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Published in:
Pain Medicine

DOI:
10.1093/pm/pnaa068

Publication date:
2020

Document Version:
Accepted author manuscript

Processing of laser evoked potentials in patients with chronic whiplash associated disorders, chronic fatigue syndrome and healthy controls: a case control study.

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Funding source

This work was financially supported by ME Research UK, a United Kingdom based national Charity funding biomedical research into Myalgic Encephalomyelitis / Chronic Fatigue Syndrome.

Conflict of interest

Eva Huysmans is a PhD fellow funded by FWO. The authors have no conflicts of interest to declare.

Running title: LEP in chronic pain patients

Abstract

Objective

Laser evoked potentials (LEPs) are among the reliable neurophysiological tools to investigate patients with neuropathic pain as they can provide an objective account of the functional status of thermo-nociceptive pathways. The goal of this study is to explore the functioning of the nociceptive afferent pathways by examining LEPs in patients with chronic whiplash associated disorders (cWAD), patients with chronic fatigue syndrome (CFS) and healthy controls (HC).

Design

Case-control study.

Setting

A single medical center in Belgium.

Subjects

LEPs of twenty-one patients with cWAD, nineteen patients with CFS and eighteen HC were analysed in this study.

Methods

All participants received brief nociceptive CO_2-laser stimuli applied to the dorsum of the left hand and left foot while recording brain activity with a 32 channel EEG. LEP signals and transient power modulations were compared between patient groups and HC.

Results

No between group differences were found for stimulus intensity, which was supraliminal for Aδ- fibers. The amplitudes and latencies of LEP wave components N1, N2 and P2 in patients with cWAD and CFS were statistically similar to those of HC. There were no significant differences between the time-frequency maps of EEG oscillation amplitude between HC and both patient populations.

Conclusions

EEG responses of heat-sensitive Aδ fibers in patients with cWAD and CFS revealed no significant differences with responses of HC. These findings thus do not support a state of generalized CNS hyperexcitability in those patients.

Keywords: chronic pain, electroencephalography, laser evoked potentials, central hyperexcitability

Introduction

A wide variety of chronic pain syndromes, like fibromyalgia (1) or irritable bowel syndrome (2, 3), are frequently associated with presumed alterations in peripheral and/or central processing of nociceptive information. One of the phenomena that is frequently evoked in this context, is central sensitization (CS). CS can be defined as an increased responsiveness of nociceptive neurons in the central nervous system to their normal or subthreshold afferent input (4).

Several questionnaires (for example the Pain Sensitivity Questionnaire (PSQ) (5) or the Central Sensitization Inventory (CSI) (6)) are available for assessing symptoms believed to be associated with CS, while quantitative sensory testing (QST) can be used to gain insight in the sensitivity of different pathways for processing nociception (7). Another option for evaluating sensitization processes is an electrophysiological assessment by the use of laser evoked potentials (LEPs) (8). QST is the better tool for evaluating hyperalgesia and related pain mechanisms, while functional abnormalities of the central nociceptive system are better evaluated with LEPs (9).

LEPs can be used to conduct a functional evaluation of the nociceptive afferent pathways. LEP amplitudes are typically reduced in patients with neuropathic pain, whereas in patients with a non-neuropathic pain condition, normal or enhanced LEP amplitudes are observed (10, 11). The observation that LEPs may be enhanced in some chronic pain conditions could, at least in part, be related to CS (10, 12). Up to now, findings regarding LEPs in patients with enhanced pain sensitivity are inconclusive. An increase in LEP amplitude has been documented in patients with fibromyalgia (13-15) and chronic tension type headache compared to healthy controls (16). However, in a large study with 199 patients with fibromyalgia, a reduced N2-P2 amplitude was found compared to healthy controls (17). In patients with migraine, there was no difference in latency nor amplitude of LEPs compared to controls (18).

In hyperalgesia ascribed to increased sensitivity, LEP amplitudes are expected to be enhanced (9). Two frequently diagnosed chronic pain syndromes with presumably sensitization of the central nervous system (19, 20), are chronic fatigue syndrome (CFS) and chronic whiplash associated disorders (cWAD). Ten to fifty percent of the persons involved in an acute whiplash trauma develop chronic pain and pain-related disability (21-23). cWAD is a debilitating condition characterized by multiple symptoms such as chronic neck pain, headache, dizziness, tinnitus, concentration disturbances, sleep difficulties and fatigue (22-25). CFS has a worldwide prevalence ranging from 0.2% to 2.6% (26) and is defined as a medically unexplained disabling fatigue that persists for more than 6 months (27). Fatigue is often accompanied by a generalized hyperalgesia and, more generally, hypersensitivity to a variety of sensory stimuli, concentration difficulties, post-exertional malaise or joint pain (28-30).

