Pain Medicine, 0(0), 2020, 1–11
doi: 10.1093/pm/pnaa068
Original research Article

Processing of Laser-Evoked Potentials in Patients with Chronic Whiplash-Associated Disorders, Chronic Fatigue Syndrome, and Healthy Controls: A Case–Control Study

Lisa Goudman, PhD,[*,†,‡] Liesbeth Daenen, PhD,[†,‡,§] Andre Mouraux, PhD,[¶] Jo Nijs, PhD,[†,‡,|] Patrick Cras, PhD,[,††,‡‡] Nathalie Roussel, PhD,[§§] Maarten Moens, PhD,[*,¶¶,***] Iris Coppieters, PhD, [†,‡,|] Eva Huysmans, MSc,[†,‡,|,†††] and Margot De Kooning, PhD[†,‡,|]**

[*]Department of Neurosurgery, Universitair Ziekenhuis Brussel, Brussels, Belgium; [†]Pain in Motion International Research Group, Vrije Universiteit Brussel, Brussels, Belgium; [‡]Department of Physiotherapy, Human Physiology and Anatomy, Faculty of Physical Education & Physiotherapy, Vrije Universiteit Brussel, Brussels, Belgium; [§]Knowledge, Information and Research Center (KIR), Group Idewe, Louvain, Belgium; [¶]Institute of Neuroscience, Université Catholique de Louvain, Brussels, Belgium; [|]Department of Physical Medicine and Physiotherapy, Universitair Ziekenhuis Brussel, Brussels, Belgium; [**]Laboratory of Neurology, Translational Neurosciences, University of Antwerp, Wilrijk, Belgium; [††]Institute Born-Bunge, University of Antwerp, Wilrijk, Belgium; [‡‡]Department of Neurology, Antwerp University Hospital, Edegem, Belgium; [§§]Department of Rehabilitation Sciences and Physiotherapy (MOVANT), Faculty of Medicine and Health Sciences, University of Antwerp, Campus, Drie Eiken, Wilrijk, Belgium; [¶¶]Center for Neurosciences (C4N), Vrije Universiteit Brussel (VUB), Brussels, Belgium; [***]Department of Radiology, Universitair Ziekenhuis Brussel, Brussels, Belgium; [†††]Department of Public Health (GEWE), Faculty of Medicine and Pharmacy, Vrije Universiteit Brussel, Brussels, Belgium

Correspondence to: Margot De Kooning, PhD, Department of Physiotherapy, Human Physiology and Anatomy, Faculty of Physical Education & Physiotherapy, VUB, Laarbeeklaan 103, 1090 Jette, Belgium. Tel: 0032 (0) 2-477-45-03; Fax: 0032 (0) 2-477-45-29; E-mail: margot.de.kooning@vub.be.

Funding sources: This work was financially supported by ME Research UK, a UK-based national charity that funds biomedical research on myalgic encephalomyelitis/chronic fatigue syndrome.

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

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 was 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 (HCs). **Design**. Case–control study. **Setting**. A single medical center in Belgium. **Subjects**. The LEPs of 21 patients with cWAD, 19 patients with CFS, and 18 HCs were analyzed 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 brain activity was recorded with a 32-channel electroencephalogram (EEG). LEP signals and transient power modulations were compared between patient groups and HCs. **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 HCs. There were no significant differences between the time–frequency maps of EEG oscillation amplitude between HCs and both patient populations. **Conclusions**. EEG responses of heat-sensitive Aδ fibers in patients with cWAD and CFS revealed no significant differences from the responses of HCs. These findings thus do not support a state of generalized central nervous system hyperexcitability in those patients.

Key Words: 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 (e.g., the Pain Sensitivity Questionnaire [PSQ] [5] or the Central Sensitization Inventory [CSI] [6]) are available for assessing symptoms believed to be associated with CS, whereas quantitative sensory testing (QST) can be used to gain insight into the sensitivity of different pathways for processing nociception [7]. Another option for evaluating sensitization processes is an electrophysiological assessment with the use of laser-evoked potentials (LEPs) [8]. QST is the better tool for evaluating hyperalgesia and related pain mechanisms, whereas 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 with healthy controls [16]. However, in a large study with 199 patients with fibromyalgia, a reduced N2-P2 amplitude was found compared with healthy controls [17]. In patients with migraine, there was no difference in latency or amplitude of LEPs compared with controls [18].

In hyperalgesia ascribed to increased sensitivity, LEP amplitudes are expected to be enhanced [9]. Two frequently diagnosed chronic pain syndromes with presumable sensitization of the central nervous system [19,20] are chronic fatigue syndrome (CFS) and chronic whiplash-associated disorders (cWADs). Ten percent to 50% 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, pain in the upper limbs, headache, dizziness, tinnitus, concentration disturbances, sleep difficulties, and fatigue [22–26]. CFS has a worldwide prevalence ranging from 0.2% to 2.6% [27] and is defined as a medically unexplained disabling fatigue that persists for more than six months [28]. Fatigue is often accompanied by a generalized hyperalgesia and, more generally, hypersensitivity to a variety of sensory stimuli, concentration difficulties, postexertional malaise, and joint pain [29–31].

