ABNORMAL ERPS AND HIGH FREQUENCY BANDS POWER IN MULTIPLE SCLEROSIS

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Event-related potentials (ERPs) and power spectral density (PSD) were registered during an auditory-oddball paradigm in 11 MS patients. These patients showed a decrease in the amplitude of P2 and N2 components and a delayed P3 latency compared to control subjects suggesting that the attentional orienting mechanism in the auditory modality is affected in MS. The PSD analysis showed that MS patients exhibited an increased power in theta, beta, and gamma bands. The combined analysis of frequency and time domain suggested diverse phenomena that occurred in the MS patient group related with the EEG background or the motivational status.

Keywords  auditory oddball, ERPs, frequency bands, multiple sclerosis, P3, PSD

INTRODUCTION
Evaluation of cognitive deficits in multiple sclerosis (MS) is currently a controversial issue. Different authors agree that a high percentage of the patients (65%) shows deterioration of their cognitive abilities (Ellger et al., 2002). However, the lesions observed in the MRI examination do not allow inferring which will be the neuropsychological scores of a patient with MS (Camp et al., 1999). Moreover, neuropsychological assessment comprises multiple mechanisms in each test and cannot evaluate precisely independent steps in
the information processing. In this sense, the detection of subtle cognitive impairment in MS can be evaluated even better with techniques such as event-related brain potentials (Leocani & Comi, 2000; Anlar et al., 2003). In the specific case of the auditory modality, several studies have demonstrated the presence of abnormalities in the latency and amplitude of ERP components in different samples of patients with MS; these include delays in the latency of the N1, P2, N2, and P3 components (Aminoff & Goodin, 2001); delay only in the latency of P2, N2, and P3 (Gil et al., 1993); no changes in either of these components (Gerschlager et al., 2000); a latency increase of P3 (Ellger et al., 2002); a latency increase and an amplitude decrease in P3 (Polich et al., 1992); no changes in P3 latency (Sailer et al., 2001), or even the absence of the P3 component (Boose & Cranford, 1996).

On the other hand, few studies have been carried out in the spectral domain and related with this pathology. In a recent study (Leocani et al., 2000), a spectral analysis of the human EEG showed a decrease in intra- and interhemispheric coherence for alpha and theta bands. They concluded that cognitive impairment in MS is related with demyelination in cortico-cortical projections within the white matter immediately underlying the cortex producing a partial functional disconnection between the implicated areas. In the case of the high frequency bands of the EEG spectrum the authors have not found any studies relating these bands with cognition in multiple sclerosis.

In other pathologies, a model has been recently suggested (Lee et al., 2003) where the gamma band is proposed as an indicator of the degree of structural integrity and how is affected in pathologies like the schizophrenia. These authors suggest that an increase in the gamma power could reflect disorganization in the brain activity typical in this pathology. Another study (Coutin-Churchman et al., 2003) that reviewed different neuropathologies and their expression in the EEG spectrum has shown that a clear sign of cortical atrophy is related with an increase of power in the high frequency bands and decrease in the lower ones.

To the best of the authors’ knowledge, there is not any study that combines spectral and temporal domain analysis with cognitive paradigm in multiple sclerosis. Therefore, the main purpose of the present study is to analyze the modulations in the EEG spectrum and ERPs components as realized before (Gómez et al., 1998) in patients with relapsing-remitting multiple sclerosis during a well-known experimental paradigm (oddball). The results will allow observing possible relationship between frequency and time modulations and suggesting some hypothesis about the etiology of this disease using combined EEG techniques.
MATERIAL AND METHODS

Eleven patients (7 females; age 37 ± 8 years) with relapsing-remitting MS form without motor disturbances and a Expanded Disability Status Scale (EDSS) score smaller than 3 (Kurtzke, 1983) participated in the study. Exclusion criteria were relapse within 1 month, use of steroids or psychoactive drugs in the last month, or presence of clear signs of depression. Patient data were compared with a group of 11 healthy subjects of similar age, gender, and educational rate. The protocol was approved by the local ethical committee (in accordance with the Declaration of Helsinki). After complete explanation of the study to the subjects, written informed consent was obtained.

