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ORIGINAL RESEARCH

Unexpectedly decreased plasma cytokines in patients with chronic back pain

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Introduction: Chronic back pain is one of the most important socioeconomic problems that affects the global population. Elevated levels of inflammatory mediators, such as cytokines, have been correlated with pain, but their role in chronic back pain remains unclear. The effectiveness of anti-inflammatory drugs seems to be limited for chronic back pain. The authors wanted to investigate the levels of inflammatory mediators in long-term medically treated patients with persistent chronic back pain.

Methods: Cytokine plasma levels of patients with chronic back pain (n=23), compared to pain-free healthy controls (n=30), were investigated by immunoassay. Patients with chronic back pain were exposed to long-term conservative medical therapy with physiotherapy and anti-inflammatories, also combined with antidepressants and/or muscle-relaxants.

Results: The patients with chronic back pain expressed lower levels of the chemokines MCP1, CCL5, and CXCL6 compared to pain-free healthy controls. Significantly lower concentrations of the anti-inflammatory cytokines, interleukin (IL)-4 and granulocyte-colony stimulating factor were also found. Interestingly, levels of proinflammatory cytokines (IL-2, IL-6, IL-1β, tumor necrosis factor alpha), IL-10, granulocyte-macrophage colony-stimulating factor, and stromal cell-derived factor 1 alpha showed no significant differences between both groups.

Conclusion: This decrease of inflammatory mediators in medically treated patients with chronic back pain is of unclear origin and might be either a long-term side effect of medical therapy or related to chronic pain. Further longitudinal research is necessary to elucidate the underlying cause of these findings.

Keywords: chronic pain, back pain, inflammation, cytokine, chemokine

Introduction

Chronic back pain, defined as pain lasting at least 3 months duration,[1] is recognized as a major public health problem, producing significant economic and social burdens.[2] Several studies have demonstrated that chronic back pain condition interferes with everyday activities and results in direct medical costs and lost productivity.[3,4] Increased serum levels of pro-inflammatory cytokines (interleukin [IL]-1β, IL-2, IL-6, and tumor necrosis factor alpha [TNF-α]) have been previously correlated with increased pain intensity in patients with different types of chronic pain.[5] Furthermore, low concentrations of the anti-inflammatory cytokines IL-4 and IL-10 were found in patients with chronic widespread pain, and the lack of anti-inflammatory cytokine activity was associated with a possible contribution to pain pathogenesis.[6]

The most common spinal degenerative problems manifest in back pain, followed by neck and head pain.[7] Degeneration of the intervertebral disc (IVD) is a widely

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recognized contributor to back pain.[8–10] Patients with discogenic low back pain showed high levels of IL-6, IL-8,[11] and IL-1β[12] in IVD tissues; IL-1β was suggested as the key regulatory cytokine in the upregulation of factors involved in innervation and vascularization of human degenerated IVD.[13] High levels of inflammatory mediators (IL-1β, TNF-α, IL-6, and IL-8) were found in degenerated and herniated IVD,[14,15] and associated with pain development during IVD herniation and degeneration.[16] Painful[12] and degenerated[17,18] IVD tissues showed higher expression of the chemokines RANTES (CCL5) and granulocyte chemotactic protein 2 (CXCL6), and high levels of monocyte chemoattractant protein 1 (MCP-1) were found in the herniated lumbar nucleus pulposus.[19] MCP-1 was also elevated in the blood at the chronic stage of complex regional pain syndrome,[20] and elevated plasma levels of CCL5 and CXCL6 were found in patients with lumbar disc degeneration.[21] Furthermore, higher plasma levels of pro-inflammatory TNF-α and IL-6 were associated with painful herniated IVD[22,23] and low back pain.[24]

People with chronic back pain, along with pain and impaired function, frequently experience anxiety and depression. Analgesics, non-steroidal anti-inflammatory drugs (NSAIDs), opioids, antidepressants and muscle-relaxants may be used for medical treatments of low back pain. Unfortunately, there is only limited evidence of the effectiveness of those drugs.[25]

In line with the observation that medications perform poorly as treatments for chronic back pain, in this study we analyzed the expression levels of cytokines in conservatively medically treated patients with chronic back pain. We hypothesized that, despite medical therapy, patients with chronic back pain will show high levels of inflammatory mediators related to back pain, and low levels of anti-inflammatory mediators.

