

EEG evidence of gender differences in a motor related CNV study

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Summary In the present study gender differences related to the contingent negative variation (CNV) were investigated. A series of two acoustic stimuli was presented to participants across a wide age range. The first stimulus was consistent throughout the experiment whereas the second one was either a high frequency or a low frequency tone. One of them had to be answered by a button press (go condition) the other did not require any response (nogo condition). Between the first and the second tone there was a time period of two seconds in which the CNV appeared as a slow negative potential shift. Within this episode data were analysed with respect to gender differences.

Statistical analysis revealed topographical differences between men and women in go conditions for both left and right index finger movements. Differences were found over frontal regions where women showed higher brain activity than men and over temporo-parietal regions where men produced higher brain activity than women. In order to explain the fact that only in “go” conditions significant gender differences occurred we introduce the phenomenon of implicit learning. Due to implicit learning assumed predictions related to S2 might have occurred from time to time. This is so, because a 50% chance for one of two different stimuli to occur leads to reasonable assumed predictions after two or more stimuli of a kind occurring in a series. The present data now provide evidence that if such assumed prediction or expectancy is directed towards an upcoming demand to act then brain activity is subject to gender differences. Further studies providing controlled sequences of “go” conditions versus “nogo” conditions have to be done to prove this idea true.

Keywords: Gender differences, CNV, EEG

Introduction

The Contingent Negative Variation (CNV) was first discovered by Walter in 1964. Since then many investigators modified the original design within various studies in

order to augment the understanding of this phenomenon. Typically, a CNV paradigm is composed of two stimuli, the first of which represents a warning stimulus (S1) whereas the second one represents an imperative stimulus (S2). The time period between the two stimuli is known to reflect preparation time or expectancy related to S2 accompanied by brain activities identified as slow negative potentials and called CNV. It has been shown that different factors do influence the CNV such as type of task (e.g. Leynes et al., 1998; Birbaumer et al., 1981; Simons et al., 1979), transcendent experiences (Travis et al., 2000) or level of catecholamine (Katsumi et al., 1999). Individual attitude as reflecting a personal state of readiness to respond in a characteristic way to a stimulus was already known by Walter et al. (1964) to be a reason for affecting the CNV in its appearance. Besides that, there were various other influences found to somehow interfere within a CNV paradigm most of which are related to attention. The CNV has further been split into two separate phases related to different functional aspects. An early phase of the CNV is thought to reflect an orienting response related to S1 (warning stimulus) (Loveless and Sanford, 1974; Rohrbaugh et al., 1980) whereas a later phase has been suggested to reflect motor preparation (e.g. Rockstroh et al., 1991). A very recent study by Lukhanina et al. (2006) investigated the effect of levodopa treatment in patients with Parkinson’s disease on the CNV. Treatment with levodopa was followed by a significant increase particularly in the late CNV phase. Lukhanina et al. (2006) stated that their results provide evidence for the important role of structures supporting

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both direct motor control and mental functions in forming both phases of contingent negative variation. However, the greater effect of levodopa on the late phase of the CNV suggests that the efferent system of the basal ganglia has a greater role in generating the late phase than in organizing the early phase of the contingent negative variation. Gomez et al. (2003, 2004) suggested a contribution from motor cortices but also posterior cortices to the genesis of the late CMV (Contingent Magnetic Variation) phase. The late CNV phase has even been compared with the readiness potential (Bereitschaftspotential, BP) in that it might also reflect motor preparation processes similar to the so called BP2 component (Deecke, 2000; Cui et al., 2000). However, it has to be emphasised that the late CNV phase has also been attributed to sensory related brain activities at posterior sites (Brunia and van Boxtel, 2001).

