PROGRAMA DE DOCTORADO EN BIOMEDICINA

DEPARTAMENTO DE FISIOTERAPIA

FACULTAD DE CIENCIAS DE LA SALUD

UNIVERSIDAD DE GRANADA

ALTERACIONES VASCULARES, INGESTA DIETÉTICA Y SU RELACIÓN CON EL DOLOR EN PACIENTES CON FIBROMIALGIA

VASCULAR ALTERATIONS, DIETARY INTAKE, AND THEIR RELATIONSHIP WITH PAIN IN FIBROMYALGIA PATIENTS

UNIVERSIDAD DE GRANADA

TESIS DOCTORAL INTERNACIONA/INTERNATIONAL PhD THESIS

ANTONIO CASAS BARRAGÁN

DICIEMBRE 2020

ALTERACIONES VASCULARES, INGESTA DIETÉTICA Y SU RELACIÓN CON EL DOLOR EN PACIENTES CON FIBROMIALGIA

Esta tesis doctoral ha sido realizada bajo la dirección de:

Prof. Dra. Dña. María Encarnación Aguilar Ferrándiz

Departamento de Fisioterapia

Facultad de Ciencias de la Salud

Prof. Dra. Dña. María Correa Rodríguez

Departamento de Enfermería

Facultad de Ciencias de la Salud

UNIVERSIDAD DE GRANADA

Editor: Universidad de Granada. Tesis Doctorales
Autor: Antonio Casas Barragán
ISBN: 978-84-1306-758-2
URI: http://hdl.handle.net/10481/66401

ÍNDICE / TABLE OF CONTENTS

ÍNDICE DE TABLAS Y FIGURAS / TABLES AND FIGURES INDEX

METODOLOGÍA/METHODS

RESULTADOS/RESULTS

ABREVIATURAS

AAV	Anastomosis arteriovenosas
CGRP	Péptido relacionado con el gen de la calcitonina
CIE	Clasificación Internacional de las Enfermedades
EDA	Aminopeptidasa degradante de la encefalina
EVA	Escala Visual Analógica del Dolor
FM	Fibromialgia
GABA	Ácido g-aminobutírico
IC	Intervalo de Confianza
NMDA	N-metil-D-aspartato
OMS	Organización Mundial de la Salud
ON	Óxido nítrico
OR	Odd Ratio
RR	Riesgo Relativo
SNA	Sistema Nervioso Autónomo
SNC	Sistema Nervioso Central
SNP	Sistema Nervioso Periférico
SNS	Sistema Nervioso Simpático
TI	Termografía infrarroja

ABBREVIATIONS

ACR	American College of Rheumatology
ANCOVA	Two-way Analysis of Covariance
AVA	Arterio-venous Anastomosis
BAI	Beck Anxiety Inventory
BMI	Body Mass Index
CBT	Cognitive Behavioural Therapy
CI	Confidence Interval
CSI	Central Sensitization Inventory
CSS	Central Sensitization Syndrome
DII	Dietary Inflammatory Index
EDA	Enkephalin-degrading Aminopeptidase
EULAR	European League Against Rheumatism
FIQ-R	Revised Fibromyalgia Impact Questionnaire
IASP	International Association for the Study of Pain
IRT	Infrared Thermography
MAOIs	Monoamine oxidase inhibitors
MDR	Monopolar Dielectric Radiofrequency
MFI	Multidimensional Fatigue Inventory
NSAIDs	Non-steroidal anti-inflammatory drugs
NO	Nitric Oxide
PNE	Pain Neuroscience Education
PPT	Pressure Pain Threshold

PSQI	Pittsburgh Quality of Sleep Questionnaire Index
SD	Standard Deviation
SNRIs	Serotonin-noradrenalin reuptake inhibitor
SSRIs	Selective serotonin reuptake inhibitor
SSS	Symptom Severity Score
VAS	Visual Analogue Scale
WMA	World Medical Association
WPI	Widespread Pain Index

RESUMEN

El síndrome de Fibromialgia se define como una enfermedad compleja caracterizada por dolor crónico músculo-esquelético generalizado y difuso y por otros síntomas asociados como hiperalgesia, alodinia, hipersensibilidad al dolor por presión, fatiga continua, rigidez articular matutina, cefaleas, migrañas, colon irritable, trastornos del sueño, ansiedad, depresión y problemas de concentración y memoria. Se clasifica tradicionalmente en dos formas, síndrome de Fibromialgia primario y síndrome de Fibromialgia secundario. En el síndrome de Fibromialgia, la etiología aún permanece incierta, sin embargo, las alteraciones a nivel del Sistema Nervioso Central han sido consideradas como el principal factor clave en el desarrollo y mantenimiento de los síntomas. No obstante, nuevas líneas de investigación han puesto de relieve la presencia de anomalías en la microcirculación sanguínea por cambios a nivel de la inervación nerviosa periférica de la microvasculatura de la piel glabra de estos pacientes y por cambios en las concentraciones séricas de factores inmunológicos e inflamatorios como mecanismos subyacentes a la fisiopatología de esta condición crónica. Este hecho implica que podría existir una respuesta periférica vascular alterada frente a estímulos térmicos que estaría relacionada con la producción de determinados biomarcadores biológicos como el óxido nítrico o las encefalinas, que intervienen en los procesos de sensibilización periférica al dolor y en el mantenimiento del estado inflamatorio. Por otra parte, se ha constatado que el perfil dietético de los pacientes puede influir en el estado basal inflamatorio y contribuir, por tanto, a los fenómenos de hipersensibilidad al dolor en poblaciones con dolor crónico, sin embargo, desde nuestro conocimiento los estudios en este ámbito en la población con FM son muy limitados. Finalmente destacar que las intervenciones terapéuticas actuales para el manejo de este síndrome se centran en tratamientos conservadores (ejercicio físico o entrenamiento de fuerza, técnicas de meditación, terapia cognitivo-conductual, mindfulness, acupuntura, hidroterapia, balneoterapia, masaje y terapias alternativas) y en tratamientos farmacológicos (tramadol, duloxetina, milnacipran, amitriptilina, pregabalina y ciclobenzaprina) con resultados no del todo concluyentes y contradictorios. Es por ello, que su desconocimiento etiológico ha limitado las estrategias terapéuticas y diagnósticas y se precisan estudios de investigación, como los desarrollados en esta tesis doctoral, que profundicen en su fisiopatología desde una perspectiva multidisciplinar.

Con estos antecedentes, las hipótesis de la presente tesis doctoral fueron: 1) Los pacientes con síndrome de Fibromialgia podrían presentar anomalías en la respuesta

vascular periférica a nivel del dorso y la palma de las manos caracterizada por una vasodilatación ante una excesiva inervación sensorial peptidérgica de las anastomosis arteriovenosas y por la dilatación pasiva de las arteriolas debido a la liberación de compuestos como el óxido nítrico de las células endoteliales que, influiría a su vez, sobre la temperatura corporal global de los pacientes con Fibromialgia. 2) En base a la presencia en suero sanguíneo de biomarcadores y neurotransmisores como el óxido nítrico y las encefalinas y su rol en la participación en los procesos de modulación del dolor, planteamos la hipótesis de que la presencia de óxido nítrico circulante así como la actividad en suero sanguíneo de las encefalinasas podrían estar asociados con las variables del dolor y sintomatología de pacientes diagnosticados con Fibromialgia. 3) Dado que las citocinas inflamatorias pueden estar involucradas en el mecanismo subyacente al síndrome de Fibromialgia, planteamos la hipótesis de que un patrón de dieta pro-inflamatoria se asociaría con la hipersensibilidad al dolor y otros síntomas asociados con la Fibromialgia.

Los objetivos principales fueron: 1) Evaluar el componente vascular periférico de la piel del dorso y palma de las manos y la temperatura corporal global como indicadores de la actividad adrenérgica del sistema nervioso simpático y de los procesos de termogénesis en personas diagnosticadas con síndrome de Fibromialgia. 2) Evaluar la presencia en suero sanguíneo de biomarcadores vasodilatadores (óxido nítrico) y la actividad de opioides endógenos (encefalinasas) como indicadores de la microcirculación sanguínea y del estado basal inflamatorio y nociceptivo y su relación con el dolor crónico y la sintomatología en personas diagnosticadas con síndrome de Fibromialgia. 3) Evaluar el potencial inflamatorio de la dieta y su relación con el dolor crónico y la sintomatología en pacientes con Fibromialgia.

Para la consecución de estos objetivos, se llevaron a cabo dos estudios observacionales de casos y controles y un estudio observacional de casos. En el primer estudio, se incluyeron 42 mujeres con síndrome de Fibromialgia y 52 mujeres sanas. En el segundo estudio, se incluyeron un total de 58 mujeres diagnosticadas con Fibromialgia. Finalmente, en el tercer estudio, se incluyeron a 95 mujeres con síndrome de Fibromialgia y 98 mujeres sanas. En primer lugar, se evaluó el flujo sanguíneo vascular periférico y la temperatura corporal central a través del patrón termográfico del dorso y palma de las manos y de la medición de la temperatura timpánica y axilar, respectivamente. Además, se midió los niveles de concentración de óxido nítrico en suero sanguíneo utilizando el método basado en quimioluminiscencia de ozono. En segundo lugar, se analizaron las

actividades séricas de la oxitocinasa y de la aminopeptidasa degradante de la encefalina usando el método de cuantificación de Bradford, así como los niveles de óxido nítrico y su asociación con el umbral y magnitud del dolor eléctrico, el nivel global de dolor autoinformado, los umbrales de dolor por presión, la sensibilización central, el impacto de la Fibromialgia y los niveles de ansiedad. En tercer lugar, analizamos el potencial inflamatorio de la dieta a través del Índice Dietético Inflamatorio y examinamos su relación con cambios sobre los umbrales del dolor por presión, intensidad global del dolor, impacto de los síntomas, fatiga, problemas de sueño y síntomas comunes de ansiedad.

Nuestros resultados indicaron que las mujeres con síndrome de Fibromialgia presentan una mayor temperatura del dorso y palma de las manos en todos los puntos evaluados ($P \leq 0.001$) y una mayor temperatura timpánica ($P=0.002$). Se observaron asociaciones significativas entre los niveles séricos de óxido nítrico y la temperatura mínima en el centro dorsal de la mano dominante ($\beta=-3.501$, $P=0.038$), la temperatura máxima ($\beta=-5.594$, $P=0.016$), mínima ($\beta=-4.090$, $P=0.036$) y media ($\beta=-5.519$, $P=0.015$) del centro de la palma de la mano no dominante y la temperatura máxima en la eminencia tenar de la mano dominante ($\beta=-5.800$, $P=0.017$), así como con la temperatura timpánica ($\beta=-9.321$, $P=0.035$) en las mujeres sanas. Se observaron asociaciones significativas entre los niveles de óxido nítrico circulante y los umbrales de dolor por presión en el occipital dominante ($\beta=0.290$, $P=0.003$) y en el occipital no dominante ($\beta=0.193$, $P=0.034$), así como en el nivel de actividad en Fibromialgia ($\beta=0.031$, $P=0.027$) en la población de mujeres diagnosticadas con Fibromialgia. También se observaron asociaciones de la actividad de la oxitocinasa con el nivel global de dolor autoinformado ($\beta=0.215$, $P=0.023$) y el umbral del dolor por presión del punto de la rodilla dominante ($\beta=2.794$, $P=0.039$). Finalmente, niveles altos de actividad sérica de la aminopeptidasa degradante de la encefalina se correlacionó con una hipersensibilidad del dolor por presión a nivel del punto de la segunda costilla dominante ($\beta=-20.096$, $P=0.049$). Por último, el análisis de regresión lineal confirmó que los umbrales del dolor por presión en el occipital ($\beta=0.234$, $P=0.036$), trapecio ($\beta=0.299$, $P=0.007$), articulación cigoapofisaria ($\beta=0.291$, $P=0.035$), segunda costilla ($\beta=0.204$, $P=0.006$), glúteo ($\beta=0.591$, $P=0.017$), trocánter mayor ($\beta=0.379$, $P=0.041$) y la rodilla ($\beta=0.482$, $P=0.011$) se asociaron con una mayor puntuación en el Índice Dietético Inflamatorio en mujeres con síndrome de Fibromialgia en comparación con las mujeres sanas. Sin embargo, no se encontraron asociaciones significativas entre la puntuación del Índice Dietético Inflamatorio y el resto de síntomas clínicos.

Las principales conclusiones fueron, en primer lugar, que las personas con síndrome de Fibromialgia presentaron una mayor temperatura en todos los puntos analizados del dorso y palma de sus manos y una mayor temperatura timpánica que las mujeres sanas. Estos resultados podrían estar relacionados, por una lado, con una posible alteración de la microcirculación sanguínea periférica por una disfunción del control neural simpático a nivel cutáneo y, por otro lado, con una mala regularización de los procesos de termogénesis que pueden influir en el metabolismo basal de las mujeres con síndrome de Fibromialgia. En segundo lugar, las mujeres con síndrome de Fibromialgia tenían niveles séricos alterados de óxido nítrico, de la actividad de la aminopeptidasa degradante de la encefalina y de la actividad de la oxitocinasa que se encontraron asociados con unos mayores niveles de hipersensibilidad al dolor por presión en algunos puntos sensibles, con la intensidad global del dolor y con un mayor impacto en la vida diaria en las mujeres con síndrome de Fibromialgia. En tercer lugar, los perfiles dietéticos pro-inflamatorios se asociaron con unos mayores niveles de hipersensibilidad al dolor por presión para la mayoría de los puntos sensibles establecidos por el Colegio Americano de Reumatología en mujeres con síndrome de Fibromialgia en comparación con las mujeres sanas. Por tanto, fomentar intervenciones dietéticas anti-inflamatorias caracterizadas por el consumo de verduras, frutas, ingesta moderada de proteínas bajas en grasas, consumo de pescado y aceite de oliva y una restricción de pan y cereales pueden mejorar los niveles globales de dolor y los síntomas asociados en poblaciones con Fibromialgia. Por lo tanto, tener en cuenta todos estos aspectos y resultados presentados en la tesis doctoral ayudaría a un mejor abordaje y manejo integral de los procesos dolorosos crónicos y de la sintomatología en personas diagnosticadas con síndrome de Fibromialgia.

ABSTRACT

Fibromyalgia syndrome is defined as a complex disease characterised by chronic widespread and diffuse musculoskeletal pain and other associated symptoms such as hyperalgesia, allodynia, hypersensitivity to pressure pain, continuous fatigue, morning joint stiffness, headaches, migraines, irritable bowel, sleep disorders, anxiety, depression, and concentration and memory problems. It is traditionally classified into two forms: primary fibromyalgia syndrome and secondary fibromyalgia syndrome. While aetiology in fibromyalgia syndrome still remains uncertain, alterations of the Central Nervous System have been considered as the main key factor in the development and maintenance of symptoms. However, new lines of research have highlighted the presence of blood microcirculation disorders due to changes in the peripheral nerve innervation of the glabrous skin microvasculature of these patients, and also due to changes in serum concentrations of immunological and inflammatory factors, as mechanisms underlying the pathophysiology of this chronic condition. This fact implies that there could be an altered peripheral vascular response to thermal stimuli that would be related to the production of certain biological biomarkers, such as nitric oxide or enkephalins, which intervene in the processes of peripheral sensitisation to pain, and in the maintenance of the inflammatory state. On the other hand, it has been found that the dietary profile of patients can influence the baseline inflammatory state and, therefore, contribute to the phenomena of pain hypersensitivity in populations with chronic pain; to our knowledge, however, related studies in the FM population are very limited. Finally, it should be noted that current therapeutic interventions for the management of this syndrome focus on conservative treatments (physical exercise or strength training, meditation techniques, cognitive-behavioural therapy, mindfulness, acupuncture, hydrotherapy, balneotherapy, massage and alternative therapies), and on pharmacological treatments (tramadol, duloxetine, milnacipran, amitriptyline, pregabalin and cyclobenzaprine), with not entirely conclusive and indeed contradictory results. That is why its aetiological ignorance has limited the therapeutic and diagnostic strategies and called for research studies, such as those developed in this doctoral thesis, which explore its pathophysiology from a multidisciplinary perspective.

With this background, the hypotheses of this doctoral thesis were: 1) Patients with Fibromyalgia syndrome could present abnormalities in the peripheral vascular response on the dorsal and palm of the hands characterised by a) vasodilation in the face of excessive

peptidergic sensory innervation of arteriovenous anastomoses, and by b) passive dilation of the arterioles due to the release of compounds such as nitric oxide from endothelial cells, which, in turn, would influence the global body temperature of patients with Fibromyalgia. 2) Based on the presence in the blood serum of biomarkers and neurotransmitters such as nitric oxide and enkephalins, and their role in pain modulation processes, we hypothesize that the presence of circulating nitric oxide, as well as the activity of enkephalinase in blood serum, could be associated with pain variables and symptoms of patients diagnosed with Fibromyalgia. 3) Since inflammatory cytokines may be involved in the underlying mechanism of Fibromyalgia syndrome, we hypothesised that a pro-inflammatory diet pattern would be associated with hypersensitivity to pain and other Fibromyalgia symptoms.

The main objectives were: 1) To assess the peripheral vascular component of the skin on the dorsal and palm of the hands and the global body temperature, as indicators of the adrenergic activity of the sympathetic nervous system and of thermogenesis processes in people diagnosed with Fibromyalgia syndrome. 2) To assess the presence in blood serum of vasodilator biomarkers (nitric oxide) and the activity of endogenous opioids (enkephalinases) as indicators of blood microcirculation and of the inflammatory and nociceptive baseline state, and its relationship with chronic pain and symptoms in people diagnosed with Fibromyalgia syndrome. 3) To assess the inflammatory potential of diet and its relationship with chronic pain and symptoms in patients with Fibromyalgia.

