ACTIVITAS NERVOSA SUPERIOR

COMPONENT N400 CAN BE DEMONSTRATED IN THE EVENT-RELATED POTENTIAL FROM A SIMPLE AUDITORY REACTION TIME PARADIGM

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Abstract

The aim of the present study was to propose and test an experimental design enabling to synchronize simultaneously both signal- and movement-related processes in a simple sensorimotor task. Seventeen healthy volunteers were asked to flex their right wrist after the presentation of an auditory signal (tone burst of 1kHz, 200 ms duration; intersignal interval between 7-9 s). In total, 2179 collected records were divided according to the same length of the signal-movement interval into 38 time groups and the average curve for each time group was calculated. The event-related potential from all these time groups exhibited two prominent negative waves over the frontocentral region. The first one, with a mean peak latency of 110 ms, exhibited characteristics of the N1 component of cortical auditory evoked potential. Analysis of data concerning the second negative wave pointed to its dependence on both sensory and motor underlying processes. The increasing slope of the wave represented its sensory part. It started in all the time groups approximately at the same time points and was not influenced by variations of S-R interval. An estimate of its peak latency was approximately 400 ms. In time groups with S-R intervals shorter than 300 ms this N400 component was located on the time axis after the movement, i.e. when the instructed task had already been accomplished. The next portion of the second negative wave, i.e. the slow increase of amplitude to its peak value and the following decrease of the wave, exhibited a clear-cut dependence on movement.

Key words: Auditory Event-related Potentials; Externally Triggered Movement; Human Auditory N400 Component

INTRODUCTION

Adults respond substantially faster than children in simple auditory reaction time tasks. Experimental investigation provides evidence that the auditory reaction time decreases with increasing age until adolescence with a much steeper slope than the central motor conduction time (Fietzek et al., 2000). This finding suggests that a more effective "short-cut" from the auditory system to motor cortical areas involved in movement programming evolves during childhood. In a recent study of Bender et al. (2006) such possibility was demonstrated in electrophysiological data. These authors analyzed the maturation of frontal contributions to the auditory event-related potential (ERP) following the warning stimulus in a contingent negative variation task in

healthy subjects. The result of high-resolution current source density mapping and a spatiotemporal source analysis of the EEG data indicate an early SMA (supplementary motor area) activation elicited by meaningful auditory stimuli from about 12 years of age on. The very early occurring intracerebral ERPs induced by a meaningful auditory stimulus in the motor cortex of adult subjects (Kukleta et al., 2006) suggested the existence of the same pathway that enable fast sensorimotor coupling. These findings initiated a detailed thinking about two alternative ways of responding to meaningful auditory stimuli – the first one with full cognitive elaboration of the signal, i.e. inner ear - subcortical auditory nuclei - temporal auditory cortices - motor associative areas, and the second one which makes use of a direct

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pathway from the subcortical auditory nuclei to the motor cortices.

The ERPs present a good instrument for investigation of the time characteristics of sequential mental processes because they are able to reflect neuronal activation in the millisecond range. Steep negative shifts from the baseline are the most striking features of these potentials. They are thought to represent synchronous activation of a sufficient number of neighboring cortical neurons (Birbaumer et al., 1990). Experimental data suggest that temporal and partially also spatial characteristics of steep negative shifts over the brain areas are related to task behavioral response (Becker & Kristeva, 1980; Deecke & Lang, 1996; Lang et al., 1994; Weinberg et al., 1974).

The ERPs are usually obtained by averaging of sufficient number of EEG responses to the same event. This procedure is imposed by an ongoing background activity which covers induced potential changes. An important aspect of this procedure is the fact that its efficacy is dependent on the precision of synchronization of averaged events. Due to common variability in the timing of single components in complex mental events, the efficacy of averaging is usually sufficient around the chosen trigger of averaging and sharply decreases on both sides of this time point. Thus in experiments where ERP induced by an externally triggered movement either stimulus-related or movement-related potential changes can be visualized reliably.