For the present study we were interested in investigating the CNV with respect to gender differences. It is well known that there are various differences between men and women related to cognitive functioning in principle including differences related to lateralization of brain activity (McGlone, 1978, 1980; Volf and Razumnikova, 1999), attention (Aartsen et al., 2004), language and mental rotation of three dimensional objects (Rescher and Rappelsberger, 1999; Volf and Razumnikova, 1999; Corsi-Cabrera et al., 1997). In the Longitudinal Aging Study Amsterdam (Aartsen et al., 2004) the results pointed to gender differences related to the level of memory functioning. Walla et al. (2001) found evidence of neurophysiological gender differences in a word recognition study measuring magnetic field changes in the brains of their participants. They interpreted their results as demonstrating that women and men use different mental strategies to achieve the same cognitive goal. In general, women were shown to have larger evoked responses in the brainstem and in the cortex compared to men (Hetrick, 1996). Other studies demonstrated eventual indications of a higher inter hemispheric collaboration in women (Corsi-Cabrera et al., 1997). To date, not much is known about gender effects related to the CNV. In 1992, Howard et al. reported about gender differences in both the degree and the direction of CNV asymmetries. Unfortunately, only 3 electrodes were used in this study and topographical differences could not be investigated to a greater extent. Facing the fact that gender differences exist in various cognitive tasks and facing the lack of more detailed descriptions of gender effects related to the CNV our idea arose to test the hypothesis that the CNV varies as a function of gender by using more electrodes in order to enable a topographical analysis.

Materials and methods

Participants

All 20 participants (10 females) reported being free of neurological or psychiatric disorders and took part as paid volunteers. They were all right handed (Oldfield, 1971) and had normal, or corrected to normal vision. None of them had known auditory deficits. The mean age of male participants was 44.8 years (SD = 20.9; range from 22 to 75) and the mean age of female participants was 43.5 (SD = 20.3; range from 20 to 72). Statistical analysis revealed no significant difference between both means ($p = 0.895$; $F = 0.018$) confirming reliability related to any group comparison.

Procedure

Participants were seated in a comfortable armchair in a quiet room during the experiment. Adjustable hand rests were adapted in order to permit a pleasant stay during the experiment. Participants were instructed not to move any other body part except the required index finger during the experiment, so that any other motor activities could be excluded as good as possible. Both index fingers had to be placed on the buttons to be pressed if required by the respective S2.

The present study included a go/nogo experimental paradigm with acoustic stimuli. The first tone (warning stimulus S1) was always the same whereas the second tone (imperative stimulus S2) varied between two different frequencies (high or low; 50% chance for each frequency). Tones were presented for 0.2 s. The inter stimulus interval (S1–S2) was 2 s, started with the onset of S1 and ended with the onset of S2. Inter trial interval varied randomly between 4 and 10 s. For S1 no answer was required but subjects were instructed to respond with a button press to the high frequency S2 and to inhibit a response to the low frequency S2. They were asked to respond as quickly and accurately as possible either by left or by right hand index finger movements according to instructions given to the subjects prior to the start of an experiment. Two alternating blocks each consisting of 160 trials were provided. A central fixation point was presented before each trial. In one block the instruction was to move the index finger of the right hand and in the other block the instruction was to move the index finger of the left hand. Trials were only accepted when responses were given between 100 ms and 1 s after presentation of S2. Trials were excluded when subjects responded to S1 or not correctly according to S2. In order to include a trial it was also required to have a correct EMG signal corresponding to the button press trigger.

Recordings

Brain potentials were continuously recorded during all single sessions. Ag/AgCl electrodes at 15 scalp locations were used. They were placed at the positions F7, F3, Fz, F4, F8, T7, C3, Cz, C4, T8, P7, P3, Pz, P4 and P8 according to the international 10–20 system (Jasper, 1958). Simultaneously, eye movements were registered from temples as well as above and below one eye for horizontal and vertical eye movement registration. Blinks were corrected offline automatically by a computer program developed by Lindinger (1990). Additional electrodes were used as ground and reference electrodes. Signals were referenced to linked mastoids. EMG was registered from the left and the right forearm. Electrodes recorded activity changes on musculus flexor digitorum superficialis with a sampling rate of 250 Hz. The EMG signal was bandpass filtered from 0.1 to 300 Hz. The EEG was registered in DC mode with a sampling rate of 250 Hz and band-pass filtered from DC–100 Hz. Electrode impedance was 3 k Ω or below for all electrodes. The EEG system was a computer supported 64-channel DC amplifier (Lindinger, 1990). The baseline period started 1 s before S1 and ended at its onset. For the time period between S1 and S2 three different time windows were differentiated, *early* (from 0.5 s to 1 s after the onset of

