms and a small negative one at 300 ms, followed by a late positive deflection that appears more marked in PS-P. As indicated by the time-course of p -values there are no differences between PS-P and PS-N.

The component 2 (C2, $Q=0.53$) is bilaterally even in the fronto-temporal region. At latencies < 300 ms two peaks with latencies similar to those found in the component 1 were present. At latencies later than 300 ms a negative peak

(at about 500 ms) more accentuated in the PS-P group is inscribed on a slight positive deflection that is nearly null in the PS-N group. As indicated by the graphic of p -values time-course there are no differences between PS-P and PS-N.

The component 3 (C3, $Q=0.91$) is localized in the centro-parietal areas. It is characterized by a positive peak at around 50 ms, followed by a negative deflection at about 100 ms, a positive peak at about 300 ms, and a negative deflection at latencies > 300 ms. As indicated by the graphic of p -values, PS-P and PS-N time-courses are significantly different across the whole time interval with a higher positive peak and a smaller late negative deflection in PS-P than in PS-N ($p < 0.01$).

The component 4 (C4, $Q=0.53$) spreads over the frontal region with a peak at the frontopolar electrodes. Like the previous component, it shows an early negative deflection followed by a positive peak at 300 ms. At lags later than 300 ms a large negative deflection more accentuated in PS-P is present. However, no significant differences between PS-P and PS-N are found.

3. Discussion

Evoked responses induced by words presentation during the Study session are in agreement with previous literature on auditory late-latency responses (for a review see Joos et al., 2014). We identified an early peak corresponding to the pre-lexical P50, followed by a series of waves likely corresponding to classical auditory N100, P300 and N400/FN400. The amplitude of P50 and P300 was smaller in PS-N than in PS-P, while the opposite occurred for N400; these differences were present across the whole scalp thus indicating that the classical analysis did not allow defining either a clear spatial segregation of the specific waves (i.e. P3a vs. P3b) nor their differential involvement in word processing in the two groups.

The analysis of the evoked responses by means of ErpI-CASSO allowed to successfully decompose the two spatio-temporally overlapping sub-components of P300 that is P3a and P3b (Polich, 2007; Heitland et al. 2013), and to separate the typical parietal N400 from the frontal FN400. In fact, peaks likely corresponding to P3b/N400 and P3a/FN400 were identified in two independent ERP components, namely C3 and C4.

The two sub-components of P300 are commonly elicited in an active oddball paradigm where infrequent target and novel unexpected stimuli are embedded in a regular train of standard stimuli (for a review see Polich, 2007). General literature suggests that P3a, also referred to as novelty P3, reflects stimulus-driven (bottom-up) attention capture, has a fronto-central scalp localization and relies on dopaminergic and serotonergic mechanisms. A peak likely corresponding to P3a (given latency and fronto-central localization) is present in C4. However, no differences between PS-P and PS-N were found, thus suggesting that the differences between the two groups cannot be related to bottom-up mechanisms.

ICA analysis identified one component (C3) with a positive peak around 300 ms from the stimulus onset, whose centro-parietal topography overlap with the typical P3b scalp topography (Polich 2007). The amplitude of P3b is typically largest over centro-parietal scalp regions, and it is generally assumed that it reflects top-down control of relevant/salient events (i.e. target), it