Materials and methods

Sample collection

Blood was collected from patients with chronic back pain and pain-free healthy controls after written informed consent and approval by ethics committee of Canton of Lucerne (Study 730–May 16, 2013) were obtained. Plasma was isolated by Ficoll density gradient (Bioconcept, Allschwil, Switzerland) centrifugation for 20 min at 800 g in Greiner Leucosep tubes (Huberlab, Aesch, Switzerland).

Inclusion/exclusion criteria

We included patients aged over 18 years old with long-term chronic back pain resistant to therapy. Any chronic back pain (cervical, thoracic, or lumbar spine origin) was considered, with or without irradiation to the extremities. Patients who had spine surgery in the past were also included. We excluded patients with acute back pain, and those who reacted to medical therapy.

Multiplex ELISA assay

The concentration of eight cytokines was analyzed in human plasma from patients with chronic back pain (n=23) and age-matched pain-free healthy controls (n=30), by Bio-Plex Pro Cytokine Chemokine and Growth Factor Assay (Bio-Rad Laboratories AG, Cressier, Switzerland). Data were collected and analyzed using a Bio-Rad BioPlex 200 instrument equipped with Bio-Plex Manager software (Bio-Rad). We measured the concentrations of (IL)-2, IL-4, IL-6, IL-10, granulocyte-colony stimulating factor (G-CSF), granulocyte-macrophage colony-stimulating factor (GM-CSF), MCP1, and TNF-α.

Quantitative ELISA assays

Quantitative determination of CXCL6, IL-1β, CCL5, and stromal cell-derived factor 1 alpha (CXCL12) (Quantikine ELISA kits – R&D Systems, Abingdon, UK) in human plasma of patients with chronic back pain (n=23) and age-matched pain-free healthy controls (n=16) was done with a DTX 880 Multiplex reader (Beckman Coulter, Nyon, Switzerland). Experiments were performed according to the respective manufacturer's protocols.

Statistical analysis

For statistical analysis and comparison between the main two groups of chronic back pain and healthy control, we used the non-parametric Mann–Whitney–Wilcoxon *U* test for independent variables. For multiple comparison and statistical analysis between groups and subgroups, we used the one-way Anova test with Tukey's post-hoc analysis. Data analysis was performed with SPSS version 24.0 for Windows (IBM Corporation, Armonk, NY, USA). Significance was indicated as $*p<0.05$; $**p<0.01$; $***p<0.001$.

Results

Patients with chronic back pain

For this study, we collected plasma from patients with chronic back pain (n=23) undergoing therapy at a pain clinic. All patients had back pain for more than 1 year, and 56% for more than 5 years. The mean age was 52.5 years old (SD =±15.9; range =26–84). Back pain was prevalently lumbar with discogenic origin, in two-thirds of the cases. The reported maximum pain intensity value was on average

7.7±1.7 (range =4–10; Numeric Pain Rating Scale, NPRS= 0–10/10). All patients were long-term treated with one or more conservative medical therapies, such as physiotherapy and NSAIDs, alone or combined with antidepressants and/or muscle-relaxants. Patients' characteristics are summarized in Table 1.

Cytokine plasma levels comparison between patients with chronic back pain and healthy controls

We measured the concentrations of eight cytokines in human plasma, by multiplex ELISA assay. In patients with chronic back pain (n=23), we found significantly lower concentrations of G-CSF (Figure 1A), IL-4 (Figure 1D), and MCP1 (Figure 1G) compared to age-matched pain-free healthy controls (n=30). We found no significant variations in GM-CSF (Figure 1B), IL-2 (Figure 1C), IL-6 (Figure 1E), IL-10 (Figure 1F), or TNF-α (Figure 1H) plasma levels.

Quantitative single ELISA assay showed significantly lower concentrations of CCL5 (Figure 2A) and CXCL6 (Figure 2B) in plasma of patients with chronic back pain (n=23), compared to age-matched pain-free healthy controls (n=16). There were no significant differences in CXCL12 (Figure 2C) or IL-1β (Figure 2D) plasma concentrations.