S1), *middle* (from 1 to 1.5 s after the onset of S1) and *late* (from 1.5 to 2 s after the onset of S1). Within each time window the relevant mean amplitude was calculated in order to serve as a dependent variable for each subject, condition and electrode.

Statistics

All included trials were grouped according to four conditions of interest: Left index finger and S2 requiring no response is referred to as condition 1 (left/nogo), left index finger and S2 requiring a motor response is referred to as condition 2 (left/go), right index finger and S2 requiring no response is referred to as condition 3 (right/nogo) and finally, right index finger and S2 requiring a motor response is referred to as condition 4 (right/go). All physiological data were normalized and following statistical analysis was done with normalised data only. This is so, because the focus of the present study is to analyse gender specific effects on brain activity and for that purpose any differences with respect to EEG amplitude between women and men have to be cancelled out. This is important because due to simple morphological gender differences such as bone thickness amplitude differences occur. Normalisation was done by setting the maximum value (potential) of each condition, each participant and each interval to 1 and correspondingly the minimum value of each condition, each participant and each time interval to 0. All values in between were recalculated relative to each maximum and each minimum value.

Mean amplitude values of all three temporal windows served as dependent variables for a 3-way Analysis of Variance (ANOVA; repeated measurements design). Thereby, the within subject factors “*condition*” for kind of condition (4 levels: left/nogo, left/go, right/nogo, and right/go),

“*time*” for latency of brain activity (3 levels: early, middle and late) and “*electrode*” for electrode location (15 levels: see electrode positions) were defined. In order to calculate a gender effect the between subject factor “*gender*” was introduced (2 levels: women and men). All data were Huynh-Feldt corrected. In addition, linear contrasts were calculated to compare conditions and to point out particular conditions with significant gender differences. To correct the increased probability of a type I error (FW error), a Bonferroni test was made.

Low resolution brain electromagnetic tomography (LORETA)

For localising brain activity LORETA (Low Resolution Brain Electromagnetic Tomography) was applied. The LORETA algorithm solves the inverse problem by assuming related orientations and strengths of neighbouring neuronal sources (however, without assuming a specific number of generating sources). The core assumption of the algorithm – which is generally well supported by animal single-unit recordings (Pascual-Marqui et al., 1994) – is mathematically implemented by finding the “smoothest” of all possible current density distributions.

Results

ANOVA (analysis of variance)

As stated above all data were normalised prior to statistical analysis. Consequently, any resulting significant gender