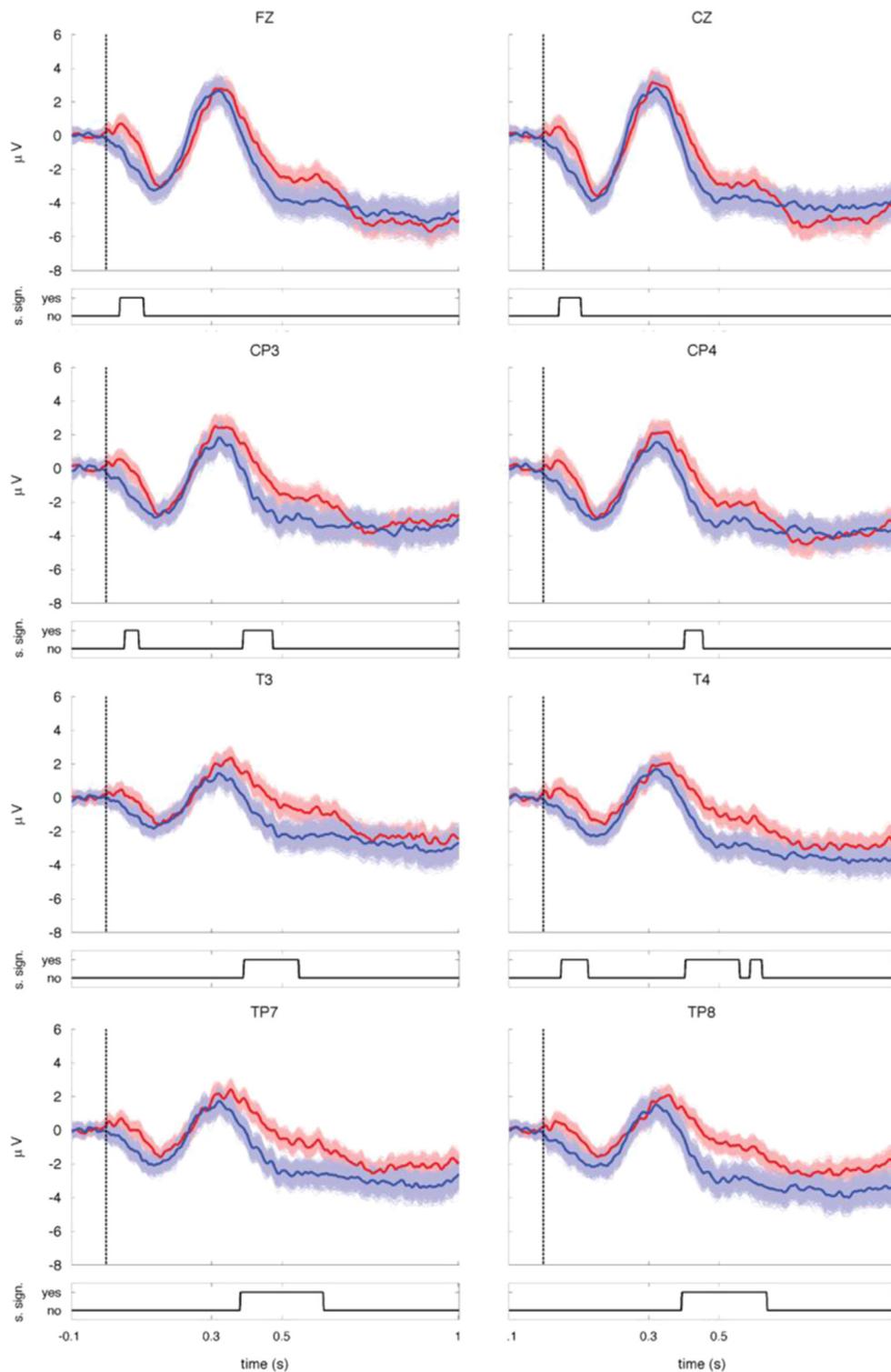


Fig. 1 – Grand-average ERPs across trials and subjects for PS-P (red lines) and PS-N (blue lines) at frontal, centro-parietal and temporal electrodes, respectively. Below each plot, the time course of p values with threshold set at $p=0.01$ relative to the comparison between the two groups performed at each time of sampling (unpaired t -test) is shown.

involves activation of attentional networks associated to memory engagement in parieto-temporal areas, (Polich, 2007). Specifically, it has been demonstrated that P3b peaks once a target has been detected and that its amplitude is proportional to the amount of attention resources voluntarily assigned to the task (Gonzalez and Polich, 2002). In particular studies with verbal

stimuli showed that studied words that receive full attention were associated with greater P300 amplitude at the centro-parietal sites and were recognized with more certainty (Curran, 2004; Curran and Cleary, 2003). Furthermore, the close association between efficiency of encoding and P300 amplitude is consistent with the view that P3b is associated with memory

engagement (Azizian and Polich, 2007). In other words, the stimulus encoding that promotes successful memory storage, retrieval and recognition of the stimulus, increases P300 amplitude. In this vein, P3b amplitude can be viewed as a good index of efficient integration of incoming information into memory representations of the stimulus.

