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Medically Unexplained Symptoms Are Linked to Chronic Inflammatory Diseases: Is There a Role for Frontal Cerebral Blood Oxygen Content?

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Keywords

Inflammation · Pain · Medically unexplained symptoms · Cerebral blood oxygen · Cerebral blood flow

Abstract

Introduction: Patients often go to the physician with medically unexplained symptoms (MUS). MUS can be autonomic nervous system-related "unspecific" symptoms, such as palpitations, heart rhythm alterations, temperature dysregulation (hand, feet), anxiety, or depressive manifestations, fatigue, somnolence, nausea, hyperalgesia with varying pains and aches, dizziness, etc. ***Methods:*** In this real-world study, we investigated MUS in a cohort of unselected outpatients from general practitioners in Italy. It was our aim to increase the understanding of MUS by using principal component analyses to identify any subcategories of MUS and to check a role of chronic inflammatory diseases. Additionally, we studied cerebral blood oxygen ($rCBO_2$) and associations with MUS and chronic inflammatory disease. ***Results:*** Participants included 1,597 subjects (50.6 ± 0.4 years, 65%/35% women/men). According to ICD-10 codes, 137 subjects had chronic inflammatory diseases. MUS were checked by a questionnaire with a numeric rating scale and cerebral blood flow with optical techniques. The analyses of men and women were stratified. Psychological symptom severity was higher in the inflamed compared to the non-inflamed group (fatigue, insomnia in women and men; recent mood changes, daytime sleepiness, anxiety, apathy, cold hands only in women; abnormal appetite and heart rhythm problems only in men). Principal component analysis with MUS provided new subcategories: brain symptoms, gut symptoms, and unspecific symptoms. Brain and gut symptoms were higher in inflamed women and men. Chronic inflammatory diseases and pain were tightly interrelated in men and women ($p < 0.0001$). In women, not in men, average frontal $rCBO_2$ content was higher in inflamed compared to non-inflamed subjects. In men, not in women, individuals with pain demonstrated a lower average frontal $rCBO_2$ content compared to pain-free men. MUS did not relate to $rCBO_2$ parameters. ***Conclusion:*** This study shows close relationships between MUS and chronic inflammatory diseases but not between MUS and $rCBO_2$ parameters.

Introduction

Immune system and central nervous system (CNS) are two selfish organs, which hierarchically control self-allocation of energy in critical states [1]. In challenging situations, either the immune system (stimulus: e.g., infectious agent) or the CNS (stimulus: e.g., psychological

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stress) initiate a program that typically blocks the other selfish organ [1–4]. In a situation with chronic peripheral inflammation, a complex program of CNS-related phenomena starts to inhibit brain function and related skeletal muscular activity in order to spare energy for immune function [5]. This leads to psychological sequelae that are often summarized under the heading of sickness behavior [6, 7], which is a forerunner of major depression in vulnerable people [2, 8]. The phenomenon is well known under controlled conditions such as, for example, administration to healthy people of lipopolysaccharide i.v. in the laboratory [9], but the situation in the real world – in the setting of a general practitioner – is not often studied.

In the environment of the general practitioner, patients appear with "medically unexplained symptoms (MUS)" that might belong to sickness behavior. MUS can be autonomic nervous system-related "unspecific" symptoms, such as palpitations, heart rhythm alterations, temperature dysregulation (hand, feet), anxiety or depressive manifestations, fatigue, somnolence, nausea, hyperalgesia with varying pains and aches, dizziness, etc. [10–13]. The physician is confronted with these typical phenomena. We aimed to study these MUS under real-world conditions in the general practice. MUS can either be related to brain function or also to peripheral bodily function; thus, we aimed to find a content structure behind these MUS. A causal platform of MUS can be chronic inflammation. Thus, we were particularly interested to see how chronic inflammatory diseases are related to MUS.

Along with the collection of MUS in these patients, we were able to study cerebral blood oxygen ($rCBO_2$) content. This part of the work was explorative because no similar studies were known that linked MUS, chronic inflammation, and $rCBO_2$ content. $rCBO_2$ content might be related to inflammation, which can be observed in acute mountain sickness [14, 15] or because of obstructive sleep apnea [16]. Acute brain hypoxia induces functional alterations in the brain, such as decreased cognitive performance and altered frontal/cortical connectivity [17]. Thus, links between MUS, chronic inflammatory diseases, and rCBO2 might exist.

This study aimed to investigate the following parts: (1) What are typical MUS under real-world conditions in general practice and what is the role of chronic inflammatory diseases on MUS? (2) Is there a content structure that can define different MUS subcategories? (3) Are the different MUS categories dependent on chronic inflammatory diseases? (4) Does chronic inflammatory diseases influence CBO_2 parameters, and are CBO_2 parameters related to MUS?

Patients and Methods

Patients

In this study, a total of 1,597 unselected participants were recruited during a visit to general medical practitioners in Italy. Since several years, participating physicians work with a health-related questionnaire and noninvasive methods like bioimpedance analysis (BIA-ACC®, BioTekna, Marcon, Italy) and optical determination of $rCBO_2$ (HemoEncephaloGraphy Technology [HEG], BioTekna, Marcon, Italy) (techniques are given below). All patients entered this explorative study without any further selection on a consecutive basis in order to create a real-world situation beyond classical clinical studies. All data were collected during one session of investigation. However, patients with severe mental/psychiatric disorders, epilepsy, cancer, established or suspected pregnancy were excluded from the study.