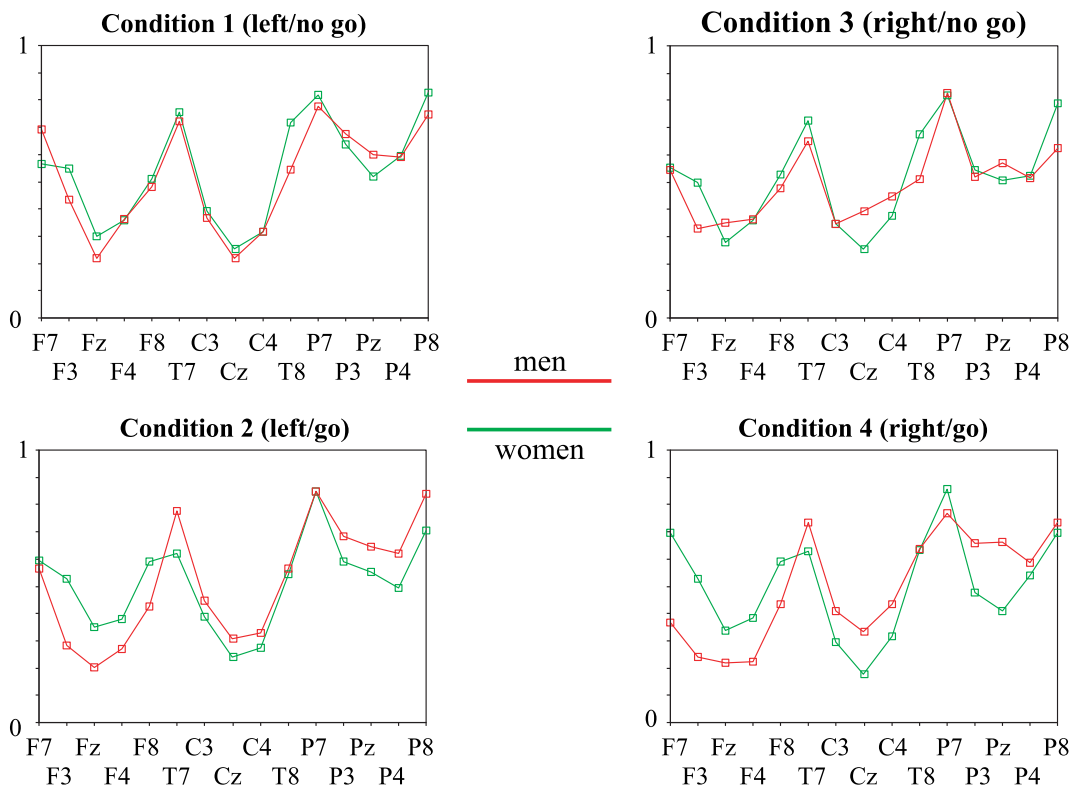


Fig. 1. Linear contrast diagrams (normalised data) for all conditions of interest showing activity distributions at all electrode locations for women and men. Note that conditions 2 and 4 which are both “go” conditions show that women produced more dominant frontal activities whereas men produced more dominant temporo-parietal activities

effects are qualitative or in other words functional in nature. Statistical analysis of normalised data revealed a significant condition * electrode * gender interaction ($p = 0.002$; $F = 1.974$; Huynh-Feldt corrected) pointing to gender specific functional brain topography differences between conditions. No significant interaction with “time” occurred.

Linear contrast

Results of linear contrast calculations revealed that condition 2 (left/go) and condition 4 (right/go) are particularly subject to gender differences ($p = 0.006$; $F = 9.913$). For both “go” conditions significant gender differences were found over frontal and parietal electrode sites. Women were found to show more activity over frontal and less activity over parietal electrode sites compared to men. Figure 1 presents all diagrams related to linear contrast calculations for conditions 1–4. In addition, Fig. 2 shows relative amplitude values (normalised data) for one frontal electrode (F3) and one parietal electrode (P3) both demonstrating pronounced differences between women and men (see Fig. 1) for conditions 2 and 4 illustrating that the female

group elicited more dominant brain activity over frontal sites compared to the male group whereas the male group elicited more dominant temporo-parietal brain activity compared to the female group in both “go”-conditions (conditions 2 and 4). Figure 2 also demonstrates that differences are similar across time. In both females and males brain activity over frontal areas (represented by F3) slightly increases over time whereas brain activity over parietal areas (represented by P3) slightly decreases over time. Figure 3 shows raw EEG curves for all electrode locations related to condition 2 (left/go).

LORETA

According to the above mentioned ANOVA calculations no significant interaction with the factor “time” occurred. Consequently, LORETA was applied for conditions 2 and 4 across all three time intervals from 0.5 to 2 s for women and men separately. Figure 4 shows that women demonstrate maximum activity over frontal sites (especially left frontal) whereas men show maximum activity over left parietal (temporo-parietal) brain areas.