To achieve these objectives, two observational case-control studies and one observational case study were conducted. The first study included 42 women with Fibromyalgia syndrome and 52 healthy women. The second study included 58 women diagnosed with fibromyalgia. Finally, the third study included 95 women with Fibromyalgia syndrome and 98 healthy women. First, peripheral vascular blood flow and core body temperature were assessed by thermographic pattern of the dorsal and palm of the hands, and by measuring the tympanic and axillary temperature, respectively. Furthermore, nitric oxide concentration levels in blood serum were measured using the ozone chemiluminescence-based method. Secondly, the serum activities of enkephalin-degrading oxytocinase and aminopeptidase were analysed using the Bradford quantification method, along with nitric oxide levels and their association with the threshold and magnitude of electrical pain, the global level self-reported pain thresholds, pressure pain thresholds, central sensitisation, the impact of Fibromyalgia, and anxiety

levels. Thirdly, we analysed the inflammatory potential of diet through the Inflammatory Dietary Index and examined its relationship with changes in pressure pain thresholds, global pain intensity, impact of symptoms, fatigue, sleep problems, and common symptoms of anxiety.

Our results indicated that women with Fibromyalgia syndrome have a higher temperature on the dorsal and palm of the hands in all points assessed ($P \leq 0.001$) and a higher tympanic temperature ($P = 0.002$). Significant associations were observed between serum nitric oxide levels and the minimum temperature in the dorsal centre of the dominant hand ($\beta = -3.501$, $P = 0.038$), the maximum temperature ($\beta = -5.594$, $P = 0.016$), minimum ($\beta = -4.090$, $P = 0.036$) and mean ($\beta = -5.519$, $P = 0.015$) of the centre of the palm of the non-dominant hand, and the maximum temperature at the thenar eminence of the dominant hand ($\beta = -5.800$, $P = 0.017$), as well as with tympanic temperature ($\beta = -9.321$, $P = 0.035$) in healthy women. Significant associations were observed between circulating nitric oxide levels and pressure pain thresholds in the dominant occiput ($\beta = 0.290$, $P = 0.003$) and in the non-dominant occiput ($\beta = 0.193$, $P = 0.034$), as well as in the level of activity in Fibromyalgia ($\beta = 0.031$, $P = 0.027$) in the population of women diagnosed with Fibromyalgia. Associations of oxytocinase activity with the global level of self-reported pain ($\beta = 0.215$, $P = 0.023$) and the pressure pain threshold of the dominant knee point ($\beta = 2.794$, $P = 0.039$) were also observed. Finally, high levels of serum activity of enkephalin-degrading aminopeptidase were correlated with a hypersensitivity of pressure pain at the level of the second dominant rib point ($\beta = -20.096$, $P = 0.049$). Lastly, linear regression analysis confirmed that pressure pain thresholds in occiput ($\beta = 0.234$, $P = 0.036$), trapezius ($\beta = 0.299$, $P = 0.007$), zygapophyseal joint ($\beta = 0.291$, $P = 0.035$), second rib ($\beta = 0.204$, $P = 0.006$), gluteal ($\beta = 0.591$, $P = 0.017$), greater trochanter ($\beta = 0.379$, $P = 0.041$) and knee ($\beta = 0.482$, $P = 0.011$) were associated with a higher score on the Inflammatory Dietary Index in women with Fibromyalgia syndrome compared to healthy women. However, no significant associations were found between the Inflammatory Dietary Index score and other clinical symptoms.

The main conclusions were, firstly, that people with Fibromyalgia syndrome had a higher temperature in all points analysed on the dorsal and palm of their hands, and a higher tympanic temperature than healthy women. These results could be related, on the one hand, to a possible alteration of peripheral blood microcirculation due to a dysfunction of sympathetic neural control at the cutaneous level and, on the other hand, to a poor

regularisation of thermogenesis processes that can influence metabolism baseline of women with Fibromyalgia syndrome. Secondly, women with fibromyalgia syndrome had altered serum levels of nitric oxide, of the enkephalin-degrading aminopeptidase activity, and oxytocinase activity that were found to be associated with higher levels of hypersensitivity to pressure pain in some tender points, with the global intensity of pain, and with a greater impact on daily life in women with Fibromyalgia syndrome. Thirdly, pro-inflammatory dietary profiles were associated with higher levels of pressure pain hypersensitivity for most tender points established by the American College of Rheumatology in women with Fibromyalgia syndrome compared to healthy women. Therefore, promoting anti-inflammatory dietary interventions characterised by the consumption of vegetables, fruits, a moderate intake of low-fat protein, the consumption of fish and olive oil, and a restriction of bread and cereals can improve overall pain levels and associated symptoms in populations with Fibromyalgia. Accordingly, bearing in mind all aspects and results presented in the doctoral thesis would help in crafting a better approach and comprehensive management of chronic painful processes and symptoms in people diagnosed with Fibromyalgia syndrome.

INTRODUCCIÓN

INTRODUCTION

1. INTRODUCCIÓN

1.1 Síndrome de Fibromialgia: concepto, prevalencia, etiopatogenia, clasificación, manifestaciones clínicas y coste socio-económico.

1.1.1 Concepto.

La Fibromialgia (FM) es un síndrome complejo, de etiología heterogénea y desconocida, caracterizado por dolor crónico músculo-esquelético generalizado y difuso, y acompañado por una amplia gama de síntomas somáticos y psicológicos tales como hiperalgesia, alodinia, fatiga persistente, rigidez articular, dolor de cabeza, migraña, problemas digestivos, intolerancia al frío, sueño no reparador, ansiedad, depresión, problemas de memoria y dificultades de concentración (Bellato et al., 2012; Giacomelli et al., 2013; Häuser et al., 2015).

Para entender cómo ha ido evolucionando el concepto y descripción de la FM a lo largo de sus más de 110 años de historia, es necesario conocer cronológicamente los hallazgos y descubrimientos más relevantes en relación al dolor y las alteraciones músculo-esqueléticas, los cuales han sido registrados en la literatura científica europea desde finales del siglo XVI (Wolfe & Walitt, 2013). En el año 1592, el médico francés Guillaume de Baillou fue el primero en describir el término reumatismo para referirse al conjunto de manifestaciones clínicas de fiebre reumática aguda y dolor muscular. Asimismo, y a diferencia del reumatismo articular, introdujo el concepto de reumatismo muscular, el cual englobaba al conjunto de trastornos músculo-esqueléticos dolorosos sin deformidad en los tejidos blandos (Inanici & Yunus, 2004). A principios del siglo XIX, el cirujano escocés William Balfour descubrió por primera vez la presencia de nódulos e informó de la sensibilidad local en el tejido por puntos anatómicos sensibles, indicando que los nódulos y los dolores musculares están causados por la inflamación que se origina en el tejido conectivo muscular (Inanici & Yunus, 2004). Años más tarde, en 1841, el pediatra francés François Louis Valleix acuñó el concepto de punto gatillo al descubrir que la palpación manual sobre varios puntos dolorosos en varias partes del cuerpo originaba un dolor referido a otras regiones del cuerpo humano y que, además, se encontraban relacionados con el recorrido y/o trayecto de diferentes nervios. Todo ello llevó a considerar al reumatismo muscular como una forma de neuralgia (Inanici & Yunus, 2004). A finales del siglo XIX, el neurólogo estadounidense Goerge Miller Beard definió el término de neurastenia como todo dolor generalizado acompañado de fatiga física y trastornos

psicológicos y mentales, atribuyendo la aparición de este conjunto de síntomas al estrés de vida diario al que nos encontramos sometidas las personas en la vida moderna (Inanici & Yunus, 2004; Wolfe & Walitt, 2013). Sin embargo, este concepto de neurastenia fue abandonado en la década de 1930 debido a su reconocimiento como un trastorno psicológico y psiquiátrico, dejando de ser una rama de estudio en el campo de la neurología (Wolfe & Walitt, 2013).

En el año 1900 emergió en la literatura científica el término fibrositis para describir toda tipología de dolor local o regional, englobando, por consiguiente, la enfermedad de FM conocida hasta el momento (Wolfe & Walitt, 2013). En 1904, el neurólogo británico Sir William Gowers mencionó por primera vez este concepto en un artículo de investigación relacionado con el dolor lumbar afirmando que tanto el lumbago como el reumatismo muscular deberían considerarse como una forma de inflamación de las fibras de los tejidos musculares y que, continuando con la analogía de la "celulitis", esta inflamación de los tejidos fibrosos de los músculos debería llamarse "fibrositis". Además, en dicho artículo de publicación, Gowers indicó una serie de síntomas relacionados con la fibrositis tales como dolor espontáneo, sensibilidad a la compresión mecánica, fatiga y trastornos del sueño, así como un agravamiento de los mismos por factores como la exposición al frío o el sobreesfuerzo muscular (Inanici & Yunus, 2004). Por tanto, se habían fijado las bases generales del término fibrositis, sin embargo, no fue hasta mediados del siglo XX cuando se inició un creciente interés por el estudio científico de la misma (Wolfe & Walitt, 2013). En este sentido, en 1951 Richard Harold Freyberg, principal contribuyente y líder en el desarrollo de la reumatología como disciplina, llevó a cabo una revisión profunda del concepto fibrositis. En su contribución estableció una división de la misma en fibrositis regional (actualmente conocida como síndrome de dolor miofascial) y fibrositis generalizada (actualmente conocida como FM), alertando también de la difícil diferenciación con el término reumatismo psicógeno caracterizado por una afectación muscular en ausencia de inflamación pero asociada con problemas de depresión y/o estrés (Freyberg, 1951). Además, y en línea con las publicaciones previamente descritas, Richard Freyberg informó que, por un lado, factores como la humedad, el frío y la lluvia podrían agravar los síntomas de la fibrositis y que, por otro lado, factores como el calor o una alta presión barométrica podrían aliviar los mismos (Freyberg, 1951; Wolfe & Walitt, 2013). Posteriormente, en 1953, Wallace Graham reavivó el interés surgido sobre la fibrositis y otras afecciones comunes como la enfermedad de FM gracias a la elaboración de un

capítulo en el conocido libro *"Artritis y afecciones afines"* (Graham, 1949). En dicha contribución, señaló la existencia de afecciones dolorosas agudas, subagudas y crónicas acompañadas de puntos sensibles locales y de dolor referido, independientemente de la enfermedad que las pudieran originar, y que involucraban a músculos, tejidos subcutáneos, tejidos ligamentosos, tejidos tendinosos y estructuras fasciales. Las causas de estas afecciones dolorosas eran infecciosas, traumáticas, ambientales y psicológicas, y el dolor referido presentaba características irritantes descritas como quemazón, punzante, insistente e insoportable (Graham, 1949). A finales del año 1968, el investigador Eugene Traut de la Universidad de Illinois (Chicago, Estados Unidos) incorporó a las características clínicas ya descritas de la fibrositis una amplia constelación de nuevos síntomas tales como dolor de cabeza, colitis, miedo, preocupación y ansiedad. Además, en relación al dolor generalizado, afirmó que el reumatismo muscular tiene su origen en varios niveles del eje espinal, considerando el dolor axial como un criterio importante en el diagnostico actual de la FM (Traut, 1968; Wolfe et al., 1990). La importante contribución científica de Traut puso de manifiesto una serie de características sistémicas importantes del síndrome de FM (fatiga, sueño, dolor de cabeza, colitis, etc.), ofreciendo, por tanto, la primera descripción casi moderna de la FM (Inanici & Yunus, 2004; Traut, 1968).

Fue en la década de 1970 cuando se proporcionó la primera descripción moderna del síndrome de FM gracias al médico y científico Hugh Smythe (Inanici & Yunus, 2004). En un elaborado capítulo de 10 páginas sobre el síndrome de fibrositis en el libro *"Artritis y afecciones afines"* describió la FM como un síndrome caracterizado por dolor generalizado acompañado de fatiga, rigidez matutina, falta de sueño, un alto recuento de puntos sensibles a la palpación anatómica (11/14; 79%) y estrés emocional (Inanici & Yunus, 2004; Smythe, 1972). Asimismo, enfatizó el importante papel de las alteraciones del sueño en pacientes con FM a través de los hallazgos con electroencefalograma reportados por su compañero y científico Harvey Moldofsky (Moldofsky et al., 1975), especificando, a su vez, un marco fisiopatológico para el síndrome de FM al describir un estatus de hiperalgesia refleja profunda junto con patrones de dolor referido, diferenciándolos de la hiperalgesia cutánea (Smythe, 1972). Estos hallazgos fueron considerados un triunfo en la medicina clínica de cabecera, conociéndose ambos autores coloquialmente como los "abuelos" de la FM (Inanici & Yunus, 2004). No obstante, y ante la falta de estudios controlados en el campo de la FM, el concepto y descripción de esta enfermedad permaneció en duda. Así pues, Yunus et al. (1981) llevaron a cabo el primer

ensayo controlado sobre las manifestaciones clínicas en FM en 50 pacientes con FM en comparación con 50 sujetos sanos emparejados por edad y sexo. El ensayo confirmó las características previas ya registradas en estudios precedentes en población con FM tales como dolor generalizado, mayor número de puntos sensibles, fatiga y problemas de sueño en comparación con los controles sanos. Además, se describieron por primera vez un conjunto de nuevos síntomas en dichos pacientes entre los que destacan dolores de cabeza de tipo tensional, migrañas, parestesias, e hinchazón subjetiva, así como la asociación con otros síndromes funcionales tales como síndrome del intestino irritable o síndrome de piernas inquietas (M. Yunus et al., 1981). Por tanto, el registro documentado de estos múltiples síntomas elevó la concepción de la FM a nivel de síndrome, el cual es aceptado actualmente por la literatura científica (Inanici & Yunus, 2004).

Finalmente, en 1990 y bajo los criterios recomendados por el Colegio Americano de Reumatología (ACR en inglés) se abandonó definitivamente la asociación existente de la FM con el síndrome de fibrositis para proporcionar una definición oficial del síndrome de FM (Wolfe & Walitt, 2013). Dicho ACR publicó los primeros criterios de clasificación y diagnóstico de la FM consistentes en: a) dolor generalizado en el lado derecho e izquierdo del cuerpo humano superior persistente en el tiempo por más de 3 meses; y b) presencia de dolor a la palpación digital de 11 o más puntos sensibles en 18 puntos específicos (Wolfe et al., 1990). Posteriormente, estos criterios se han ido actualizando a lo largo de los años (2010, 2011, 2016, 2019), mejorando, por consiguiente, el conocimiento y descripción del síndrome de FM (Arnold et al., 2019; Wolfe et al., 2010, 2016).

1.1.2 Prevalencia.

Numerosos estudios en la literatura científica han intentado establecer la prevalencia del síndrome de FM en diferentes poblaciones a nivel mundial. La prevalencia estimada en población general difiere entre los estudios, reflejando, por consiguiente, la dificultad existente a la hora de fijar con exactitud la prevalencia real de dicho síndrome (Branco et al., 2010; Cabo-Meseguer et al., 2017; Heidari et al., 2017; Queiroz, 2013). Esta variación de las cifras de prevalencia puede explicarse por la heterogeneidad observada en la literatura científica de los diferentes estudios epidemiológicos en relación al diseño, los criterios de reclutamiento, las cohortes de pacientes y al tamaño muestral de los mismos (Fitzcharles et al., 2018).

En líneas generales, la literatura científica indica que entre un 0.4% y un 9.3% de la población general mundial padece FM, fijándose la tasa de prevalencia media en un porcentaje aproximado del 2.7% (Queiroz, 2013). En una reciente revisión sistemática y meta-análisis sobre el estudio de prevalencia de la FM en población general y en función de las diferentes regiones establecidas por la Organización Mundial de la Salud (OMS), la prevalencia de la enfermedad se sitúa en un 2.64% en la región de Europa, en un 2.41% en la región de América, en un 4.43% en la región del Mediterráneo Oriental y en un 1.62% en la región del Pacífico Occidental (Heidari et al., 2017). Estudios epidemiológicos sobre la prevalencia de la FM en el continente Europeo muestran variaciones de la misma entre diferentes países europeos como, por ejemplo, la prevalencia observada en Francia fijada en un 2.2%, en Portugal en un 3.7%, en Alemania en un 5.8% o en Italia en un 6.6% (Cabo-Meseguer et al., 2017). En la población española la tasa de prevalencia media se estima en un 2.4%, con una mayor afectación en mujeres (4.2%) que en hombres (0.2%) en una proporción de 21 a 1 (Cabo-Meseguer et al., 2017). Asimismo, y en línea con esta revisión anterior, un estudio reciente elaborado por Gayà et al. (2020) sitúa la tasa de prevalencia de FM en la población general española en un 2.45%, con un mayor porcentaje de afectación en mujeres (4.49%) que en hombres (0.29%) (Gayà et al., 2020). Además, las consultas por FM en el territorio español a nivel de servicios sanitarios de atención no especializada refleja un porcentaje de entre el 2.1% y el 5.7% de las visitas totales en comparación con las consultas registradas a nivel de atención especializada en reumatología que suponen entre el 10% y el 20% de la totalidad (Cabo-Meseguer et al., 2017).