The HEG device has been registered at the Italian Ministry of Health – National Classification of Medical Devices. The HEG device has been validated and CE-certified as a noninvasive medical device used for diagnostic and monitoring purposes and has been used in the EU since 2004. Before the measurements, patient received all information and gave oral consent. The retrospective use of the patient data obtained with the above devices in routine medical evaluations for anonymous analysis and publication has been approved earlier for another study by the Ethics Committee of the University Research Institute of Maternal and Child Health and Precision Medicine, National and Kapodistrian University of Athens, Athens, Greece (ethics document without registration number can be obtained through the corresponding author). The ethical review for the study was exempt, as this previous study with the same design has been approved in another EU country (Greece). The procedures were in accordance with the ethical standards of the responsible committee on human experimentation and with the Helsinki Declaration of 1975, as revised in 1983. Reporting of the study conforms to STROBE statements along with references to STROBE and the broader EQUATOR guidelines [18]. Due to the nature of the study, with electronic submission of information and due to ethical guidelines, the authors did not receive several critical patient-related data including blood material and exact diagnoses.

The mean age of the 1,597 participants was 50.6 ± 0.4 years, 64.8%/35.2% were female/male, weight was 69.5 ± 0.4 kg, body height was 1.67 ± 0.01 m, and mean body mass index was 24.7 ± 0.1 kg/m^2. Physicians did not communicate the exact diagnosis, but they categorized their patients in the electronic submission form using ICD-10 codes necessary in Italy for administrative and insurance purposes. This ICD-10-dependent classification yielded a subgroup of patients with chronic inflammatory diseases, and, in addition, C-reactive protein serum levels supported the subgroup classification (blood values not communicated). This yielded a subgroup of 137 patients with chronic inflammatory diseases that was not different in age (50.4 ± 0.4 year) and sex (64%/36% women/men) compared to the non-inflamed group (n = 1,460; 53.6 ± 1.3 year; 74%/26%).

Questionnaire Variables

Chronic stress has been associated with autonomic nervous system-related "nonspecific" symptoms, such as palpitations, heart rhythm alterations, temperature dysregulation (hand, feet), anxiety, or depressive manifestations, etc. Similarly, chronic activation of the immune system has been linked to "nonspecific", "sickness behavior"-type manifestations, such as fatigue, somnolence, nausea, hyperalgesia with varying pains and aches, dizziness, etc. [10–13]. These very common clinical manifestations associated with chronic stress and inflammation bringing many patients to the general practitioner. Physicians, despite a full physical examination and many laboratory evaluations, failed to come up with a concrete diagnosis, and the term "medically unexplained symptoms" has been commonly used to describe a cluster of such manifestations [10–13]. We use the abbreviation MUS for the rest of the text.

Similar to another study [19], we used a published questionnaire in the Italian language that included questions for MUS focusing on general signs and symptoms such as fatigue, mood alterations, sleep alterations (waking-up, insomnia), daytime sleepiness, anxiety, apathy, panic attacks, heart rhythm alterations, changes in eating behavior and appetite, nocturnal eating, abdominal symptoms of irritable bowel syndrome (bloating, nausea, flatulence, obstipation, etc.), cold hand and feet, nocturnal sweating, awakening with bad mood, feelings of unjustified guilt, feeling of anhedonia, and weight loss. In addition, we asked for general pain all over the body without naming a special location.

For every MUS item, grading of severity was estimated with a numeric rating scale with a minimum of 0 points and a maximum of 10 points provided as an electronic visual analog scale. In addition, overall pain was scored similarly using an electronic visual analog scale (minimum = 0, maximum = 10).

Measurement of Parameters of Frontal Cerebral Oxygen Content

Cerebral blood oxygenation ($rCBO_2$) was measured using the HemoEncephaloGraphy technology (HEG; BioTekna, Marcon, Italy). HEG is a noninvasive hemodynamic medical device, which measures in real time the changes in hemoglobin concentration, cerebral blood flow (CBF), and oxygen of the prefrontal cortex ($rCBO_2$). This HEG device is based on near-infrared spectroscopy complementary to other imaging methods using the hemodynamic response such as functional magnetic resonance imaging (fMRI) and positron emission tomography (PET). The technique of near-infrared spectrometry has been extensively evaluated elsewhere [20].

The HEG medical device, thanks to the noninvasive plethysmographic measurement technology with a sensor positioned on the patient's forehead, allows in just 5 min to easily detect cerebral blood oxygenation of the prefrontal cortex ($rCBO_2$) independent of skull thickness or amount of tissue between the frontal skin and cerebral vessel. For the measurements, the patient is seated in a comfortable position with the forearms resting on a table. The technique is described in Figure 1.