In patients with cWAD and CFS, hyperalgesia (i.e., increased responsiveness of nociceptive neurons in the central nervous system (CNS) would enhance the pain response to noxious stimuli [32]) to pressure-evoked pain and electrically evoked pain has been revealed [19,33,34]. 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 [19,35]. However, a real evaluation of the nociceptive afferent pathways (for which LEPs are better suited) has not yet been performed in patients with CFS (chronic pain syndrome with fatigue as the primary symptom) and cWAD (chronic pain syndrome). 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 electroencephalogram (EEG) activity (event-related synchronization and desynchronization) [36].

Methods

Subjects

Twenty-one patients with cWAD, 21 patients with CFS, and 20 healthy persons participated in the present study (Table 1). Healthy controls had a median age (interquartile range [IQR]) of 46.8 (27.73–51.21) years. Patients with cWAD and CFS had a median age (IQR) of 45.8 (40.41–51.08) years and 43.9 (35.90–48.41) years, respectively. Thirty-three participants obtained a higher education degree, 20 participants successfully finished secondary school as their highest education, and three participants completed primary school. For three participants, the educational level was not known. All participants provided written informed consent before 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

Table 1. Demographics of all participants, separated by group

	CON (N = 18)	cWAD (N = 21)	CFS (N = 20)	*P* Value
Age, y	46.8 (27.73–51.21)	45.8 (40.41–51.08)	43.9 (35.90–48.41)	0.501*
Sex	M: 7 (38.9%) F: 11 (61.1%)	M: 11 (52.4%) F: 10 (47.6%)	M: 2 (10%) F: 18 (90%)	0.014†
VAS current pain (0–100), mm		28 (20–53)	49 (19–54.5)	0.508‡
VAS last 7 d (0–100), mm		51 (35–62)	50 (31.25–61)	0.789‡
PCS (0–52)		18.1 (10.69)	17.95 (11.73)	0.967§
BDI (0–63)		11 (10–20.5)	16 (11–22)	0.265‡
PDI (0–70)		35 (14.34)	38.9 (11.46)	0.341§
Time since accident, y		4.7 (4.34)		

For age, VAS pain and BDI median values with Q1 and Q3 are reported. For sex, the exact counts with percentages are reported. For time since accident, PCS and PDI mean values with standard deviations are provided.

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 analog scale.

*Kruskal Wallis test.

†Chi-square test.

‡Mann-Whitney *U* test.

§Independent *t* test.

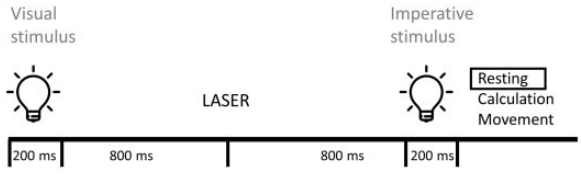

Figure 1. Study protocol. A visual stimulus of 200 ms preceded the nociceptive laser stimulus. After 800 ms, a second visual stimulus was provided. The second visual stimulus was followed by a resting period, a movement task, or a calculation task. In this study, only laser-evoked potentials followed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

defined by the Quebec Task Force classification [23]. Chronicity was defined as complaints persisting for at least three 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 Centers for Disease Control and Prevention criteria [28]. This implies that any other medical condition possibly explaining the debilitating fatigue and pain was excluded before establishing the diagnosis of CFS. Hence, all patients within the CFS group underwent comprehensive cardiovascular, neurological, psychiatric, and hematological 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 six 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 nonopioid 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 [37]. 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 diseases. Finally, to limit confounding of the study findings, we strived toward three groups with a comparable age distribution.

The demographics of the three groups were compared with Kruskall-Wallis tests and chi-square 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 interstimulus interval ranged between 10 and 15 seconds in order to avoid habituation effects to painful stimulation [38]. A visual warning signal lasting 200 ms preceded each laser stimulus and was initiated one second before stimulus administration (Figure 1). Eight hundred milliseconds 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 [39–41]. Only LEPs recorded in the rest condition were analyzed in this manuscript.

At the end of each block, participants were asked to rate the intensity elicited by the laser stimuli using a visual analog scale (VAS) ranging from "no detection" (VAS = 0) to "maximum pain" (VAS = 100). At the middle of the scale (VAS = 50), an anchor marked the borderline between the nonpainful and painful domains of sensation [42]. 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 [41,43]. 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 analysis of variance (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 scored on a five-point Likert scale (0 = not at all, 4 = all the time) [44]. Scores $\geq$30/52 indicate a clinically relevant level of catastrophizing [45]. The internal consistency, test–retest reliability, and validity are acceptable [46,47].

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 [48].

The Pain Disability Index provides an indication of the impact of pain on daily living activities. This questionnaire consists of seven 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 [49]. The Dutch version of this questionnaire is a valid tool with good internal consistency and test–retest reliability [50].