The greater amplitude of P3b during the study phase of PS-P individuals could thus indicate a stronger memory representation of the words in PS-P than in PS-N participants. Selective attention at encoding has been indeed associated with better consolidation of perceptual representation that would be then more easily accessed during subsequent implicit or explicit retrieval (Turk-Browne et al. 2006).

Evidence from research on animals and humans suggest that the P3b wave may represent an electrophysiological correlate of the phasic Locus Coeruleus (LC) response (Nieuwenhuis et al., 2005, 2011; Polich and Criado, 2006), thus pointing to a possible involvement of the LC-Noradrenergic system in P3b generation. Even though there has been little research in humans, genetic evidence that links P3b amplitude to a series of polymorphisms that code for noradrenaline synthesis and expression in the human brain (Liu et al., 2009) as well as a recent pupillometry study evaluating P3b and pupil diameter as LC-noradrenergic system markers (Murphy et al. 2011), reinforce the P3b-LC hypothesis in humans. In line with this hypothesis, we can thus assume that the decreased P3b amplitude found in PS-N participants may be related to an altered efficiency in the LC-Noradrenergic function.

The ICA method allowed detecting significant differences between PS-P and PS-N also for the negative deflection that is present in C3 at long latencies (>300). The topographic, as well as, its time course likely resemble those typical of the parietal lexically-induced N400 (Kutas and Federmeier, 2011). N400 is consistently elicited by words or word-like stimuli, either in full sentences (Kutas and Van Petten, 1990) or in isolation or pairs (Holcomb and Neville, 1990). It is found across paradigms (Kutas and Van Petten, 1990; Holcomb and Neville, 1990; Rugg et al., 1995; Curran and Dien, 2003), both in the visual and auditory modalities (Holcomb and Neville, 1990) as well as in signed languages (Kutas et al., 1987), suggesting task and modality independence. In general, it is assumed that the N400 could reflect meaning retrieval (Kutas and Federmeier, 2011). In particular, N400 reflects processing at the form-meaning interface whereby word-based form information is mapped onto meaning, with larger N400s for less efficient mappings (Holcomb and Grainger, 2007). Typically, when a word is preceded by a supportive context, i.e. a lexical associate, the so-called N400 effect, namely a reduction in the amplitude of the N400 deflection, is reliably observed (Kutas and Federmeier, 2011). Extensive literature

agrees that the N400 effect is at least partially determined by the degree to which the context predicts the target (e.g. Federmeier, 2007; Van Berkum et al., 2005; Kutas et al., 2006). Accordingly, ERP studies of recognition memory extensively described a word-repetition effect, also called “old/new” effect, which consists in the reduction of the word-elicited N400 around midline parietal electrode locations, in association with the awareness that a word has recently been experienced (Van Strien et al., 2005). The old/new effect has been demonstrated to be modulated by memory strength (i.e. increasing the repetition number) and has been indicated as a possible neural correlate of implicit memory processes (Grill-Spector et al., 2006). Similar findings of a reduction in N400 have been recently found in a masked priming experiment (Eddy et al., 2014). Altogether these findings indicated the N400 effect as a good marker of the efficiency of word meaning processing.

Our result showed a greater amplitude of N400 in PS-N than in PS-P. This finding, together with results on P3b, suggests that the scarce proficiency in processing and memorizing words of PS-N would be also associated with a low efficiency in meaning analysis.