In patients with cWAD and CFS, hyperalgesia (i.e. increased responsiveness of nociceptive neurons in the CNS would enhance the pain response to noxious stimuli (31)) to pressure-evoked pain and electrically-evoked pain has been revealed (19, 32, 33). Evidence thus suggests that cWAD and CFS are associated with functional changes in the central nervous system, including changes that lead to enhanced responses to nociceptive input (19, 34). However, a real evaluation of the nociceptive afferent pathways (wherefore LEPs are better suited) has not yet been performed in patients with CFS and cWAD. This was the aim of the present study, in which we recorded LEPs in 21 patients with cWAD, 21 patients with CFS and 20 healthy controls. We predicted that, in both patient groups, increased LEP amplitudes might be observed, in line with the findings that LEP amplitudes may be enhanced in patients demonstrating hyperalgesia, ascribed to increased sensitivity (9). In addition to characterizing the elicited responses in the time domain, we also performed a time-frequency analysis to characterize the transient stimulus-evoked modulations of oscillatory EEG activity (event-related synchronization and desynchronization) (35).

Methods

Subjects

Twenty-one patients with cWAD, twenty-one patients with CFS and twenty healthy persons participated in the present study (Table 1). Healthy controls had a median age of 46.8 years (Q1-Q3: 27.73 – 51.21). Patients with cWAD and CFS had a median age of 45.8 years (Q1-Q3: 40.41 – 51.08) and 43.9 years (Q1-Q3: 35.90 – 48.41) respectively. Thirty-three participants obtained a higher education degree, twenty participants successfully finished secondary school as highest education and three participants primary school. For three participants, the educational level is not known. All participants provided written informed consent prior to participation. The study was conducted according to the revised Declaration of Helsinki (1998). The study protocol was approved by the Ethics Committee of the University of Antwerp (approval number B300201214521).

Patients with cWAD were recruited through advertisement on the website of our research group (Pain in Motion), from the medical database of the local Red Cross medical care unit and from a medical database obtained from previous studies. The criteria for inclusion were experiencing chronic symptoms resulting from a whiplash trauma (e.g., motor vehicle accident or fall) and fulfilling diagnostic criteria of WAD grade I to III as defined by the Quebec Task Force classification (23). Chronicity was defined as complaints persisting for at least 3 months. Subjects were excluded if they were classified as WAD grade IV (23).

Patients with CFS were recruited through advertisement on the website of our research group, via private physician practices and from a medical database obtained from previous studies. Only patients who were diagnosed by a physician were eligible to participate. Diagnosis established by the physician was performed according to the 1994 Center for Disease Control and Prevention criteria (27). This

implies that any other medical condition possibly explaining the debilitating fatigue and pain was excluded prior to establishing the diagnosis of CFS. Hence, all patients within the CFS group underwent comprehensive cardiovascular, neurological, psychiatric and haematological screening.

Healthy sedentary control participants were recruited among the university college staff, family members and acquaintances of the researchers. Controls were not allowed to participate if they ever experienced a whiplash trauma, suffered from persistent pain or neck-shoulder-arm symptoms, or had sought medical help for neck-shoulder-arm symptoms in the past 6 months. Healthy control participants were excluded if they were suffering from an acute or chronic disease, or when they were experiencing pain on the day of the assessment.

All participants were asked to discontinue non-opioid analgesic and anti-inflammatory drugs 48 hours before testing. This interval was chosen based on ethical consideration and based on the fact that analgesic effects are mostly limited in time (36). Participants were instructed to avoid physical exertion and to refrain from consuming nicotine, alcohol and caffeine 24 hours before testing. Additionally, participants were excluded if they were pregnant, or if they suffered from any cardiovascular or neurological disease. Finally, to limit confounding of the study findings, we strived towards three groups with a comparable age distribution.

Demographics of the three groups were compared with Kruskall-Wallis tests and chi-squared tests, depending on the normality and variance of the data.

Procedure

Study participation required one study visit at the Institute of Neuroscience (Université catholique de Louvain, Brussels, Belgium) during which LEPs were recorded. Before the start of the assessments,

patients with CFS and cWAD completed three questionnaires: the Pain Catastrophizing Scale (PCS), the Beck Depression Inventory (BDI) and the Pain Disability Index (PDI). Then, the EEG recording started. Three blocks of 10 laser stimuli were delivered on the left foot and left hand (randomized order; total number of stimuli: 60). The inter stimulus interval ranged between 10 and 15 seconds in order to avoid habituation effects to painful stimulation (37). A visual warning signal lasting 200 ms preceded each laser stimulus and was initiated 1 second before stimulus administration (Fig. 1). Eight-hundred ms after the laser stimulus, a second visual signal of 200 ms was delivered. After this second visual stimulus, participants had a resting period until the next visual stimulus was delivered. These two visual stimuli were presented because, in addition to recording LEPs at rest, the experiment also included two other conditions in which LEPs were recorded while participants prepared for a mental calculation task and a movement task (raising the right index finger). In these conditions, the first visual stimulus represented a warning stimulus and the second visual stimulus an imperative stimulus (38-40). Only LEPs recorded in the rest condition were analysed in this manuscript.