The present paper is focused on ERPs changes related to internal cognitive processes that occur during experimental tasks before movement, as for instance the selection of one from the possible alternatives of movement, the decision to start mental counting or the decision to start self-initiated movement (Kukleta et al., 1996; Kukleta, Rektor, & Lamarche, 1996; Kukleta & Lamarche, 1996, 1998). The aim of the present study was to propose and test an experimental design that enable to synchronize simultaneously both signal- and movement-related processes in a simple reaction time task. The idea of the experimental design was to collect a sufficient number of trials, to divide them into groups according to the same length of signal-movement intervals and, finally, to create averaged curves for each of these groups. In this way we expected to obtain complex ERP depicting the sequence of the underlying neuronal processes from stimulus onset to movement execution. The experiment was done in 1999 and its preliminary results were published shortly afterwards in the form of short communication (Kukleta & Lamarche, 2000). At present time, our interest in these results was revivified in context with the above-cited findings of very early EEG responses to a meaningful auditory stimulus in the frontal lobes and age-dependent frontal involvement in the processing of such stimuli.

METHODS

Seventeen right-handed volunteers (5 men, 12 women, aged 22 - 25 years) participated in the experiment representing repetitive flexing of the right wrist after the presentation of an auditory stimulus. The participants were informed that the experiment was not designed to measure the reaction time and that it is not necessary to respond as quickly as possible. The auditory signal was a tone burst of 1 kH, 200 ms in duration, and 70 dB, generated by a loudspeaker placed at a distance of 1.5 m from the participant's head. One hundred and fifty single records were collected from each participant in five blocks separated by 3-min resting periods. An inter-signal interval varied randomly between 7 and 9 seconds. During the recording the subject laid comfortably on a bed. The EEG from Fz, Cz, C3, C4, and Pz sites, EOG from the supraorbital and zygomatic sites, and EMG from the flexor digitorum communis were recorded using surface electrodes (Nihon Cohden cups). Each data frame had the duration of 5 sec and was digitized at a frequency of 200 Hz. The auditory stimulus was triggered by a system interrupt, working with a microsecond precision, exactly 1000 ms after the start of the recording. All the records were taken with binaural reference. The data processing was performed offline using artifact-free EEG records only. In all experimental blocks, the analysis window was 5 sec. All the recorded signals from these periods were amplified by a Nihon Kohden electroencephalograph (Neurotop, bandpass 0.03 - 100 Hz) and transferred online through a Cambridge Electronic Design interface (CED 1401) to a compatible PC. The stored data were elaborated offline using the SIGAVG program (Cambridge Electronic Design Limited). Before the EEG analysis, both the EOG traces and the EMG trace were displayed on the screen and thoroughly inspected. All the records with signs of eyelid movements and saccadic eye displacements were excluded from further analysis (the selection was done by an experienced person). Then, the intervals between the signal onset and the EMG onset were measured, groups with the same signal-movement (S-M) intervals were created, and the average wave for each time group was calculated. The program package Statistica '99 (StatSoft, Tulsa, USA) was used for statistical evaluation of the results.

RESULTS

In total, 2179 single records from 2550 collected records were used for the analysis. The remaining recordings (14.5 %) were removed owing to the presence of eyelid or eye movement artifacts within the pre-movement period. As demonstrated by the evaluation of EMG parameters, the subjects responded to the signal by slow movements (Fig. 1 presents the average curves calculated from all the 2179 records).
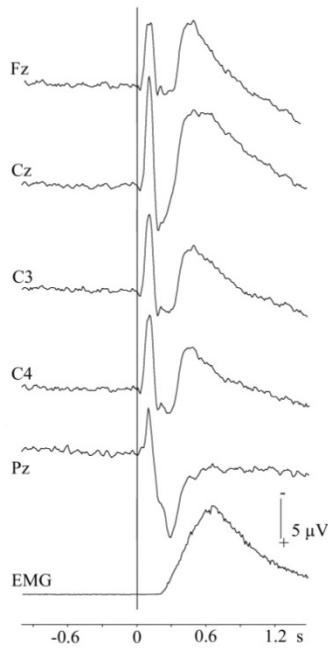
Figure 1. Grand average of surface potentials accompanying the wrist flexion triggered by an auditory signal. Electrode positions Fz, Cz, C3, C4, and Pz, rectified and squared EMG from the flexor digitorum communis. The zero point on the time axis corresponds to the onset of auditory signal. In total, 2179 records were averaged.