A negative deflection likely corresponding to the FN400 (Curran and Hancock, 2007), a negative potential named after its shape (maximum over frontal brain regions and peak at about 400ms after stimulus onset) was found in C4. Parietal N400 and FN400 are strikingly similar, but have different distribution across the scalp: midfrontal for FN400 potentials and centro-parietal for typical N400 potentials. Moreover, the FN400 has been regarded as an index of familiarity rather than a general meaning-retrieval one (Paller et al., 2007).

As indicated by the results, no amplitude differences were found between PS-P and PS-N thus suggesting that a familiarity effect that is independent from the specific learning phase cannot be involved in priming differences.

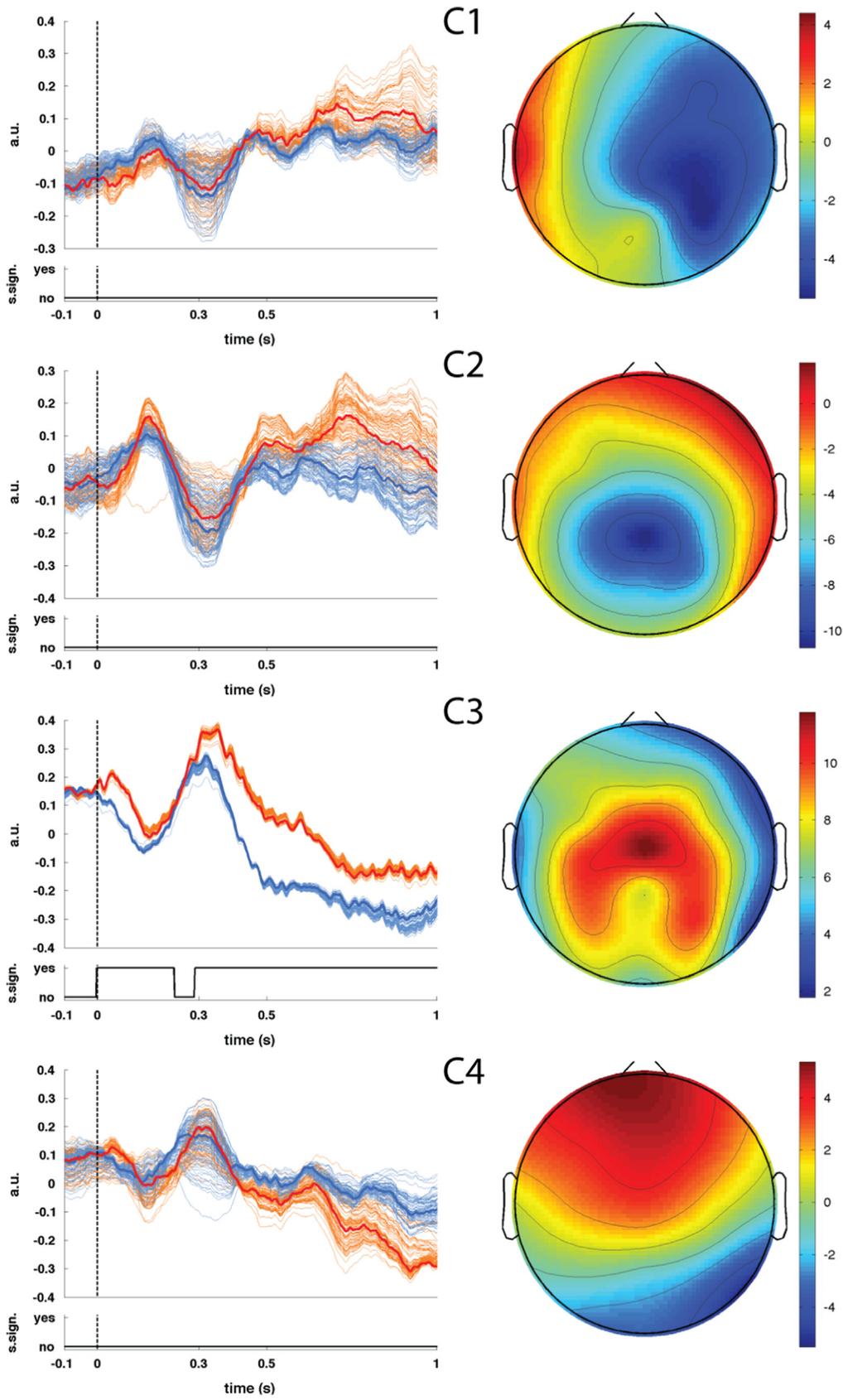
In conclusion, our study suggests that inadequacy of different processes during word encoding, such as attention and/or meaning retrieval, would generate weak general semantic representations thus making the studied words less accessible during subsequent implicit retrieval.