Furthermore, we divided the chronic pain group into 14 subgroups (according to type of pain, cause of pain, spine surgery operation, medical therapy, and pain history), and we performed a multiple comparison statistical analysis to analyze if there were differences between subgroups and to compare them to the healthy control group. For all analyzed cytokines, there were no significant differences between chronic pain subgroups. The multiple comparison analysis confirmed statistical significance between chronic back pain and the healthy control group for G-CSF, IL4, MCP1 (Table 2), CCL5, and CXCL6 (Table 3). These cytokines were significantly different in almost all subgroups compared to the healthy control group. Tables 2 and 3 show, for each subgroup, the mean ± standard deviation and the statistical significance (*p*-value) of comparison with the healthy control group.

Discussion

Contrary to our expectations of positively correlating chronic back pain to increased levels of pro-inflammatory

Table 1 Medically treated patients with chronic back pain

Demographics and clinical variables	Subgroups	Chronic back pain (n=23), % (n)	Females, % (n)	Males, % (n)
Number of patients			61 (14)	39 (9)
Age	≤45 years old	35 (8)	29 (4)	44 (4)
	>45 years old	65 (15)	71 (10)	56 (5)
Type of pain	Lumbar	74 (17)	71 (10)	78 (7)
	Cervical	13 (3)	14 (2)	11 (1)
	Multiple types[a]	13 (3)	14 (2)	11 (1)
	Additional radiating pain[b]	70 (16)	71 (10)	67 (6)
Origin of nociceptive pain	Discogenic	61 (14)	71 (10)	44 (4)
	Facetogenic	22 (5)	21 (3)	22 (2)
	Osteochondrosis	9 (2)	7 (1)	11 (1)
	Vertebrogenic	4 (1)	(0)	11 (1)
	Fracture	4 (1)	(0)	11 (1)
Post-operation pain[c]		43 (10)	50 (7)	33 (3)
Conservative therapy	Medication and/or physiotherapy	100 (23)	100 (14)	100 (9)
	NSAIDs	52 (12)	43 (6)	67 (6)
	NSAIDs+other drugs[d]	43 (10)	57 (8)	22 (2)
	Physiotherapy	4 (1)	(0)	11 (1)
Pain history	1–5 years	43 (10)	43 (6)	44 (4)
	6–10 years	30 (7)	29 (4)	33 (3)
	>10 years	26 (6)	29 (4)	22 (2)
Maximum pain intensity NPRS 0–10/10[e]		7.7±1.7	7.2±2.3	7.9±1.4

Notes: [a]Multiple types: 2 or more type of pain–lumbar and/or cervical and/or thoracic. [b]There is local pain at the spine area, but additional unspecific radiation to extremities. [c]Post-operation pain: indicates chronic pain following spine surgery operation in the pain area. [d]Other drugs: antidepressants and/or muscle-relaxants. [e]Average ± standard deviation.

Abbreviations: NSAIDs, non-steroidal anti-inflammatory drugs; NPRS, Numeric Pain Rating Scale.

