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Effect of neural therapy on NGF and BDNF serum levels in patients with chronic pain. A pilot study

Efecto de la terapia neural sobre los niveles séricos del NGF y el BDNF en pacientes con dolor crónico. Estudio piloto

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Abstract

Introduction: Neurotrophins (NT) are a family of proteins consisting of the nerve growth factor (NGF), the brain-derived neurotrophic factor (BDNF) and NT-3 and NT-4/5. These proteins play an essential role in neuronal survival, differentiation, and proliferation.
Objectives: To analyze the variations of NGF and BDNF serum levels in patients with chronic pain after undergoing neural therapy and to establish the effects of this type of intervention on their quality of life.
Materials and methods: Prospective pilot study conducted in 10 patients with chronic pain treated with neural therapy between July 2017 and April 2018 in Bogotá D.C., Colombia. Three consultations were performed (one in which the intervention was initiated, and two follow-up visits every three weeks). During each consultation, the patients' quality of life was assessed using the SF-12 scale and their NGF and BDNF serum levels were measured. Data were analyzed by means of descriptive statistics, using medians and interquartile ranges for quantitative variables, and absolute frequencies and percentages for qualitative variables.
Results: The median score on the SF-12 scale tended to improve in the first and second follow-up visits compared with the baseline score (pre-intervention), particularly during the first follow-up visit (consultation No. 1: 34.5; follow-up No. 1: 39.5, and follow-up No. 2: 38). Median NGF serum levels had a downward trend after the intervention, particularly in the first follow-up visit (157.6, 42.95, and 237.8, respectively), and in the case of BNDF, an overall downward trend was also found (29.96, 19.24 and 20.43, respectively). An improvement in quality of life related to the decrease in the serum levels of both neurotrophins was observed.
Conclusion: Neural therapy intervention reduced NGF and BDNF serum levels and improved the quality of life of the participants. Therefore, the behavior of these neurotrophins could become a biomarker for the diagnosis, treatment, and follow-up of patients with chronic pain.
Keywords: Nerve Growth Factors; Pain Management; Local Anesthetics; Quality of Life; Pain (MeSH).

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Resumen

Introducción. Las neurotrofinas (NT) son una familia de proteínas conformada por el factor de crecimiento nervioso (NGF), el factor neurotrófico derivado del cerebro (BDNF) y las neurotrofinas NT-3 y NT-4/5; estas proteínas tienen un papel esencial en la supervivencia, diferenciación y proliferación neuronal.
Objetivos. Analizar las variaciones de los niveles séricos del NGF y el BDNF en pacientes con dolor crónico luego de recibir terapia neural y establecer los efectos de este tipo de intervención en su calidad de vida.
Materiales y métodos. Estudio piloto prospectivo realizado en 10 pacientes con dolor crónico tratados con terapia neural entre julio de 2017 y abril de 2018 en Bogotá D.C., Colombia. Se realizaron 3 consultas (una en la que se inició la intervención y dos de control cada tres semanas) y en cada una se evaluó la calidad de vida mediante el cuestionario de salud SF-12 y se midieron los niveles séricos del NGF y el BDNF. Los datos se analizaron mediante estadística descriptiva, utilizando medianas y rangos intercuartiles para las variables cuantitativas, y frecuencias absolutas y porcentajes para las cualitativas.
Resultados. La mediana de puntaje del cuestionario SF-12 tendió a mejorar en el primer y segundo control comparada con la puntuación inicial (antes de la intervención), en particular en el primer control (consulta 1: 34.5; control 1: 39.5, y control 2: 38). La mediana de los niveles séricos del NGF tendió a disminuir luego de la intervención, en particular en el primer control (157.6, 42.95 y 62.2, respectivamente), y en el caso del BNDF, la tendencia global también fue hacia la disminución (29.96, 19.24 y 20.43, respectivamente). Se observó una mejora en la calidad de vida relacionada con la disminución de los niveles séricos de ambas neurotrofinas.
Conclusión. La intervención de terapia neural produjo una reducción en los niveles séricos del NGF y el BDNF y mejoró la calidad de vida de los participantes; por tanto, el comportamiento de estas neurotrofinas podría convertirse en un biomarcador para el diagnóstico, tratamiento y seguimiento de pacientes con dolor crónico.
Palabras clave: Factores de crecimiento nervioso; Manejo del dolor; Anestésicos locales; Calidad de vida; Dolor (DeCS).