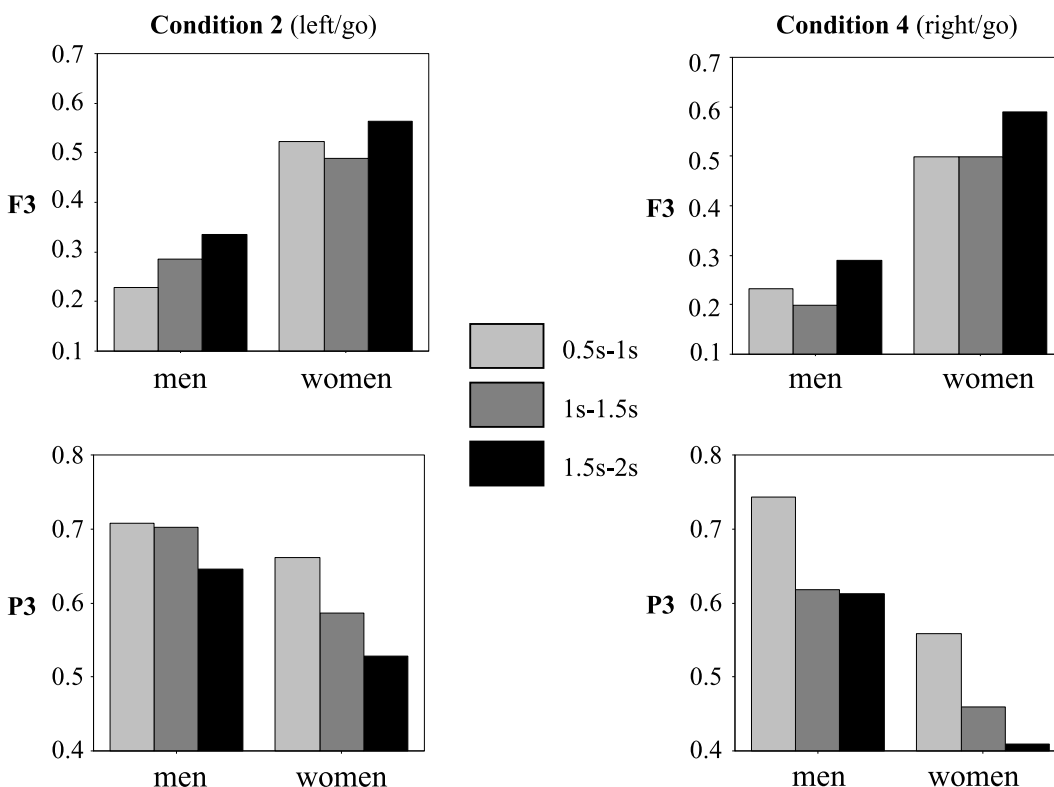


Fig. 2. Bar diagrams showing relative activity distributions (normalised data) at selected electrode sites F3 and P3 for all three time intervals and for women and men separately. Note that across all three time intervals at F3 women produced higher brain activity than man whereas at P3 men produced higher brain activity than women