4. Experimental procedures

4.1. Participants

Participants were 20 volunteers (11 males, 9 females; age 22 ± 1.87 , mean \pm SD) selected from a pool of students at the University of Pisa, who decided to participate in the experiment to obtain an extra credit for a Physiology Lab. Participants were right-handed (Edinburgh Inventory for Handedness (Oldfield,

Fig. 2 – Best activity templates (BATs) of extracted ERP components. The figure shows the four components (each one in a different panel). The label of each panel identifies the component. For each component the plot of the activity templates and their topographical distribution on the scalp are shown. In the plots, the sheaves of light lines correspond to the activity templates and the BATs are highlighted with thick lines. Red and blue lines refer to PS-P and PS-N, respectively. Below each plot, the time course of *p* values relative to the comparison between the two BATs performed at each time of sampling (paired *t* test) is shown. The significance threshold was set at $p=0.01$ (dashed line). The activation maps indicate the weight (color scale) of the component at each electrode site, thus globally each map shows the spatial distribution of the corresponding component over the scalp.



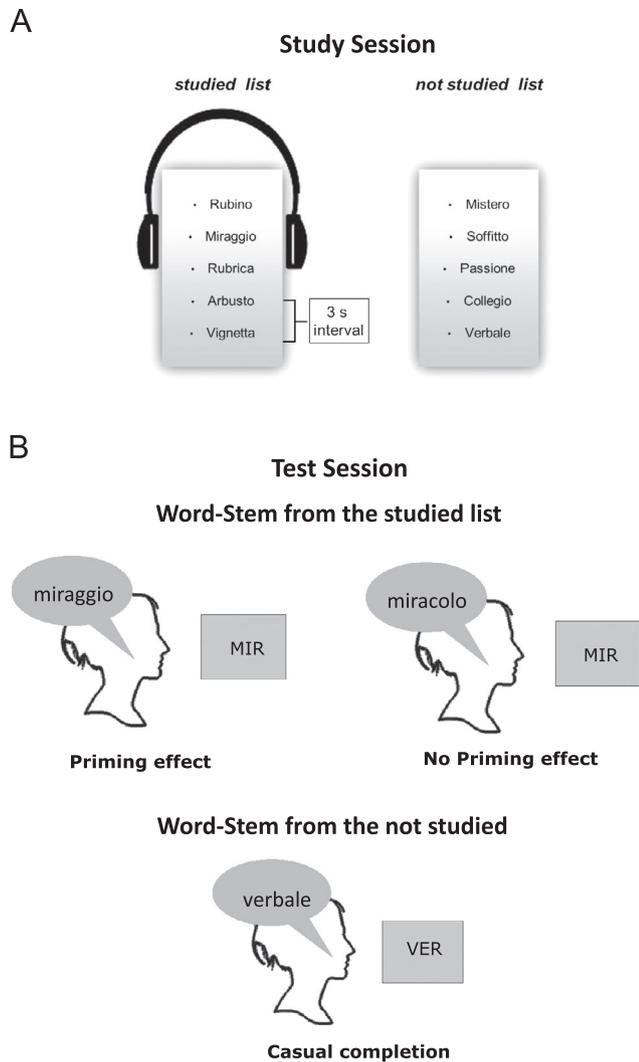


Fig. 3 – Schematic representation of the experimental sessions (A) **Study session**: on the left, a section of the word list listened by participants is shown. The time interval between two successive words is 3 s. On the right, a section of the word list not listened by participants and whose stems were employed as filler stems in the test phase is shown. (B) **Test session**: a scheme of word-stem completion task is shown. All word-stems were visually presented. Priming effect occurs if the word-stem from a word of the studied list is completed with a studied word, while no priming occurs if it is completed with a not studied word (upper line). Casual completion occurs if the word-stem from a word of the not studied list (filler stem) is completed with a word of the not studied list (bottom line).