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Introduction

Neurotrophins, also known as neurotrophic factors, were first described in the 1960's by Rita Levi-Montalcini, as reported by Bradshaw *et al.*[1] Currently, it is known that they are proteins that modulate the processes of neuronal differentiation, maturation, growth, regeneration, survival and death and that they regulate the mechanisms involved in synaptic processes and neuronal plasticity.[2-5] Likewise, they induce differentiation of progenitor cells for the formation of new neurons.

Neurotrophin production was initially identified in tissues considered as targets, on which actions of the nervous system are performed; however, it is now commonly accepted that its production is both central and peripheral.[6] It has also been demonstrated that these molecules belong to a family of growth factors and that they bind to receptors in neuronal cells.

There are two types of receptors to which neurotrophins can bind: p75 and the Trk family (tyrosine kinase), which, once activated, trigger intracellular signaling cascades that end with the expression of the genes responsible for neuronal response.[7-9]

The neurotrophins described in humans to date are nerve growth factor (NGF), brain-derived neurotrophic factor (BDNF), neurotrophin 3 (NT-3) and neurotrophin 4/5 (NT-4/5).[10-12] Others such as NT-6 and NT-7 have recently been found in some animal species.[13]

NGF is released by the target cells of the nervous system. It plays a fundamental role in the survival and maintenance processes of sympathetic and sensory neurons, has a high affinity for TrkA receptors, and is considered an inflammatory mediator and pain modulator.[14]

BDNF acts on neurons of the central and peripheral nervous system and aims to maintain the survival of existing neurons and promote the growth and differentiation of new ones, keeping synaptic mechanisms in optimal conditions. It has been identified in the hippocampus, the cerebellum, the cerebral cortex, the ventral tegmental area, and the basal forebrain, which are areas involved in the mechanisms of learning, memory, and motivation. Likewise, BDNF is one of the most active neurotrophins in neurogenesis, has also been detected in several peripheral tissues, and its receptors are TrkB and p75.[15]

NGF and BDNF are responsible for the optimal maintenance of the communication mechanisms of the nervous system, both central and peripheral, and therefore alterations in their concentration are observed in pathological states.[16,17]

In 2018, Patatel *et al.*[18] confirmed that NGF is an important peripheral pain mediator, especially in relation to inflammation, since elevated levels are associated with hyperalgesia. Likewise, García-Cosamalón *et al.*[19] reported an increase in NGF and BDNF serum levels and their receptors in cases of pain associated with diseases of the intervertebral disc; moreover, this increase was related to the activity of pro-inflammatory cytokines.

On the other hand, neural therapy is a medical treatment that involves the use of local anesthetics such as procaine or lidocaine in low doses, taking into account their cell membrane stabilizing action, dielectric capacity and other properties that influence the nervous system and are the object of research. When applied to specific sites, determined based on each patient's medical history, this therapy modifies interferences in signaling systems and restores the proper functioning of the biological program.[20-22]

Considering the above, the objectives of this study were to analyze changes in NGF and BDNF serum levels in patients with chronic pain after receiving neural therapy and to determine the impact of this type of intervention on their quality of life.

Materials and methods

Design and study population

A prospective pilot study was conducted in 10 patients with chronic pain who attended the Neural Therapy Consultation Service of the master's degree in Alternative Medicine offered by the Faculty of Medicine of the Universidad Nacional de Colombia between July 2017 and April 2018. Data were collected during 3 consultations, one at the start of the intervention and two at follow-up consultations every 3 weeks. At each visit, the patients' quality of life was assessed using the SF-12 health questionnaire and their NGF and BDNF serum levels were measured.

The following inclusion criteria were taken into consideration for patient selection: being treated by the neural therapy service for chronic pain;[23] being between 18 and 60 years of age; attending the 3 consultations; agreeing to participate in the study; and signing the informed consent form. In contrast, patients with hemorrhagic syndromes; immunocompromised; with cancer undergoing chemotherapy or radiotherapy; with convulsive or degenerative disorders; heavy alcohol users, drug addicts, or those who were under the influence of sedatives or hypnotics at the time of consultation; patients in treatments involving the use of needles; patients with chronic kidney disease and chronic metabolic disease; and those who did not consent to the administration of the tests were all excluded.

Procedures

During the first consultation, each patient was interviewed to obtain their medical history and establish the therapy to be implemented according to the parameters of neural therapy. Quality of life was evaluated before the intervention by doing a clinical assessment and administering the SF-12 questionnaire; a blood sample was also taken to measure pre-intervention NGF and BDNF serum levels. Before the end of this first consultation, the neural therapy intervention was performed.