Statistical Analyses

This was an explorative study and patients were included consecutively. A prior sample size was not determined due to the explorative nature of the study. All data are given as mean ± SEM. Normal distribution was checked using Kolomogorov-Smirnov test (IBM SPSS Statistics for Windows, Version 26.0. Armonk, NY: IBM Corp.). Data are presented as vertical bar graphs with error bars when data are normally distributed. Frequencies were compared with the χ^2 test and group medians were compared by the non-parametric Mann-Whitney U test (both IBM SPSS Statistics). Spearman rank correlation analysis was used to study the interrelation between variables (IBM SPSS Statistics). $p < 0.05$ was the significance level.

In order to define a hidden content structure for MUS of the 19-item questionnaire, principal component analysis was applied (IBM SPSS Statistics). The sampling adequacy for each variable was examined on the anti-image correlation matrix and all variables had high scores. The validity of the principal component analysis for the data was assessed by the Kaiser-Meyer-Olkin measure of sampling adequacy, which was 0.913. The hypothesis that the correlation matrix is an identity matrix was rejected by the Bartlett's test ($p < 0.0001$). Factors were then extracted, and factors with an eigenvalue greater than 1 were included and a varimax rotation with Kaiser normalization was used to enhance factor loading. Data for women and men are presented in a stratified form without a direct comparison of the two groups.

Results

Chronic Inflammation Related to Subjective Symptoms

MUS in Non-Inflamed and Chronically Inflamed Subjects

The applied questionnaire focuses on MUS associated with "nonspecific" autonomic nervous system-related symptoms, "sickness behavior" type manifestations, and others [10–13]. In a direct comparison of non-inflamed patients and patients with chronic inflammatory diseases, many symptoms appeared more often in subjects with inflammation. For every symptom, a patient was able to judge severity on a visual analog scale. Women with chronic inflammatory diseases scored higher in 9 of 19 items (Table 1). Significant differences in women were found for fatigue, recent mood changes, insomnia, daytime sleepiness, anxiety, apathy, cold hands, bad mood during awakening, and feelings of unjustified guilt (Table 1). Men with chronic inflammatory diseases scored higher in 4 of 19 items. Significant differences in men were found for fatigue, insomnia, noticeable problems with heart rhythm, and abnormal appetite (low or binge) (Table 1).

In order to check whether one can group the 19 heterogeneous symptoms into a smaller number of subcategories, principal component analysis was performed in the entire group of women and men. With this technique, four different subcategories regrouped the 19 mixed symptoms, as demonstrated in Table 2. These new factors were used in further analyses.

The first two subcategories mainly include brain symptoms (factor 1 and factor 2), and they explain together 29.466% of the variation (third row in Table 2). In further analyses, a combined factor – called "brain symptoms" – was generated as the sum of factor 1 and