1971) median=0.8, range 0.6–1), reported normal/corrected-to-normal vision and no hearing nor neurological problems.

Written informed consent was obtained from all participants.

4.2. Procedure

The experimental procedure consisted of two sessions: Study and Test (Fig. 3). Participants performed both Study and Test sessions individually.

In both sessions, auditory stimuli were presented one at a time. The auditory stimuli were Italian nouns (6–9 letters) of low/medium frequency (De Mauro et al., 1993; Lorenzi et al., 2006). All words were digitized at 44.1 kHz with a 16-bit sampling rate. Their average intensity was normalized to 64 dB.

The instructions for the Study and Test sessions were provided before the start of each session.

4.2.1. Study session

Before starting the Study session, participants were informed that they were about to take part in an experiment to assess their attention. Following the application of the EEG electrodes, they comfortably seated in front of a computer keyboard, and were instructed to listen to words presented by means of earphones and to press a keypad as quickly as possible when they had completed listening. This task ensured that participants attentively listened to the words during the experiment. Subjects were also instructed to keep their eyes shut throughout the whole Study session.

Following a brief training, participants listened to a list of 36 words (Studied list) repeated 4 times (Table 1). The order of words within each of the 4 repetitions was randomized. There was a 2000 ms inter-stimulus interval (ISI) between the offset of the last word and the presentation of the next. The delay guaranteed a period of time in which the N400 could be clearly examined without the risk of contamination from sensory responses evoked by the following words. Participants were asked to remain as relaxed as possible throughout the test phase trials to minimize EEG artifacts due to head and body movements.

4.2.2. Test session

During the Test session, participants comfortably seated in front of a computer screen positioned at a distance of about 57 cm. In order to reduce head movements and to maintain the distance from the screen fixed, participants positioned their head on a suitable support. Sixty word stem that is 4 buffer stems, 36 from the Studied list (critical stems) and 20 filler stems from a not studied word list, were presented in black on a light grey background for 3 s.

The buffer stems were presented at the beginning of the list, the critical stems were presented in a random order with respect to the Study session, and the filler stems were distributed randomly among the critical stems (Table 2).

Participants were instructed to complete each stem with the first word that came to mind as fast as possible. The instructions emphasized the need to provide “the first word that came to mind” because of the limited time available. Responses were collected via microphone. No mention of the Test session or its nature was made until it started.

Even though stem-completion tasks reflect involuntary retrieval and are usually uncontaminated by voluntary recall strategies (Beauregard et al., 1999; Fay et al., 2005), possible explicit memory occurrence was evaluated by asking subjects, at the end of the Test session, to do a voluntary recall of the studied words. Priming effects were calculated only on items that participants did not show voluntary retrieval for.

Table 1 – Shows the list of the 36 Italian words listened by participants during the Study session (studied list). The list was presented 4 times in a randomized order.

Studied list							
1	Dottrina	10	Veterano	19	Frazione	28	Lettere
2	Lanugine	11	Furfante	20	Locuzione	29	Bastone
3	Solido	12	Grossista	21	Ottava	30	Sapore
4	Astinenza	13	Follicolo	22	Velista	31	Coperta
5	Radicale	14	Lamina	23	Corallo	32	Perito
6	Tintura	15	Rubrica	24	Latrina	33	Impiegato
7	Garanzia	16	Arbusto	25	Merenda	34	Foresta
8	Margine	17	Miraggio	26	Ritocco	35	Tempera
9	Parabola	18	Vignetta	27	Tendenza	36	Antenna

Table 2 – Shows the list of the 60 word stems used in the Test session: 4 buffer stems (light grey) were presented at the beginning of the list, 36 stems from the Studied list (black) and 20 filler stems from a not studied list (grey) were presented in a random order.