ERPs were recorded during a typical auditory oddball task. Subjects listened to standard (1000 Hz, probability = 0.80) and deviant (2000 Hz, probability = 0.20) tones presented binaurally through headphones, with a 50-ms plateau and a 10-ms rise/fall time, in random order. The interstimulus interval was 1 second and the intensity of auditory stimuli was of 70 dB. Two blocks with 200 trials each were presented. Subjects seated in a sound-attenuated room and they were instructed to count target deviant tones. At the end of each block, the subjects were asked how many target tones they counted. Electrodes attached at the external canthi of the eyes and the inferior and superior areas of the ocular orbit recorded horizontal and vertical eye movements, respectively. The EEG was recorded from 13 scalp electrodes located according to the 10–20 system: Fz, Cz, Pz, F4, C4, P4, O2, T6, F3, C3, O1, P3, T5. All the EEG recordings were made with reference to the right mastoid and re-referenced off-line to both mastoids. The EEG channels were amplified with a 0.01–100 Hz band-pass filter and digitized at a rate of 250 Hz. Artifact rejection software rejected automatically trials with amplitudes higher than ±75 µV in any EEG or EOG channel.

Average amplitude measurements were made with respect to the –100–0 ms baseline for the P2 and N2 components in the latency windows defined by ±10 ms from the peak observed for the component in the grand-average across subjects (P2: 165–185; N2: 185–205). These data were obtained from the difference wave resulting from subtracting the standard ERP from the deviant one. The latency of the P3 component was measured also in the grand average of the deviant stimulus individually for each subject by two different experimenters at the highest positive peak between 275 to 500 ms at the Cz recording.

To obtain the spectral analysis, a Fast-Fourier Transformation (cosine window, 10%) was applied to the same epochs that were averaged to obtain the
ERP data. The resulting power spectral density was computed for the following bands: delta (0.5–4 Hz); theta (4–8 Hz); alpha (8–12 Hz); beta (12–30 Hz), and Gamma (30–45 Hz).

The statistical analysis of P2-N2 amplitude and P3 latency were computed in the Cz electrode and using a \( t \)-paired test. In the case of the spectral bands, the analysis was performed comparing the potential values for individual subjects in the midline electrodes: Fz, Cz, and Pz, by means of an ANOVA with the following factors: Group (2 levels: Patient and Control) and Electrode (3 levels: Fz, Cz and Pz). For spectral analysis, a logarithmic transformation was applied to all values to get a normal distribution in these data (Gotman, 1990). Post-hoc analyses were performed using the Tukey honest significance difference test.

Additionally, neuropsychological exploration was carried out in the patient group to detect attentional impairments. The tests employed were Stroop and SDMT (Symbol Digit Modality test).

RESULTS

The behavior response (counted targets during the experiment) from patients and controls was always over 90%. In the MS group, the latency of the P3 showed a significant delay compared to the control group (\( t = 3.000, p = .007 \)) (see Table 1A). The mean value for the P3 latency in MS patients was 372 ms ± 36, and of 337ms ± 14 for the control group. These latency values imply a delay of the P3 peak of 35 ms in the MS group with respect to the control group at the Cz electrode (see Figure 1).

For the P2 and N2 latencies, the paired \( t \)-test revealed smaller amplitude for both components in the MS group with respect to controls (\( t = 2.80; p = .011 \)). The average value for the whole time window (comprising P2 and N2: 165–205 ms) in MS group was –1.07 ± 2.27 \( \mu \)V and of –3.95 ± 2.56 \( \mu \)V for controls (see Figure 1).