At the end of each block, participants were asked to rate the intensity elicited by the laser stimuli using a visual analogue scale (VAS) ranging from “no detection” (VAS = 0) to “maximum pain” (VAS=100) at the appropriate ends. At the middle of the scale (VAS=50) an anchor marked the borderline between the non-painful and the painful domain of sensation (41). Kruskal-Wallis tests were used to compare VAS values between the three groups.

Sample size calculation

An a priori sample size calculation was performed for the whole protocol, based on results from previous studies (40, 42). For reaching an effect size of 0.5, at least 20 subjects per group (total of 60

subjects) were required to detect a within, between and interaction effect of repeated measures Anova, at the 5% level with a power of 80%.

Questionnaires

The Pain Catastrophizing Scale questionnaire was used to measure the level of pain catastrophizing. This questionnaire consists of 13 pain-related cognition items that needed to be scored on a 5-point Likert scale (0 = not at all, 4= all the time) (43). Scores ≥30/52 indicate a clinically relevant level of catastrophizing (44). The internal consistency, test-retest reliability and validity are acceptable (45, 46).

The Beck Depression Inventory was used to evaluate depression in all participants. Total scores range between 0 and 63, with higher scores indicating more severe depression. This questionnaire is a reliable and valid tool for the assessment of depressive symptoms in chronic pain patients (47).

The Pain Disability Index provides an indication of the impact of pain on daily living activities. This questionnaire consists of 7 items scored on an 11-point Likert scale (0 = no disability, 10 = completely disabled). Total scores range from 0 to 70, with higher scores indicating higher levels of perceived disability. Differences of 8.5 to 9.5 points are considered clinically relevant (48). The Dutch version of this questionnaire is a valid tool with good internal consistency and test re-test reliability (49).

Total scores on the questionnaires were used in the analyses. Comparisons between patient groups were made with independent t-tests and Mann Whitney U tests depending on the normality and variance of the data.

Laser stimulation

Laser stimuli were delivered by a CO_2 laser designed and built in the Department of Physics (Université catholique de Louvain, Louvain-La-Neuve, Belgium) (41). Stimuli were applied on the dorsum of the left hand (C6-C7 skin dermatomes) and left foot (L5-S1 skin dermatomes). The CO_2 laser system generates a highly collimated infrared beam (wave length: 10.6 um). The power output is continuously adjustable between 1 and 25 W. Heat pulse duration was 20 ms. Laser beam diameter was 4 mm. The laser stimulus is highly reproducible (variation **<**1%). The stimulation site was visualized with a He-Ne laser beam aligned with the CO_2 laser beam. To avoid skin burns and nociceptor fatigue (50), the location of the impact on the skin was moved slightly between 2 successive stimulations. In order to obtain reproducible LEPs, stimulus intensity was adjusted to elicit a clear painful pinprick sensation on the left hand. For each subject, the intensity threshold to elicit detections related to the activation of Aδ- fibers was determined once (before applying the 60 stimuli) by measuring the reaction time to the laser stimulation. A series of increasing and decreasing stimulus intensities were given to the participants on the left hand dorsum. Participants were asked to push a button as fast as possible when they felt the laser stimulation. Laser stimulus intensity was set at the intensity that repetitively elicited a sensation detected with a reaction time below 600 ms. With this procedure, the stimulus intensity was supraliminal for Aδ- fiber activation, as confirmed by the reaction times compatible with peripheral nerve conduction velocities within the range of myelinated small fibers.

LEP recording

Subjects were seated in a comfortable chair in a silent room. They were asked to relax muscles and gaze at a light diode. LEPs were recorded from 32 Ag-AgCl scalp electrodes placed according to the International 10-20 system for electrode positioning and sampled at a sampling rate of 1000 Hz. The digitized signals were referenced to the average of all scalp electrodes. A bipolar recording of electro-

oculographic (EOG) signals was simultaneously measured using surface electrodes placed diagonal over the right eye to monitor eye-blink artefacts. The electrode impedance was kept below 10 kΩ with a target below 5kΩ.