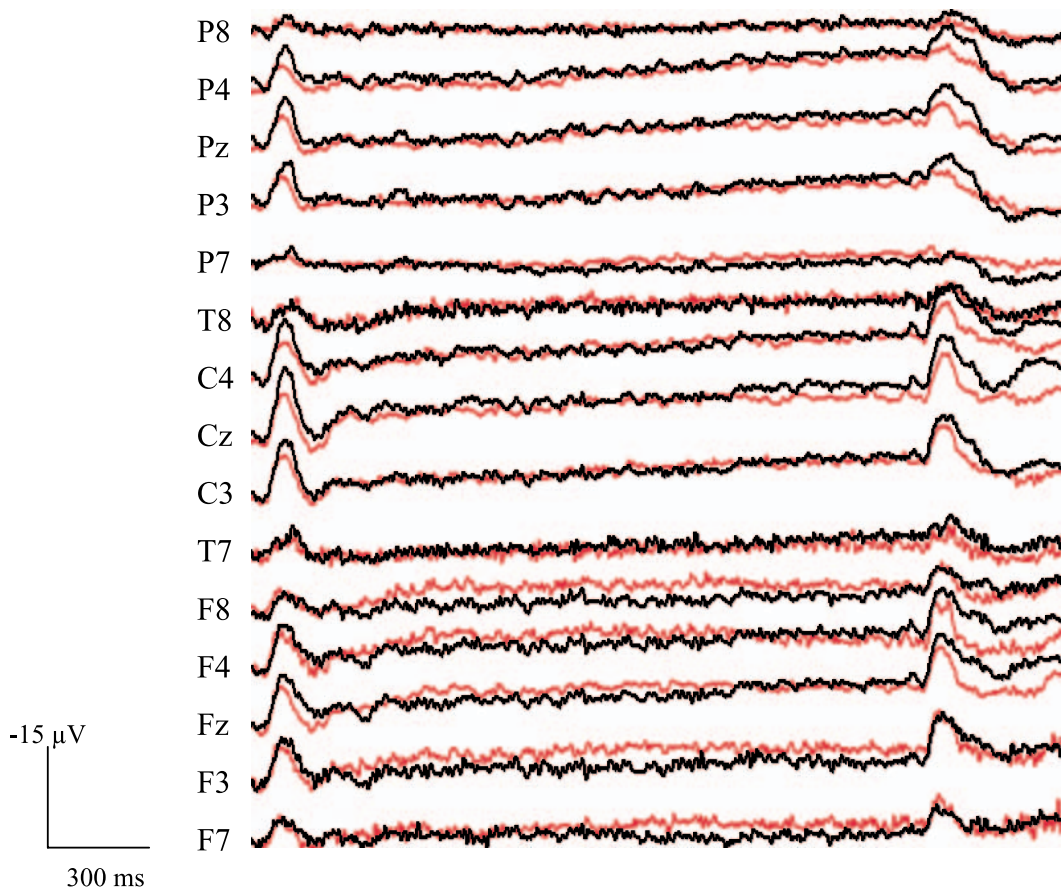


Fig. 3. Grand average EEG curves showing raw data of all electrodes in condition 2. Curves in black were produced by males and curves in red were produced by females. Gender differences over frontal electrodes are quite obvious whereas significant differences over parietal electrodes can only be seen after normalisation or LORETA calculation (see Fig. 4)

Discussion

The aim of this study was to investigate differences related to gender during an acoustic CNV paradigm (S1 and S2) related to a go/nogo motor response task. The acoustic stimulus S1 represented the so called warning stimulus whereas the acoustic stimulus S2 represented the imperative stimulus telling each study participant to move (go) or not to move (nogo) the index finger of either the left or the right hand. Significant gender differences related to brain activities (functional differences; normalised data) which occurred between S1 and S2 (from 0.5 s after S1 until onset of S2) were found in condition 2 and in condition 4. In both conditions the task was to “go” for the respective index finger movement. In those conditions frontal electrodes showed higher brain activation in women (especially left frontal) whereas temporo-parietal and central electrodes showed higher brain activation in men (Figs. 1, 2, and 4). The applied LORETA approach proved to be an excellent tool for visualising brain activities across the entire cortex

(tomography). Figure 4 nicely shows the dominant frontal activity distribution in women and the dominant temporo-parietal activity distribution in men for both “go” conditions across the whole time range from 0.5 to 2 s after S1. In contrast, conditions 1 and 3 in which no movements were required did not show any differences between women and men. This finding is interesting because the relevant time intervals which were analysed were before S2 which contained the essential information to move or not to move. Therefore, at any time before S2 participants did not yet know whether they had to make a motor response or not. At first this phenomenon seems strange and lacking any understandable logic but as can be seen later there is a scientifically acceptable interpretation which we will explain in detail.