Un factor determinante a tener presente en el desarrollo del síndrome de FM es la edad. El rango de edad comprendido entre los 30 y los 50 años de edad es el rango de afectación más frecuente en el síndrome de FM aunque puede aparecer también después de los 50 años de edad (Queiroz, 2013). En el continente Europeo la prevalencia de FM es baja en los adultos jóvenes, aumentado, por el contrario, en el rango de edad comprendido entre los 35 y los 44 años de edad y hasta los 74 y 85 años edad (Branco et al., 2010). En la población española la FM suele manifestarse entre los 40 y 49 años de edad, alcanzando su prevalencia máxima entorno a los 60 y 69 años de edad (Cabo-Meseguer et al., 2017; Gayà et al., 2020).

Otras de las cuestiones más relevantes a tener en cuenta en la FM son las diferencias ligadas al género. La FM se presenta en mayor proporción en las mujeres, cuantificándose su prevalencia general media a nivel mundial en un 4.2% frente al género masculino cuya prevalencia se estima en un 1.4%. Por tanto, a nivel mundial, las mujeres son más susceptibles de padecer FM que los hombres en una proporción 3 a 1 (Queiroz, 2013). La prevalencia que presentan las mujeres en manifestar el síndrome de FM se sitúa en un 5.2% a nivel Europeo con una proporción general de mujeres a hombres inferior a 2 (Branco et al., 2010). Estudios realizados en Estados Unidos indican una prevalencia de FM mucho más alta en mujeres (3.28%) que en hombres (1.06%) (Walitt et al., 2015). Por otra parte, la proporción de FM entre mujeres y hombres difiere entre continentes. Sudamérica es el continente que presenta una mayor proporción de afectación en FM en mujeres en comparación con los hombres en una proporción de 12 a 1, seguido del continente Asiático con una proporción de 5 a 1, del continente Americano con una proporción de 4 a 1 y del continente Europeo con una proporción de 3 a 1 (Cabo-Meseguer et al., 2017). En España, la prevalencia de FM para el sexo femenino es del 15.8% y para el sexo masculino del 2.2% (Cabo-Meseguer et al., 2017). Además, el sexo femenino en población española se encuentra asociado de una manera muy significativa con el desarrollo del síndrome de FM con una Odd Ratio (OR) de 10.156 y un Intervalo de Confianza (IC) al 95% de 5.066 a 20.352 (Gayà et al., 2020).

Dentro de los principales factores de riesgo en la FM destacan una serie de comorbilidades como la obesidad, diabetes mellitus, el síndrome del intestino irritable, síndrome de la vejiga dolorosa, cistitis intersticial, enfermedades celiacas, alteraciones de la articulación temporo-mandibular, dolores de cabeza y migraña (Fitzcharles et al., 2018; Heidari et al., 2017). En este sentido, la obesidad se ha identificado como un factor de riesgo importante para el desarrollo de la FM con una tasa de prevalencia del 45% (Dias et al., 2017), observándose una fuerte asociación con dicho síndrome en el territorio español con una OR de 1.689 y un IC al 95% de 1.036 a 2.755 (Gayà et al., 2020). En relación a la diabetes mellitus, la prevalencia total del síndrome de FM se estima alrededor del 14.80% con un IC al 95% de 11.10 a 18.40 (Heidari et al., 2017). Con respecto al síndrome del intestino irritable, la tasa de prevalencia global de FM en estos pacientes se ha estimado en un 12.90% con un IC al 95% de 12.70 a 13.10 (Heidari et al., 2017). Los dolores de cabeza, migraña y cefalea tensional también han mostrado unas altas tasas de prevalencia en pacientes con FM con un porcentaje del 36.4%, 28.5% y 59%, respectivamente (M. De

Tommaso et al., 2009; Marina De Tommaso et al., 2011). Por último, en otros tipos de condiciones de dolor crónico como alteraciones en la articulación temporo-mandibular, dolor lumbar crónico, cistitis intersticial, síndrome de la vejiga dolorosa, endometriosis y encefalomielitis se han reportado elevadas tasas de prevalencia de FM que oscilan entre el 20% y el 65% (Fitzcharles et al., 2018).

En referencia a factores socioeconómicos, socioculturales y educativos destaca un amplio consenso en la comunidad científica en el desarrollo del síndrome de FM en personas con bajos niveles económicos, bajos niveles socioculturales y bajos niveles educativos (Branco et al., 2010; Cabo-Meseguer et al., 2017; Queiroz, 2013).

Por último, cabe indicar que parece existir discrepancias en la literatura científica con respecto a las tasas de prevalencia de FM y su asociación con el estado civil de la persona o la residencia de la misma en núcleos urbanos o zonas rurales. Mientras unos estudios afirman que el divorcio o residir en zonas rurales se asocian significativamente con el desarrollo de la FM (Branco et al., 2010; Cabo-Meseguer et al., 2017), otros estudios muestran altas tasas de prevalencia de FM independientemente del estatus civil de la persona (casados/casadas, viudos/viudas, divorciados/divorciadas) y si se reside en los núcleos urbanos de las grandes ciudades (Gayà et al., 2020; Queiroz, 2013).

1.1.3 Etiopatogenia.

A pesar de que han transcurrido más de 110 años desde que William Gowers describiera las primeras características del síndrome de FM a través del concepto “fibrositis”, la fisiopatología y los mecanismos subyacentes de la misma siguen siendo desconocidos (Chinn et al., 2016; Coskun Benlidayi, 2019; Häuser et al., 2015). Las alteraciones del Sistema Nervioso Central (SNC) se han considerado como el principal factor clave en el desarrollo del síndrome de FM (Chinn et al., 2016; Russell & Larson, 2009). Sin embargo, la literatura científica actual propone una serie de factores que juegan un papel importante en la fisiopatología de esta condición crónica tales como alteraciones vasculares, disfunciones neurógenas, factores inflamatorios, factores infecciosos, factores nutricionales y factores genéticos, entre otros (Coskun Benlidayi, 2019; Häuser et al., 2015; Peck et al., 2020; Russell & Larson, 2009).

A continuación se describen los principales hallazgos fisiopatológicos, según el componente estructural o funcional afectado:

1. **Sensibilización central del dolor.** La sensibilización central (SC) se define como un fenómeno fisiológico producido por cambios en el procesamiento a nivel del SNC que provocan alteraciones en la función neuronal, originando, por consiguiente, un estado de hiperexcitabilidad (hipersensibilidad) ante estímulos nocivos y no nocivos (Neblett et al., 2017; Nijs et al., 2017). A nivel clínico, la SC se manifiesta por la presencia de alodinia (sensación dolorosa frente a un estímulo no doloroso, como por ejemplo, el tacto), hiperalgesia (sensibilidad excesiva frente a un estímulo que en condiciones normales es doloroso, como por ejemplo, la presión), expansión del campo del dolor (dolor que se expande más allá del área de los nervios periféricos) y dolor prolongado en el tiempo después de que haya cesado un estímulo doloroso determinado (principalmente palpitante, ardor, hormigueo o entumecimiento) (Nijs et al., 2010). El elemento clave de la teoría que explica el fenómeno de SC reside en que los impulsos de dolor mantenido van a originar una alteración a nivel de las vías ascendentes y descendentes del SNC, por lo que dolores persistentes pueden provocar cambios a nivel del SNC y Sistema Nervioso Periférico (SNP) (Kindler et al., 2011; Nijs et al., 2010).

   Investigaciones científicas han informado de una descompensación en las dos principales vías de la nocicepción del SNC en población con FM (Chinn et al., 2016; Russell & Larson, 2009). Ante el estado de dolor prolongado tan característico en pacientes con FM, los receptores nociceptivos del tejido periférico se encuentran sobreestimulados enviando constantemente señales dolorosas a las fibras nerviosas A delta y fibras nerviosas C, que son las encargadas de transmitir los impulsos nociceptivos hasta las neuronas del asta dorsal de la médula espinal. La activación continuada de estas fibras nerviosas nociceptivas produce la liberación de neurotransmisores y neuropéptidos como la substancia P, el glutamato, el péptido relacionado con el gen de la calcitonina (CGRP), el factor de crecimiento nervioso y el aspartato, los cuales modulan las descargas eléctricas post-sinápticas en el asta dorsal de la médula espinal. Los neurotransmisores y neuropéptidos incrementados proceden a llevar a cabo las correspondientes respuestas post-sinápticas causando una hiperestimulación de los receptores de N-metil-D-aspartato (NMDA) de las neuronas de segundo orden del asta dorsal de la médula espinal. La activación de los

receptores NMDA alteran notablemente la función normal que deben ejercer las neuronas post-sinápticas, originando cambios en las membranas celulares, permitiendo la entrada de calcio y activando la proteína quinasa. Estas sustancias neuroquímicas sensibilizan a las neuronas de amplio rango dinámico ("Wide-Dinamic Range"), las cuales se vuelven hiperexcitables y originan los fenómenos descritos previamente como alodinia e hiperalgesia (Nijs & Van Houdenhove, 2009; M. B. Yunus, 2007). Como consecuencia de este proceso, se produce la activación de las neuronas de segundo orden que estimulan una serie de áreas y estructuras de la corteza cerebral (tálamo, hipotálamo, corteza pre-frontal, corteza cingulada anterior, corteza insular y corteza somatosensorial) que juegan un papel transcendental en el procesamiento del dolor en todas sus dimensiones posibles (sensorial, afectiva y valorativa) (Nijs & Van Houdenhove, 2009; M. B. Yunus, 2007). Este mecanismo que ha sido observado en población con FM se define como "pro-nocicepción" (Bellato et al., 2012; Chinn et al., 2016; Russell & Larson, 2009).

Por otra parte, el fenómeno de SC en pacientes con FM no implica únicamente a las fibras nerviosas nociceptivas encargadas de transmitir la información dolorosa al asta dorsal de la médula espinal, sino que también involucra a los mecanismos inhibitorios descendentes del dolor, los cuales se encuentran alterados. La limitación de la actividad de las vías descendentes del dolor (sistema reticular, tronco del encéfalo e hipotálamo) deriva en una liberación insuficiente de neurotransmisores como serotonina, norepinefrina, encefalinas y ácido g-aminobutírico (GABA), que provocan una alteración en la modulación de la respuesta dolorosa a la periferia. Este mecanismo que ha sido observado en población con FM se define como "anti-nocicepción" (Bellato et al., 2012; Chinn et al., 2016; Nijs et al., 2011; Russell & Larson, 2009; M. B. Yunus, 2007).

Finalmente, estudios científicos han informado tanto en diferentes poblaciones con dolor crónico como en poblaciones con FM que el fenómeno de SC característico en pacientes con esta condición ve incrementada su capacidad de respuesta ante una amplia gama de estímulos como el frío, el calor, la luz, el sonido, los estímulos eléctricos, las sustancias químicas, la ansiedad, el pánico, la depresión y las emociones (Häuser et al., 2015; Nijs et al., 2011; Sluka & Clauw, 2016). Por tanto, la presencia de estos factores y su interconexión con los procesos fisiológicos descritos previamente indican la necesidad de tener presente en todo momento el modelo biopsicosocial del dolor (Nijs & Van Houdenhove, 2009).

2. **Factores vasculares.** Otros de los hallazgos informados en pacientes con FM son la presencia de anomalías morfológicas en el diámetro y densidad de los pequeños vasos sanguíneos (capilares) y la existencia de alteraciones de la microcirculación sanguínea. Pacientes con síndrome de FM presentan síntomas vasoespásticos similares a los acontecidos en el Fenómeno de Raynaud, resultando en un aumento del tono simpático de los vasos sanguíneos y en una estimulación del sistema vasoconstrictor (Morf et al., 2005). La activación continuada de los sistemas vasoconstrictores alfa-2-adrenérgico, endotelina-1, tirosinacinasa, serotonina y angiotensina II produce una disfunción del tejido endotelial acompañada de una disminución de la densidad capilar y del flujo sanguíneo microcirculatorio, reduciendo, por consiguiente, la actividad de los vasos sanguíneos musculares primarios y el aporte nutricional y de oxígeno hacia los tejidos (Choi & Kim, 2015; Morf et al., 2005).

3. **Factores neurógenos y neurovasculares.** Otros de los mecanismos subyacentes en el síndrome de FM son las alteraciones a nivel neurógeno. El Sistema Nervioso Autónomo (SNA) desempeña una función muy importante en el control de la temperatura central del cuerpo humano. En ese sentido, estudios científicos muestran disfunciones de los sistemas nerviosos simpático y parasimpático del SNA, de las fibras nerviosas sensitivas aferentes en la zona de conexión neurovascular y de las anastomosis arteriovenosas (AAV) de los pacientes con FM (Albrecht et al., 2013). En concreto, esta afectación de las AAV podría generar una alteración de la inervación sensorial de los vasos sanguíneos de pequeño calibre del tejido cutáneo, causando cambios en el flujo sanguíneo e isquemia en el tejido músculo-esquelético (Albrecht et al., 2013).

4. **Factores inmunológicos e inflamatorios.** Otra hipótesis que subyace a la patología de FM es la función del sistema inmune y su interacción con los procesos inflamatorios (Peck et al., 2020; Sluka & Clauw, 2016). Las células del sistema inmune de sujetos con FM son altamente plásticas lo que podría alterar los niveles de concentración de citocinas pro-inflamatorias y anti-inflamatorias a nivel sistémico o a nivel local de los tejidos. La liberación de citocinas pro-inflamatorias al torrente sanguíneo como, por ejemplo, interleucina IL-1β, interleucina IL-6, interleucina IL-8 o el factor de necrosis tumoral alfa afectarían a las redes neuronales durante la

interrelación del sistema nervioso con las células inmunitarias, lo que estimularía a los nociceptores y conduciría a un aumento de la SC, de la sensibilización periférica y a una neuroinflamación (Peck et al., 2020; Sluka & Clauw, 2016)

5. **Factores genéticos.** Finalmente, otra posible causa asociada al desarrollo de la FM son las diferentes variantes genéticas (Häuser et al., 2015; Peck et al., 2020). La literatura científica ha documentado de manera notable con múltiples estudios que la herencia de genes relacionados con el dolor y la predisposición y agregación familiar contribuyen en hasta un 50% en el desarrollo de afecciones asociadas al dolor crónico como es el caso del síndrome de FM (Peck et al., 2020). En este sentido, se ha observado que familiares de primer grado de pacientes con FM presentan un riesgo 8.5 veces mayor de desarrollar esta enfermedad con un IC al 95% de 2.8 a 26 (Clauw, 2014). Asimismo, existen numerosos genes y polimorfismos de genes asociados con la hipersensibilidad al dolor (por ejemplo, genes GRM6, GABRB3, SNPs, entre otros) debido a los cambios producidos en la expresión y función de múltiples proteínas responsables de la modulación del dolor (Peck et al., 2020). Además, hay genes asociados con el desarrollo de la fatiga (polimorfismo del gen IL-6 rs1800795), la rigidez articular (gen MTHFR), con el sueño (polimorfismo del gen DAT1), la ansiedad (polimorfismo del gen T102C) o con trastornos cognitivos (gen MYT1L) (D'Agnelli et al., 2019; Peck et al., 2020).

En resumen, la FM es un síndrome de etiología compleja e incierta, aunque la participación del SNC constituye un elemento clave en las manifestaciones clínicas de estos pacientes. No obstante, la influencia de diversos factores vasculares, neurógenos, inmunológicos, inflamatorios y genéticos generan, en la actualidad, un gran interés en el campo de la investigación en FM para intentar dilucidar cuál de ellos constituye la causa real de esta afectación (Häuser et al., 2015).

1.1.4 Clasificación.

El dolor generalizado por todas partes del cuerpo humano es una condición muy presente en personas diagnosticadas con FM, considerándose también como uno de los criterios más importantes para la clasificación de la misma, tanto para pacientes con síndrome de FM primario como con síndrome de FM secundario (Wolfe et al., 1990). El dolor crónico se define como un dolor que se mantiene o persiste en el tiempo durante más

de 3 meses, por lo que pierde la función fisiológica de alerta de la nocicepción aguda (Treede et al., 2019). El dolor crónico genera en las personas que lo padecen angustia, sufrimiento y discapacidad, derivando en un alto impacto en la calidad y expectativa de vida (Gary J. Macfarlane et al., 2017). Además, es uno de los problemas más frecuentes de salud pública a nivel mundial, demandando una alta presencia en la atención médica (Goldberg & McGee, 2011; Gary J. Macfarlane et al., 2017).

El grupo de trabajo de la Asociación Internacional para el Estudio del Dolor (IASP en inglés), y en colaboración con la OMS, elaboró en 2019 un nuevo sistema de clasificación para el dolor crónico que incluyeron nuevos conceptos taxonómicos relacionados con las principales afecciones que cursan con dicha condición y que además han sido incorporados a la Clasificación Internacional de las Enfermedades (CIE-11) (Treede et al., 2019). Dentro de la CIE-11 el dolor crónico tiene asignado un código específico (código G89.4), ya que ha sido valorado como una entidad patológica en sí misma (Treede et al., 2019). El grupo de trabajo de la IASP, por un lado, definió el dolor crónico músculo-esquelético primario como un dolor recurrente en músculos, tendones, huesos y articulaciones caracterizado por angustia emocional y discapacidad funcional y no atribuible directamente a una enfermedad previa. Por otro lado, definió el dolor crónico músculo-esquelético secundario como todo dolor persistente a nivel del sistema músculo-esquelético derivado de una enfermedad subyacente como, por ejemplo, dolor crónico relacionado con los procesos cancerígenos, dolor crónico post-traumático, dolor crónico post-quirúrgico, dolor crónico neuropático o dolor crónico visceral (Perrot et al., 2019; Treede et al., 2019). En relación con esta división establecida, la FM es considerada como un dolor crónico músculo-esquelético secundario debido a la presencia de numerosas patologías neuropáticas, reumáticas y trastornos autoinmunes, excluyendo, por tanto, la vertiente de considerar el dolor crónico generalizado como el único factor definitorio de este síndrome (Häuser & Fitzcharles, 2018). Asimismo, el código de diagnóstico del síndrome de FM dentro de la clasificación del dolor crónico de la CIE-11 es el M79.7 (Perrot et al., 2019).