Time-domain analysis of LEPs

Offline data pre-processing was performed with the Letswave 6 EEG toolbox (http://letswave.org). EEG recordings were filtered (0.3-30 Hz, Butterworth filter) and segmented into epochs of 6 seconds (-3 to +3 s relative to the laser stimulus onset). Electroculographic artefacts were removed using an Independent Component Analysis. Independent Components having a frontal scalp distribution and a time course compatible with eyeblink artefacts were removed (1-5 ICs). Afterwards, all epochs were visually inspected to remove remaining artefacts (± 100 μV). Finally, epochs were baseline corrected, using the time interval from -1.5 to -1 seconds as reference (i.e. before the onset of the warning visual stimulus). For each subject, epochs were averaged according to the location (foot versus hand). LEP components were identified on basis of their latency and polarity and labelled according to Valeriani et al. 2012 (10). Peak latencies of N2 and P2 components amplitudes were measured at the vertex (Cz versus average reference (51)) and defined as the largest negative and positive deflection, between respectively 150-350 ms and 300-500 ms (52). The LEP N1 component was evaluated at the T8 electrode with the Fz electrode as reference (51) within a targeted time frame of 100-300 ms poststimulus (52, 53).

The amplitude and latencies of the N1, N2 and P2 waves of LEPs obtained following stimulation of the hand and foot were compared with two sample t-tests to evaluate differences in each population versus the control group. In total 4 tests were performed: CON versus CFS on the hand (1), CON versus CFS on the foot (2), CON versus cWAD on the hand (3) and CON versus cWAD on the foot (4). Bonferroni correction was used to account for multiple testing procedures. In addition to comparing latencies and

amplitudes of the N1, N2 and P2 waves of LEPs, we also compared the entire LEP waveforms obtained in the different groups using 4 point-by-point t-tests. The steps are described in van den Broeke et al. (54-56) and briefly overviewed here. As a first step, LEP waveforms of two conditions (CFS vs. HC and cWAD vs. HC) were compared by point-by-point two sample t-tests. Then, adjacent samples in time above the critical t-value for parametric two-sided tests were identified and clustered. Additionally, an estimate of the magnitude of each cluster was calculated by summing up the t-values constituting each cluster. Then, a reference distribution of maximum cluster magnitude was obtained by random permutation testing (1000 times) of the LEP waveforms of the different conditions. The last step entails calculating the proportion of random partitions that has a larger cluster-level statistic than the observed one. This analysis was conducted separately for hand and foot stimulation, as the latencies of the elicited responses may be expected to differ when stimulating the hand and foot, because of the differences in peripheral conduction distance. Clusters were considered significant if $p<0.01$.

Time-frequency analysis of LEPs

A time-frequency analysis of the recorded EEG signals was performed to characterize and compare non-phase-locked stimulus-induced changes in the power of ongoing EEG oscillations. A short-time fast Fourier transform (STFFT) (57) with a fixed Hanning window of 500 ms was used to express the oscillation amplitude of each single trial, as a function of time and frequency. The analysis was performed using the signal recorded at Cz vs. average reference and T8 vs. Fz. The obtained single-trial time-frequency maps were then averaged across trials. The resulting time-frequency maps represent the average oscillation amplitude as a function of time and frequency, regardless of phase. A baseline correction was then performed using the interval ranging from -1750 to -1250 ms relative to stimulus onset (i.e. before the onset of the first visual stimulus), to identify decreases (event-related

desynchronization, ERD) and increases (event-related synchronization, ERS) of oscillation amplitude relative to baseline (58).

Comparison of the time-frequency maps obtained in the different groups was performed using 4 point-by-point t-tests to evaluate differences in each patient group compared to healthy controls. Such as for the time-domain analysis of LEP waveforms, this analysis was conducted separately for hand and foot stimulation. Clusters were considered significant if $p<0.01$.

All statistical analyses were performed with Letswave 6 and R Studio version 0.99.903. Normality was controlled with the Shapiro Wilk test and QQ-plots and equality of variances by Levene's tests.

Results

Group characteristics and self-reported pain

Twenty-one patients with CFS, 21 patients with cWAD and 20 healthy controls participated in this study. Data from 1 person with CFS and data from 1 healthy control were lost due to recording/processing issues with the EEG data. One HC participant was excluded after study participation for the reason that he reported neck pain (VAS 31) on the day of testing wherefore all analyses are based on 18 healthy controls. In the CFS group one patient terminated the experiment half way due to upcoming headache. From this person, only LEP recordings with foot stimulation were available.