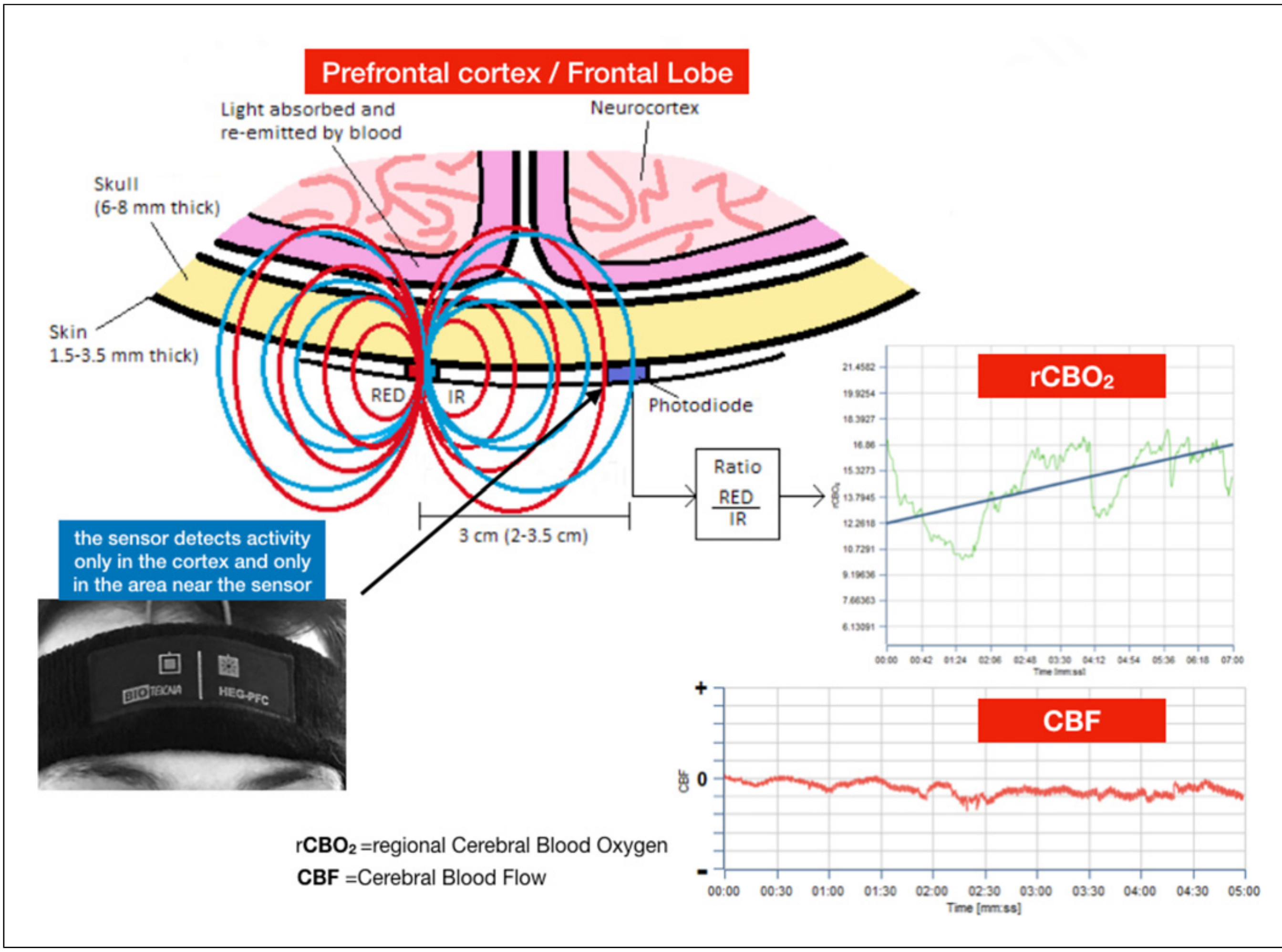

Fig. 1. The noninvasive plethysmographic HemoEncephaloGraphy (HEG) technology. The unit of measure for cerebral oxygen ($rCBO_2$) is measured in scalar units from 0 to 200. With the help of the receiver photodiode, $rCBO_2$ is measured as the ratio between absorption of reflected red light with 660 nm wavelength (RED) and absorption of reflected infrared light (IR) at 940 nm wavelength. The red and IRs are alternately shown to the brain tissue and the absorption of reflected light is measured with a photodiode. There is a large difference in the attenuation of red light (RED) between oxygen-rich and oxygen-deficient hemoglobin, while IR is minimally modified. With the HEG device, 3 parameters of the frontal CBO_2 were studied in 5 min of continuous recording: mean frontal CBO_2 content (average CBO_2), increase/decrease in frontal CBO_2 over time (5-min CBO_2 slope), and change in frontal CBO_2 over time (CBO_2 standard deviation [SD] over 5 min).

factor 2. Factor 3 is mainly linked to symptoms of the gastrointestinal tract ("gut symptoms"), and factor 4 to eating behavior and nighttime sweating ("unspecific symptoms"). All factors together explain 47.320% of the entire variation (Table 2, third row: "cumulative percentage of variance"). Factor analysis allows creating new variables used in further analyses.

Thus, we compared the new factors in non-inflamed compared to chronically inflamed patients (Table 3). Significant differences in women were found for the combination of factor 1 and factor 2 ("brain symptoms"), factor 1 alone, factor 2 alone, and factor 3 (Table 3). Men differed only in the combination of factor 1 and factor 2 ("brain symptoms"), factor 2 alone, and factor 3 (Table 3). For women and men, no differences were observed for factor 4, which described nocturnal food intake, nocturnal sweating, and abnormal appetite ("unspecific symptoms").

In summary, chronically inflamed women and men often demonstrate a higher score for different MUS. MUS can be regrouped into four reasonable categories that were higher in chronic inflammatory diseases compared to non-inflamed subjects.

Table 1. Symptom comparison of women and men without versus with chronic inflammation

Symptoms	Non-inflamed	Chronic inflammation	p value*
	women, n = 934 (men, n = 526)	women, n = 101 (men, n = 36)	women (men)
Fatigue (score from 0 to 10)	3.98±0.11 (2.91±0.15)	5.58±0.34 (5.06±0.54)	0.000005 (0.000141)
Recent mood changes (0–10)	3.10±0.11 (2.18±0.14)	4.84±0.38 (3.33±0.56)	0.000006 (n.s.)
Insomnia (0–10)	3.05±0.11 (2.44±0.14)	5.06±0.40 (4.67±0.64)	0.000001 (0.000127)
Daytime sleepiness (0–10)	1.87±0.09 (1.45±0.11)	2.97±0.34 (2.47±0.60)	0.000739 (n.s.)
Anxiety (0–10)	3.71±0.12 (2.43±0.14)	5.51±0.37 (3.58±0.59)	0.000002 (n.s.)
Apathy (0–10)	1.09±0.08 (1.05±0.10)	2.26±0.33 (2.06±0.55)	0.000057 (n.s.)
Panic reactions (0–10)	0.61±0.06 (0.30±0.06)	1.18±0.26 (1.28±0.45)	n.s. (n.s.)
Noticeable problems with heart rhythm (0–10)	1.75±0.09 (1.02±0.10)	2.36±0.33 (3.33±0.61)	n.s. (0.000001)
Abnormal appetite (low or binge) (0–10)	1.74±0.10 (1.03±0.10)	2.68±0.35 (2.64±0.54)	n.s. (0.000059)
Nocturnal food intake (0–10)	0.25±0.04 (0.26±0.05)	0.34±0.15 (0.58±0.31)	n.s. (n.s.)
Acidic stomach (0–10)	3.19±0.12 (2.29±0.14)	4.10±0.38 (3.67±0.60)	n.s. (n.s.)
Irritated bowel (0–10)	1.88±0.10 (1.18±0.12)	2.81±0.38 (2.50±0.61)	n.s. (n.s.)
Obstipation (0–10)	1.92±0.10 (1.03±0.10)	2.39±0.35 (2.00±0.57)	n.s. (n.s.)
Cold hands (0–10)	2.33±0.11 (1.56±0.12)	3.81±0.39 (2.42±0.59)	0.000317 (n.s.)
Nocturnal sweating (0–10)	1.53±0.09 (0.90±0.09)	1.59±0.30 (1.28±0.46)	n.s. (n.s.)
Bad mood during awakening (0–10)	1.08±0.08 (0.95±0.10)	2.30±0.34 (1.44±0.46)	0.000060 (n.s.)
Feelings of unjustified guilt (0–10)	1.85±0.10 (1.14±0.11)	3.13±0.38 (1.31±0.47)	0.000610 (n.s.)
Anhedonia (0–10)	1.00±0.08 (0.85±0.10)	1.85±0.30 (2.14±0.56)	n.s. (n.s.)
Unexplained weight loss (0–10)	0.26±0.02 (0.47±0.07)	0.66±0.19 (0.69±0.33)	n.s. (n.s.)
Average of all symptoms	**6.24±0.13 (4.65±0.18)**	**8.10±0.40 (7.50±0.70)**	**0.000008 (0.000024)**