En resumen, el síndrome de FM se clasifica en dos formas: síndrome de FM primario y síndrome de FM secundario (Häuser et al., 2015):

1. El **síndrome de FM primario** se define como el conjunto de personas que en ausencia de una entrada nociceptiva continua identificable experimentan un proceso doloroso como daño o inflamación. A su vez, estos pacientes pueden desarrollar afecciones dolorosas regionales como dolores de cabeza, trastornos temporomandibulares, dismenorrea y cistitis intersticial, así como síntomas psicológicos incluyendo ansiedad y depresión (Clauw, 2014).

2. El **síndrome de FM secundario** se define como el conjunto de personas que en presencia de una entrada nociceptiva continua identificable experimentan un proceso doloroso centralizado. Es decir, estos pacientes presentan dolor concomitante a una enfermedad secundaria tales como patología reumática (osteoartritis, artritis reumatoide, lupus), trastornos autoinmunes y anemia de las células falciformes (Clauw, 2014; Häuser et al., 2015).

1.1.5 Manifestaciones clínicas.

En relación a las manifestaciones clínicas del síndrome de la FM, el síntoma más característico es un dolor crónico generalizado y difuso por todo el cuerpo focalizado en la región axial, región corporal superior derecha, región corporal superior izquierda, región corporal inferior derecha y región corporal inferior izquierda, aunque, en ocasiones, el dolor puede iniciarse en sitios específicos como la zona lumbar o la zona cervical (Bair & Krebs, 2020; Wolfe et al., 1990). Generalmente el dolor crónico músculo-esquelético generalizado que experimentan las personas con síndrome de FM suele estar acompañado de síntomas neuropáticos como disestesias, parestesias, hormigueo y entumecimiento en las extremidades superiores e inferiores y sensación de quemazón o ardor en una o más articulaciones (Clauw, 2014). Además, y muy frecuentemente, aparecen síntomas relacionados con estados de centralización del dolor tales como la alodinia (dolor ante un estímulo que, normalmente, no es doloroso) o la hiperalgesia (excesiva sensibilidad ante un estímulo normalmente doloroso) y síntomas comórbidos de origen también en el SNC como la fatiga, la cual a menudo es informada como severa, y los trastornos del sueño (Clauw, 2014).

Otro conjunto de síntomas que informan con frecuencia los pacientes con FM, y que se incluyen en los marcos de diagnóstico actuales de esta enfermedad, son los síntomas somáticos. Dentro de ellos destacan los dolores de cabeza, dolores abdominales,

distensiones abdominales, dolores en la articulación temporo-mandibular, náuseas, mareos y diarreas (Bair & Krebs, 2020; Wolfe et al., 2016).

Otros síntomas que clínicamente también acompañan al síndrome de FM son los síntomas cognitivos y psicológicos. Entre esta variedad de síntomas, los pacientes con FM normalmente informan de afectación de la memoria, dificultad en la atención, dificultad para centrarse en un objetivo concreto o concentrarse en realizar una tarea o actividad de la vida diaria (informalmente conocido como «fibro niebla»), estados de ánimo alterados, ansiedad y depresión (Bair & Krebs, 2020; Clauw, 2014).

Finalmente, en la práctica clínica, pueden aparecer síntomas asociados con una hiperreactividad sensorial tales como sensibilidad a la luces brillantes, sensibilidad a ruidos o sonidos elevados y sensibilidad a los olores fuertes (Bair & Krebs, 2020).

1.1.6 Coste Socioeconómico.

El conjunto de manifestaciones clínicas previamente descritas en el síndrome de FM junto con todos los trastornos asociados a la misma generan de manera considerable en las personas que la padecen un sufrimiento personal y una pérdida funcional en el desempeño de las actividades de la vida diaria y de la actividad laboral, así como importantes costes socioeconómicos y sanitarios directos e indirectos (Schaefer et al., 2016).

En referencia a los costes que generan patologías con dolor crónico como es el caso del síndrome de FM, estudios realizados en Estados Unidos informan que el gasto medio anual por paciente diagnosticado con síndrome de FM se sitúa en un rango comprendido entre los 9.575 dólares y los 18.671 dólares, casi 5 veces más (aproximadamente 3.291 dólares) que el conjunto de personas que presentan otro tipo de síndrome doloroso (Berger et al., 2007; Knight et al., 2013). Asimismo, se ha observado que el gasto socio-sanitario se ve incrementado en aquellos pacientes en el que los síntomas de la FM son más graves. En este sentido, Chandran et al. (2012) llevaron a cabo en Estados Unidos un estudio en función de la gravedad de los síntomas en los pacientes diagnosticados con FM e informaron que el costo sanitario en pacientes con síntomas leves fue de 10.219 dólares, en pacientes con síntomas moderados fue de 26.217 dólares y en pacientes con síntomas graves fue de 42.456 dólares (Chandran et al., 2012).

La literatura científica también registra diversos estudios socioeconómicos en el síndrome de FM realizados en el continente Europeo (Knight et al., 2013; Pérez-Aranda et al., 2019; Perrot et al., 2012; Rivera et al., 2009). Al igual que en el país estadounidense, en Francia también se evaluó la asociación existente entre el gasto económico sanitario y la gravedad de los síntomas en FM. En concreto, Perrot et al. (2012) indicaron que el costo anual en pacientes con FM con sintomatología leve fue de 6.436 euros, en pacientes con sintomatología moderada fue de 7.800 euros y en pacientes con sintomatología grave fue de 11.862 euros (Perrot et al., 2012). En España, en un estudio multicéntrico realizado por Rivera et al. (2009) donde se analizó los costes socioeconómicos y sanitarios directos e indirectos en el síndrome de FM se observó que el gasto total anual por paciente diagnosticado con FM fue de 9.982 euros, de los cuales 3.245,8 euros correspondieron a costos directos y 6.736,2 euros correspondieron a costos indirectos. En relación a los costos directos, el gasto en las visitas médicas fue de 847 euros, en las pruebas diagnósticas fue de 473,5 euros, para los tratamientos farmacológicos fue de 439,2 euros, para los tratamientos no farmacológicos fue de 1.368,1 euros y para otras intervenciones sanitarias fue de 117,9 euros. Asimismo, en referencia a los costos indirectos, es decir, los costos laborales, el gasto por reducción de la jornada laboral fue de 913,1 euros, para una baja laboral fue de 3.556,2 euros y la pensión por invalidez permanente fue de 2.266,9 euros (Rivera et al., 2009). Por tanto, la carga socioeconómica relacionada con el síndrome de FM es un factor muy importante a tener en cuenta. Finalmente, y en línea con estos resultados, otro estudio realizado entre los años 2012 y 2014 en el territorio español reafirmó esos elevados costes en relación a los costos directos (consultas en atención primaria, atención especializada y pruebas diagnósticas específicas) y a los costos indirectos (Pérez-Aranda et al., 2019).

En resumen, y dada la condición de dolor crónico y la afectación que produce el síndrome de FM a nivel personal o laboral, esta patología causa un gran impacto en la atención sanitaria y genera un elevado costo económico para contrarrestar los efectos de la misma (Schaefer et al., 2016).

1.2 Alteraciones neurovasculares y su relación con los síntomas del síndrome de Fibromialgia.

Desde finales del siglo XIX, el estudio de la actividad y el control del flujo sanguíneo despertaron un gran interés en todos los investigadores fisiólogos que se ha mantenido hasta la actualidad. Numerosos factores como las alteraciones del Sistema Nervioso Autónomo, la excesiva presencia de sustancias circulantes en el torrente sanguíneo, la estimulación mecánica de los vasos sanguíneos, la respuesta miogénica de las arteriolas a las variaciones de la presión intraluminal, la concentración de metabolitos en los vasos sanguíneos y tejidos y las fuerzas de cizallamiento pueden influir en la perfusión óptima del flujo sanguíneo a los tejidos cuando estos demandan oxígeno (Secomb, 2008). A nivel microvascular, la mayor resistencia al flujo sanguíneo ocurre entre las arteriolas que proceden de una arteria principal y las vénulas de drenaje. La regulación del sistema de la red microvascular está controlada por la actividad de un conjunto de hormonas pero, sobre todo, por el SNA que se encarga de regular el tono vascular (Bagher & Segal, 2011). En este sentido, varios estudios científicos han afirmado que en sujetos diagnosticados de FM se han observado alteraciones en la morfología de sus capilares en cuanto a su diámetro y densidad, acompañada de una menor actividad microcirculatoria (Choi & Kim, 2015; Morf et al., 2005). Estas anomalías se deben a que los pacientes con síndrome de FM presentan, al igual que los sujetos con Fenómeno de Raynaud, síntomas de frialdad periférica persistentes que aumentan el tono vascular simpático mediado por el SNA y que provocan un mal funcionamiento vascular (espasmo) (Morf et al., 2005). Esta vasoconstricción sostenida activa a los receptores alfa-2-adrenérgicos del Sistema Nervioso Simpático (SNS), los cuales son potentes vasoconstrictores e interfieren, por un lado, en las dilataciones y formaciones de los capilares y, por otro lado, en el tono vasomotor del tejido muscular esquelético por lo que altera el mecanismo que asegura a dicho tejido una correcta provisión de sangre y oxígeno (Carlson et al., 2008; Choi & Kim, 2015).

Por otro lado, nuevos hallazgos científicos indican que esta alteración en la microcirculación sanguínea en pacientes con FM puede explicarse por cambios en la inervación de las anastomosis arteriovenosas del tejido cutáneo de las palmas de las manos (Albrecht et al., 2013). Las AAV son conexiones directas entre las pequeñas arteriolas y vénulas, cuya función principal es el transporte del calor desde el núcleo del cuerpo

humano hasta las áreas superficiales. Se encuentran localizadas en las mucosas y en las zonas de la piel glabra (sin pelo) del lecho ungeal de los dedos, nariz, orejas, pies y, sobre todo, en la profundidad de la dermis de las regiones hipotenares (100 AAVs/cm^2) de las palmas de las manos (Walløe, 2016). Las AAV, junto con las ateriolas cutáneas, se encuentran inervadas por fibras pequeñas de inervación simpática vasoconstrictora y de inervación sensorial vasodilatadora, aunque la actividad termorreguladora en esta área se encuentra gobernada por el sistema vasoconstrictor adrenérgico. Asimismo, intervienen en la regulación de la temperatura corporal central por lo que mantienen al organismo en una zona termoneutral adecuada que para el ser humano desnudo y en reposo oscila alrededor de los 26ºC y los 36ºC (Walløe, 2016). El concepto de zona termoneutral fue introducido entre los años 1940 y 1950 por Scholander et al. (1950) definiéndose como: "el metabolismo basal constante de un mamífero en reposo cuando la temperatura ambiente se encuentra en un intervalo entre los 37ºC de la temperatura corporal central hasta una temperatura inferior dependiente del aislamiento del cuerpo del propio mamífero" (Scholander et al., 1950). Por encima del extremo superior de la zona termoneutral la tasa metabólica aumenta debido a la producción del sudor. Por debajo del extremo inferior de la zona termoneutral la tasa metabólica aumenta proporcionalmente a la diferencia entre la temperatura ambiente y la temperatura central del organismo a consecuencia de la producción de calor generado por los temblores y el trabajo muscular activo por acción del frío (Walløe, 2016). Las AAV actúan como esfínteres que se abren o se cierran generando una disminución o aumento de flujo sanguíneo en la zona, derivando la sangre directamente hacia los plexos venosos de las extremidades. Ante un descenso de la temperatura (exposición al frío) el hipotálamo, como centro principal del control de la temperatura del cuerpo humano, recoge esta información y envía impulsos nerviosos a todos los axones simpáticos adrenérgicos de las AAV, comprometiendo su luz con la finalidad de proveer de flujo sanguíneo al tejido cutáneo para satisfacer las demandas metabólicas básicas ante este evento estresor (Daanen, 2003; Walløe, 2016). En relación a las AAV, un estudio llevado a cabo por Albrecht et al. (2013) en mujeres con síndrome de FM informó de la presencia de alteraciones en la respuesta vascular periférica. A través de biopsias realizadas en la piel de la eminencia hipotenar de las palmas de las manos descubrieron una excesiva inervación sensorial peptidérgica y una subrepresentación simpática noradrenérgica de las AAV de mujeres diagnosticadas con FM en comparación con controles sanos. Asimismo, encontraron que esta inervación sensorial peptidérgica de las AAV expresaban una hiperreactividad del receptor alfa-2-adrenérgico, el cual produce

la liberación simpática de noradrenalina/norepinefrina que median a favor de una constricción de la musculatura lisa de la túnica media de los vasos sanguíneos. Es importante recordar que las AAV regulan el flujo sanguíneo en el cuerpo humano para asegurar una óptima termorregulación (Walløe, 2016), por lo que una alteración del control del sistema nervioso conllevaría a una desregularización de la temperatura corporal central y a una disminución de la microcirculación sanguínea periférica (por cierre de las arteriolas y vénulas), restringiendo al tejido músculo-esquelético de una oxigenación y nutrición óptima, favoreciendo el depósito de metabolitos y el acúmulo de ácido láctico que explicaría el dolor profundo y fatiga generalizada característica en los pacientes con síndrome de FM (Albrecht et al., 2013). Finalmente, estos autores también indicaron que esta alteración neurovascular en las AAV no sólo explicaría los síntomas en sujetos con FM, sino que también explicaría la exacerbación de los mismos ante los cambios de temperatura (Albrecht et al., 2013).

Por último, el Sistema Nervioso Periférico a través de alteraciones de sus fibras nerviosas de pequeño calibre parece también estar involucrado en el desarrollo de los síntomas del síndrome de FM (Grayston et al., 2019). Actualmente, no existe un concepto o definición estándar para la neuropatía de las fibras nerviosas pequeño calibre, sin embargo, se sospecha de una neuropatía cuando en al menos dos de las siguiente tres pruebas diagnósticas se evidencia un deterioro de las fibras nerviosas pequeñas: a) test de examen neurológico; b) prueba sensorial cuantitativa; y c) biopsia cutánea acompañada siempre de dolor y disestesias por parte del paciente (Devigili et al., 2008). En una neuropatía de las fibras nerviosas pequeñas de pequeño calibre se produce un deterioro mucho más predominante en fibras nerviosas aferentes periféricas mielinizadas (A-delta) y en las fibras nerviosas aferentes periféricas no mielinizadas (fibras C). Ambas fibras aferentes están involucradas en la percepción dolorosa (alodinia e hiperalgesia), en la percepción sensitiva (parestesias) y en la percepción térmica (frío y calor) (Pickering et al., 2020). Además, se encargan de inervar a las glándulas sudoríparas que establecen un nexo de unión con el SNA (Lefaucheur et al., 2015; Oaklander et al., 2013). La prevalencia de la neuropatía de las fibras nerviosas pequeñas en población con síndrome de FM es del 49%, subrayando el impacto negativo que ejerce el SNP sobre el dolor y los síntomas de estos pacientes (Grayston et al., 2019). Esta alteración en las fibras, altera la función de los vasos sanguíneos de pequeño calibre a través de la producción de una respuesta neuropéptida alterada y la desregulación en la activación de receptores α adrenérgicos (Grayston et al.,

2019). De esta manera, esta microvasculopatía neurogénica podría explicar, al menos parcialmente, los déficits de perfusión a nivel músculoesquelético, el dolor profundo, intolerancia al ejercicio y un síntoma común como es "niebla mental" o “brain fog” característicos de la fibromialgia. En esta línea, estudios previos a través de microneurografía han evidenciado en sujetos con FM cambios en la velocidad de la conductividad de las fibras nerviosas periféricas C y una hiperreactividad de los nociceptores C y de las fibras nerviosas mecanosensibles de tipo 1 (Evdokimov et al., 2019). Por otro lado, y en línea con la conexión que presenta el SNP con el SNA a través de las glándulas sudoríparas, la literatura científica muestra disfunciones en el SNA de los pacientes diagnosticados con FM por lo que puede llegar a afectar a las funciones de sudoración y regulación de la pérdida de calor corporal, alterando los procesos de la termorregulación corporal global (Elmas et al., 2016).

1.3 Biomarcadores pro-inflamatorios y nociceptivos: el rol del Óxido Nítrico y Catecolaminas en los síntomas generales de hipersensibilidad al dolor en pacientes con síndrome de Fibromialgia.