There was a significant sex difference between both patient groups whereby 2 males and 18 females were included in the CFS group and 11 males and 10 females in the cWAD group ($\chi2(2)=8.499$, $p=0.014$). No differences with the control group (consisted of 7 males and 11 females) were found. Participants had a median age of 46.8 years in the healthy group, 43.9 years in the CFS group and 45.8

years in the cWAD group without differences between the groups ($\chi2(2)=1.381$, $p=0.501$). The two patient groups reported similar levels of self-reported pain over the last 7 days with a median score of 50 (Q1-Q3: 31-61) in the CFS group and 51 (Q1-Q3: 35-62) in the cWAD group ($W=179$, $p= 0.789$). Pain catastrophizing scores of 18.1 (SD: 10.69) and 18.0 (SD: 11.73) out of 52 were found in respectively the cWAD and CFS group ($t(38)=0.0414$, $p=0.967$). Additionally, no statistically significant differences were found in scores on the PDI ($t(38)=-0.964$, $p=0.341$) with mean scores of 35 (SD: 14.3) in the cWAD group and 39 (SD: 11.5) in the CFS group. In the cWAD group a median score of 11 out of 63 was found on the BDI and in the CFS group 16 out of 63 without between group differences ($W=141.5$, $p=0.265$). Three patients with cWAD and one patient with CFS were taking opioids. None of the healthy participants was taking opioids. Two patients with cWAD indicated that they were not taking pain medication on the day of the experiment. Group characteristics and self-reported measurements are listed in Table 1.

Aδ fiber related threshold and pain intensity ratings

The intensities at which stimuli delivered to the left hand elicited responses consistently detected with reaction times shorter than 600 ms, were not significantly different between the groups (cWAD 223 (Q1-Q3: 213 - 237) mJ vs. CFS 222 (Q1-Q3: 217 – 229) mJ vs. controls 228 (Q1-Q3: 224 - 230) mJ ; $\chi2(2)=1.313$, $p=0.519$). VAS pain intensity ratings following laser stimulation between the three groups were not significantly different for stimulating the hand ($\chi2(2)=0.659$, $p=0.719$ (CON: 46(Q1-Q3: 17-53), CFS: 48 (Q1-Q3: 25-57), cWAD: 50 (Q1-Q3: 30-56))), nor the foot ($\chi2(2)=0.273$, $p=0.872$ (CON: 50 (Q1-Q3: 17-53), CFS: 45 (Q1-Q3: 27-59), cWAD 48 (Q1-Q3: 25-61))).

Time-domain analysis of LEPs

Grand average LEP waveforms (averaged across locations and groups) recorded at the vertex (Cz) and at the contralateral temporal electrode (T8) are presented in Fig. 2. In all three groups, the laser stimulus elicited a clear negative-positive complex (N2-P2) maximal at the scalp vertex. This complex was flanked by additional responses triggered by the two visual stimuli, which were presented -1000 ms before the laser stimulus and 800 ms after the laser stimulus. The magnitude of the second visual ERP was considerably reduced as compared to the first visual ERP in all groups. The N2-P2 complex of LEPs was preceded by an earlier N1 wave, visible at electrode T8. Visual stimuli have a dominant negative peak that is highest in the occipital region, while the negative peak for the laser stimulus is highest at the vertex.

The N1 component of the LEP elicited by stimulation of the hand had an average amplitude of -4.2 μV (SD: 2.7 μV) with a latency of 229 ms (SD: 60 ms) poststimulus (T8). The N2 was identified at a latency of 231 ms (SD: 50 ms) with an amplitude of -2.9 μV (SD: 2.6 μV) and the P2 was identified at 383 ms (SD: 60 ms) with an average amplitude of 4.7 μV (SD: 3.0 μV) (Cz). The N2-P2 component had an average peak-to-peak amplitude of 7.6 μV (SD: 4.4 μV) (Cz). The N1 component of the LEP waveform measured on the foot had an average amplitude of -3.4 μV (SD: 2.8 μV) with a latency of 226 ms (SD: 43 ms) poststimulus (T8). The N2 was identified at a latency of 235 ms (SD: 57 ms) with an amplitude of -3.3 μV (SD: 3.1 μV) and the P2 was identified at 411 ms (SD: 51 ms) with an average amplitude of 4.0 μV (SD: 3.0 μV) (Cz). The N2-P2 component had an average peak-to-peak amplitude of 7.2 μV (SD: 5.45 μV) (Cz). No differences could be revealed between LEP parameters on the foot for controls vs CFS, nor for controls vs cWAD (Table 2). For stimulation of the hand, only a significant difference in the latency of the N1 was found between HC and CFS ($t(35)=2.25$, $p=0.03$). However, this result did not pass the multiple correction procedure. The point-by-point analysis of the LEP waveforms revealed no significant clusters ($t=0$, $p=1$).