Subsequently, two follow-up consultations took place every three weeks when, once again, NGF and BDNF serum levels were measured, and quality of life was assessed using the SF-12 questionnaire. Based on clinical evaluation, instrument scores, and symptomatic manifestations of pain in each patient, neural therapy was adjusted during each intervention.

Finally, the serum values of the two neurotrophins tested in the 10 patients and the scores obtained in the SF-12 questionnaire in each of the 3 consultations were analyzed.

Instruments

SF-12 Health Questionnaire

The SF-12 questionnaire was created in 1996 as a shorter version of the SF-36 health questionnaire; it assesses the same items as the original through more concise questions, yet the scoring and interpretation are same. The instrument is administered in 5 to 10 minutes and is composed of 12 questions or items that evaluate positive and negative aspects of health status related to the following considerations:

General health perception: to assess patients' personal perception of their health.
Physical functioning: to assess the extent to which health status limits moderate or strenuous physical activity.
Role - physical: to assess the extent to which physical health interferes with work and other daily activities.
Pain: to assess the severity of pain and its impact on usual work, both at home and away.
Energy/vitality: to compare the sensation of energy and vitality with the sensation of tiredness and exhaustion.
Social functioning: to assess the degree to which physical or emotional health problems interfere with the usual social life.
Role - emotional: to assess the extent to which emotional problems interfere with work or other daily activities.
Mental health: to assess overall mental health, including aspects such as depression, anxiety, behavior control, and general well-being.

The response options of this instrument are presented using Likert scales (with scores from 2 to 6 depending on the item). The total score ranges from 0 to 100, and the lowest score implies a worse health-related quality of life. Scores from items of the same dimension are averaged to create specific scores, and unanswered items are not considered.[24-26]

Serum analysis

Commercially available enzyme-linked immunosorbent assay (ELISA) kits were used to measure NGF and BDNF serum levels: the ab99978 kit was used for BDNF and the ab99986 kit for NGF; both are produced by Abcam®. All measurements were made according to the manufacturer's instructions.

Statistical analysis

Data were analyzed by means of descriptive statistics, using medians and interquartile ranges (IQR) for quantitative variables, and absolute frequencies and percentages for qualitative variables. Data for the variables BDNF, NGF and SF-12 questionnaire score were presented separately depending on the consultation (consultation 1, follow-up 1 and follow-up 2).

Ethical considerations

The study took into account the ethical principles for medical research in human subjects established by the Declaration of Helsinki[27] and the scientific, technical and administrative standards for health research of Resolution 8430 of 1993 of the Ministry of Health of Colombia.[28] The research project was approved by the Ethics Committee of the Faculty of Medicine of the Universidad Nacional de Colombia according to Minutes No. 017-205-16 of September 22, 2016.

All patients who took part in the study read, accepted, and signed the informed consent. The confidentiality of patients' identities, personal information, and test results contained in their medical records was maintained.

Results

The median age of the participants was 40.5 years (IQR: 33-45) and most patients were female (80%). All patients consulted for pain in various body areas with a median duration of 9 years (IQR: 5-16) and all reported having received treatment with conventional or alternative medicine previously (Table 1).

Table 1. Characteristics of the study participants.

Patient	Age (years)	Sex	Type of pain	Pain duration
1	53	Female	Abdominal and lumbar pain	2 months
2	33	Female	Thoracolumbar pain	19 years
3	39	Male	Low back pain	16 years
4	26	Female	Pelvic pain and dysmenorrhea	10 years
5	42	Female	Pain in the left breast due to chronic mastitis	5 months
6	45	Female	Headache	5 years
7	40	Female	Joint pains	11 years
8	41	Female	Low back pain	25 years
9	27	Female	Joint pains	8 years
10	56	Male	Lower limb pain	7 years

Source: Own elaboration.

The median score obtained by participants in the SF-12 questionnaire tended to improve in the follow-up visits, being more evident in the first one. At the initial consultation, it was 34.5 (IQR: 30-37); in the first follow-up, 39.5 (IQR: 34-43), and in the second follow-up, 38 (IQR: 36-41) (Figure 1).