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) [42]. Stimuli were applied on the dorsum of the left hand (C6-C7 skin dermatomes) and left foot (L5-S1 skin dermatomes). For patients with cWAD, the left foot served as a distant location. The CO_2 laser system generates a highly collimated infrared beam (wave length: 10.6 μm). The power output is continuously adjustable between 1 and 25 W. Heat pulse duration was 20 ms. The 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 [51], the location of the impact on the skin was moved slightly between two 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 was 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 1,000 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 diagonally 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 preprocessing 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 six seconds (–3 to +3 seconds 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 eye-blink 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 the reference (i.e., before the onset of the warning visual stimulus). For each subject, epochs were averaged according to the location (foot vs hand). LEP components were identified on the basis of their latency and polarity and labeled according to Valeriani et al. [10]. Peak latencies of N2 and P2 component amplitudes were measured at the vertex (Cz vs average reference [52]) and defined as the largest negative and positive deflections between, respectively, 150–350 ms and 300–500 ms [53]. The LEP N1 component was evaluated at the T8 electrode with the Fz electrode as reference [52] within a targeted time frame of 100–300 ms poststimulus [53,54].

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 vs the control group. In total, four tests were performed: CON vs CFS on the hand (1), CON vs CFS on the foot (2), CON vs cWAD on the hand (3), and CON vs 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 four point-by-point *t* tests. The steps are described in van den Broeke et al. [55–57] and briefly overviewed here. As a first step, LEP waveforms of two conditions (CFS vs CON and cWAD vs CON) 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 the *t* values constituting each cluster. Then, a reference distribution of maximum cluster magnitude was obtained by random permutation testing (1,000 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) [58] 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 –1,750 to –1,250 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 [59].

Comparison of the time–frequency maps obtained in the different groups was performed using four point-by-point *t* tests to evaluate differences in each patient group compared with 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 was controlled 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 one person with CFS and data from one healthy control were lost due to recording/processing issues with the EEG data. One CON participant was excluded after study participation for the reason that he reported neck pain (VAS 31) on the day of testing, so all analyses are based on 18 healthy controls. In the CFS group, one patient terminated the experiment halfway 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; two males and 18 females were included in the CFS group and 11 males and 10 females in the cWAD group ($\chi^2(2) = 8.499$, $P = 0.014$). No differences with the control group (seven 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 ($\chi^2(2) = 1.381$, $P = 0.501$). The two patient groups reported similar levels of self-reported pain over the last seven days with a median score (IQR) of 50 (31–61) in the CFS group and 51 (35–62) in the cWAD group ($W = 179$, $P = 0.789$). Pain catastrophizing scores (SD) of 18.1 (10.69) and 18.0 (11.73) out of 52 were found in, respectively, the cWAD and CFS groups ($t(38) = 0.0414$, $P = 0.967$). Additionally, no statistically

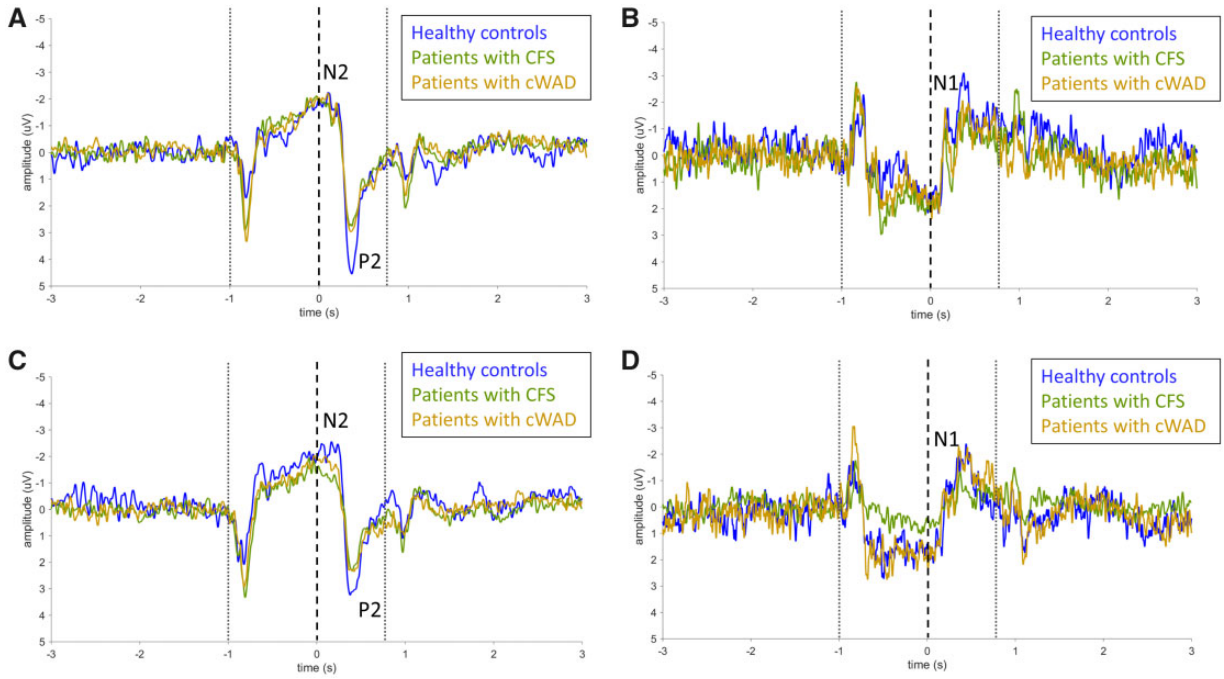

Figure 2. Group-level laser-evoked potentials in the time domain, separated by pathology. The two dotted lines represent the onset of the visual-evoked potentials. The dashed line represents the laser onset. The peaks of the laser-evoked potentials are indicated on the figure. A) Average waveforms at 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.