According to Hetrick (1996) we can assume, that there is no gender specific difference related to sensory processing of S1 because of its acoustic nature. In addition, all data were analysed starting from 0.5 s after S1, which means

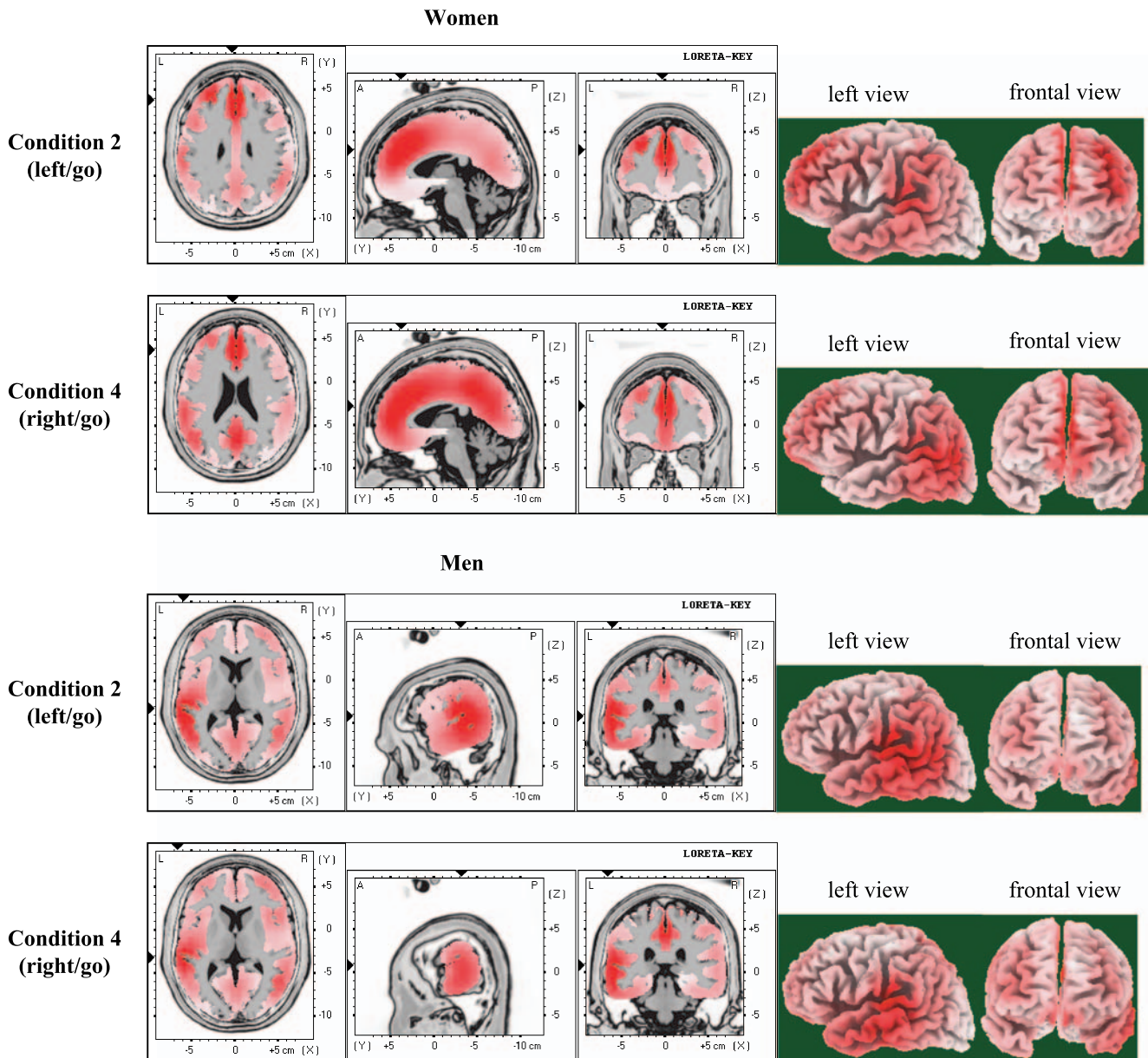


Fig. 4. LORETA (Low Resolution Brain Electromagnetic Tomography) showing activity distributions across the whole time period from 0.5 to 2 s after stimulus S1. Note, that women show maximum activity at frontal areas compared whereas men show maximum activity at temporo-parietal areas