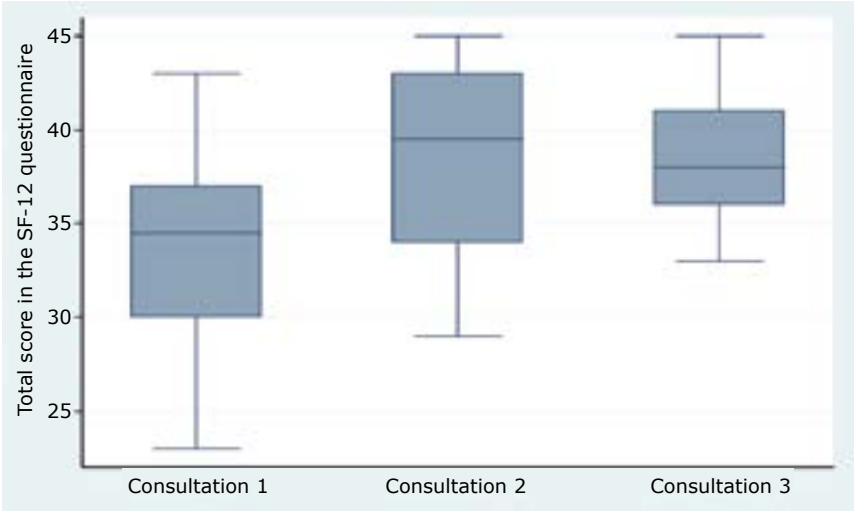

Figure 1. Box-and-whisker plot of the scores obtained by the participants in the SF12 health questionnaire.
Source: Own elaboration.

The median NGF serum levels tended to decrease after the intervention, particularly in the first follow-up. During the initial consultation, it was 157.6 (IQR: 51.9-237.8); in the first follow-up, 42.95 (IQR: 25.9-90.4), and in the second follow-up 62.2 (IQR: 0-223.7) (Figure 2).

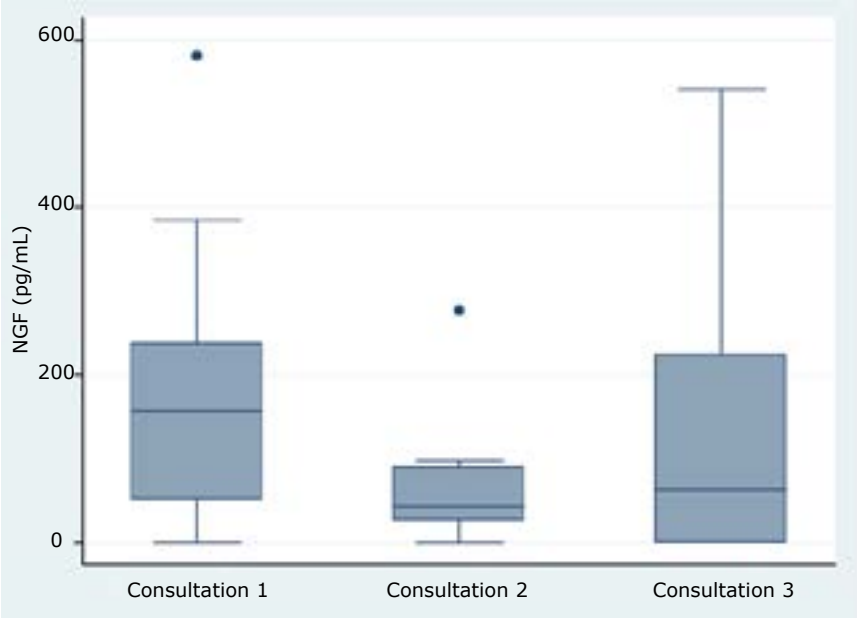

Figure 2. Box-and-whisker plot of variations in nerve growth factor levels in the participants.
Source: Own elaboration.

BDNF serum levels also had an overall downward trend. In the initial consultation, the value was 29.96 (IQR: 26.76-33.4); in the first follow-up, 19.24 (IQR: 16.43-24.52), and in the second follow-up, 20.43 (IQR: 12.81-35.18) (Figure 3).

After analyzing the results, it was found that there was a correlation between the decrease in BDNF and NGF levels in the first follow-up in 9 patients, which was less evident in the second follow-up, and that there was a general correlation between decreased BDNF and NGF levels before the intervention versus the first and second follow-ups in 7 patients. Overall, there was an improvement in quality of life and a decrease in neurotrophin serum levels.