Time-frequency analysis of LEPs

The grand average time-frequency maps of the amplitude of ongoing EEG oscillations recorded at the scalp vertex (Cz) is presented in Fig. 3. These maps show marked increases of low-frequency activity (<8 Hz) when stimulating the hand between -950 and -550 ms (actively elicited by the first visual stimulus) and between 150 and 500 ms poststimulus (actively elicited by the laser component). Additionally, a long-lasting decrease of alpha-band oscillations was observed between -600 and -50 ms and from 350 ms poststimulus onwards. For stimulation of the foot, increases of low-frequency activity were revealed between -920 and -620 ms (visual-evoked activity) and between 160 ms and 520 ms (laser-evoked activity). Additionally, decreases of alpha-band oscillations could be detected between -700 ms and -20 ms and from 370 ms onwards.

Point-by-point comparison could not reveal any significant clusters after stimulus onset between healthy controls and patient populations, nor for LEPs elicited by stimulation of the hand, nor for LEPs elicited by stimulation of the foot (t=0, p=1).

Discussion

Previous reportings revealed signs of hyperalgesia, indications for central sensitization and suggestions for functional changes in the CNS in patients with cWAD and CFS (19, 34). However, the tool that is best suited to explore the functioning of the nociceptive afferent pathways, i.e. LEPs, has not yet been used to confirm this conclusion. This study evaluated the function of nociceptive Aδ fibers and their afferent pathways in the central nervous system (CNS) by measuring laser-evoked brain potentials elicited by stimulation of the hand and foot in patients with cWAD, patients with CFS and healthy controls. Laser stimulation elicited clear LEPs in all three groups. The responses elicited by laser stimulation were on average greater when stimulating the hand as compared to the foot, a finding already reported by Truini et al. (2005) (59). Latencies and amplitudes of the N1, N2 and P2 waves of LEPs were not significantly different across the three groups. There were also no significant differences

between the time-frequency maps of EEG oscillation amplitude between healthy controls and both patient populations.

Our negative findings are in line with previous studies showing no differences in LEP latencies in patients with fibromyalgia (42), chronic tension type headache (16), tempo-mandibular disorders (60) and migraine (18) compared to healthy volunteers.

If a state of CNS hyperexcitability exists in these conditions, it would appear that this state cannot be readily detected using LEPs. It might thus be that LEPs are not the most suitable tool to differentiate between healthy participants and patients with cWAD and CFS. Up to now, there is no gold standard to determine whether a patient has central hyperexcitability. Quantitative Sensory Testing or the Central Sensitisation Inventory questionnaire can provide us with some indication which may be useful to clinicians, however they need to be interpreted very cautiously (6, 61, 62).

In patients with fibromyalgia, some studies have reported increased amplitudes of the N2 and P2 waves of LEPs (42, 63). The authors explained these findings as a consequence of either central sensitization or hypervigilance, defined as a state of exaggerated attention towards threatening information (14, 42, 63). In this study pain catastrophizing was measured, which is a construct that comprises elements of rumination (i.e., an anxious preoccupation with pain and the inability to inhibit pain-related thoughts and fears), magnification (i.e., the tendency to amplify the significance of pain with respect to implications for one's global health), and helplessness (i.e., despair surrounding perceived inability to control one's pain experience) (43, 64). All patients had low levels of pain catastrophizing, which could possibly explain why LEPs of similar amplitude were obtained in the three groups. Nevertheless, the PCS scores are in line with previous studies .

Indirect perceptual assessments of the function of the CNS by measuring pain thresholds have shown hyperalgesia to different kinds of painful stimuli (pressure-evoked pain, electrically-evoked pain) in patients with cWAD and in patients with CFS (19, 32, 33). Hyperalgesia is a typical manifestation of sensitisation of the nociceptive system, including central sensitization, whereby increased responsiveness of nociceptive neurons in the CNS would enhance the pain response to noxious stimuli (31). Despite this perceptual evidence supporting a state of sensitization in these patients, we did not observe any enhancement of the EEG responses elicited by nociceptive laser stimuli. Because these EEG responses are triggered by the activation of quickly-adapting heat-sensitive Aδ fibers (AMH-2; (65)), our results suggest that the CNS response to transient input conveyed by AMH-2 is not amplified in these patients.

In this study, patients with cWAD were included if they were classified as Quebec Task Force classification grades I to III whereby classification degrees I, II and III are characterised by respectively neck complaints (I) with musculoskeletal signs (II) and neurological signs (III) (66). In all subgroups, the neck and/or shoulder region could be considered as the primary affected region. Consequently, the foot and hand regions serve as remote or secondary affected regions in these patients, contrary to patients with CFS, where there is a generalised hypersensitivity. This entails measurements were conducted on body locations other than the primary affected region. As previously suggested by Valeriani et al. (2006), functional changes in the nociceptive system might differently affect the responses to stimuli delivered to painful versus non-painful regions (67).