that sensory processing related to S1 should be finished by then and all exogenous components reflecting early acoustic information processing do not anymore influence the appearance of the EEG curve characteristics (Alain and Izenberg, 2003). It is therefore concluded that the gender related effect which we found is related to the CNV which has previously been defined as reflecting brain activity associated with expectancy for S2 (Walter et al., 1964) in such a paradigm as it was used in the present study. Now, for such expectation it is suggested that the phenomenon of so called implicit (subconscious) learning (for example

Reber, 1989) might have been involved. Due to implicit learning a kind of “directed expectation” towards one or the other S2 stimulus might have occurred. In the present study an equal number of acoustic S2 signals was chosen for “go” and for “nogo” conditions. Although actually random a certain pattern (two or three S2 of one kind) at least from time to time might have elicited “directed expectation” to one or the other S2 stimulus. In light of this idea an additional post hoc analysis was done. Analysing the sequential structure of S2 conditions according to classical information theory the random sequences used

in our study indeed enabled a certain amount of “directed expectation”. The frequency of possible S2 pairs (go–go, go–nogo, nogo–go, nogo–nogo) was uneven (although random) and the amount of entropy (information: $H = \sum_{i=1}^n -1/n * \sum_{i=1}^n \text{ld } n_i$) resulted in $H_2 = 1.85$ bit/symbol instead of 2.00 bit for an even distribution. Taking triples of S2 into account the amount of entropy again gets reduced to $H_3 = 2.66$ bit instead of 3.00 bit. We can therefore assume that implicit learning theoretically could have led to a certain assumed prediction of S2 stimuli and hence could have elicited “directed expectation” towards one or the other S2 (go or nogo). Cleeremans (1993) provided evidence to support the notion that participants become increasingly sensitive to sequential structure and that this sensitivity extends back as far as three elements in a sequence.

In spite of this interesting background there still is the question of why such differences related to gender occurred only in “go” conditions? Accepting the above mentioned idea of “directed expectation” we could simply conclude that only if implicit learning leads to expectation for S2 being a “go” stimulus then gender differences do occur. This would mean that the CNV although mainly associated with expectancy does also include brain activity related to the motor system which in fact has been previously described as already mentioned in the introduction section. It might be that at least parts of the S2 sequence were assumed to be predictable and in case of a “go” prediction requiring a motor response women and men used different functional strategies reflected by different brain activities. The lack of a significant “time” effect on neurophysiological differences between women and men leads to the further interpretation that brain activity related to S1 orienting responses (reflected by early CNV stages) as well as brain activity related to motor preparation (reflected by later stages of the CNV) differed with respect to gender.

Having this idea in mind, the findings of the present study can further be interpreted. In short, the main finding was higher frontal activation in women and higher temporo-parietal activation in men (Fig. 4). This could lead to the interpretation that women include more frontal brain areas related to implicit learning and motor preparation reflecting higher level cognitive involvement compared to men. Especially, the prefrontal cortex is known to be involved in cognitive control, willful behaviour and anticipate attention (Brunia and van Boxtel, 2001; Miller, 2000). On the other hand, men demonstrate higher temporo-parietal brain activity contribution reflecting brain processes related to rather sensory processing. However, the present study highlights

gender differences related to the well known CNV and does add to its understanding because it provides evidence of a possibly close connection to another well known potential, the so called “Bereitschaftspotential” (BP; Kornhuber and Deecke, 1965) which is meant to reflect the preparation of a voluntary movement. Finally, it would be interesting to design an experiment with controlled occurrence of certain sequences of S2, first in order to confirm our present finding and second to define functional links between the CNV and the BP more precisely.

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