significant differences were found in scores on the PDI ($t(38) = -0.964$, $P = 0.341$), with mean scores (SD) of 35 (14.3) in the cWAD group and 39 (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 were 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 were 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 <600 ms (IQR) were not significantly different between the groups (cWAD 223 [213–237] mJ vs CFS 222 [217–229] mJ vs controls 228 [224–230] mJ; $\chi^2(2) = 1.313$, $P = 0.519$). VAS pain intensity ratings following laser stimulation between the three groups (IQR) were not significantly different for stimulating the hand ($\chi^2(2) = 0.659$, $P = 0.719$; CON: 46 [17–53], CFS: 48 [25–57], cWAD: 50 [30–56]) or the foot ($\chi^2(2) = 0.273$, $P = 0.872$; CON: 50 [17–53], CFS: 45 [27–59], cWAD: 48 [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 Figure 2. In all three groups, the laser stimulus elicited a clear negative–positive complex (N2-P2) maximally at the scalp vertex. This complex was flanked by additional responses triggered by the two visual stimuli, which were presented –1,000 ms before the laser stimulus and 800 ms after the laser stimulus. The magnitude of the second visual ERP was considerably reduced as compared with 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, whereas 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 (SD) of –4.2 (2.7) μV with a latency of 229 (60) ms poststimulus (T8). The N2 was identified at a latency (SD) of 231 (50) ms with an amplitude of –2.9 (2.6) μV, and the P2 was identified at 383 (60) ms with an average amplitude of 4.7 (3.0) μV (Cz). The N2-P2 component had an average peak-to-peak amplitude (SD) of 7.6 (4.4) μV (Cz). The N1 component of the LEP waveform measured on the foot had an average amplitude (SD) of –3.4 (2.8) μV with a latency of 226 (43) ms poststimulus (T8). The N2 was identified at a latency (SD) of 235 (57) ms with an amplitude of –3.3 (3.1) μV, and the P2 (SD) was identified at

Table 2. LEP latencies and amplitudes, separated by group for nociceptive stimulation at the hand and foot

	LEP Hand					LEP Foot				
	CON (N = 18)	CFS (N = 19)	cWAD (N = 21)	CON vs CFS	CON vs cWAD	CON (N = 18)	CFS (N = 20)	cWAD (N = 21)	CON vs CFS	CON vs cWAD
N1 amp, μV	−4.47 (2.37)	−3.5 (2.75)	−4.67 (2.81)	$t(35) = -1.15$, $P = 0.26$	$t(37) = 0.24$, $P = 0.81$	−3.17 (2.83)	−3.89 (1.93)	−3.09 (3.35)	$t(30) = 0.90$, $P = 0.38$	$t(37) = -0.08$, $P = 0.93$
N2 amp, μV	−3.41 (3.25)	−2.73 (2.67)	−2.54 (1.7)	$t(33) = -0.69$, $P = 0.50$	$t(25) = -1.02$, $P = 0.31$	−4.31 (4.77)	−2.7 (1.93)	−2.92 (2.08)	$t(22) = -1.33$, $P = 0.20$	$t(22) = -1.14$, $P = 0.26$
P2 amp, μV	5.56 (2.83)	4.48 (3.02)	4.27 (3.11)	$t(35) = 1.13$, $P = 0.27$	$t(37) = 1.35$, $P = 0.18$	4.72 (4.29)	3.59 (2.79)	3.65 (1.8)	$t(29) = 0.95$, $P = 0.35$	$t(22) = 0.99$, $P = 0.33$
N2P2 amp, μV	8.97 (5.29)	7.21 (3.29)	6.81 (4.32)	$t(28) = 1.21$, $P = 0.24$	$t(33) = 1.38$, $P = 0.18$	9.03 (8.57)	6.30 (3.25)	6.57 (3.09)	$t(21) = 1.27$, $P = 0.22$	$t(21) = 1.15$, $P = 0.26$
N1 lat, msec	252 (56)	212 (53)	224 (65)	$t(34) = 2.25$, $P = 0.03$	$t(37) = 1.42$, $P = 0.16$	235 (46)	216 (44)	227 (40)	$t(35) = 1.30$, $P = 0.20$	$t(34) = 0.58$, $P = 0.57$
N2 lat, msec	229 (54)	241 (46)	225 (50)	$t(33) = -0.71$, $P = 0.48$	$t(35) = 0.20$, $P = 0.84$	227 (59)	247 (49)	232 (64)	$t(33) = -1.13$, $P = 0.27$	$t(37) = -0.24$, $P = 0.81$
P2 lat, msec	374 (59)	388 (55)	388 (70)	$t(34) = -0.77$, $P = 0.44$	$t(37) = -0.68$, $P = 0.50$	404 (53)	413 (52)	414 (51)	$t(35) = -0.54$, $P = 0.59$	$t(35) = -0.58$, $P = 0.56$

For all parameters, mean values with standard deviations are provided. Both patient groups were compared with healthy controls using *t* tests.
Amp = amplitude; CFS = chronic fatigue syndrome; CON = controls; cWAD = chronic whiplash-associated disorders; lat = latency.