One of the limitations of this study is that there was no equal sex distribution, with a male/female ratio in favour for females in the CFS group. This inequality is in line with the knowledge that CFS primarily affects young women with a male/female ratio of 1:4, as reported in previous studies (50, 51). Future studies could expand on possible sex differences. Additionally, in this study only LEP amplitudes and latencies were evaluated. An evaluation of cerebral cortex excitability through habituation could be

performed in future studies to further elucidate the pathophysiology of cWAD and CFS. Finally, only laser stimuli supraliminal for Aδ- fiber activation were used. Future studies could also incorporate non-nociceptive stimuli in the protocol and compare relative differences between nociceptive and non-nociceptive stimuli between healthy participants and chronic pain patients.

Conclusion

Although previous studies have suggested that a characteristic of patients with cWAD and CFS may be a state of central sensitization, and although such a state could be expected to result in an enhancement of the EEG responses elicited by the selective activation of skin nociceptors, comparison of the EEG responses to the transient activation of heat-sensitive Aδ fiber skin nociceptors in a group of patients with cWAD and CFS with those obtained in healthy controls revealed no significant differences.

Acknowledgements

The authors would like to thank prof. Leon Plaghki for the extensive help with the processing of the data and critical review of the manuscript.

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Tables

Table 1. Demographics of all participants, separated by group. For age, VAS pain and the BDI median values with Q1 and Q3 are reported. For sex the exact counts with percentages are reported. For the time since accident, PCS and PDI mean values with standard deviations are provided. [a] Kruskal Wallis test; [b] Chi-square test; [c] Mann-Whitney U test [d] independent t test. Abbreviations. BDI: Beck Depression Inventory, CFS: chronic fatigue syndrome, CON: controls, cWAD: chronic whiplash associated disorders, PCS: pain catastrophizing scale, PDI: pain disability index, VAS: visual analogue scale.

Table 2. LEP latencies and amplitudes, separated by group for nociceptive stimulation at the hand and foot. For all parameters, mean values with standard deviations are provided. Both patient groups were compared to healthy controls with t-tests. Abbreviations. Amp: amplitude, CFS: chronic fatigue syndrome, CON: controls, cWAD: chronic whiplash associated disorders, lat: latency.

Figures

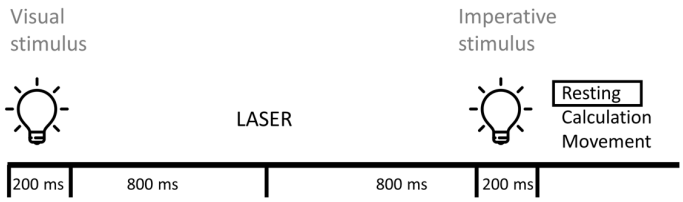

Figure 1. Study protocol. A visual stimulus of 200 ms preceded the nociceptive laser stimulus. After 800 ms, a second visual stimulus was provided. The second visual stimulus was followed by a resting period, a movement task or a calculation task. In this study, only LEPs followed by a resting period were used. The time interval between two successive laser stimuli ranged from 10 to 15 seconds.