Concerning the spectral content, an ANOVA was calculated for each frequency band, the results showed that Beta (12–30 Hz) and Gamma (30–45 Hz) had a statistically significant higher power spectral density in MS patients with respect to controls (see Figure 2 and Table 1B). Beta band showed a statistical difference for the group factor \( F(1,10) = 9.21, p = .012 \). All electrodes were different among groups but the biggest difference was obtained in the frontal electrodes (MS-beta: 1.63 ± 0.61 \( \mu \)V\(^2\), Control-beta: 1.05 ± 0.22 \( \mu \)V\(^2\)).
Figure 1. A and B. Event-related brain potentials (ERPs) elicited at Cz in the deviant and standard conditions for MS patients and the control group, respectively. C. Difference wave (deviant—standard) for both groups. In all cases, arrows indicate the incoming of the tone.
Table 1. A) Mean amplitude for P2-N2 complex expressed in µV and P3 latency indexed in ms. ERPs calculations were obtained in Cz derivation. B) Power Spectral Density (PSD) of all bands: delta (0.5–4 Hz), theta (4–8 Hz), alpha (8–12), beta (13–30 Hz) and gamma band (30–45 Hz) (µV²) in healthy controls and Multiple Sclerosis patients. Values for PSD calculations were obtained in Fz derivation. (S.D. = Standard Deviation).

1A. ERP values

<table>
<thead>
<tr>
<th></th>
<th>Controls</th>
<th>Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>P2-N2 Mean</td>
<td>−3.95</td>
<td>−1.07</td>
</tr>
<tr>
<td>S.D.</td>
<td>2.56</td>
<td>2.27</td>
</tr>
<tr>
<td>P3</td>
<td>337</td>
<td>372</td>
</tr>
<tr>
<td>S.D.</td>
<td>14</td>
<td>36</td>
</tr>
</tbody>
</table>

1B. Spectral values

<table>
<thead>
<tr>
<th></th>
<th>Controls</th>
<th>Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Delta Mean</td>
<td>6.11</td>
<td>5.82</td>
</tr>
<tr>
<td>S.D.</td>
<td>1.32</td>
<td>1.73</td>
</tr>
<tr>
<td>Theta Mean</td>
<td>3.53</td>
<td>3.72</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.80</td>
<td>1.39</td>
</tr>
<tr>
<td>Alpha Mean</td>
<td>3.58</td>
<td>4.82</td>
</tr>
<tr>
<td>S.D.</td>
<td>1.68</td>
<td>2.20</td>
</tr>
<tr>
<td>Beta Mean</td>
<td>1.05</td>
<td>1.63</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.22</td>
<td>0.61</td>
</tr>
<tr>
<td>Gamma Mean</td>
<td>0.49</td>
<td>0.90</td>
</tr>
<tr>
<td>S.D.</td>
<td>0.13</td>
<td>0.67</td>
</tr>
</tbody>
</table>

For gamma band the main factor “group” was also statistically significant $F(1,10) = 7.71$, $p = .019$. A descriptive analysis showed a bigger value for patients compared to control and again localized in frontal areas (MS-gamma: 0.90 ± 0.67 µV², Control-gamma: 0.49 ± 0.13 µV²).

Figure 2. Difference wave of power spectral density at Fz resulting from subtracting the values of the control group from the MS patients.
Delta and alpha were more pronounced in MS subjects than in controls but none of these spectral band values were statistically different.

Neuropsychological evaluation showed low values in both tests for the patient group (Stroop: 40.40 ± 11.97; SDMT: 36.20 ± 12.05). The Stroop and SDMT patient scores were 1 and 1.4 standard deviation lower compared to the control population.

As a summary, the amplitude of the P2 and N2 components showed a decrease in MS patients when compared to control subjects. Moreover the latency of the component P3 showed a delay for the patient group with respect to controls. The EEG frequency analysis showed an increase of MS patients compared to the control group in the spectral power for the theta, beta, and gamma bands.

**DISCUSSION**

The latency delay of the P3 component is in agreement with that observed in previous studies (Polich et al., 1992; Ellger et al., 2002). This phenomenon could be explained by a delay in the latency of the previous components P2-N2 (further explanation later). However, it is also possible that this delay reflects deterioration of the frontal-central P3 subcomponent that forces a delay in all the P3 complex comparing MS patients and the control group (see Figure 1A). Regarding the physiological basis, this delay may be explained by an impairment of neural conduction related to the demyelination process (Fuhr & Kappos, 2001).