Stem list					
LIM	OTT	VET	FRA	LET	LOC
TES	COL	COP	TEN	COR	STI
SEL	ANT	SOF	CAT	FAV	RIL
FAN	TIN	DOC	FOR	ARB	SAP
AST	MIS	IMP	MAR	LAM	GRO
MIR	PAR	VER	RAD	GAR	SOL
FUR	DOT	FOL	TOV	CON	TRI
INF	LEG	TEM	LAN	VIG	PER
LAT	BOT	DIS	VEL	PIT	MAL
BAS	RUB	RIT	PAS	MER	FED

4.3. Priming score computation

For each participant, we measured the magnitude of Implicit Memory by calculating the Priming Score that is the rate of correct completions, namely completions with words of the Studied list, relative to casual completions that is completions with words that are “correct” in principle but belong to the not studied list.

4.4. EEG recording and preprocessing

During the Study session, electroencephalogram (EEG) was recorded by means of a 40-channel DC-coupled monopolar amplifier (Nuamps, Neuroscan, Compumedics, El Paso, TX). Scalp EEG signals were acquired with a sampling rate of 500 Hz by electrodes having contact impedance below 5 k Ω and referenced to the FCz potential. For the offline preprocessing, a re-referencing to the average potential of the two earlobes (A1 and A2) was done in order to obtain monopolar-like recordings (Piarulli et al. 2010). Two horizontal, at the outer left and right canthus, and two vertical, above and below the right eye, electrooculogram (EOG) electrodes, recorded eye movements and blinks. Horizontal (electrodes at the outer left and right canthus) and vertical (electrodes above and below the right eye) electrooculogram (EOG) signals recorded eye movements and blinks, respectively. Movement artifacts and temporary declines of signal quality

(instability or loss of contact with the scalp during recordings) were detected in line with previous specifications (Menicucci et al., 2009). Signals were filtered both with a comb notch filter (10th order, 50 Hz-centered, 3.5 Hz-wide) and with a bandpass one (0.1–20 Hz). Each filter was applied in both forward and reverse directions so as to avoid time biases. Ocular artifacts were detected by computing a moving-window cross-correlation between the frontal EEG channels and the EOG signals: high values of cross-correlation marked putative ocular artifacts. We considered cross correlation values as high if greater than a threshold derived by computing the same moving window cross-correlation between phase-randomized surrogated (Theiler, 1994) frontal EEG channels and the EOG. Furthermore, we considered only noteworthy artifacts, that is those producing on frontal EEG channels fluctuations greater than 50 μ V and lasting a least 70 ms. Trials containing signal segments marked as artifact-corrupted were tagged and, after visual inspection, most of them were definitively discarded (Artoni et al., 2012a). The visual inspection was aimed at retain the trials in which above-threshold EEG signals associated to high EEG/EOG correlation was due to genuine high-amplitude brain-related signals contaminating EOG: the most common case was that of full-fledged alpha spindles that ‘contaminated’ EOG.

During the recordings the onset of each auditory stimulus was digitally marked so as to enable the extraction of ERP epochs (trials). Each trial corresponded to the portion of signals ranging from 100 ms before to 1000 ms after the onset of the stimulus.

4.5. Spatial decomposition of event-related potentials

The characterization of ERPs recorded during the Study session comprises, in turn, two main sections: a conventional, averaging-based, ERP analysis and an ICA-based ERP decomposition (Menicucci et al., 2013).

Within the conventional ERP analysis a grand-average of the trials for each electrode was performed. The assumption was that averaging enhances activity triggered by stimulus compared to spontaneous ongoing activity. Before averaging, each trial underwent two standardizations: a baseline correction by subtracting the mean value of the trial pre-stimulus [–0.1, 0] s and an inter-subjects EEG amplitude normalization which was performed by dividing the trials of each subject by the mean standard deviation of the related pre-stimulus intervals (Menicucci et al., 2013, Martini et al., 2012; Rodriguez et al., 1999).