Data are given as means ± SEM. Data of men are given in parentheses. *Mann-Whitney U test; correction of the p value according to Bonferroni: sig. p value = 0.05/25 = 0.002 (number 25 includes also variables in Table 3).

Interrelation of Pain and Chronic Inflammation

Patients with chronic inflammatory diseases scored higher on the overall pain score, which was true for women and men (Fig. 2). In addition, in women and men, inflammation positively correlated with pain (women: $R_{\text{Rank}} = 0.172$, $p < 10^{-6}$; men: $R_{\text{Rank}} = 0.215$, $p < 10^{-5}$). Furthermore, pain score positively correlated with all four factors of Table 3 in women and men (all p value below 10^{-8}) and, of course, with individual MUS (data not shown). This shows the tight interrelation of pain and inflammation and pain and reported MUS.

Chronic Inflammation Related to Frontal Blood Oxygen Content

The data above clearly linked chronic inflammatory diseases and pain to subjective symptoms. We were able to define subjective factors related to brain symptoms and gut symptoms. Brain and gut symptoms were tightly linked to inflammation. Beyond subjective symptoms, we expected that chronic inflammatory diseases were also related to objectively measurable parameters of frontal cerebral oxygen content.

Frontal $rCBO_2$ and Chronic Inflammatory Diseases

Three frontal CBO_2 parameters were studied over 5 min of continuous recording: the average frontal CBO_2 content (avg.CBO_2), the increase/decrease of frontal CBO_2 over time (CBO_2 slope over 5 min), and the variation of frontal CBO_2 over time (CBO_2 standard deviation [SD] over 5 min). In the first analysis in women and separately in men, different results were observed for CBO_2 slope (women vs. men: −1.05 ± 0.7 vs. −1.52 ± 0.12 scalar unit x time, $p < 0.001$) and CBO_2 SD (2.46 ± 0.11 vs. 3.67 ± 0.68 scalar units), but avg.CBO_2 was similar (33.53 ± 0.53 vs. 35.04 ± 0.93 scalar units). This prompted us to study women and men in stratified analyses.

In the subchapters above, we recognized that MUS and the derived factors were related to chronic inflammatory diseases in women and men. The question appeared to be whether frontal CBO_2 parameters were also related to inflammation. Indeed, in women, the average frontal CBO_2 was lower in non-inflamed compared to subjects with chronic inflammatory diseases that was not significant in men (Fig. 3).

Table 2. Factor analysis of 19 diverse MUS derived from 1,597 subjects

Final statistics of the principal components analysis				
	factor 1	factor 2	factor 3	factor 4
Eigenvalue	5.556	1.308	1.090	1.036
% of variance	15.495	13.971	10.749	7.106
Cumulative percentage of variance	15.495	29.466	40.214	47.320
Factor matrix after varimax rotation				
Anhedonia	0.730			
Feelings of unjustified guilt	0.644			
Bad mood during awakening	0.628			
Apathy	0.592			
Panic reactions	0.417			
Insomnia		0.660		
Fatigue		0.624		
Recent mood changes		0.589		
Anxiety		0.560		
Daytime sleepiness		0.527		
Noticeable problems with heart rhythm		0.434		
Obstipation			0.603	
Unexplained weight loss			0.596	
Irritated bowel			0.562	
Acidic stomach			0.546	
Cold hands			0.434	
Nocturnal food intake				0.723
Nocturnal sweating				0.539
Abnormal appetite (low or binge)				0.459

The different symptoms loaded variably on four factors. Factor loadings less than 0.400 are not shown.