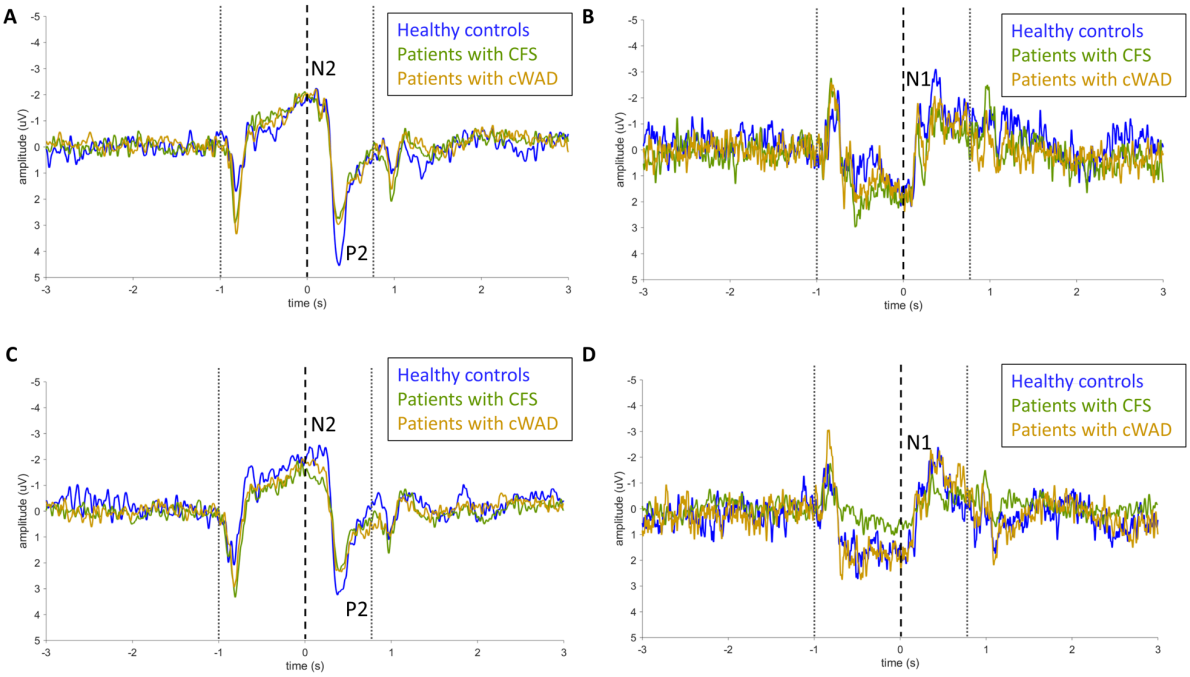

Figure 2. Group-level laser evoked potentials in the time domain, separated by pathology. The two gray dotted lines are representing the onset of the visual evoked potentials. The black dotted line is representing the laser onset. The peaks of the LEP are indicated on the figure.

A: Average waveforms at Cz (vs. average reference) for hand stimulation.

B: Average waveform at T8 (vs. Fz) for hand stimulation.

C: Average waveforms at Cz (vs. average reference) for foot stimulation.

D: Average waveforms at T8 (vs. Fz) for foot stimulation.

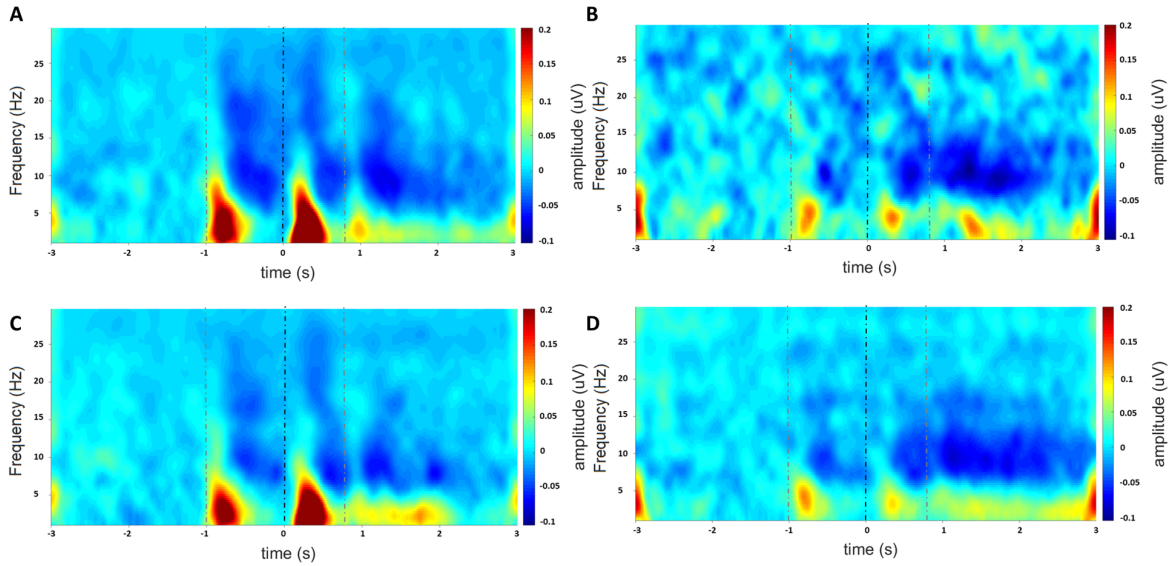

Figure 3. Group level average time-frequency maps in all patients. Visual stimulus onsets (gray dotted lines) and laser onset (black dotted line) are indicated on the figure.

A: Average time-frequency map at Cz (vs. average reference) for hand stimulation.

B: Average time-frequency map at T8 (vs. Fz) for hand stimulation.

C: Average time-frequency map at Cz (vs. average reference) for foot stimulation.

D: Average time-frequency map at T8 (vs. Fz) for foot stimulation.