As evident, the execution of the experimental task was accompanied by ERP comprising under frontocentral electrodes two prominent negative waves – the first with peak latency about 110 ms and the second with peak latency about 480 ms. Under the Pz electrode the second negative wave was absent. The great variability of the S-M intervals (range from 0.2 s to 1.1 s) enabled distribution of the records into 38 time groups. The S-M difference between two neighboring groups was 0.01 s in 34 consecutive time groups, and 0.03 s in groups Nos. 35, 36, and 37. Group 38 gathered the remaining trials with the longest S-M intervals (the numbering of the groups started with the shortest S-M interval). The mean number of records in one group was 56.9±22.2 (median 51.5, minimum 21, maximum 126 records). On average 13 subjects contributed by their trials to a time group (minimum 8, maximum 17 subjects). As can be seen in Fig. 2, the average curves computed from the time groups exhibited comparable negative waves as grand average curves. A more detailed analysis of these averages was focused on the second negative wave. We were especially interested in the latency of its onset, its maximal amplitude and latency of this peak, the velocity of potential change measured by the increase of amplitude within a 0.1 s window after the onset (slope val-

ue), and in the relationship between the wave and the movement.

The onset of the second negative shift could be measured in time groups from Fz, C3, and C4 records. On the curves derived from the Cz electrode, the beginning of the second negative shift was not discernible in approximately one quarter of the cases, so these curves were not included in the analysis. Results presented in Fig. 2 and Table 1 show that the second negative shift occured at the same time after the stimulus. A comparison of the onset latencies in subgroups, created by division of time group values into four subgroups with increasing S-R intervals, corroborated this possibility statistically ($p > 0.05$ in all the cases; ANOVA, $F_{(3, 32)} = 1.7$, 1.7, and 0.7, respectively). An interelectrode comparison of the mean onset latencies did not show any difference, either ($p > 0.05$ in all the cases; t-test for dependent samples). As far as the interelectrode relationship of onset values was concerned, the partial correlations obtained by the multiple regression test revealed a highly significant association between C3 and C4 (beta 0.76; $p < 0.0001$). No other significant association among the onset values was demonstrated. Taken together, these findings suggest that the second negative shift was not related to the movement execution.

The peak latency of the second negative wave and its maximum amplitude were measured on smoothed average curves. Their mean values are presented in Table 1. Significant interelectrode differences of peak latencies were found between Cz and Fz, Cz and C3, Cz and C4 and between Fz and C3 ($p < 0.0025$ in all the cases; t-test for dependent samples). The peak values exhibited a high correlation with S-R intervals in all the cases ($p < 0.00001$; Spearman correlation). The maximal amplitude was the largest under the Cz electrode. Using t-test for dependent samples highly significant differences between all pairs of electrodes were found ($p < 0.0001$) only with exception of C3 – C4 ($p > 0.05$).

The mean slope value of the second negative shift, which was measured after its onset in the time window of 0.1 s, was the largest under the Cz electrode. A t-test for dependent samples demonstrated significant differences between all pairs of electrodes ($p < 0.0001$ in the case of Fz-C3, Fz-C4, Cz-C3, Cz-C4; $p < 0.003$ in the case of Fz-Cz) with the only exception of C3-C4 values ($p > 0.05$).

In a comparison of the mean amplitudes calculated in the time windows of 0.1 s before and 0.1 s after the movement onset (indicator: increase of negativity), eight time groups with the shortest S-M intervals were excluded because of overlapping of the 0.1 s premovement period with the first negative wave. This comparison demonstrated a significant increase of negativity in the second window ($p < 0.0001$ in all the cases; t-test for dependent samples).
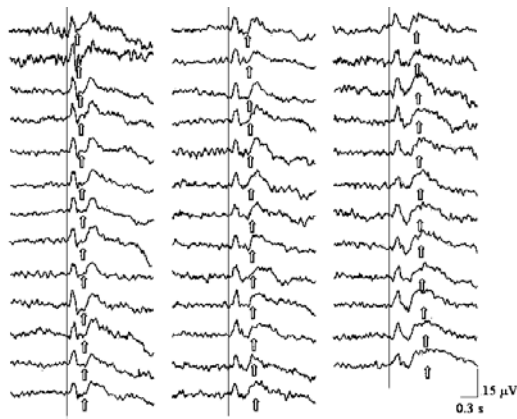
Figure 2. Average curves of 38 time groups. Electrode position Fz. The zero point on the time axis corresponds to the onset of auditory signal; flashes under each curve mark the onset of movement-related EMG activity. The number of averaged records is mentioned in the text.