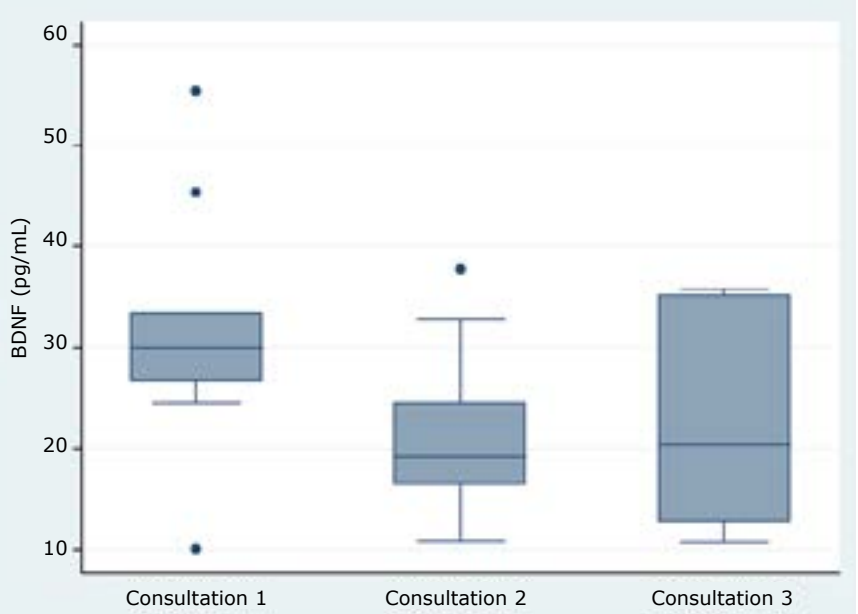

Figure 3. Box-and-whisker plot of variations in brain-derived growth factor levels in the participants.
Source: Own elaboration.

Discussion

Considering that the nervous system is the most elaborate communication system in humans and that its plasticity allows it to interact with other systems, there is an increasing interest in analyzing variations in neurotrophin serum levels when using neural therapy to treat patients with chronic pain.

The present study found that interventions with this type of therapy resulted in an overall improvement in pain symptoms, which were assessed using the parameters of the SF-12 questionnaire, as well as in a decrease in the median NGF and BDNF serum levels, bearing in mind that, compared with the measurement before the intervention, they showed a downward trend in the two follow-up consultations. These findings coincide with those reported by authors such as McKelvey *et al.*[29] and Miller *et al.*[30] in patients who have experienced improvements after undergoing pain treatments that inhibit the action of these neurotrophins.

The literature has reported that the clinical course of patients undergoing chronic pain treatment varies depending on factors related to the initial causes of the manifestations, which could explain the variations in the quality of life and behavior of the serum levels of neurotrophins in these patients.[31] Furthermore, many aspects about neurotrophins, which are associated with biological processes involving the nervous system, are still unknown.[32]

Similarly, there is evidence of elevated BDNF serum levels in post-traumatic situations related to multiple traumas involving painful processes, such wars, which are known as post-traumatic brain injury and post-traumatic stress disorders.[33]

It should be noted that the effects of neural therapy are not always evident since the beginning; in fact, pain may worsen at first and subsequently improve.[34]

As stated by Fisher,[35] one of the fundamental pillars of neural therapy is the autonomic system. It controls communication in all human biological systems through circuits (communication and feedback) organized vertically, creating an information network that runs from the periphery to the central levels.

The findings of this study lead us to consider that, after improving biological conditions by removing the interfering fields that keep patients in a constant state of inflammation, neural therapy not only improves symptoms and, thus, quality of life, but also lowers neurotrophin serum levels because the irritation caused by the inflammatory process is reduced. This has been reported in studies comparing analgesics and other conventional strategies, which have shown a decrease in neurotrophin levels as a result of treatment.[36-38]

Therefore, analyzing the behavior of neurotrophin serum levels following neural therapy interventions could be used as a biomarker for therapeutic success and follow-up of chronic pain patients.[39] This is corroborated by Tu *et al.*,[40] who evaluated the response of neurotrophins with interventions such as acupuncture and obtained results similar to those of the present study.

Conclusions

The neural therapy intervention resulted in a reduction in NGF and BDNF serum levels, which had increased in response to each patient's pathological processes and improved their quality of life. Consequently, it is considered that the behavior of serum levels of these neurotrophins could be a biomarker for the diagnosis, treatment, and follow-up of patients with chronic pain. However, further studies with larger samples and longer follow-up times are necessary to confirm the findings reported here.

Conflicts of interest

None stated by the authors.

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