La inflamación es un mecanismo de defensa del cuerpo humano que protege a nuestro organismo y, por tanto, es vital para la supervivencia. Sin ella no sería posible reaccionar ante una agresión endógena y/o exógena y tampoco se podría proporcionar los mecanismos necesarios para la reparación de los tejidos ante un daño o lesión en los mismos (Porth CM, 2011). Un correcto trabajo entre el sistema inmune, el sistema nervioso y las vías de coagulación es indispensable para que se produzca una adecuada respuesta inflamatoria, la cual se encuentra modulada por una serie de mensajeros denominados mediadores inflamatorios (Porth CM, 2011). Estos mediadores inflamatorios pueden ser liberados al torrente sanguíneo por células del sistema inmune como los macrófagos, linfocitos, mastocitos y leucocitos o por células plasmáticas. Los mediadores inflamatorios más característicos liberados por las células inmunes son las citocinas pro-inflamatorias y anti-inflamatorias, el factor de crecimiento nervioso, los neuropéptidos, los radicales libres de oxígeno, los derivados del ácido araquidónico y las aminas vasoactivas. Por otro lado, los mediadores inflamatorios a destacar a nivel del plasma son los factores de coagulación, las proteínas de fase aguda y las proteínas de complemento (Porth CM, 2011).

Las citocinas son pequeños polipéptidos que tienen un papel muy importante en la respuesta inflamatoria. Son liberadas al flujo sanguíneo principalmente por células del sistema inmune (macrófagos, monocitos y células T), aunque también pueden ser liberadas por células no pertenecientes al sistema inmune (células de Schwann, fibroblastos, microglía y astrocitos). A su vez se dividen en citocinas pro-inflamatorias y anti-inflamatorias con funciones inmunológicas y fisiológicas en el organismo (Bjurstrom et al., 2016). Las citocinas pueden actuar como mediadores entre las células gliales y las neuronas a través de las vías centrales y periféricas. En condiciones de normalidad, la glía tiene un papel protector sobre el SNC, sin embargo, en condiciones patológicas, la glía se excita y produce la liberación al torrente sanguíneo de citocinas pro-inflamatorias, las cuales sensibilizan a los nociceptores y a las neuronas del tejido nervioso permitiendo una mayor entrada de estímulos dolorosos a nivel central de forma directa e indirecta, facilitando así el fenómeno de SC (Graeber, 2010; Watkins et al., 2007).

La literatura a través de una gran variedad de estudios científicos ha informado de altos niveles en las concentraciones sanguíneas de citocinas pro-inflamatorias en diferentes poblaciones con dolor crónico. En pacientes con osteoartritis de rodilla se ha observado un estado basal pro-inflamatorio ante un aumento en el plasma sanguíneo de interleucinas IL-1β, IL-6 e IL-8 (Lundborg et al., 2010). En patologías como el síndrome del dolor regional complejo se ha mostrado un aumento significativo en las concentraciones de citocinas pro-inflamatorias IL-1β e IL-6 (Alexander et al., 2005). Estudios científicos transversales y longitudinales en poblaciones con dolor lumbar crónico y radiculopatía lumbar han indicado elevadas concentraciones de interleucinas IL-6 en el líquido cefalorraquídeo de estos pacientes (Nagashima et al., 2009; Zin et al., 2010). Finalmente, en pacientes con neuralgia postherpética se ha evidenciado un estatus basal pro-inflamatorio ante los elevados niveles de interleucinas IL-8 en el torrente sanguíneo en comparación con controles sanos (Kotani et al., 2004).

Estudios muy recientes en población con síndrome de FM han sugerido una inflamación sistémica crónica subyacente a la patología (Coskun Benlidayi, 2019). En este sentido, pacientes con FM muestran elevados niveles de citocinas pro-inflamatorias en el torrente sanguíneo entre las que destacan la interleucina IL-1β, IL-6, IL-8 y el factor de necrosis tumoral alfa (Mendieta et al., 2016; Peck et al., 2020). Estas concentraciones de citocinas pro-inflamatorias favorecen el desarrollo de síntomas como el dolor generalizado,

la fatiga, la hiperalgesia, la alodinia y la pérdida del sueño debido a la sensibilización que producen sobre los nociceptores (Andrés-Rodríguez et al., 2020; Rodriguez-Pintó et al., 2014; Üçeyler et al., 2011). Por tanto, y dado que las citocinas pro-inflamatorias pueden contribuir a la iniciación y/o progresión de las distintas manifestaciones clínicas, la regulación de los niveles de concentración en el plasma sanguíneo ha sido considerado como uno de los objetivos principales en el tratamiento del síndrome de FM (Coskun Benlidayi, 2019).

El estudio del manejo del dolor crónico en todas sus dimensiones ha generado grandes avances científicos en el conocimiento fisiopatológico del mismo, centrándose la mayor parte de las investigaciones en la identificación y análisis de los biomarcadores sanguíneos más relevantes (Gunn et al., 2020). Un biomarcador se define como toda sustancia que mide de manera objetiva tanto el estado biológico normal como cualquier proceso patogénico, así como la respuesta a un determinado método o tratamiento (Atkinson et al., 2001). En definitiva, el uso de los biomarcadores en el ámbito de la investigación científica y en el práctica clínica se centra en identificar pacientes con riesgo a desarrollar una patología, en el diagnóstico de una enfermedad, en evaluar la respuesta generada a un intervención y en el desarrollo de fármacos en etapa temprana. Sin embargo, encontrar de manera exitosa biomarcadores mecanicistas asociados al dolor es realmente complicado debido a la experiencia subjetiva que yace en todo proceso doloroso y al modelo biopsicosocial que concierne al dolor crónico (Kalso, 2004).

Uno de los biomarcadores circulantes más importantes en los procesos inflamamatorios es el óxido nítrico (ON). El ON es un radical libre derivado de la L-arginina que actúa como mensajero intercelular en los procesos inflamatorios y que interviene además en los mecanismos de respuesta inmunitaria, modulación del dolor y regulación del sistema circulatorio (vasodilatación) (Cury et al., 2011; Lewis et al., 1993; Schulman, 1997). El ON se concentra en el asta dorsal de la médula espinal con una función muy bien definida sobre los circuitos de transmisión nerviosa (neurotransmisor). Este biomarcador se puede liberar al torrente sanguíneo por el estímulo de múltiples factores humorales (aumento de la actividad plaquetaria, hipofibrinolisis, hiperviscosidad, deformidad de los hematíes, la activación de leucocitos y el estrés oxidativo), por el *"shear stress"* (tensión que ejerce la sangre en dirección tangencial sobre la superficie del endotelio generando una deformidad de la misma) y por la actividad prolongada de las

fibras nerviosas C en el asta dorsal de la médula espinal. La sobreactividad de algunos de estos factores y procesos produce la liberación y difusión del ON a través de la hendidura sináptica o bien fuera de las neuronas, por lo que puede llegar a excitar a los astrocitos y la microglía (Cury et al., 2011; Gratt & Anbar, 2005). La estimulación de la actividad normal de la glía provoca la liberación sistémica de citocinas pro-inflamatorias, especies reactivas de oxígeno y prostaglandinas que sensibilizan a los nociceptores y neuronas del tejido nervioso prolongando la hiperexcitabilidad de la médula espinal y favoreciendo la sensibilización central y el dolor crónico (Nijs et al., 2017). Asimismo, el ON es el principal factor relajante del músculo liso vascular por la acción que ejerce sobre el radical guanosín monofostato cíclico. Si el ON se difunde de manera excesiva a un área cercana de la piel se origina una hipertermia local por lo que puede influir en el proceso de disipación del calor por convección y provocar una desregularización de la temperatura general corporal, a favor de un aumento de la misma, y del metabolismo basal del cuerpo humano (Cheung, 2015; Gratt & Anbar, 2005). Además, Albrecht et al. (2013) en su estudio sobre las AAV de la piel glabra de la palma de las manos de pacientes con FM informaron que la liberación de compuestos como el ON puede dar lugar a una dilatación pasiva de las arteriolas y de las AAV, lo cual estimularía térmicamente las ramas sensoriales peptidérgicas de la piel y desencadenaría la activación del "reflejo axónico local" que produce la liberación al torrente sanguíneo de una gran cantidad de substancia P y receptores del CGRP (potentes vasodilatadores) con importantes efectos pro-nociceptivos e implicados en el dolor de tipo inflamatorio (Albrecht et al., 2013).

La literatura científica no ha mostrado un claro consenso en relación a la asociación existente entre los niveles de ON en el plasma sanguíneo y los síntomas del dolor característicos en sujetos con síndrome de FM. Si bien algunos estudios científicos encontraron que altas concentraciones de ON en suero sanguíneo se correlacionaron positivamente con distintas variables clínicas de FM como el dolor de cabeza, los umbrales del dolor a la presión, la percepción subjetiva dolor evaluado a través de la Escala Visual Analógica del Dolor y la capacidad funcional (Çimen et al., 2009; Rus et al., 2016; Sendur et al., 2009), otros estudios científicos no encontraron correlaciones significativas entre los niveles de concentración de ON y las características clínicas como el dolor subjetivo auto-percibido y el impacto global de los síntomas en la FM (Koca et al., 2018; Ozgocmen et al., 2006).

Otros de los biomarcadores y neurotransmisores circulantes a tener en cuenta en los procesos inflamatorios y nociceptivos son las catecolaminas y las aminopeptidasas. Por un lado, las catecolaminas son un conjunto de neurotransmisores monoamínicos derivados de aminoácidos aromáticos y producidos por la médula suprarrenal y por las fibras postganglionares del SNS. Dentro de este grupo destacamos la dopamina, la norepinefrina y la epinefrina. Las catecolaminas son secretadas al torrente sanguíneo ante una respuesta al estrés mediada por el SNS con un papel fundamental sobre las vías descendentes del dolor ya que modulan la respuesta dolorosa hacia la periferia (Rus et al., 2018). Por otro lado, las aminopeptidasas son metaloenzimas de zinc que favorecen la división de los aminoácidos en el extremo distal de las proteínas y de los péptidos. Entre las principales funciones de las aminopeptidasas destacan la regulación de la oxitocina, la secreción gonodal y de hormonas tiroideas y la regulación del metabolismo de las encefalinas (Ortega et al., 2013).

Así pues, las encefalinas son opioides endógenos con importantes efectos analgésicos que regulan los procesos de nocicepción y funciones cognitivas y psicológicas como la ansiedad, la depresión, el estrés y el estado de ánimo. Las encefalinas son sintetizadas en el asta dorsal de la médula espinal por diferentes tipos de neuronas con la finalidad de regular los procesos dolorosos producidos tras una lesión periférica (Henry et al., 2017). Son metabolizadas por enzimas denominadas encefalinasas entre las que destacan la aminopeptidasa degradante de la encefalina (EDA) y la oxitocinasa que metaboliza a la Met-encefalina y oxitocina endógena con efectos analgésicos, ansiolíticos y anti-estrés (Neumann & Slattery, 2016; Xin et al., 2017). Es decir, tanto la EDA como la oxitocinasa se encuentran asociadas con los mecanismos de regulación de la nocicepción y con los procesos cognitivos y psicológicos ya que participan en el metabolismo de las encefalinas (Martínez-Martos et al., 2019).

Bajo nuestro conocimiento, algunos estudios científicos han determinado una asociación entre el metabolismo de las catecolaminas y el síndrome de FM pero con resultados contradictorios pues los hallazgos de los niveles plasmáticos de dopamina, norepinefrina y epinefrina varían entre los estudios mostrando un aumento o un descenso de los mismos e incluso sin variaciones en sujetos con FM en comparación con controles sanos (Bote et al., 2012; Giske et al., 2008; Kadetoff & Kosek, 2010; Rus et al., 2018; Torpy et al., 2000). Asimismo, sólo un estudio ha investigado las asociaciones existentes

entre la actividad de las encefalinasas en el plasma sanguíneo y la sintomatología en FM (Martínez-Martos et al., 2019). En este estudio de casos y controles, la actividad a nivel sérico de las aminopeptidasas fue analizada en un total de 75 mujeres diagnosticadas con esta patología. Los resultados reportados indicaron que no existen asociaciones significativas entre los niveles de actividad de las encefalinasas en el suero sanguíneo y el dolor global auto-percibido por los pacientes evaluado a través de la Escala Visual Analógica del Dolor así como con respecto a la gravedad de los síntomas de Fibromialgia analizada a través del cuestionario de Impacto de Fibromialgia validado al español (Martínez-Martos et al., 2019).

Tal y como se ha comentado en apartados anteriores, la SC es un fenómeno característicos en sujetos con síndrome de FM (Chinn et al., 2016; Clauw, 2014; Stisi et al., 2007). Este fenómeno puede estar mediado por una serie de biomarcadores, neurotransmisores y neuropéptidos como el ON, la serotonina, la norepinefrina, la dopamina, la epinefrina, las encefalinasas, la substancia P o el CGRP, entre otros (Bellato et al., 2012; Martínez-Martos et al., 2019; Rus et al., 2018; Russell & Larson, 2009). A pesar de los resultados contradictorios reflejados en la literatura científica, se recomiendan futuras investigaciones pues cambios en los niveles de actividad de las encefalinasas y catecolaminas en el plasma sanguíneo pueden modificar los eventos reguladores en los que están involucrados, resultando, finalmente, en un estado patológico en los pacientes con FM (Martínez-Martos et al., 2019).

1.4 Estado basal pro-inflamatorio en poblaciones con dolor crónico e ingesta dietética.

Recientemente se ha evidenciado el posible papel determinante del estilo de vida, concretamente la calidad de la dieta, en pacientes con patologías asociadas a estados de dolor crónico (Tick, 2015; Witkamp & van Norren, 2018). Investigaciones previas postulan que un patrón dietético saludable podría ejercer una importante función en la modulación del dolor y, por tanto, podría considerarse como estrategia para controlar los síntomas en la práctica clínica (Tick, 2015; Witkamp & van Norren, 2018). No obstante, la evidencia disponible en relación a la asociación entre la ingesta dietética y modificaciones en la nocicepción debido a una disminución del estado inflamatorio es muy limitada (Tick, 2015; Witkamp & van Norren, 2018).

Intervenciones dietéticas centradas en un bajo consumo de carbohidratos y una alta adherencia a la dieta mediterránea en la reducción del dolor crónico, el estatus inflamatorio y el estrés oxidativo han reportado resultados prometedores en diferentes poblaciones con dolor crónico (Kaushik et al., 2020). En personas adultas con osteoartritis de rodilla la inclusión en un programa dietético consistente en un bajo consumo de carbohidratos durante 12 semanas evidenció una reducción del dolor evocado y del estrés oxidativo (Strath et al., 2020). En otro estudio llevado a cabo también en población con osteoartritis de rodilla se observó una reducción del dolor, rigidez, mejoría en el rango del movimiento articular y en la realización de las actividades de la vida diaria en los pacientes que recibieron un programa de dieta antioxidante caracterizado por el consumo de 40 gramas de polvo de arándanos deshidratados cada día en un periodo de 4 meses en comparación con aquellos que consumieron un placebo (Du et al., 2019).

Revisiones sistemáticas recientes han informado que diferentes intervenciones dietéticas pueden contribuir a mejorar los síntomas incluido el dolor en pacientes diagnosticados con FM (Bair & Krebs, 2020; Rossi et al., 2015; Silva et al., 2019). Sin embargo, los estudios existentes en este campo muestran resultados contradictorios que pueden deberse al bajo tamaño muestral de los mismos, al empleo de diferentes métodos de estimulación para provocar la liberación de citocinas al torrente sanguíneo y a la utilización de diferentes métodos de análisis que generan dificultad a la hora de extraer resultados y conclusiones generalizables (Bair & Krebs, 2020; Rossi et al., 2015; Silva et al., 2019). Por un lado, estudios científicos han indicado que una dieta vegana rica en lactobacterias disminuye los niveles de dolor y mejora la calidad del sueño en pacientes con FM (Kaartinen et al., 2000). Asimismo, otro estudio en pacientes que siguieron un plan de dieta vegetariana mostró una mejora en la gravedad e impacto de los síntomas de la FM (Donaldson et al., 2001). Sin embargo, por otro lado, y en contraste con estos hallazgos anteriores, un estudio realizado en pacientes con FM vinculando la dieta vegetariana con la posibilidad de mejorar los niveles de fatiga, insomnio, sueño y dolor no encontró asociaciones significativas entre esta intervención y las variables cínicas en FM (Azad et al., 2000).

Por otro lado, el Índice Dietético Inflamatorio (DII en inglés) es un índice dietético recientemente validado que se ha desarrollado para predecir los niveles de inflamación del paciente (Shivappa et al., 2014). Las puntuaciones del DII se asocian con varios

marcadores inflamatorios entre los que se incluyen la proteína C reactiva, IL-1, IL-2, IL-6, homocisteína y el fibrinógeno (Shivappa et al., 2014, 2015, 2017; Wirth et al., 2014). Sin embargo, el potencial de la dieta en pacientes con síndrome de FM no ha sido previamente examinado. Dado el estado pro-inflamatorio basal que presentan los sujetos con FM debido a las elevadas concentraciones de citocinas pro-inflamatorias en el suero sanguíneo, el empleo de intervenciones dietéticas anti-inflamatorias caracterizadas por un consumo abundante de verduras y frutas, de antioxidantes dietéticos, por una ingesta moderada de proteínas bajas en grasas así como de ácidos grasos monoinsaturados y por un consumo limitado de pan y cereales (especialmente los cereales refinados), carnes rojas y lácteos podría ser una estrategia prometedora para el control del estado basal pro-inflamatorio y en consecuencia la mejora de la sintomatología, fundamentalmente el dolor crónico, en pacientes con esta patología (Bjørklund et al., 2018; Kaushik et al., 2020; Rus et al., 2017; Sears, 2015; Sears & Ricordi, 2011). La falta de resultados pone de manifiesto la necesidad de futuras investigaciones que exploren los posibles vínculos entre el potencial inflamatorio de la dieta y la reducción de los síntomas relacionados con el síndrome de FM.