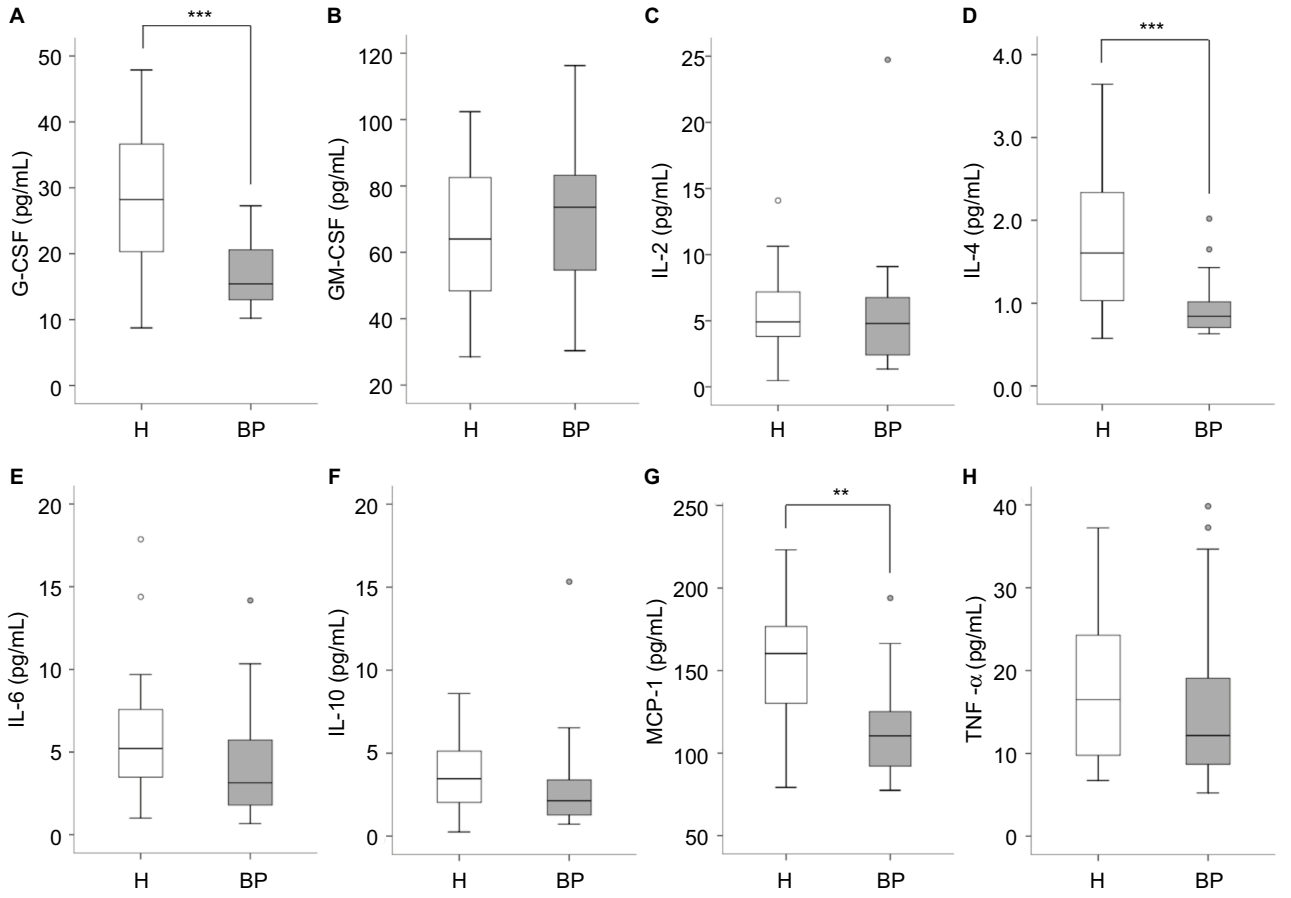

Figure 1 Multiplex ELISA assay. Box plots show significantly lower levels of G-CSF (**A**), IL-4 (**D**), and MCP-1 (**G**) detected in plasma of medically treated patients with chronic back pain (n=23), compared to pain-free healthy controls (n=30). No significant differences were obtained for GM-CSF (**B**), IL-2 (**C**), IL-6 (**E**), IL-10 (**F**), or TNF-α (**H**). The line across the box indicates the median. **$p<0.01$; ***$p<0.001$.

Abbreviations: G-CSF, granulocyte-colony stimulating factor; GM-CSF, granulocyte-macrophage colony-stimulating factor; IL, interleukin; MCP-1, monocyte chemoattractant protein 1; TNF-α, tumor necrosis factor alpha; H, healthy controls; BP, back pain.