Table 3. Factor comparison of women and men without versus with chronic inflammation

Factors**	Non-inflamed	Chronic inflammation	*p* value*
	women, *n* = 934 (men, *n* = 526)	women, *n* = 101 (men, *n* = 36)	women (men)
Factor 1 + factor 2 (brain symptoms)	23.07±0.62 (16.72±0.82)	37.04±2.27 (30.67±3.90)	8.00×10^{-10} (1.32×10^{-5})
Factor 1 (factor 1 alone)	5.63±0.27 (4.29±0.34)	10.71±1.14 (8.22±1.87)	1.67×10^{-6} (n.s.)
Factor 2 (factor 2 alone)	17.45±0.43 (12.43±0.56)	26.33±1.40 (22.44±2.30)	3.55×10^{-9} (5.28×10^{-6})
Factor 3 (gut symptoms)	9.58±0.28 (6.53±0.35)	13.77±1.04 (11.28±1.68)	8.00×10^{-5} (0.001)
Factor 4 (unspecific symptoms)	3.51±0.16 (2.19±0.18)	4.61±0.56 (4.50±1.03)	n.s. (n.s.)

Data are given as means ± SEM. Data of men are given in parentheses. *Mann-Whitney U test; correction of the *p* value according to Bonferroni: sig. *p* value = 0.05/25 = 0.002 (number 25 includes also variables in Table 1). **Factors are calculated by summing up the individual visual analog scale values for the symptoms of Table 1 allocated to the factor given in Table 2.

In women and in men, CBO_2 slope over 5 min and CBO_2 SD did not differ between non-inflamed and chronically inflamed subjects (data not shown). In addition, the individual symptom scores and the factors of Table 3 were not related to one of the CBO_2 parameters, neither in women nor in men (data not shown).

Frontal Cerebral Blood Oxygen and Pain

In the subchapters above, we recognized that inflammation and pain were related to each other in women and men. The question appeared to be whether frontal CBO_2 parameters were also related to pain. Indeed, in men, the average frontal CBO_2 was lower in men with

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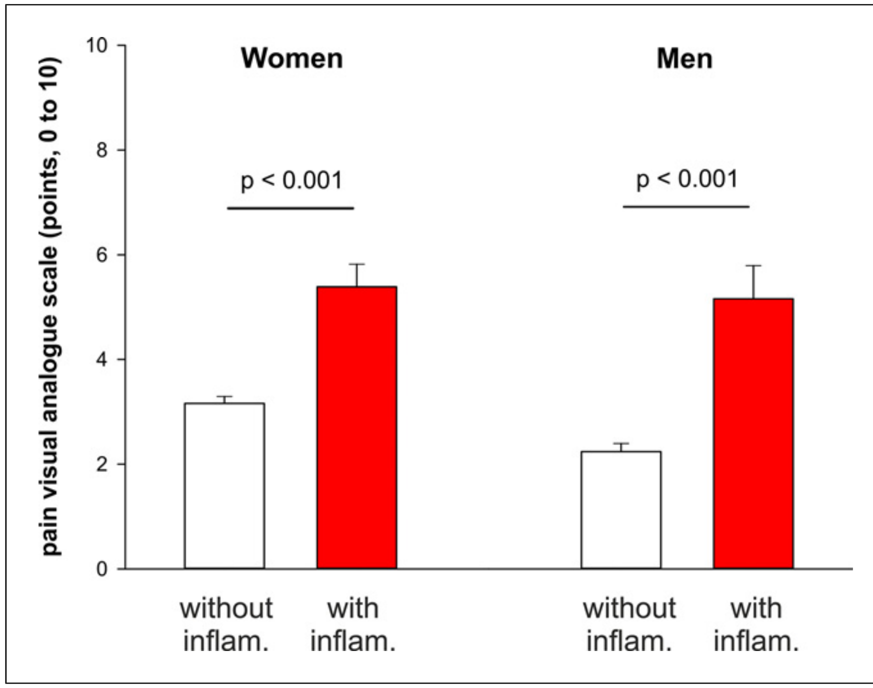

Fig. 2. Pain and inflammation. Data are given as means ± SEM. Red bars represent subjects with chronic inflammatory diseases.