1.5 Diagnóstico y abordaje terapéutico del síndrome de Fibromialgia.

1.5.1 Diagnóstico.

En 1990 el ACR estableció los primeros criterios de diagnóstico para el síndrome de FM (Wolfe et al., 1990). Dichos criterios fueron:

1. Historia clínica de dolor generalizado durante al menos 3 meses en el lado izquierdo y derecho del cuerpo humano, por encima y por debajo de la cintura y en el esqueleto axial (raquis cervical, raquis dorsal o raquis lumbar).

2. Dolor a la palpación digital con una fuerza aproximada de 4 Kg en al menos 11 de los 18 puntos sensibles bilaterales que se muestran a continuación:

    I. Occipital: localizado en las inserciones del músculo suboccipital.
    II. Cervical: localizado en las caras anteriores de los espacios intertransversos de C5-C7.
    III. Trapecio: localizado en el punto medio del borde superior del músculo trapecio.

IV. Supraespinoso: localizado por encima de la espina de la escápula próximo al borde medial.
V. Segunda costilla: localizado en la segunda unión costocondral.
VI. Epicóndilo lateral: localizado a 2 cm distal del epicóndilo.
VII. Glúteo: localizado en el cuadrante externo del músculo glúteo superior.
VIII. Trocánter mayor: localizado posteriormente a la prominencia ósea del trocánter.
IX. Rodilla: localizado en la almohadilla de grasa próxima a la línea articular.

Durante el transcurso de los años, los criterios inicialmente propuestos por el ACR no abordaban ciertas realidades observadas en la categorización y evaluación de los pacientes con FM porque, en primer lugar, no se habían considerado características clínicas claves como la fatiga, los síntomas somáticos y los síntomas cognitivos y, en segundo lugar, porque no había una escala de gravedad de los síntomas que ayudase a registrar el nivel de los mismos entre diferentes pacientes con FM. Por ello, en 2010 el ACR actualizó los criterios diagnósticos e incluyó dos principales escalas: el índice de dolor generalizado (WPI en inglés) y la escala de gravedad de los síntomas de la FM (SSS en inglés) (Wolfe et al., 2010). Por un lado, el WPI consiste en una escala acompañada de un dibujo donde el paciente anota las áreas en las que refiere dolor durante la última semana. La puntuación total del índice oscila entre los 0 puntos y los 19 puntos. Por otro lado, la SSS evalúa la gravedad de los síntomas a través de un cuestionario que contiene 2 dimensiones: a) una primera dimensión que engloba síntomas de fatiga, sueño no reparador y síntomas cognitivos y en la cual el paciente tiene que indicar el nivel de gravedad de los síntomas durante la última semana a través de una escala Likert con un rango de puntuación desde los 0 puntos (ningún problema) hasta los 3 puntos (problemas graves y persistentes); y b) una segunda dimensión donde se evalúan los síntomas somáticos a través de una escala Likert con un rango de puntuación desde los 0 puntos (ningún síntoma) hasta los 3 puntos (gran número de síntomas). La puntuación total de la SSS oscila entre los 0 puntos y los 12 puntos y se obtiene de la sumatoria de los puntos del primer dominio junto con los puntos del segundo dominio (Wolfe et al., 2010).

En resumen, esta actualización de los criterios diagnósticos del ACR en 2010 estableció que un paciente se encontraba diagnosticado de FM si cumplía con las 3 condiciones siguientes (Wolfe et al., 2010):

1. Puntuación en el WPI≥7 y puntuación en la SSS≥5 o puntuación en el WPI entre 3-6 puntos y puntuación en la SSS≥9.
2. Los síntomas deben de estar presentes con el mismo nivel durante al menos 3 meses.
3. El paciente no tiene una patología previa diagnosticada que pudiera explicar su dolor.

Posteriormente, en el año 2016, nuevamente el ACR actualizó los criterios diagnósticos tras la revisión y evaluación de una serie de informes científicos publicados entre los años 2010 y 2016 con la finalidad de determinar la validez, utilidad, fiabilidad y modificaciones posibles de los criterios fijados en el año 2010 (Wolfe et al., 2016). Esta nueva actualización es la utilizada en la actualidad y considera que una persona puede ser diagnosticada de FM si cumple con las siguientes condiciones (Wolfe et al., 2016):

1. Presenta dolor generalizado en al menos 4 de las 5 regiones siguientes: región corporal superior derecha, región corporal superior izquierda, región corporal inferior derecha, región corporal inferior izquierda y región axial.
2. Los síntomas deben de estar presentes con el mismo nivel durante al menos 3 meses.
3. Puntuación en el WPI≥7 y puntuación en la SSS≥5 o puntuación en el WPI entre 4-6 puntos y puntuación en la SSS≥9.
4. El diagnóstico de FM es completamente válido independientemente de otras enfermedades previas diagnosticadas en el paciente.

Finalmente, destacar que en el año 2018 una asociación de trabajo público-privada denominada ACTTION junto con la Sociedad Estadounidense del Dolor propuso nuevos criterios diagnósticos para el síndrome de FM, englobado dentro de un proyecto a gran escala destinado a desarrollar un sistema de diagnóstico útil y consistente para todos los trastornos de dolor crónico (Arnold et al., 2019). Los criterios diagnósticos establecidos por este grupo de trabajo fueron:

1. Dolor generalizado en múltiples sitios, en concreto, dolor en al menos 6 de las 9 regiones siguientes: cabeza, miembro superior izquierdo, miembro superior derecho, tórax, abdomen, raquis vertebral superior, raquis vertebral inferior, miembro inferior izquierdo y miembro inferior derecho.
2. Los síntomas deben de estar presentes con el mismo nivel durante al menos 3 meses y deben ir acompañados de fatiga y problemas de sueño de una gravedad moderada a severa.
3. Hay que tener en cuenta una serie de síntomas que aunque no se consideran necesarios para el diagnóstico de la FM apoyan al mismo. Dichos síntomas son: sensibilidad de los tejidos blandos, rigidez, hipersensibilidad ambiental e hipervigilancia.

A pesar de esta gama de criterios diagnósticos mencionados anteriormente, la literatura científica propone la utilización de una serie de métodos con fines evaluativos y de diagnóstico para el síndrome de FM (Bair & Krebs, 2020). En la evaluación y exploración inicial se debe de incluir una historia clínica completa que registre un listado de uso de fármacos, afecciones médicas previas, antecedentes cardiovasculares (hipertensión, dislipemia), antecedentes familiares, factores predisponentes al desarrollo de los síntomas como son los estilos de vida (inactividad física o sedentarismo), la obesidad, el nivel socioeconómico y la afectación psicológica. Del mismo modo, se debe registrar la presencia de comorbilidades como la hipersensibilidad química, la cistitis intersticial (síndrome de la vejiga dolorosa) y la fatiga crónica ya que pueden ayudar a lograr los objetivos propuestos en el tratamiento. Se debe también anotar cualquier tipo de problema neurológico, infeccioso, reumatológico o endocrino para que puedan ser evaluados posteriormente con mayor profundidad (Bair & Krebs, 2020; Häuser et al., 2015). Por otra parte, en el proceso de evaluación se puede proporcionar informes o cuestionarios autoadministrados como la Escala Visual Analógica del Dolor (EVA), el Cuestionario Revisado de Impacto de la Fibromialgia (FIQ-R en inglés), el Inventario de Sensibilización Central (CSI en inglés), el Cuestionario de Calidad del Sueño de Pittsburgh (PSQI en inglés), el Inventario Multidimensional de Fatiga (MFI en inglés) o el Inventario de Ansiedad de Beck (BAI en inglés). Para evaluar las alteraciones vasculares se pueden utilizar diferentes intervenciones no invasivas como la capilaroscopia en la punta de los dedos de la mano, la termografía infrarroja o el láser Doppler (Choi & Kim, 2015; Costa et al., 2019; Morf et al., 2005). De igual forma, para evaluar la temperatura corporal global se

puede utilizar un termómetro infrarrojo en el conducto auditivo externo del oído ya que proporciona una medida precisa de la temperatura corporal central debido a la relación de la arteria timpánica con el hipotálamo (Gasim et al., 2013). Por último, los hallazgos a través de imágenes radiográficas o de neuroimagen no presentan una gran utilidad para fines diagnósticos o de pronóstico en el síndrome de FM (Bair & Krebs, 2020).

1.5.2 Abordaje terapéutico del síndrome de Fibromialgia.

La Liga Europea Contra el Reumatismo (EULAR en inglés) a través de la revisión de numerosos ensayos clínicos de revisiones sistemáticas propuso, en 2016, una serie de recomendaciones básicas para el tratamiento del síndrome de FM en función de los niveles de evidencia y de las fuerzas de recomendaciones que las sostienen (G. J. Macfarlane et al., 2017). Dichos criterios de fuerzas de recomendación y niveles de evidencia establecidos en la guía EULAR fueron los siguientes: **Grado A**. Recomendación fuerte. Evidencia de que el beneficio/eficacia de la intervención o procedimiento es significativamente mayor que los posibles efectos adversos; **Grado B**. Evidencia de que el beneficio/eficacia de la intervención o procedimiento es mayor que los posibles efectos adversos, por lo tanto, el tratamiento puede llevarse a cabo sin ningún tipo de problema; **Grado C**. Evidencia de que el beneficio/eficacia de la intervención o procedimiento se encuentra en equilibrio con los posibles efectos adversos, aunque tampoco existe evidencia en contra del tratamiento. No se recomienda realizar la intervención en la práctica clínica; **Grado D**. Evidencia de que los posibles efectos adversos de la intervención o procedimiento superan a los beneficios/eficacia. No se recomienda llevar a cabo la intervención o tratamiento; **Grado I**. La evidencia existente es pobre o insuficiente para decantarse a favor o en contra de la intervención o procedimiento. En relación a los niveles de evidencia: **Nivel Ia**. Datos derivados de ensayos controlados aleatorizados de revisiones sistemáticas con meta-análisis; **Nivel Ib**. Datos derivados de ensayos clínicos aleatorizados de revisiones sistemáticas sin meta-análisis; **Nivel IIa**. Datos derivados de estudios observacionales comparativos con diferencias estadísticas; **Nivel IIb**. Datos derivados de estudios observacionales no comparativos; **Nivel IIc**. Datos derivados de estudios de reporte de casos; **Nivel III**. Datos derivados de ensayos clínicos aleatorizados con resultados inconsistentes, de pequeños estudios, de informes, de registros o del consenso de expertos. **Nivel IV**. No hay estudios identificativos en la literatura científica.

Las principales recomendaciones específicas citadas en la guía EULAR de 2016 se desarrollan a continuación (G. J. Macfarlane et al., 2017):

1. Terapia farmacológica:

I. El tramadol es un tipo de fármaco opioide débil que regula la liberación de neurotransmisores como la substancia P, la acetilcolina y la noradrenalina que se recomienda para el tratamiento del dolor crónico en FM con un Riesgo Relativo (RR) de 1.77 y un IC al 95% de 1.26 a 2.48. **Grado A-Nivel Ib**.

II. La amitriptilina es un fármaco antidepresivo tricíclico que proporcionado a dosis bajas de unos 25 mg por día puede ayudar a reducir el dolor y la fatiga y a mejorar la calidad del sueño con un RR de 1.60 y un IC al 95% de 1.15 a 2.24. **Grado A-Nivel Ia**.

III. La duloxetina y el milnacipran son otro tipo de fármacos antidepresivos que ayudan a reducir el dolor y disminuir la fatiga pero que se recomiendan en aquellos pacientes con síndrome de FM que presentan un dolor severo. RR de 1.38 y un IC al 95% de 1.25 a 1.51. **Grado A-Nivel Ia**.

IV. La pregabalina es un fármaco anticonvulsivo que presenta resultados positivos sobre la disminución del dolor y la mejora de la calidad del sueño pero que se debe de administrar en pacientes diagnosticados con FM con un dolor intenso. RR de 1.37 y un IC al 95% de 1.22 a 1.53. **Grado A-Nivel Ia**.

V. La ciclobenzaprina, con estructura muy similar a la amitriptilina, es un fármaco que ayuda a mejorar la eficiencia del sueño pero que debe recomendarse en aquellas personas con problemas de sueño derivados verdaderamente del síndrome de FM. RR de 4.8 y un IC al 95% de 3 a 11. **Grado A-Nivel Ia**.

2. Terapia no farmacológica:

I. El ejercicio aeróbico, ya sea en tierra firme o en agua, mejora el dolor y sintomatología en pacientes con síndrome de FM por la secreción de beta-endorfinas y citocinas anti-inflamatorias IL-4 e IL-5 con efectos analgésicos. Del mismo modo,

el entrenamiento de la fuerza muscular con pesas también es beneficioso en estos pacientes a pesar de que se pueda producir un aumento del dolor al principio de la ejecución del mismo con un RR de 0.65 y un IC al 95% de -0.09 a 1.39. **Grado A-Nivel Ia**.

II. La terapia cognitivo-conductual (CBT en inglés) es una modalidad de tratamiento que ayuda a pacientes con FM a controlar el dolor y mejorar la calidad de vida a través de una terapia de conversación pedagógica con el paciente para ayudarle a afrontar los pensamientos o ideas negativas y cambiar su forma de comportarse ante un evento adverso. RR de -0.29 y un IC al 95% de -0.49 a -0.17. **Grado A-Nivel Ia**.

III. Las terapias de componente múltiple se basan en la combinación del ejercicio físico, educación del paciente, técnicas de relajación u otros tratamientos específicos como Tai Chi o masaje con resultados beneficiosos sobre el dolor generalizado y la fatiga en personas con síndrome de FM, con un RR de -0.37 y un IC al 95% de -0.62 a -0.13. **Grado A-Nivel Ia**.

IV. Las técnicas de acupuntura tradicional o acupuntura eléctrica cuando se combinan con otras modalidades de tratamiento pueden disminuir los síntomas de dolor en pacientes con FM. Asimismo, sesiones de hidroterapia o spa pueden mejorar la sintomatología de estos pacientes con una duración de hasta unas 14 semanas con un RR de -0.78 y un IC al 95% de -1.42 a -0.13. **Grado A-Nivel Ia**.

V. Las terapias de mindfulness (atención plena) centradas en la reducción del estrés así como diversas modalidades de técnicas de meditación como el Qigon, Yoga o Tai Chi pueden ayudar a mejorar el dolor, la fatiga y la calidad del sueño en pacientes con FM. RR de -0.23 y un IC al 95% de -0.46 a -0.01 para el dolor; RR de -0.61 y un IC al 95% de -0.95 a -0.27 para el sueño; RR de -0.66 y un IC al 95% de -0.99 a -0.34 para la fatiga. **Grado A-Nivel Ia**.

Por otra parte, en esta guía EULAR también se establecieron una serie de recomendaciones generales basadas todas ellas en el consenso y en la opinión unánime de los expertos (**Nivel III**) (G. J. Macfarlane et al., 2017). Así pues, se indicó que para un buen abordaje del síndrome de FM se requiere de un diagnóstico precoz. Además, es necesaria una evaluación integral del dolor, de la función y del contexto social y se debe de

proporcionar al paciente toda la información posible, incluyendo cualquier tipo de material escrito con la finalidad de ayudarlo en el entendimiento de su patología. En general, la gestión del síndrome de FM debe de seguir un enfoque gradual y siempre atendiendo a la complejidad y heterogeneidad de los síntomas debido al procesamiento anormal del dolor y las características concomitantes de esta condición. De la misma manera, las intervenciones terapéuticas deberían centrarse primariamente en el empleo de terapias no farmacológicas adaptadas a la intensidad del dolor, funcionalidad, fatiga, alteraciones del sueño, comorbilidades y preferencias del paciente. Es decir, se recomienda que la toma de decisiones sea consensuada con el paciente (G. J. Macfarlane et al., 2017).

1.5.2.1 Tratamiento farmacológico.

El uso de terapias farmacológicas en condiciones de dolor crónico como el síndrome de FM lleva implícito un proceso de reevaluación y revisión constante para garantizar la continuidad de estos tratamientos, especialmente, aquellos destinados a la cognición y fatiga (Häuser et al., 2017). La mayoría de los fármacos empleados para el tratamiento de la FM siguen mostrando resultados insatisfactorios en la actualidad, principalmente, por la falta de eficacia que presentan, por el elevado riesgo de efectos secundarios que reportan y por la adicción a los mismos (Häuser et al., 2017). En este sentido, en la guía revisada para el manejo de la FM de la EULAR de 2016 no recomiendan el uso de terapias farmacológicas que incluyan fármacos anti-inflamatorios no esteroideos (NSAIDs en inglés), fármacos inhibidores de la monoaminooxidasa (MAOIs en inglés), fármacos inhibidores de la recaptación de serotonina (SSRIs en inglés), hormona del crecimiento y, sobre todo, el uso de oxibato de sodio (rechazado por la Agencia Europera del Medicamento y por la Agencia de Alimentos y Medicamentos de los Estados Unidos), opiodes fuertes y corticoesterioides (G. J. Macfarlane et al., 2017).