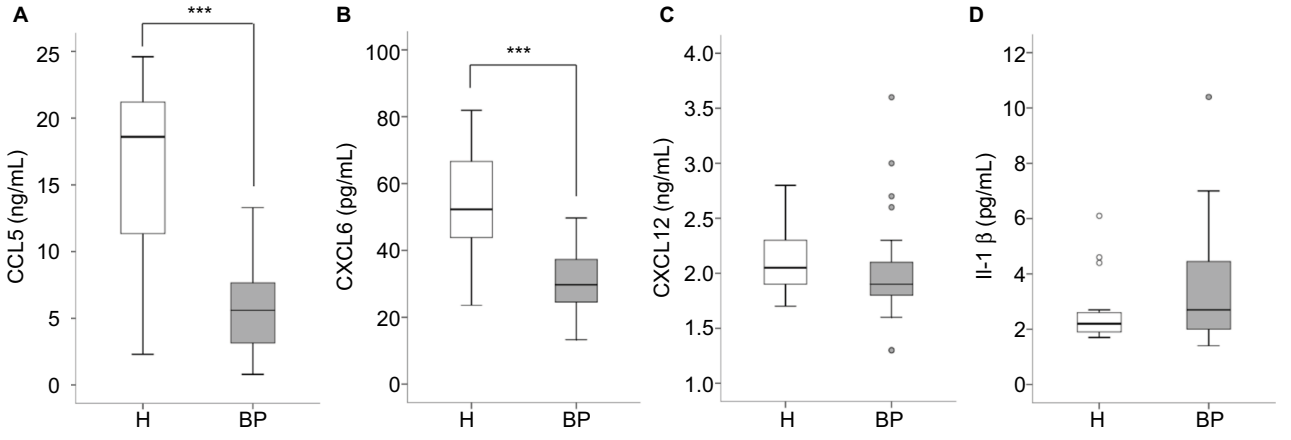

Figure 2 Quantitative ELISA assay. Box plots show significantly lower concentrations of CCL5 (**A**) and CXCL6 (**B**) detected in plasma of medically treated patients with chronic back pain (n=23), compared to pain-free healthy controls (n=16). No significant differences were found for CXCL12 (**C**) and IL-1β (**D**). The line across the box indicates the median. ***$p<0.001$.

Abbreviations: CCL5, RANTES; CXCL6, granulocyte chemotactic protein 2; CXCL12, stromal cell-derived factor 1 alpha; H, healthy controls; BP, back pain.

cytokines, in this study we found that plasma levels of pro-inflammatory cytokines were comparable between medically treated patients with chronic back pain and pain-free healthy controls. Furthermore, patients with chronic back pain showed significantly lower plasma levels of chemotactic and anti-inflammatory cytokines.

Table 2 Multiple comparative statistical analysis between chronic back pain groups vs healthy controls