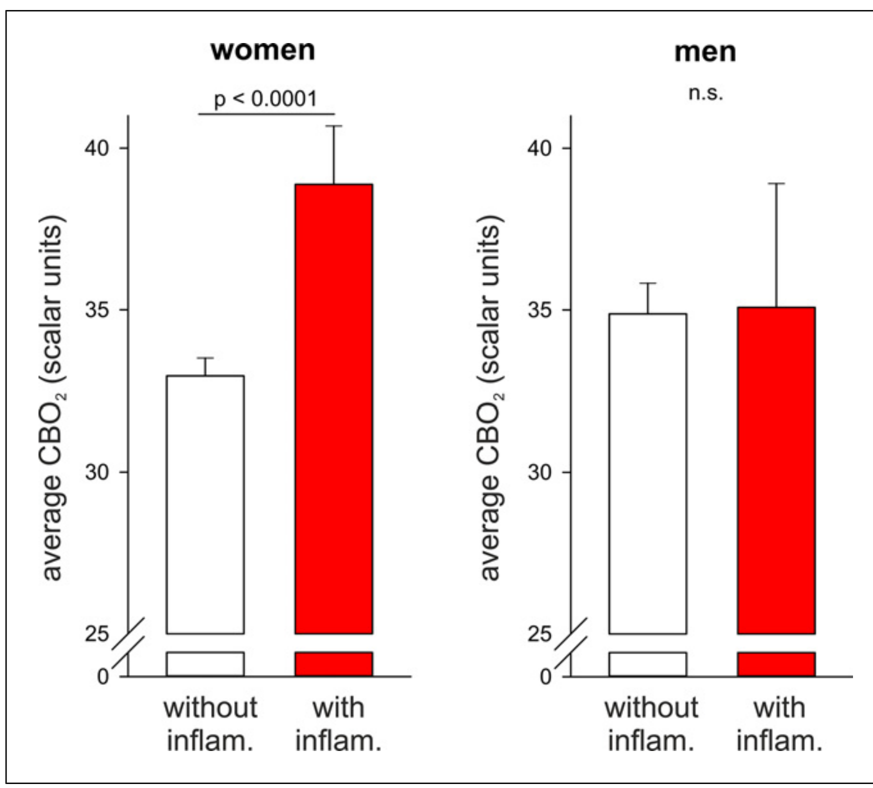

Fig. 3. Average frontal CBO^2 in non-inflamed and chronically inflamed women and men. Red bars represent results of patients with chronic inflammatory diseases. Data are given as means ± SEM because data were normally distributed.

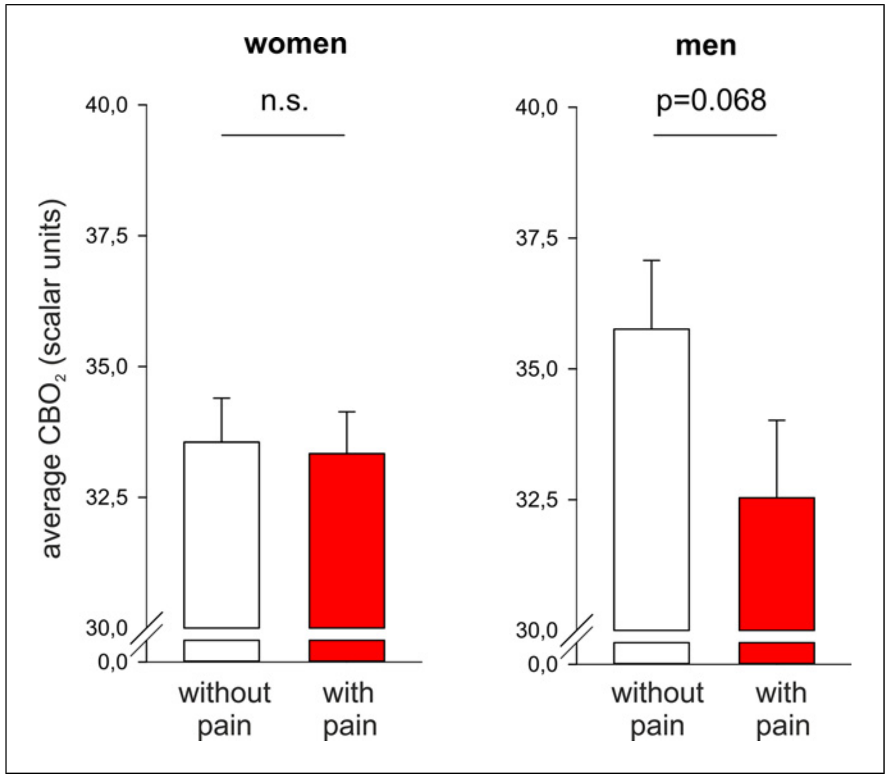

Fig. 4. Average frontal CBO[2] in patients without and with pain in women and men. Red bars represent results of patients with chronic pain. Data are given as means ± SEM because data were normally distributed.

chronic pain compared to subjects without pain, which reached the level of a statistical trend, which was not observed in women (Fig. 4). No differences were found for the other CBO_2 parameters in the two groups with and without pain, neither in women nor in men (data not shown).

Discussion

This real-world study on patients from general practitioners in Italy found a close interrelation between MUS and chronic inflammatory diseases in women and men. The results are presented in a stratified form for women and men separately; thus, no direct comparisons of women and men were carried out. Factor analysis provides four different factors by regrouping the 19 MUS. Newly described factors like "brain symptoms" and "gut symptoms" are higher in chronically inflamed than non-inflamed women and men. Chronic inflammatory diseases and pain are tightly interrelated in women and men, and pain is interrelated to brain symptoms and gut symptoms in women and men. In women, not in men, the average frontal CBO_2 content was higher in chronically inflamed compared to non-inflamed subjects. In men, not in women, individuals with pain demonstrated a lower average frontal CBO_2 content compared to men without pain.

Our study reports on information of typical MUS that is routinely recorded in the general practice of associated physicians in Italy. These doctors also apply the noninvasive techniques of the optical frontal CBO_2 measurement. In multiple sessions, they were trained to correctly perform the different test procedures. As predefined in ethical guidelines, doctors transmit the information electronically without personal data of the patients to one of us (D.B.). This anonymous submission of data led to several publications using similar techniques [21–26]. In the context of new developments, the optical measurement of CBO_2 parameters was added to the armamentarium in this cross-sectional study.