411 (51) ms with an average amplitude of 4.0 (3.0) μV (Cz). The N2-P2 component had an average peak-to-peak amplitude (SD) of 7.2 (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 CON 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) are presented in Figure 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 reports have revealed signs of hyperalgesia, indications for central sensitization, and suggestions for functional changes in the CNS in patients with cWAD and CFS [19,35]. However, the tool that is best suited to explore the functioning of the nociceptive afferent pathways, that is, 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 with the foot, a finding already reported by Truini et al. [60]. 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

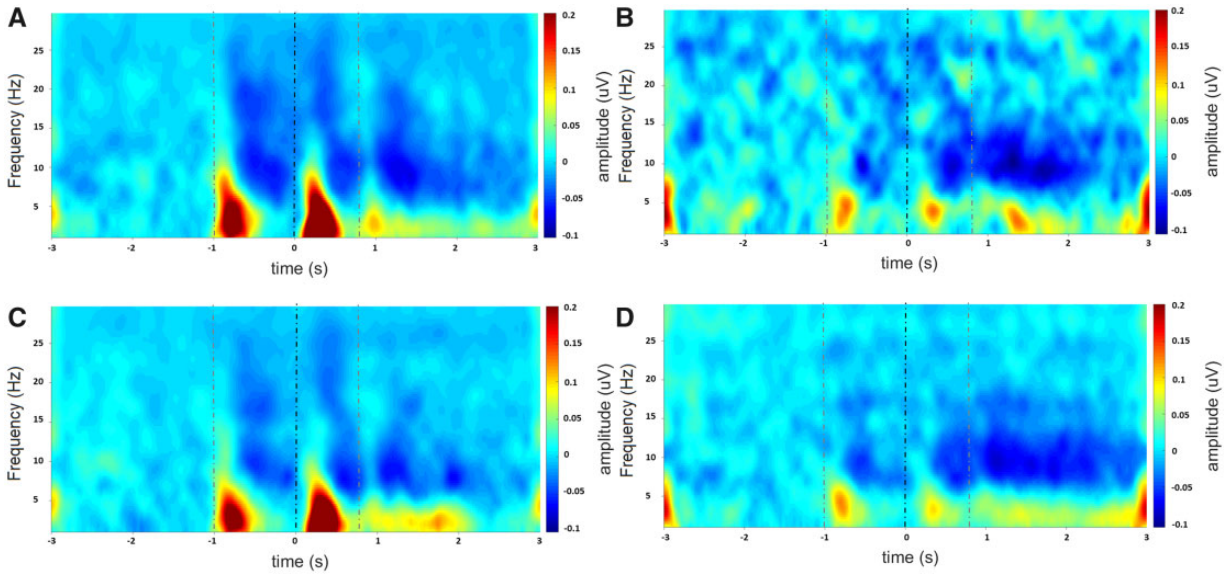

Figure 3. Group-level average time–frequency maps for all patients. Visual stimulus onsets (dotted lines) and laser onset (dashed line) are indicated on the figure. A) Average time–frequency map at Cz (vs average reference) for hand stimulation. B) Average time–frequency map at T8 (vs Fz) for hand stimulation. 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.

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 [43], chronic tension type headache [16], temporomandibular disorders [61], and migraine [18] compared with 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 has been no gold standard to determine whether a patient has central hyperexcitability. Quantitative sensory testing or the Central Sensitization Inventory questionnaire can provide us with some indication, which may be useful to clinicians; however, they need to be interpreted very cautiously [6,62,63].

In patients with fibromyalgia, some studies have reported increased amplitudes of the N2 and P2 waves of LEPs [43,64]. The authors explained these findings as a consequence of either central sensitization or hypervigilance, defined as a state of exaggerated attention toward threatening information [14,43,64]. 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) [44,65]. 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 [66–68].

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 [19,33,34]. Hyperalgesia is a typical manifestation of sensitization of the nociceptive system, including central sensitization, whereby increased responsiveness of nociceptive neurons in the CNS would enhance the pain response to noxious stimuli [32]. 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) [69], our results suggest that the CNS response to transient input conveyed by AMH-2 is not amplified in these patients. It might be possible that these results, obtained by specifically targeting the Aδ fibers with a methodology based on reaction times, do not lead to the same conclusion as a protocol that is based on subjective pain intensity ratings to determine the intensity of nociceptive stimuli. However, further research is needed to explore this question.

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 characterized by, respectively, neck complaints (I) with musculoskeletal signs (II) and neurological signs (III) [70]. Patients did not undergo new radiographic or neuroimaging evaluations when they were included in

the study. Therefore, it might be possible that some patients demonstrate intradiscal injuries or facet joint-related issues. However, the potential presence of these findings presumably does not induce an influence on the outcome of these patients. A systematic review exploring factors predicting the outcome after a whiplash trauma concluded that factors identified as not being associated with the prognosis of whiplash were postinjury magnetic resonance imaging (MRI) or radiological findings [71]. Furthermore, a systematic literature review of imaging features in asymptomatic populations revealed that the prevalence of disk degeneration in asymptomatic individuals increased from 37% of 20-year-old individuals to 96% of 80-year-old individuals [72], in whom the causal relationship between radiographic findings and the accidental trauma would be difficult to define. Nevertheless, in all subgroups (grade I, II, III), the neck and/or shoulder region could be considered 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 generalized hypersensitivity. This entails measurements that 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 vs nonpainful regions [73].