Groups		G-CSF (pg/mL)	GM-CSF (pg/mL)	IL-2 (pg/mL)	IL-4 (pg/mL)	IL-6 (pg/mL)	IL-10 (pg/mL)	MCP-1 (pg/mL)	TNF-α (pg/mL)
	Healthy controls (n=30)	27.8±10.5	65.2±20.1	5.5±2.8	1.7±0.9	5.9±3.7	3.6±1.9	150.9±38.9	18.0±8.9
	Chronic back pain (n=23)	17.0±5.0***	70.7±22.3	5.5±4.8	0.9±0.3***	4.5±3.6	3.1±3.1	115.3±29.8**	16.2±10.8
1	Lumbar pain (n=17)	16.1±4.9***	68.3±21.9	4.6±2.5	0.9±0.4***	4.2±3.3	2.5±1.8	114.2±26.6*	16.4±12.0
2	Cervical pain (n=3)	15.9±4.1	81.5±34.0	11.5±11.8	0.8±0.2	5.9±7.2	6.4±7.8	135.8±51.9	10.7±3.9
3	Multiple types of pain[a] (n=3)	22.6±4.5	73.4±15.0	4.4±2.1	1.0±0.4	4.3±2.5	3.0±1.3	100.6±21.4	20.2±7.8
4	Additional radiating pain[b] (n=16)	17.5±5.3***	67.8±22.8	4.8±2.3	0.9±0.2***	3.9±2.7	2.3±1.2	109.6±19.0**	15.7±9.6
5	No radiating pain (n=7)	15.8±4.6***	77.3±20.9	7.0±8.3	1.1±0.5	5.8±5.3	4.8±5.2	128.1±45.6	17.2±14.1
6	Discogenic pain origin (n=14)	15.9±5.07***	76.0±18.6	5.3±5.9	0.9±0.4***	3.5±3.1	3.2±3.9	104.3±22.1***	16.9±12.4
7	Other origins of pain[c] (n=9)	18.6±4.9*	62.6±26.0	5.7±2.8	1.0±0.3**	5.9±4.1	2.9±1.6	132.3±33.4	15.0±8.4
8	Post-operation pain[d] (n=10)	18.7±5.5**	73.8±23.3	7.5±6.5	1.0±0.3**	4.2±3.2	4.2±4.3	113.7±27.8	20.8±11.6
9	No operation (n=13)	15.6±4.4***	68.3±22.0	3.9±2.3	0.9±0.4***	4.7±4.1	2.2±1.5	116.4±32.4	12.6±9.1
10	NSAIDs (n=12)	16.8±5.0***	75.3±20.1	6.2±6.1	1.0±0.4***	5.0±4.2	3.8±4.0	120.6±30.8	15.8±11.6
11	NSAIDs+other drugs[e] (n=10)	16.6±5.3***	64.2±25.0	4.3±2.9	0.8±0.2***	3.3±2.5	1.8±0.8	103.7±23.1**	14.7±9.1
12	Pain history 1–5 years (n=10)	18.6±6.3**	69.5±30.1	7.5±6.3	0.9±0.3***	4.9±4.5	4.2±4.3	116.8±37.8	15.9±10.2
13	Pain history 6–10 years (n=7)	15.3±3.8***	69.5±14.6	2.8±1.3	0.9±0.3**	3.1±1.9	1.9±1.3	105.2±13.1*	12.6±8.4
14	Pain history >10 years (n=6)	16.2±3.6**	74.1±16.6	5.2±3.1	1.0±0.5	5.4±3.6	2.6±1.8	124.5±29.7	20.7±14.3

Notes: Mean ± standard deviation (*p*-value: *$p<0.05$; **$p<0.01$; ***$p<0.001$). [a]Multiple types: 2 or more type of pain–lumbar and/or cervical and/or thoracic. [b]There is local pain at the spine area, but additional unspecific radiation to extremities. [c]Facetogenic, osteochondrosi, vertebrogenic, fracture. [d]Post-operation pain: indicates chronic pain following spine surgery operation in the pain area. [e]Other drugs: antidepressants and/or muscle-relaxants.

Abbreviations: G-CSF, granulocyte-colony stimulating factor; GM-CSF, granulocyte-macrophage colony-stimulating factor; IL, interleukin; MCP-1, monocyte chemoattractant protein 1; TNF-α, tumor necrosis factor alpha; NSAIDs, non-steroidal anti-inflammatory drugs.

Table 3 Multiple comparative statistical analysis between chronic back pain groups vs healthy controls

Groups		CCL5 (ng/mL)	CXCL6 (pg/mL)	CXCL12 (ng/mL)	IL1β (pg/mL)
	Healthy controls (n=16)	16.4±6.9	53.4±17	2.1±0.3	2.7±1.3
	Chronic back pain (n=23)	5.8±3.7***	31.1±8.3***	2.0±0.5	3.4±2.1
1	Lumbar pain (n=17)	6.0±3.8***	31.9±6.5***	2.0±0.5	3.4±2.3
2	Cervical pain (n=3)	4.0±1.7***	24.7±11.7***	2.5±0.9	4.1±1.6
3	Multiple types of pain[a] (n=3)	6.6±5.3*	32.9±14.6	1.9±0.3	2.5±1.1
4	Additional radiating pain[b] (n=16)	6.1±3.6***	31.4±9.2***	1.9±0.4	3.0±1.5
5	No radiating pain (n=7)	5.0±4.0***	30.4±6.2***	2.3±0.7	4.3±3.0
6	Discogenic pain origin (n=14)	4.8±3.7***	30.2±9.8***	2.0±0.4	3.1±2.3
7	Other origins of pain[c] (n=9)	7.4±3.3***	32.4±5.6***	2.1±0.7	3.8±1.8
8	Post-operation pain[d] (n=10)	5.9±3.9***	32.2±11.1***	1.9±0.3	3.2±1.2
9	No operation (n=13)	5.7±3.7***	30.2±5.7***	2.2±0.6	3.5±2.7
10	NSAIDs (n=12)	6.0±3.6***	32.9±8.1***	2.1±0.6	4.2±2.6
11	NSAIDs+other drugs[e] (n=10)	5.6±4.1***	29.0±8.9***	1.9±0.4	2.4±0.9
12	Pain history 1–5 years (n=10)	4.0±2.1***	29.1±6.3***	1.9±0.7	3.4±1.9
13	Pain history 6–10 years (n=7)	6.1±4.1***	29.4±11.6***	2.2±0.4	2.7±1.1
14	Pain history >10 years (n=6)	8.6±4.0**	36.4±5.0*	2.1±0.4	4.3±3.2