En contraposición, los fármacos más utilizados para el tratamiento de los síntomas en pacientes diagnosticados con FM suelen incluir fármacos inhibidores de la recaptación de la noradrenalina (SNRIs en inglés) entre los que destacan el tramadol, la duloxetina y el milnacipran, los cuales son considerados para pacientes con dolor severo; fármacos antidepresivos como la amitriptilina; fármacos antiepilépticos como la pregabalina; y fármacos para problemas de sueño como la ciclobenzaprina (Häuser et al., 2017; G. J. Macfarlane et al., 2017). Numerosos revisiones sistemáticas y meta-análisis avalan con resultados prometedores el empleo de estos fármacos mencionados anteriormente en

población con FM (Häuser et al., 2009, 2011, 2013; Lunn et al., 2014; Mease et al., 2009; Murakami et al., 2017; Nishishinya et al., 2008; Roskell et al., 2011; Wiffen et al., 2013). En este sentido, una revisión sistemática realizada en el año 2014 mostró que la duloxetina disminuyó el dolor en los pacientes con FM alrededor de un 50% en comparación con el grupo control que recibió placebo. Además, otro estudio elaborado por Murakami et al. (2017) indicó que la duloxetina es una buena opción de tratamiento farmacológico en pacientes con FM con fatiga severa y depresión, mostrando, además, que dicho fármaco es seguro y eficaz por el seguimiento y revisión realizado al año del inicio del estudio (Murakami et al., 2017). En relación al fármaco milnacipran, un ensayo controlado aleatorizado de doble ciego demostró la eficacia del milnacipran en el alivio del dolor, en la mejora del bienestar general y en el incremento de la función física de los pacientes con síndrome de FM en comparación con el grupo control placebo (Mease et al., 2009). De la misma manera, otro estudio científico mostró que el uso de milnacipran presentó efectos positivos sobre la intensidad del dolor y la fatiga (Häuser et al., 2013). Con respecto al tramadol, un meta-análisis realizado en 2011 evidenció que la prescripción de dicho fármaco con una dosis de 37.5 mg/4 veces al día durante 3 meses mejoraba el dolor en pacientes con FM alrededor de un 30% en comparación con el grupo control placebo (Roskell et al., 2011). Por otra parte, la literatura científica muestra efectos beneficios del uso de la pregabalina en sujetos con FM (Wiffen et al., 2013). Así pues, un meta-análisis de 4 ensayos controlados aleatorizados con tratamiento de pregabalina en pacientes diagnosticados con FM mostró mejoras significativas sobre el dolor, los problemas de sueño y la calidad de vida en comparación con los controles placebo (Häuser et al., 2009). Por último, el empleo de la amitriptilina como tratamiento en la FM también ha sido avalado por numerosas revisiones sistemáticas en la literatura científica. Nishishinya et al. (2008) indicaron que la prescripción de 25mg por día de amitriptilina en pacientes con FM tenía resultados positivos sobre el dolor, sueño y fatiga a las 6-8 semanas de tratamiento, pero no se mantuvieron a las 12 semanas de seguimiento (Nishishinya et al., 2008). Además, una revisión sistemática con meta-análisis realizada en 2011 afirmó que el uso de amitriptilina aliviaba el dolor alrededor de un 30%, aunque los efectos reportados sobre la fatiga y el sueño fueron medios o bajos (Häuser et al., 2011).

1.5.2.2 Tratamiento no farmacológico.

Una gran variedad de métodos de tratamiento han sido estudiados y desarrollados a lo largo de estos últimos años con la finalidad de mejorar la sintomatología y calidad de

vida en los pacientes que padecen FM (Aman et al., 2018). En relación al estudio proporcionado por la EULAR del manejo del síndrome de FM y bajo el consenso unánime de los expertos que conformaron el grupo de trabajo, se establecieron unas recomendaciones para el uso de terapias no farmacológicas en función de una escala de graduación (fuertemente recomendado; débilmente recomendado; no recomendado; en contra) (G. J. Macfarlane et al., 2017). En primer lugar, dicho grupo de trabajo recomendó fuertemente la realización de ejercicio físico, entrenamiento de fuerza con pesas y ejercicio aeróbico en pacientes con FM por el efecto beneficioso sobre el dolor, la función física, la discapacidad y el bienestar personal. En segundo lugar, y en un menor rango de recomendación que la realización del ejercicio físico, indicaron diferentes modalidades de terapias que incluyeron técnicas de meditación (Qigon, Yoga o Tai Chi) por sus efectos positivos en el sueño, la fatiga y la calidad de vida, terapias de mindfulness por las mejorías observadas sobre el dolor y la calidad de vida, terapia cognitivo-conductual por los efectos sobre el dolor, discapacidad y estado de ánimo a corto plazo, en incluso, levemente a largo plazo, y terapias complementarias como la acupuntura o hidroterapia por los buenos datos observados en la disminución del dolor, fatiga y calidad de vida. En tercer lugar, este grupo de trabajo no recomendó el empleo por su baja o nula eficacia de terapias como el masaje, hipnoterapia o terapias alternativas como la homeopatía. Finalmente, mostraron su disconformidad rotunda por precaución y seguridad de los pacientes con FM del empleo de la quiropráctica (quiropraxia) (G. J. Macfarlane et al., 2017).

A continuación se detallan una variedad de intervenciones terapéuticas conservadoras aplicadas en población con FM en base a las publicaciones de revisiones sistemáticas y meta-análisis de la literatura científica:

1. **Ejercicio**. En relación a los programas de ejercicio físico, la literatura científica indica efectos positivos sobre la sintomatología de pacientes con FM tanto en la realización de actividades de ejercicio aeróbico como en los programas de entrenamiento de fuerza o resistencia (A. Busch et al., 2002; A. J. Busch et al., 2013). Por un lado, una revisión sistemática de 47 estudios con ensayos clínicos aleatorizados de diferentes programas de ejercicio aeróbico con un tiempo de duración igual o mayor a 30 minutos (si los ejercicios se realizaban una vez al día) o un tiempo de duración igual o mayor a 10 minutos (si los ejercicios se realizaban dos veces al día) evidenció mejorías significativas sobre la intensidad del dolor y la

función física en pacientes diagnosticados con FM (A. Busch et al., 2002). Por otro lado, otra revisión sistemática que incluyó 5 ensayos controlados aleatorizados centrados en un programa de entrenamiento de resistencia de 8 o más repeticiones por ejercicio durante 2 o 3 veces a la semana informó de una reducción del dolor de unos 3.3 cm sobre la escala numérica del dolor así como de una mejoría de la funcionalidad de los pacientes con FM en comparación con el grupo control que recibió placebo (A. J. Busch et al., 2013).

2. **Acupuntura**. El uso de acupuntura combinada con otras modalidades de tratamiento ha sido analizada en poblaciones con FM. Una revisión sistemática de alta calidad constituida por 9 ensayos clínicos aleatorizados con un total de 395 pacientes con FM mostró que el empleo de acupuntura tradicional sobre los puntos específicos establecidos durante un tiempo de 20 a 30 minutos en un periodo de 3 a 13 semanas combinada con terapias estándar generaba una reducción del dolor alrededor del 30% en comparación con aquellos pacientes que solamente recibieron acupuntura tradicional. De la misma manera, el uso de acupuntura eléctrica se correlacionó con una disminución del dolor en un 22% y con una mejora sobre la fatiga generalizada en un 11% (Deare et al., 2013).

3. **Terapia cognitivo-conductual**. Con respecto a esta modalidad de tratamiento, la literatura científica muestra una revisión sistemática de 23 ensayos controlados aleatorizados con más de 2000 pacientes diagnosticados de FM (Bernardy et al., 2013). Las sesiones consistían en una serie de charlas pedagógicas con el paciente a través de habilidades comunicativas entendibles con la finalidad de proporcionarle estrategias de afrontamiento y control sobre su dolor y patología. La duración media observada en todos los estudios fue de 18 horas distribuidas en 10 sesiones en un periodo de 10 semanas. El uso de la CBT mostró ser efectiva a corto y largo plazo en cuanto a la reducción del dolor y la mejora de la discapacidad y del estado de ánimo en comparación con el grupo control de pacientes con FM que recibieron tratamiento habitual o que se encontraban en una lista de espera para poder participar en los proyectos de investigación (Bernardy et al., 2013).

4. **Mindfulness**. La terapia de mindfulness basada en la reducción del estrés y centrada en la atención plena sobre el cuerpo es otra de las intervenciones terapéuticas

actuales más empleadas en el síndrome de FM (Lakhan & Schofield, 2013; Marikar Bawa et al., 2015). En este sentido, una revisión sistemática y meta-análisis elaborada por Lauche et al. (2013) sobre la intervención del mindfulness en pacientes con FM con sesiones de duración de 2 a 3.5 horas en un periodo de 8 a 10 semanas y con actividades individualizadas en el domicilio de una duración de 30 a 45 minutos mostró mejorías inmediatas en el dolor tras el tratamiento con mindfulness en comparación con el grupo control de sujetos con FM que recibió cuidados de salud habituales o terapias activas constituidas por técnicas de educación, de apoyo, de relajación y de stretching (Lauche et al., 2013).

5. **Terapias de meditación**. El empleo de técnicas de meditación en diferentes modalidades como el Yoga, Tai Chi o Qigon han experimentado un notable crecimiento en el campo de la FM (Prabhakar et al., 2019). En reciente meta-análisis de revisiones sistemáticas que incluyó 7 ensayos controlados aleatorizados y un total de 362 pacientes con FM indicó que los tratamientos de meditación como el Yoga, Qigon y Tai Chi aplicados en un periodo de 4 a 12 semanas con una duración total distribuida en 16 horas mejoraban la calidad del sueño y la fatiga a corto y largo plazo en los pacientes con FM en comparación con los pacientes que recibieron tratamiento habitual o terapias activas (ejercicio aeróbico, educación para a salud y estiramientos) (Langhorst et al., 2013).

6. **Hidroterapia y balneoterapia**. La hidroterapia y la baleneoterapia son otras de las terapias complementarias empleadas en la patología de FM (Lauche et al., 2015). Varias revisiones sistemáticas analizaron las terapias de baños de agua o lodo a una temperatura de 36ºC a 37ºC o ligeramente superior (de 40ºC a 45ºC) con un tiempo medio de duración de 4 horas distribuidas a lo largo de varias semanas. Como resultados se observaron mejorías en las variables del dolor y el estado de calma con efectos a largo plazo mantenidos hasta la cuarta semana. Además, no se reportaron diferencias significativas en cuanto a una mayor o menor efectividad del uso de la balneoterapia o la hidroterapia por separado (Langhorst et al., 2009; Naumann & Sadaghiani, 2014).

7. **Masaje**. El masaje es una terapia conservadora que ha sido investigada en el manejo de la FM a través de revisiones literarias (Yuan et al., 2015). En un meta-análisis

elaborado en 2014 que incluyó 9 ensayos clínicos aleatorizados con un total de 404 pacientes diagnosticados con FM evaluó diferentes técnicas de masoterapia con un tiempo de duración de 25 a 30 minutos y unas 5 sesiones de media en comparación con grupos controles que recibieron una variedad de terapias como la estimulación eléctrica transcutánea, técnicas de relajación, acupuntura y cuidados básicos de salud. Dicho meta-análisis indicó que el uso del masaje por sí solo no se asociaba con una mejoría en las características clínicas del dolor. No obstante, hubo efectos positivos a corto plazo sobre el dolor, la ansiedad y la depresión aunque, sin embargo, los ensayos clínicos que sustentaron estos resultados fueron de una baja calidad metodológica y no mostraron evidencia sobre los efectos del seguimiento (Li et al., 2014).

8. **Quiropráctica (quiropraxia)**. La quiropraxia es una de las técnicas a evitar en el ámbito de la FM (G. J. Macfarlane et al., 2017). Los estudios científicos disponibles sobre la misma son escasos, encontrándose únicamente un estudio piloto, un estudio cuasialeatorio y un ensayo clínico controlado sin resultados significativos y contraproducentes. Por tanto, ante la falta de evidencia y con el objetivo de salvaguardar el estado de salud de los pacientes con FM, el empleo de técnicas quiroprácticas no está recomendado en esta población (Ernst, 2009).

9. **Homeopatía**. Por último, la homeopatía es otra de las variantes terapéuticas que la guía de la EULAR no recomienda utilizar en pacientes con FM (G. J. Macfarlane et al., 2017). Los únicos estudios disponibles son un total de 4 ensayos clínicos aleatorizados en 2 revisiones sistemáticas con un tratamiento que consistía en el consumo de medicamentos homeopáticos como la arnica montana, la bryonia alba y el rhus toxicodendon 1 vez al día, a lo largo de 3 meses y con un seguimiento cada 4-8 semanas a través de entrevistas clínicas. Dicha terapia no mostró efectos beneficiosos sobre la sintomatología en pacientes con FM en comparación con el grupo control placebo (Boehm et al., 2014; Perry et al., 2010).

1.5.2.3 Nuevos enfoques terapéuticos y evaluativos desde la Fisioterapia.

1.5.2.3.1 Fisioterapia mediante Educación en Dolor y electroterapia.

A pesar del amplio conjunto de tratamientos terapéuticos no farmacológicos aplicados en poblaciones con síndrome de FM, recientemente se han propuesto nuevas

líneas de tratamiento e investigación en Fisioterapia para el control y el abordaje de la sintomatología en pacientes con FM tales como la neuroeducación del dolor (PNE en inglés) o técnicas de electroterapia como la radiofrecuencia dieléctrica monopolar (MDR en inglés) (Barrenengoa-Cuadra et al., 2020; Ibáñez-Vera et al., 2020; Malfliet et al., 2017).

Por un lado, la neuroeducación del dolor es un enfoque terapéutico centrado en la educación y en el proceso de aprendizaje por parte del paciente sobre los mecanismos de neurofisiología, neuroanatomía y neurobiología del dolor, así como del procesamiento a nivel del SNC (Malfliet et al., 2017). Reconceptualizar el término del dolor en base a que todo proceso doloroso no tiene por qué estar asociado a un daño tisular sino que puede deberse a una alteración en el procesamiento de la información nociceptiva a nivel cerebral, es uno de los objetivos primordiales en la intervención de la PNE. En esta reconceptualización es necesario proporcionar al paciente información detallada y clarificadora sobre la fisiopatología del dolor y los procesos de sensibilización central y periférica, apoyada siempre de soportes de información escrita (folletos, guías de fácil lectura, etc.) y acompañada de ejemplos y metáforas ya que pueden ayudar a disminuir la hiperactividad del Sistema Nervioso Simpático, del sistema inmunológico, del sistema endocrino y del sistema músculo-esquelético (Moseley, 2003). La PNE, además, persigue que el paciente tenga una alta participación en todo el proceso de neuroeducación por lo que, para ello, debe de ser consciente del origen de sus síntomas, aceptar el diagnóstico de la enfermedad, afrontar la patología en diferentes tareas o actividades de la vida diaria, identificar factores externos agravantes (creencias, entorno cultural, capacidad de aprendizaje, sobreinformación clínica, etc.), trabajar la conciencia corporal y la atención, y adherirse a diferentes programas aeróbicos, de higiene del sueño o dietéticos (Aman et al., 2018; Barrenengoa-Cuadra et al., 2020). Todo este proceso ayudará al paciente a reducir las regiones de dolor, la gravedad de los síntomas, la catastrofización y el impacto sobre la discapacidad funcional (Barrenengoa-Cuadra et al., 2020; García-Ríos et al., 2019; Malfliet et al., 2017).

La literatura científica recoge una amplia variedad de estudios científicos y revisiones sistemáticas con intervención de PNE en diferentes poblaciones con dolor crónico (Andias et al., 2018; Bodes Pardo et al., 2018; Louw et al., 2016; Watson et al., 2019; Wood & Hendrick, 2019). En este sentido, una revisión sistemática elaborada por

Louw et al. (2016) que incluyó 13 ensayos clínicos aleatorizados con un total de 629 pacientes con diferentes condiciones de dolor crónico (dolor lumbar crónico inespecífico, radiculopatía lumbar, dolor de cuello, síndrome de fatiga crónica y síndrome de FM) indicó que la aplicación de diversos programas de PNE son efectivos en la disminución del dolor, en la mejoría de la discapacidad y funcionalidad, en el incremento del movimiento articular y en la disminución de los síntomas psicológicos (Louw et al., 2016).

En relación a población con FM también existen numerosas investigaciones científicas con esta modalidad de tratamiento de PNE (Amer-Cuenca et al., 2020; Barrenengoa-Cuadra et al., 2020; Serrat et al., 2020; van Ittersum et al., 2014). En un estudio llevado a cabo por Barrenengoa-Cuadra et al. (2020) en un centro hospitalario de Bilbao (España) en un total de 85 pacientes con FM, indicó que la intervención de PNE acompañada de un programa de trabajo corporal y atención plena mejoraba la puntuación del dolor en el WPI, en la SSS y en el FIQ. Además, estos beneficios se mantuvieron en las visitas a los 6 meses y a los 12 meses, observándose beneficios a medio y largo plazo (Barrenengoa-Cuadra et al., 2020). Por otra lado, un reciente ensayo clínico aleatorizado de 12 semanas de duración en 169 pacientes diagnosticados con FM analizó los posibles efectos de una intervención de PNE junto con CBT, mindfulness y técnicas de meditación en comparación con un grupo control que recibió asesoramiento de ejercicio aeróbico, educación básica de salud y tratamiento farmacológico con duloxetina y amitriptilina. Los datos observados indicaron importantes efectos en la disminución del impacto funcional (d=1.13), en el dolor autopercibido (d=0.66), en los síntomas de depresión (d=0.69), en la actividad funcional (d=0.53), en la fatiga (d=0.77), en la ansiedad (d=0.99) y en la catastrofización del dolor (d=1.21) (Serrat et al., 2020). Finalmente, otro reciente ensayo controlado aleatorizado de simple ciego evaluó la dosificación de una intervención de PNE y sus efectos sobre las variables del dolor y características clínicas en 103 pacientes con síndrome de FM (Amer-Cuenca et al., 2020). El programa de PNE consistía en diferentes diapositivas elaboradas en power point con información sobre la fisiología del SNC, las características del dolor agudo y crónico, el origen del dolor en el SNC y la cronificación de todo proceso doloroso a través de los fenómenos de SC, neuroplasticidad del SNC y neuromatriz del dolor. La aleatorización estaba configurada en 4 grupos de intervención: el grupo 1 recibió 45 minutos del programa de PNE 1 vez por semana a lo largo de 6 sesiones; el grupo 2 recibió 45 minutos de PNE 1 vez por semana en un total de 2 sesiones; el grupo 3 recibió 6 sesiones de 15 minutos de PNE 1 vez a la semana; y el grupo 4 recibió

un programa de educación biomédica durante 45 minutos en un total de 2 sesiones. Los investigadores observaron diferencias significativas en todos los grupos en relación a las puntuaciones del dolor con un tamaño del efecto largo ($\eta 2p = 0.170$) a los 3 meses de seguimiento a favor del grupo que recibió un mayor tiempo y número de sesiones de PNE (Amer-Cuenca et al., 2020).