One of the limitations of this study is that there was no equal sex distribution, with a male/female ratio in favor of 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.

Conclusions

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.

Acknowledgments

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

References

1. Cagnie B, Coppieters I, Denecker S, Six J, Danneels L, Meeus M. Central sensitization in fibromyalgia? A systematic review on structural and functional brain MRI. Semin Arthritis Rheum 2014;44(1):68–75.
2. Verne GN, Price DD. Irritable bowel syndrome as a common precipitant of central sensitization. Current Rheumatology Reports 2002;4(4):322–8.
3. Cashman MD, Martin DK, Dhillon S, Puli SR. Irritable bowel syndrome: A clinical review. Current Rheumatology Reviews 2016;12(1):13–26.
4. International Association for the Study of Pain (IASP). IASP terminology. 2017. Available at: http://www.iasp-pain.org/Education/Content.aspx? ItemNumber=1698#Centralsensitization (accessed December 19, 2019).
5. Ruscheweyh R, Verneuer B, Dany K, et al. Validation of the pain sensitivity questionnaire in chronic pain patients. Pain 2012;153(6):1210–8.
6. Mayer TG, Neblett R, Cohen H, et al. The development and psychometric validation of the Central Sensitization Inventory. Pain Pract 2012;12(4):276–85.
7. Fillingim RB, Loeser JD, Baron R, Edwards RR. Assessment of chronic pain: Domains, methods, and mechanisms. J Pain 2016;17(Suppl 9):T10–20.
8. Morton DL, Sandhu JS, Jones AK. Brain imaging of pain: State of the art. J Pain Res 2016;8(9):613–24.
9. Treede RD, Lorenz J, Baumgärtner U. Clinical usefulness of laser-evoked potentials. Neurophysiol Clin 2003;33(6):303–14.
10. Valeriani M, Pazzaglia C, Cruccu G, Truini A. Clinical usefulness of laser evoked potentials. Neurophysiol Clin 2012;42(5):345–53.
11. Garcia-Larrea L, Convers P, Magnin M, et al. Laser-evoked potential abnormalities in central pain patients: The influence of spontaneous and provoked pain. Brain 2002;125(12):2766–81.
12. Truini A, Biasiotta A, La Cesa S, et al. Mechanisms of pain in distal symmetric polyneuropathy: A combined clinical and neurophysiological study. Pain 2010;150(3):516–21.
13. Gibson SJ, Littlejohn GO, Gorman MM, Helme RD, Granges G. Altered heat pain thresholds and cerebral event-related potentials following painful CO2 laser stimulation in subjects with fibromyalgia syndrome. Pain 1994;58(2):185–93.
14. Lorenz J, Grasedyck K, Bromm B. Middle and long latency somatosensory evoked potentials after painful laser stimulation in patients with fibromyalgia syndrome. Electroencephalogr Clin Neurophysiol 1996;100(2):165–8.
15. de Tommaso M, Ricci K, Libro G, et al. pain processing and vegetative dysfunction in fibromyalgia: A study by sympathetic skin response and laser evoked potentials. Pain Pract Treat 2017;2017:9747148.
16. de Tommaso M, Shevel E, Pecoraro C, et al. Topographic analysis of laser evoked potentials in chronic tension-type headache: Correlations with clinical features. Int J Psychophysiol 2006;62(1):38–45.
17. de Tommaso M, Nolano M, Iannone F, et al. Update on laser-evoked potential findings in fibromyalgia patients in light of clinical and skin biopsy features. J Neurol 2014;261(3):461–72.