With regard to P2 and N2 components, the study found decrements in the amplitude in the difference wave (deviant—standard) instead of delay effects described in previous studies (Aminoff & Goodin, 2001; Gil et al., 1993). The latency analysis was not carried out in the present study because in some patients these components were not clearly identifiable as referred in other studies (Onofrj et al., 1991). However, if the latencies of the P2 and N2 components of MS and control groups are compared in the ERP grand average for the deviant stimulus a delay of 35 ms between the two groups is observed (see Figure 1). This observation suggests that the latency delay observed in P3 may not be intrinsic to this component per se, but may take place during the time window of P2 and N2 components, that is, along the whole processing pathway. The reduced amplitude observed for these components may result also from the deteriorated inputs (neuronal death) to the brain areas analyzing the auditory stimuli.
The P2 and N2 components have been related with mechanisms involved in the orienting of the auditory attention (Ahveninen et al., 2002). Given that the selected patients had an EDSS score smaller than 3, the present data indicate that the possible impairment of these mechanisms controlling orienting of auditory attention in MS can occur even in early phases of the pathological condition, and could remain undetectable by means of clinical exploration.

The possible impairment of this orienting mechanism is supported by studies pointing out that the automatic reorientation to changes in the acoustic context is located in regions of the temporal and frontal cortex (Yago et al., 2001), and the frontal cortex is usually affected in MS patients (Zorzon et al., 2001; Sailer et al., 2001).

Indeed, the Stroop test indicated that the patient group showed lower values compared to the control group in what it has been associated with cortical atrophy (Suderlund et al., 2004). Moreover, Symbol Digit Modality Test (SDMT) also reflects attentional impairment in these subjects (Klonoff et al., 1991; Jansen & Cimprich, 1994) supporting our conclusions with ERP data.

With regard to the frequency analysis, a significant increment of the high frequencies of the EEG appeared (beta: 12–30 Hz and gamma: 30–45 Hz), contrasting with the absence of statistically significant differences in delta and alpha bands. It must be remarked that for the beta and gamma frequency bands there are not significant contributions from the ERPs (Vázquez-Marrufo et al., 2001), and the obtained modulation in MS patients must be considered as frequency changes rather than phasic ERPs changes. In the same sense, it must be remarked that given that the ERPs in controls have larger amplitude than in MS patients, any contribution from the ERPs to the power spectral density values should be in the opposite sense to those obtained in the theta, beta, and gamma bands.

It is also possible to discard a contribution from alpha harmonics as responsible for the changes in the power of the high bands because no statistical difference was found for the alpha band between controls and multiple sclerosis patients.

The statistical analysis did not show differences in the spectral power for beta and gamma bands among the anterior (Fz) and posterior (Pz) locations. However, it has been proved that beta and gamma bands have frontal generators (Gómez et al., 2004). Therefore, the increment of spectral power in high-frequency bands in MS subjects could indicate a fronto-cortical atrophy related with partial white matter disconnection as was described by different authors (Sailer et al., 2001; Piras et al., 2003) but not strictly related with depression symptoms as has been suggested by other authors (Bakshi et al.,
These results are also in concordance with the integrative neuroscience model (Lee et al., 2003) that considers an increment of gamma synchrony as a good indicator of disorganization between cerebral structures in the schizophrenia.

An alternative explanation comes from the fact that the increase of gamma power has been related also with the motivational status (Karmos et al., 2002). Indeed, MS patients try to compensate their impairment with a higher arousal during the experiment what carries usually a fatigue status at the end of the experiment. Nevertheless, the wide range for the increment in the spectral domain (12–45 Hz) could reflect more a general deterioration of the high frequency generators than a concrete effect of the motivational status.

About the relationship between ERP and spectral domain, the decrease in the phasic ERP auditory responses and the increased EEG background would be that the latter offers a noisy background to the neural inputs to ERP generators, debilitating the already impaired neural response due to the demyelination process. Therefore, the combined effects of damaged neural connection and increased background activity would produce the reduced auditory response in MS patients.

It seems that both techniques (ERPs and spectral power) agree to show an abnormal frontal function probably caused by the lost of connectivity reflected as an increase of the high frequency bands power in frontal regions and an impaired auditory orienting mechanism indexed by alterations in amplitude and latency in ERPs components (P2-N2 and P3).

The combined study with ERPs and Power Spectral Density can suggest an alternative hypothesis about the etiology of the disease and help each other in the interpretation of results with different physiological meaning.

REFERENCES