Notes: Mean ± standard deviation (*p*-value: *$p<0.05$; **$p<0.01$; ***$p<0.001$). [a]Multiple types: 2 or more type of pain–Lumbar and/or Cervical and/or Thoracic. [b]There is local pain at the spine area, but additional unspecific radiation to extremities. [c]Facetogenic, Osteochondrosi, Vertebrogenic, Fracture. [d]Post-operation pain: indicates chronic pain following spine surgery operation in the pain area. [e]Other drugs: antidepressants and/or muscle-relaxants.

Abbreviations: CCL5, RANTES; CXCL6, granulocyte chemotactic protein 2; CXCL12, stromal cell-derived factor 1 alpha; IL, interleukin; NSAIDs, non-steroidal anti-inflammatory drugs.

We quantified, by immunoassay, a circulating concentration of possible biomarkers related to pain, inflammation, and degeneration of the IVD, and we found that, in plasma of medically treated patients with chronic back pain, there were significantly lower concentrations of chemokines, such as MCP1, CCL5, and CXCL6. Furthermore, pro-inflammatory cytokines, such as (IL)-2, IL-6, IL-1β, IL-10, TNF-α, GM-CSF, and CXCL12 showed no significant differences between chronic back pain patients and pain-free healthy controls. This is a surprising result, because in several studies a high

expression of pro-inflammatory and chemotactic cytokines has been found in herniated and degenerated IVD,[11,12,15,17,18,26,27] and correlated with the pathogenesis of pain.[12,16,28] Elevated plasma levels of MCP-1 have been observed in the blood at the chronic stage of complex regional pain syndrome,[20] while high levels of CCL5 and CXCL6 have been found in patients with lumbar disc degeneration.[21] However, in those studies the intervention with medical treatments was mostly not specified. In a study where one of the exclusion criteria was the use of analgesic drugs, elevated serum levels of TNF-α and IL-6 were shown in individuals with back pain, due to herniated lumbar disc.[23]

Interestingly, we also showed significantly lower plasma levels of anti-inflammatories cytokines IL-4 and G-CSF in medically treated patients with chronic back pain. The anti-inflammatory cytokines IL-4 and IL-10 have been demonstrated to have potential as treatments for persistent inflammatory pain,[29] but low concentrations of these two cytokines have been found in patients with chronic widespread pain and associated with a possible contribution to pain pathogenesis.[6] G-CSF, hematopoietic growth factors for neutrophils, could have immune-stimulatory effects, and serum levels are often elevated in response to infection;[30] however, G-CSF has also been proven to be an anti-inflammatory immune-modulator.[31]