18. Valeriani M, de Tommaso M, Restuccia D, et al. Reduced habituation to experimental pain in migraine patients: A CO(2) laser evoked potential study. Pain 2003;105(1):57–64.
19. Nijs J, Meeus M, Van Oosterwijck J, et al. In the mind or in the brain? Scientific evidence for central sensitisation in chronic fatigue syndrome. Eur J Clin Invest 2012;42(2):203–12.
20. Van Oosterwijck J, Nijs J, Meeus M, Paul L. Evidence for central sensitization in chronic whiplash: A systematic literature review. Eur J Pain 2013;17(3):299–312.
21. Cote P, Hogg-Johnson S, Cassidy JD, Carroll L, Frank JW. The association between neck pain intensity, physical functioning, depressive symptomatology and time-to-claim-closure after whiplash. J Clin Epidemiol 2001;54(3):275–86.
22. Radanov B, Sturzenegger M. The effect of accident mechanism and initial findings on the long-term outcome of whiplash injury. J Musculoskelet Pain 1996;4(4):47–60.
23. Spitzer W, Skovron M, Salmi L, et al. Scientific monograph of Quebec Task Force on Whiplash Associated Disorders: Redefining 'whiplash' and its management. Spine (Phila Pa 1976) 1995;20:1–73.
24. Carroll L, Holm L, Ferrari R, Ozegovic D, Cassidy JD. Recovery in whiplash-associated disorders: Do you get what you expect? J Rheumatol 2009;36(5):1063–71.
25. Lovell M, Galasko C. Whiplash disorders—a review. Injury 2002;33(2):97–101.
26. Sterling M. Whiplash-associated disorder: Musculoskeletal pain and related clinical findings. J Man Manip Ther 2011;19 (4):194–200.
27. Prins JB, van der Meer JW, Bleijenberg G. Chronic fatigue syndrome. Lancet 2006;367(9507):346–55.
28. Fukuda K, Straus SE, Hickie I, Sharpe MC, Dobbins JG, Komaroff A. The chronic fatigue syndrome: A comprehensive approach to its definition and study. International Chronic Fatigue Syndrome Study Group. Ann Intern Med 1994;121 (12):953–9.
29. Selvaraj N, Jaryal A, Santhosh J, Deepak KK, Anand S. Assessment of heart rate variability derived from finger-tip photoplethysmography as compared to electrocardiography. J Med Eng Technol 2008;32(6):479–84.
30. VanNess JM, Stevens SR, Bateman L, Stiles TL, Snell CR. Postexertional malaise in women with chronic fatigue syndrome. J Women's Health (Larchmt) 2010;19(2):239–44.
31. Vos-Vromans DC, Smeets RJ, Huijnen IP, et al. Multidisciplinary rehabilitation treatment versus cognitive behavioural therapy for patients with chronic fatigue syndrome: A randomized controlled trial. J Intern Med 2016;279 (3):268–82.
32. Woolf CJ. Central sensitization: Implications for the diagnosis and treatment of pain. Pain 2011;152(Suppl):S2–15.
33. Curatolo M, Petersen-Felix S, Arendt-Nielsen L, Giani C, Zbinden A, Radanov B. Central hypersensitivity in chronic pain after whiplash injury. Clin J Pain 2001;17(4):306–15.
34. Scott D, Jull G, Sterling M. Widespread sensory hypersensitivity is a feature of chronic whiplash-associated disorder but not chronic idiopathic neck pain. Clin J Pain 2005;21(2):175–81.
35. Herren-Gerber R, Weiss S, Arendt-Nielsen L, et al. Modulation of central hypersensitivity by nociceptive input in chronic pain after whiplash injury. Pain Med 2004;5(4):366–76.
36. Mouraux A, Iannetti GD. Across-trial averaging of event-related EEG responses and beyond. Magn Reson Imaging 2008;26 (7):1041–54.
37. Meeus M, Nijs J, Van de Wauwer N, Toeback L, Truijen S. Diffuse noxious inhibitory control is delayed in chronic fatigue syndrome: An experimental study. Pain 2008;139(2):439–48.
38. Bromm B, Lorenz J. Neurophysiological evaluation of pain. Electroencephalogr Clin Neurophysiol 1998;107(4):227–53.
39. Gomez CM, Marco J, Grau C. Preparatory visuo-motor cortical network of the contingent negative variation estimated by current density. Neuroimage 2003;20(1):216–24.
40. Bender S, Resch F, Weisbrod M, Oelkers-Ax R. Oelkers-Ax R. Specific task anticipation versus unspecific orienting reaction during early contingent negative variation. Clin Neurophysiol 2004;115(8):1836–45.
41. Le Pera D, Brancucci A, De Armas L, et al. Inhibitory effect of voluntary movement preparation on cutaneous heat pain and laser-evoked potentials. Eur J Neurosci 2007;25(6):1900–7.
42. Plaghki L, Delisle D, Godfraind JM. Heterotopic nociceptive conditioning stimuli and mental task modulate differently the perception and physiological correlates of short CO2 laser stimuli. Pain 1994;57(2):181–92.
43. de Tommaso M, Federici A, Santostasi R, et al. Laser-evoked potentials habituation in fibromyalgia. J Pain 2011;12 (1):116–24.
44. Sullivan MJL, Bishop SR, Pivik J. The Pain Catastrophizing Scale: Development and validation. Psychol Assess 1995;7 (4):524–32.
45. Sullivan M. The Pain Catstrophizing Scale: User Manual. 1995. Available at: http://sullivan-painresearch.mcgill.ca/pdf/pcs/PCSManual_English.pdf (accessed July 15, 2019).
46. Osman A, Barrios FX, Kopper BA, Hauptmann W, Jones J, O'Neill E. Factor structure, reliability, and validity of the Pain Catastrophizing Scale. J Behav Med 1997;20(6):589–605.
47. Lame IE, Peters ML, Kessels AG, Van Kleef M, Patijn J. Test–retest stability of the Pain Catastrophizing Scale and the Tampa Scale for Kinesiophobia in chronic pain over a longer period of time. J Health Psychol 2008;13(6):820–6.
48. Bishop SR, Edgley K, Fisher R, Sullivan M. Screening for depression in chronic low back pain with the Beck Depression Inventory. Can J Rehabil 1993;7:143–8.
49. Soer R, Reneman MF, Vroomen PC, Stegeman P, Coppes MH. Responsiveness and minimal clinically important change of the Pain Disability Index in patients with chronic back pain. Spine (Phila Pa 1976) 2012;37(8):711–5.
50. Soer R, Köke AJA, Vroomen PCAJ, et al. Extensive validation of the Pain Disability Index in 3 groups of patients with musculoskeletal pain. Spine (Phila Pa 1976) 2013;38(9):E562–8.
51. Schwarz S, Greffrath W, Busselberg D, Treede RD. Inactivation and tachyphylaxis of heat-evoked inward currents in nociceptive primary sensory neurones of rats. J Physiol 2000;528(3):539–49.
52. Valentini E, Hu L, Chakrabarti B, Hu Y, Aglioti SM, Iannetti GD. The primary somatosensory cortex largely contributes to the early part of the cortical response elicited by nociceptive stimuli. Neuroimage 2012;59(2):1571–81.
53. Hatem SM, Hu L, Rage M, et al. Automated single-trial assessment of laser-evoked potentials as an objective functional diagnostic tool for the nociceptive system. Clin Neurophysiol 2012; 123(12):2437–45.
54. Cruccu G, Aminoff MJ, Curio G, et al. Recommendations for the clinical use of somatosensory-evoked potentials. Clin Neurophysiol 2008;119(8):1705–19.
55. van den Broeke EN, Lambert J, Huang G, Mouraux A. Central sensitization of mechanical nociceptive pathways is associated with a long-lasting increase of pinprick-evoked brain potentials. Front Hum Neurosci 2016;10:531.
56. van den Broeke EN, de Vries B, Lambert J, Torta DM, Mouraux A. Phase-locked and non-phase-locked EEG responses to pinprick stimulation before and after experimentally-induced secondary hyperalgesia. Clin Neurophysiol 2017;128(8):1445–56.