As evident, the largest increase of negativity was observed under the Cz electrode. All the interelectrode differences with the only exception of the Fz-Cz pair were statistically significant (p<0.001 in all the cases; t-test for dependent samples).

Table 1. Mean values and standard deviations of selected parameters of the second negative wave following the auditory signal over various brain areas

	Fz	Cz	C3	C4
Wave onset (ms)	311±28	-	302±33	305±30
Wave peak (ms)	494±67	546±114	515±82	514±107
Min-max values (ms)	400-690	410-930	410-720	410-940
Spearman R (N 38)	0.82	0.83	0.87	0.81
Wave amplitude (µV)	-8.9±2.0	-11.0±2.9	-6.4±1.9	-5.9±2.0
Slope value (µV/s)	90.1±20.9	99.6±21.3	77.2±17.9	81.0±18.4
Increase of negativity (µV)	3.2±2.8	4.6±2.4	3.0±1.8	2.3±2.3

Note: Spearman R – correlation between wave peak and S-R interval

DISCUSSION

The study demonstrated that both stimulus-related and movement-related neuronal activations underlying the simple sensorimotor task could be synchronized simultaneously and that ERP depicting the whole task could be created. The ERP resulting from the procedure consisted of two negative waves. The first one, with a mean peak latency of 110 ms, exhibited characteristics of the N1 component of the cortical auditory evoked potential which is known to be generated in temporal auditory cortices and other regions (mainly frontal). A detailed analysis of the data concerning the second negative wave pointed to its dependence on both sensory and motor underlying processes. The increasing slope of the wave represented its sensory part. This negative steep shift of the baseline started in all the time groups approximately at the same time points and was not influenced by variations of the S-R interval. As the onset of

the EMG activity varied in a range from 200 ms to 1100 ms (mean S-M value 389±118 ms), the movement had been initiated before the onset of the negative shift in the time groups with short S-M intervals and after the onset of this shift in the time groups with long S-M intervals. The next portion of the wave, i.e. the slow increase of the amplitude to its peak value and the following decrease of the wave, exhibited a clear-cut dependence on the movement. An estimate of the peak latency of the sensory part of the second negative wave which was derived from the minimal peak amplitude values of the wave and other relevant data was approximately 400 ms. The components of auditory ERPs of such a latency are classified as slow or late and are thought to be related to lexical integration processes (Brown & Hagoort, 1993; Kutas & King, 1995; Osterhout & Holcomg, 1992). The presence of N400 component of auditory ERP in a sensorimotor task using a very simple auditory stimulus was not surprising when we realized how the behavioral significance of the stimulus was obtained. The stimulus was given a meaning through verbal instruction, which had to be retained in the working memory and, very probably, repeatedly recalled throughout the experiment.

When comparing the supposedly sensory and motor ERP components obtained in the experiment, a noticeable difference in their configuration became evident. Rapid shifts of the baseline, which were typical of sensory components, were missing in the motor component. The most probable reason for this discrepancy was the fact that the synchronization efficiency of sensory and motor processes was not the same. The onset of the auditory stimulus which was used for sensory synchronization was triggered by a system interrupt, working with a microsecond precision, exactly 1000 ms after the start of the recording. On the other hand, movement synchronization was based on grouping trials with approximately identical S-R intervals and the precision of synchronization was of the order of milliseconds in this case.

From a more general point of view, N400 auditory component in time groups with the S-R interval shorter than 300 ms, i.e. when the instructed task had already been accomplished, opens the discussion on the behavioral significance of this ERP component and also related mental processes. Although meaningless in the cases mentioned, its presence in all such time groups suggested that it represented an obligatory step in the sequence of underlying events. In the context of the above-cited discussion about alternative pathways of sensorimotor coupling, the question about alternative modes of cognitive evaluation of stimulus significance inevitably arises. It is probable that a short-cut pathway allowing very rapid response to an auditory stimulus makes use of a different, less time-consuming way of its evaluation. Our experimental setting using the same

stimulus of unequivocal significance from the beginning to the end of the experimental session, which was moreover repeated many times, belonged probably to such situations. The presence of an N400 component in all time groups, regardless to the phase of the task, could simply show that a full-value evaluation of the stimulus meaning cannot in any case be omitted.

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