In this real-world approach, we confirmed a clear interrelation between chronic inflammatory diseases and MUS, which was particularly obvious in women but also in men. The average symptom severity was higher in patients with chronic inflammatory diseases compared to non-inflamed women and men. This study confirms short-time experimental studies with injected lipopolysaccharide, which increases the inflammatory load for a short time [9, 27–30]. These studies were prepared by many experiments in rodents (e.g., summarized in [31]).

In the present study, we planned to recategorize the 19 investigated MUS into meaningful factors that to be used in further analyses. Principal component analysis provided four different factors leading to different grouping of symptoms. Factor 1 and factor 2 were combined to a new category called “brain symptoms,” which is complemented by a subcategory of “gut symptoms” and a subcategory of “unspecific symptoms.” Brain symptoms and gut symptoms are higher in subjects with chronic inflammatory diseases than non-inflamed women and men. Whether or not these new brain and gut subcategories will be used in future analyses must stand the test of time.

The link between pain and inflammation goes back to the old work of neurogenic inflammation [32]. More links between inflammation and pain started to emerge in the 1990s when glial activation on the spinal level became an important element [33]. In our present study in the real-world environment, we observed that pain related to MUS in women and men.

In order to link aspects of cortical oxygen content to inflammation and pain, the optical technique of CBO_2 measurement was applied. In women, not in men, average frontal CBO_2 content was higher in patients with chronic inflammatory diseases compared to non-inflamed subjects. In men, not in women, patients with pain demonstrated – in the form of a trend – a lower average frontal CBO_2 content compared to men without pain. To the best of our knowledge, we do not know of other studies that have examined CBO_2 parameters in outpatient subjects. Thus, the meaning of these dichotomous findings in women and men is presently unclear. One can only speculate that in women, the aspect of chronic inflammatory diseases plays a stronger role for CBO_2 data (higher prevalence of these diseases), whereas in men, pain has a more important role.

Nevertheless, either inflammation in women and pain in men is interrelated with CBO_2 content in the frontal cortex. Similar to studies looking on acute brain hypoxia [14–17], the inflammation-induced CBO_2 change in women and the pain-induced CBO_2 changes in men might be linked to symptoms. However, we were not able to see this direct interrelation because CBO_2 parameters were not related to individual symptoms, brain symptoms, or gut symptoms. This fact surprised us because we expected that CBO_2 changes related to subjectively measured symptom severity. A careful literature search in PubMed revealed a substantial lack of information concerning the interrelation of inflammation and CBO_2. Thus, we cannot easily interpret our data, but this is different for the pain aspects.

Indeed, chronic pain was related to lower cerebral blood flow in patients with spinal cord injury [34, 35]. Short acute pain to the gingiva also reduced prefrontal cerebral hemodynamics [36]. Thus, our results with lower frontal CBO_2 content in men with pain fit into the concept of lowered cortical hemodynamics. It remains unclear why this is absent in women.

In general, it is not clear why results are contrasting in women and men. However, women usually have more chronic inflammatory diseases [37]. Thus, the link between chronic inflammation and symptoms or CBO_2 parameters might be more obvious in women than men.

In conclusion, we saw a clear link between MUS and inflammation in women and in men. A newly described subcategory “brain symptoms” and “gut symptoms” are higher in chronically inflamed than non-inflamed women and men. Chronic inflammatory diseases and pain are tightly interrelated in women and men, and pain is interrelated to brain and gut symptoms in women and men. In this first study, we did not see an interrelation between CBO_2 parameters and MUS, i.e., brain symptoms or gut symptoms. Future studies need to address this link in women and men in larger groups with chronic inflammation.

Statement of Ethics

The HEG device has been registered with the Italian Ministry of Health – National Classification of Medical Devices. The HEG device has been validated and CE-certified as a noninvasive medical device used for diagnostic and monitoring purposes and has been used in the EU since 2004. Before the measurements, patient received all information and gave oral consent. The retrospective use of the patient data obtained with the above-mentioned devices in routine medical evaluations for anonymous analysis and publication has been approved earlier for another study by the Ethics Committee of the University Research Institute of Maternal and Child Health and Precision Medicine, National and Kapodistrian University of Athens, Athens, Greece (ethics document without registration number can be obtained through the corresponding author). The ethical review for the study was exempt as this previous study with the same design has been approved in another EU country (Greece). The procedures were in accordance with the ethical standards of the responsible committee on human experimentation and with the Helsinki Declaration of 1975, as revised in 1983. Reporting of the study conforms to STROBE statements along with references to STROBE and the broader EQUATOR guidelines [18]. Due to the nature of the study with electronic submission of information and due to ethical guidelines, the authors did not receive several critical patient-related data including blood material and exact diagnoses.

Conflict of Interest Statement

D.B. is director of research and development at BioTekna, which sells the HemiEncephaloGraphy (HEG) device used in this study.

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Author Contributions

R.H.S. created tables and figures, and he statistically analyzed the data. R.H.S. wrote the draft version of the manuscript. D.B. discussed and corrected the tables, figures, statistical analysis, and text.

Data Availability Statement

The data that support the findings of this study are not publicly available due to privacy reasons but are available from the corresponding author upon reasonable request.

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