57. van den Broeke EN, Mouraux A, Groneberg AH, Pfau DB, Treede RD, Klein T. Characterizing pinprick-evoked brain potentials before and after experimentally induced secondary hyperalgesia. J Neurophysiol 2015;114(5):2672–81.
58. Zhang ZG, Hu L, Hung YS, Mouraux A, Iannetti GD. Gamma-band oscillations in the primary somatosensory cortex—a direct and obligatory correlate of subjective pain intensity. J Neurosci 2012;32(22):7429–38.
59. Pfurtscheller G, Lopes da Silva FH. Event-related EEG/MEG synchronization and desynchronization: Basic principles. Clin Neurophysiol 1999;110(11):1842–57.
60. Truini A, Galeotti F, Romaniello A, Virtuoso M, Iannetti GD, Cruccu G. Laser-evoked potentials: Normative values. Clin Neurophysiol 2005;116(4):821–6.
61. Romaniello A, Cruccu G, Frisardi G, Arendt-Nielsen L, Svensson P. Assessment of nociceptive trigeminal pathways by laser-evoked potentials and laser silent periods in patients with painful temporomandibular disorders. Pain 2003;103(1):31–9.
62. Kennedy DL, Kemp HI, Ridout D, Yarnitsky D, Rice AS. Reliability of conditioned pain modulation: A systematic review. Pain 2016;157(11):2410–9.
63. van den Broeke EN. Central sensitization and pain hypersensitivity: Some critical considerations. F1000Res 2018;7:1325.
64. Lorenz J. Hyperalgesia or hypervigilance? An evoked potential approach to the study of fibromyalgia syndrome. Z Rheumatol 1998;57(Suppl 2):19–22.
65. Burns LC, Ritvo SE, Ferguson MK, Clarke H, Seltzer Z, Katz J. Pain catastrophizing as a risk factor for chronic pain after total knee arthroplasty: A systematic review. J Pain Res 2015;8:21–32.
66. Carriere JS, Thibault P, Milioto M, Sullivan M. Expectancies mediate the relations among pain catastrophizing, fear of movement, and return to work outcomes after whiplash injury. J Pain 2015;16(12):1280–7.
67. Rivest K, Cote JN, Dumas JP, Sterling M, De Serres SJ. Relationships between pain thresholds, catastrophizing and gender in acute whiplash injury. Man Ther 2010;15(2):154–9.
68. Meeus M, Nijs J, Van Mol E, Truijen S, De Meirleir K. Role of psychological aspects in both chronic pain and in daily functioning in chronic fatigue syndrome: A prospective longitudinal study. Clin Rheumatol 2012;31(6):921–9.
69. Treede RD, Meyer RA, Raja SN, Campbell JN. Evidence for two different heat transduction mechanisms in nociceptive primary afferents innervating monkey skin. J Physiol 1995;483(3):747–58.
70. Sterling M. A proposed new classification system for whiplash associated disorders—implications for assessment and management. Man Ther 2004;9(2):60–70.
71. Sarrami P, Armstrong E, Naylor JM, Harris IA. Factors predicting outcome in whiplash injury: A systematic meta-review of prognostic factors. J Orthop Traumatol 2017;18(1):9–16.
72. Brinjikji W, Luetmer PH, Comstock B, et al. Systematic literature review of imaging features of spinal degeneration in asymptomatic populations. Am J Neuroradiol 2015;36(4):811–6.
73. Valeriani M, Tonali P, Le Pera D, et al. Modulation of laser-evoked potentials by experimental cutaneous tonic pain. Neuroscience 2006;140(4):1301–10.