There is a possibility that reduced plasma levels of some cytokines and/or chemokines are related to chronic back pain; however, the findings could be the result of the exposure to long-term conservative medical therapy. Plasma cytokine levels have been shown to be altered by various environmental and personal factors: medical treatments,[34–37,39,40] depression,[41,42] physical activity,[43] and alcohol and nicotine consumption.[44] In our study, patients had a long-term history of chronic pain and long exposure to physiotherapy and pharmaceutical treatments with NSAIDs, also combined with other drugs (antidepressants and muscle-relaxants). Such a multimodal conservative therapy is a standard approach to treating chronic back pain of different origin. The drugs which are commonly prescribed for chronic low back pain[32,33] can reduce cytokine expressions, as has been demonstrated for antidepressants[34,35] and NSAIDs.[36,37] In patients with herniated IVD, higher plasma levels of TNF-α decreased after treatment with an opioid pain medication (tramadol),[22] while natural phyto-pharmaceutical components, such as curcuma and epigallocatechin 3-gallate, have been recently shown to reduce IVD inflammation in vitro.[20,25] It is possible that pain still persists, while NSAIDs and other anti-inflammatory molecules lower the levels of pro-inflammatory cytokines. We show, in another study,[38] that people with spinal cord injury (SCI) had, despite higher infection rates and elevated serum C-reactive protein concentrations, lower plasma levels of TNF-α and other cytokines when compared with age matched able bodied healthy controls. Similar to the chronic back pain patients, persons with SCI consume above-average NSAIDs, antidepressants, and muscle-relaxants. We further analyzed chronic back pain subgroups in a multiple statistical comparison analysis to test if different conditions, such as type and origin of pain, spine surgery operation, medical therapy, and history of pain, influence cytokines plasma levels. We found no differences between chronic back pain subgroups and, in most cases, the analysis confirmed a significant decrease of cytokines in chronic back pain subgroups compared to the healthy control group.

On the other hand, not all studies could correlate cytokines with chronic pain. Andrade et al[45] observed that local IL-1β and IL-6 cytokine expression of lumbar disc hernia patients, suffering from chronic sciatic pain, did not differ from those of the painless healthy control group. Such a lack of correlation between systemic and local cytokine levels and pain is in line with the observation that some anti-inflammatory drugs perform poorly as a treatment for chronic pain. The minor effects of most current medications[46] could be because inflammation is not the only causal variable for back pain. Indeed inflammation is viewed as only one of the aspects, according to the recognized bio-psycho-social model of pain.[47] Several studies revealed that chronic pain is related to a pain memory encoded within the nervous system,[48,49] and neural modifications could be a possibility to reverse the pain memory circuits and alleviate chronic pain.[50,51]

A limitation of this study is the small number of samples due to recruitment of long-term chronic pain patients resisting to every type of pain therapy. Generally, back pain evolves to chronic pain in approximately one third of the affected individuals,[52] and the further two-thirds of patients with different grades of chronicity are satisfied with medical therapy.[53] Further analysis of a larger patient sample and, even better, a longitudinal study could be very interesting in the future. A minor limitation of our study was the unequal gender distribution in the healthy control group for one of the immunoassays (Multiplex ELISA assay). However, this is unlikely to have introduced bias because there were no differences between males and females in the chronic back pain group and in the other immunoassay (Quantitative ELISA assay).

Conclusion

We cannot conclude if the observed reduced plasma levels of inflammatory mediators in medically treated patients with chronic back pain are related to the chronic pain or are due to

the long-term effects of medications. Inflammatory mediators could be altered by both physiological and environmental factors. Our results support the idea that inflammation is not the only cause of chronic back pain, and that other factors are involved in the process of pain. In view of that, more studies are needed to discover the underlying reason for the decrease in the studied biomarkers, which may ultimately lead to better pain management.

In addition, since chronic back pain is considered to be a disease of the central nervous system,[54] we suggest that it could be worthwhile to analyze biomarkers implicated in the regulation of the central nervous system and the risk of developing chronic back pain. For example, a polymorphism of the potassium channel alpha subunit KCNS1 is one of the first prognostic indicators of chronic pain risk;[55] the calcium channel gamma subunit gene CACNG2 significantly affects susceptibility to chronic pain following nerve injury,[56] and the brain-derived neurotrophic factor BDNF regulates neuronal function and induces expression of pain-associated cation channels.[15]

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Author contributions

SC: substantial contributions to conception and design, data acquisition, data analysis and interpretation, drafting the article; DP: data acquisition, data analysis, critically revising the article; AB: data analysis and interpretation, critically revising the article; GL: patient recruitment and consent, sample management, critically revising the article; JVS: substantial contributions to conception and design, data interpretation, critically revising the article, final approval of the version to be published, agreement to be accountable for all aspects of the work. All authors read and approved the final manuscript.

Disclosure

The authors report no conflicts of interest in this work.

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