The ICA-based ERP decomposition consisted in deriving a special combination of the different EEG channel signals in order to separate components originating from different brain sources (the ERP components). Thus, the ICA-based ERP decomposition modeled ERPs as the sum of temporally independent components (that is with statistically independent time course) arising from distinct, spatially fixed, brain processes. This stands on the fact that the ERP derives from the superposition of stimulus-related brain activations distributed over the cortex. More in details, the components were estimated by means of a data-driven approach named group-level ICASSO (Himberg et al. 2004), specifically modified for the treatment of ERP trials (ErpICASSO) and whose computational details are available in a previous paper of ours (Artoni et al. 2012b).

4.6. Decomposition of event-related potentials

In order to model ERPs as the sum of temporally independent components we determined the number of underlying components, as well as to estimate their time course and scalp distribution. The model estimation was based on an ICA approach (Hyvarinen 1999) and consists in estimating a demixing matrix W that, applied to trials, transforms them into the independent components. We used ErpICASSO since it is an ICA algorithm that provides a reliability measure of each component. Specifically, deriving the confidence interval for each time point of each component allowed us to assess the statistical significance of the differences between the activity templates, namely to compare the different conditions.

ErpICASSO was applied on the concatenated trials of all subjects in order to perform a unique group-level extraction of the common components. This approach implied the assumptions that all subjects have similar brain responses, and possible differences in attention and or imagery capabilities could modulate the time course of the components without affecting their scalp distribution. In this way we had a direct matching of components between subjects thus avoiding complex semi-automatic clustering procedures for associating the corresponding components related to different subjects (Jung et al., 2001). As just mentioned, both the determination of the independent components and the reliability measure of each component were based on the same approach. In detail, ErpICASSO consists of performing multiple decompositions, each one of different samples of the trials obtained with a bootstrapping approach (DiCiccio and Efron, 1996) and within ErpICASSO, the components extracted from each decomposition are used to estimate the component variability, based on the principles of bootstrapping. The result of this procedure is the quality index Q , which ranges from 0 to 1: high value of Q indicates high component reliability (Himberg et al., 2004). The number of underlying components was determined based on the Q index as we assumed that the right order of the model (namely that with the proper number of components) should have high-quality components, that is components with high Q . Effectively, we assumed that the order of the model better describing the ERP data was that with the higher Q index averaged over components. This criterion has been previously verified on ad-hoc created multichannel

electromyography datasets with a predetermined number of embedded sources with added noise (Artoni et al. 2013).

4.7. Extraction of activity template with ErpICASSO

Each component has two main features: the activity template and the activation map over the scalp. ErpICASSO provides a direct estimate of the latter since it corresponds to the weights in the mixing matrix (inverse matrix of W) while for the former a further step is required since ErpICASSO directly provides the time course of the reference components by demixing the trials. The activity templates are obtained by averaging the reference components over trials. In particular, as ErpICASSO was performed at group level from each component we derived two distinct activity templates, one from PS-P and one from PS-N. Effectively, the extraction of the two activity templates relied on the correspondence between concatenated trials and component time course, and each activity template was derived as the average of the trial-by-trial segmented component epochs related to each group, separately.

For each ICA decomposition, ErpICASSO generates a demixing matrix, a set of independent components and, consequently, two activity templates per component. Thus, in order to provide a unique result representative of the multiple decomposition runs, ErpICASSO selects a run with outputs corresponding to an intermediate solution with respect to the cluster of estimates of the same component (see the methodological works –Himberg et al., 2004; Artoni et al., 2012b – for further details). In this sense ErpICASSO provides the best activity templates (BATs) with the related activation map (inverse of the matrix W). We also derived a point-by-point confidence interval of each BAT from the point-by-point variability of the activity templates related to the component. This allowed assessing statistically significant differences in BAT time courses between groups.

Differences between groups were investigated by performing unpaired t-tests on each sampling point.

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