Por otro lado, hay grupos de investigación que en la actualidad están aplicando diferentes modalidades de tratamiento con agentes físicos a través de técnicas de electroterapia como la estimulación eléctrica nerviosa transcutánea, el láser de baja intensidad, los ultrasonidos o las ondas electromagnéticas (Honda et al., 2018). Desde un punto de vista terapéutico, el empleo de la estimulación eléctrica a través de señales electromagnéticas dieléctricas con radiofrecuencia genera importantes efectos fisiológicos en el organismo debido a su elevada capacidad vasodilatadora y calorífica a nivel molecular sobre los tejidos orgánicos (Rodríguez-Martín, 2014). La MDR presenta un rango de frecuencia situado entre los 600 y los 930 kHz con señales eléctricas transmitidas de forma pulsátil y modulada manualmente de forma continua para reducir el impacto térmico y evitar la adaptación de los receptores cutáneos, además de focalizar la energía en la profundidad de los tejidos sin dañar las estructuras suprayacentes (Albornoz-Cabello et al., 2016).

La aplicación de MDR origina los siguientes efectos fisiológicos en nuestro organismo:

a. El paso de la onda electromagnética origina una energía cinética que conlleva a una vasodilatación, a una elevación de la temperatura y a un aumento local del flujo sanguíneo, permitiendo la eliminación de sustancias nocivas (Albornoz-Cabello et al., 2016; Rodríguez-Martín, 2014).

b. Inhibición de las fibras nerviosas aferentes periféricas que están involucradas en la percepción del dolor, y que según la teoría del “Control Gate”, daría lugar a una inhibición del dolor puesto que el estímulo sensorial eléctrico proporcionado por la MDR impide que los estímulos nociceptivos progresen hacia los centros superiores del sistema nervioso (Albornoz-Cabello et al., 2016; Rodríguez-Martín, 2014).

c. Se produce una mayor permeabilidad de la membrana celular que ayuda a la eliminación de la inflamación que compromete a las raíces nerviosas (Albornoz-Cabello et al., 2016; Rodríguez-Martín, 2014).

d. Liberación de opioides endógenos al torrente sanguíneo con función sobre las vías descendentes del sistema nervioso, lo cual bloquea la segregación de neuropéptidos como la substancia P con importantes efectos pro-nociceptivos en el organismo (Albornoz-Cabello et al., 2016; Rodríguez-Martín, 2014).

Los estudios científicos observados en la literatura científica a nivel de MDR en población con dolor crónico son escasos pero con resultados esperanzadores (Hochsprung et al., 2018; Ibáñez-Vera et al., 2020). En este sentido, en estudio piloto en 24 pacientes con esclerósis múltiple donde se aplicó MDR con una frecuencia de 800 a 900 kHz durante 20 minutos, 5 días a la semana a lo largo de 3 semanas para valorar el impacto del dolor en diferentes áreas, se observaron resultados positivos en las puntuaciones medias y máximas del dolor según el cuestionario del dolor Brief Pain Inventory, en la afectación del dolor en el ámbito laboral, en la afectación del dolor en el ámbito personal, y en la calidad del sueño y el descanso nocturno (Hochsprung et al., 2018). En población con FM, y bajo nuestro conocimiento, un único estudio reciente ha evaluado el empleo de la MDR con resultados prometedores en el alivio del dolor en sujetos diagnosticados con FM (Ibáñez-Vera et al., 2020). Este ensayo controlado aleatorizado a simple ciego en 66 pacientes con FM estaba configurado en 3 grupos: grupo experimental (8 sesiones de 20 minutos de MDR con una frecuencia de 870 kHz, intensidad de 30 amperios y una emisión pulsada al 50% sobre el músculo trapecio siguiendo los criterios del ACR), grupo de simulación (igual dispositivo de radiofrecuencia pero sin la activación del dispositivo emisor de corriente), y grupo control (recibieron tratamientos habituales, programa de mantenimiento de actividad física y continuación de su plan de tratamiento farmacológico). Los datos del estudio indicaron diferencias significativas en el dolor local ($p=0.025$) y en el índice de impacto de la FM ($p=0.031$), aunque con efecto a corto plazo (Ibáñez-Vera et al., 2020). Por tanto, estos resultados alentadores obtenidos en pacientes con FM abren una línea investigativa para evaluar la influencia real a largo plazo del tratamiento con MDR.

1.5.2.3.2 Nuevas herramientas para la evaluación del dolor y la reactividad vascular en Fisioterapia.

En la disciplina de la Fisioterapia, el síndrome de FM se percibe como una enfermedad problemática debido a que no se encuentra ajustada a un modelo biomédico clásico, es decir, no existe una etiología específica, la patología no presenta unos marcadores claros y el diagnóstico se produce por descarte de otras afecciones (Roitenberg & Shoshana, 2019). Sin embargo, los fisioterapeutas disponen de una serie de herramientas de evaluación del dolor y sintomatología que han demostrado satisfactorias propiedades psicométricas en pacientes con FM (Roitenberg & Shoshana, 2019). Entre ellas destacan: 1) la EVA es uno de los instrumentos más importantes en la evaluación del dolor en pacientes con FM presentando una sensibilidad y especificidad del 80% (Marques et al., 2008); 2) la algometría de presión es un instrumento muy utilizado para valorar la sensibilidad al dolor con una gran fiabilidad y consistencia interna (α=0.93) (Chesterton et al., 2007); 3) el FIQ-R indica información del deterioro físico, la capacidad para realizar un trabajo o tarea, la fatiga, la rigidez y el dolor en pacientes con FM demostrando una alta confiabilidad con un coeficiente de correlación intraclase de 0.82 (Salgueiro et al., 2013); 4) el CSI evalúa síntomas característicos de este fenómeno mostrando también una alta fiabilidad de 0.91 (Cuesta-Vargas et al., 2016); 5) el PSQI es una herramienta que evalúa la eficiencia y eficacia del sueño en pacientes con FM presentando una alta fiabilidad con un coeficiente de correlación de 0.80 (Hita-Contreras et al., 2014); 6) el MFI, que evalúa la fatiga, es otro de los instrumentos que ha demostrado tener buenas propiedades psicométricas en pacientes con FM (coeficiente de correlación intraclase entre 0.65 a 0.91) (Munguía-Izquierdo et al., 2012); y 7) el BAI valora síntomas de salud psicológica y ansiedad y presenta también una buena confiabilidad con una alta consistencia interna de 0.93 (Magán et al., 2008).

Por otro lado, para valorar la reactividad vascular la termografía infrarroja (TI) es un método óptimo que proporciona una adecuada relación entre los cambios del flujo sanguíneo a nivel periférico y las propiedades térmicas del tejido de la piel (Fujimasa et al., 2000.; Sagaidachnyi et al., 2014). La TI es una herramienta que se basa en la teoría de la transferencia del calor. La teoría propone que un elevado calor en una región del cuerpo se relaciona con una mayor afluencia sanguínea y que, por el contrario, las regiones frías del cuerpo se relacionan con una menor afluencia de flujo sanguíneo (Sagaidachnyi et al.,

2014). Además, la TI es un método no invasivo que presenta una gran sensibilidad (90%) y una gran especificidad (86%) (Mirbod & Sugiura, 2017).

1.6 Justificación de la tesis.

Las pacientes con síndrome de FM presentan un complejo conjunto de síntomas caracterizado fundamentalmente por la presencia de dolor crónico músculo-esquelético generalizado. La evidencia científica disponible sugiere la existencia de diversas hipótesis que intentan dar explicación a la etiopatogenia de las manifestaciones clínicas de la enfermedad, concretamente al origen del dolor crónico característico del síndrome de FM. Numerosos estudios señalan que la presencia de factores vasculares, factores inflamatorios y biomarcadores nociceptivos a nivel de suero sanguíneo y factores dietéticos podrían ser determinantes ya que todos ellos participan en la modulación de la percepción dolorosa y en los mecanismos del procesamiento del dolor a nivel del SNC. Hasta la fecha no existe evidencia científica que haya investigado las alteraciones neurovasculares a nivel periférico en la piel glabra del dorso y palma de las manos de los sujetos con FM, así como la temperatura corporal central y su posible relación con las anomalías de la microvasculatura cutánea y los procesos de termorregulación corporal global que pudieran explicar el dolor crónico y las diferentes manifestaciones clínicas propias de esta patología. Además, el papel vasodilatador que ejerce el ON sobre los capilares periféricos y la alta presencia metabólica de encefalinasas en suero sanguíneo puede influir en los mecanismos de nocicepción e hiperalgesia, explicando síntomas como el dolor, ansiedad, depresión y estrés. Finalmente, dado que las citocinas inflamatorias pueden estar involucradas en el mecanismo subyacente al síndrome de FM, una dieta pro-inflamatoria se asociaría con la hipersensibilidad al dolor y otros síntomas asociados de dicha enfermedad. Un patrón dietético saludable podría ejercer una importante función en la modulación del dolor y, por tanto, podría considerarse como estrategia para controlar los síntomas en la práctica clínica. No obstante, la evidencia disponible en relación a la asociación entre la ingesta dietética y modificaciones en la nocicepción debido a una disminución del estado inflamatorio es muy limitada.

La hipótesis inicial para el primer estudio de la tesis doctoral fue que los pacientes con síndrome de FM podrían presentar anomalías en la respuesta vascular periférica a nivel del dorso y la palma de las manos caracterizada por una vasodilatación ante una excesiva inervación sensorial peptidérgica de las anastomosis arteriovenosas y por la dilatación

pasiva de las arteriolas debido a la liberación de compuestos como el óxido nítrico de las células endoteliales que, influiría a su vez, sobre la temperatura corporal global de los pacientes con FM.

La hipótesis inicial para el segundo estudio de la tesis doctoral fue que en base a la presencia en suero sanguíneo de biomarcadores y neurotransmisores como el óxido nítrico y las encefalinas y su rol en la participación en los procesos de modulación del dolor, planteamos la hipótesis de que la presencia de óxido nítrico circulante así como la actividad en suero sanguíneo de las encefalinasas podrían estar asociados con las variables del dolor y sintomatología de pacientes diagnosticados con FM.

La hipótesis de partida para el tercer estudio de la tesis doctoral fue que dado que las citocinas inflamatorias pueden estar involucradas en el mecanismo subyacente al síndrome de FM, planteamos la hipótesis de que una dieta pro-inflamatoria se asociaría con la hipersensibilidad al dolor y otros síntomas asociados con la FM.

1. INTRODUCTION

1.1 Fibromyalgia syndrome: concept, prevalence, etiopathogenesis, classification, clinical manifestations, and socioeconomic cost.

1.1.1. Concept

Fibromyalgia (hereinafter also FM for brevity) is a complex syndrome, of heterogeneous and unknown aetiology, characterised by chronic widespread and diffuse musculoskeletal pain, and accompanied by a wide range of somatic and psychological symptoms such as hyperalgesia, allodynia, persistent fatigue, joint stiffness, headache, migraine, digestive problems, cold intolerance, unrefreshing sleep, anxiety, depression, memory problems and concentration difficulties (Bellato et al., 2012; Giacomelli et al., 2013; Häuser et al., 2015).

To understand how the concept and description of FM has evolved over its 110 years of history, we must explore chronologically the most relevant findings and discoveries relating to pain and musculoskeletal alterations, which have been recorded in the European scientific literature since the end of the 16th century (Wolfe & Walitt, 2013). In 1592, the French physician Guillaume de Baillou was the first to use the term rheumatism to describe the set of clinical manifestations of acute rheumatic fever and muscle pain. Likewise, and unlike joint rheumatism, he introduced the concept of muscular rheumatism, which encompassed the set of painful musculoskeletal disorders without deformity in the soft tissues (Inanici & Yunus, 2004). In the early 19th century, Scottish surgeon William Balfour first discovered the presence of nodules and reported local tenderness in the tissue from tender anatomical points, indicating that nodules and muscle pain are caused by inflammation originating from muscle connective tissue (Inanici & Yunus, 2004). A few years later, in 1841, the French paediatrician François Louis Valleix coined the concept of "trigger point", as he discovered that manual palpation of several painful points in various parts of the body caused pain referred to other regions of the human body and that, moreover, they were related to the itinerary and/or path of different nerves. All of this led to considering muscular rheumatism as a form of neuralgia (Inanici & Yunus, 2004). At the end of the 19th century, the American neurologist George Miller Beard defined the term neurasthenia as all generalised pain accompanied by physical fatigue and psychological and mental disorders, attributing the appearance of this set of symptoms to the daily stress endured by people leading modern lives. (Inanici & Yunus,

2004; Wolfe & Walitt, 2013). However, this concept of neurasthenia was abandoned in the 1930s due to its recognition as a psychological and psychiatric disorder, disappearing as a branch of study in neurology (Wolfe & Walitt, 2013).

In 1900, the term fibrositis emerged in scientific literature to describe all types of local or regional pain, thus encompassing the FM disease known to date (Wolfe & Walitt, 2013). In 1904, the British neurologist Sir William Gowers first mentioned this concept in a research article related to low-back pain, stating that both low-back pain and muscular rheumatism should be considered a form of inflammation of the fibres of muscle tissues and that, continuing with the "cellulite" analogy, this inflammation of muscle fibrous tissues should be called "fibrositis". Furthermore, Gowers indicated in his article a series of symptoms related to fibrositis such as spontaneous pain, sensitivity to mechanical compression, fatigue and sleep disorders, as well as their aggravation by factors such as exposure to cold or muscle overexertion (Inanici & Yunus, 2004). Therefore, the general bases of the term fibrositis had been established. However, it was not until the middle of the 20th century that a growing interest in its scientific study began (Wolfe & Walitt, 2013). In this context, Richard Harold Freyberg, the main contributor and leader in the development of rheumatology as a discipline, carried out an in-depth review of the fibrositis concept in 1951. In his contribution, he established a division of the same into regional fibrositis (currently known as myofascial pain syndrome) and generalised fibrositis (currently known as FM), also warning of the difficult differentiation with the term psychogenic rheumatism characterised by muscle involvement in the absence of inflammation, but associated with depression and/or stress problems (Freyberg, 1951). Moreover, and in line with the previously described publications, Richard Freyberg reported that, on the one hand, factors such as humidity, cold and rain could aggravate the symptoms of fibrositis and that, on the other hand, factors such as heat or a high barometric pressure could alleviate them (Freyberg, 1951; Wolfe & Walitt, 2013). Later, in 1953, Wallace Graham rekindled interest in fibrositis and other common conditions such as FM disease by writing a chapter in the well-known book " *Arthritis and Related Conditions* (Graham, 1949)." In this contribution, he pointed out the existence of acute, subacute, and chronic painful conditions accompanied by local tender points and referred pain, regardless of the disease that could originate them, and which involved muscles, subcutaneous tissues, ligamentous tissues, tendon tissues and fascial structures. The causes of these painful conditions were of an infectious, traumatic, environmental and psychological

nature, and the referred pain had irritating characteristics described as a burning, stabbing, insistent and unbearable feeling (Graham, 1949). At the end of 1968, the researcher Eugene Traut from the University of Illinois (Chicago, United States) incorporated a wide constellation of new symptoms such as headache, colitis, fear, worry and anxiety into the clinical characteristics of fibrositis. Furthermore, in relation to generalised pain, he stated that muscular rheumatism has its origin in several sections of the spinal axis, considering axial pain as an important criterion in the current diagnosis of FM(Traut, 1968; Wolfe et al., 1990). Traut's important scientific contribution revealed a number of important systemic features of FM syndrome (fatigue, sleepiness, headache, colitis, etc.), thus offering the first near-modern description of FM(Inanici & Yunus, 2004; Traut, 1968).

It was in the 1970s that the first modern description of FM syndrome was provided thanks to physician and scientist Hugh Smythe. (Inanici & Yunus, 2004) In an elaborate 10-page chapter on fibrositis syndrome in the book *"Arthritis and Related Conditions* ," he described FM as a syndrome characterised by generalised pain accompanied by fatigue, morning stiffness, lack of sleep, a high count of tender points upon anatomical palpation (11/14; 79%), and emotional stress (Inanici & Yunus, 2004; Smythe, 1972). Likewise, he emphasised the important role of sleep disturbances in FM patients through the EEG findings reported by his colleague and scientist Harvey Moldofsky (Moldofsky et al., 1975), specifying, in turn, a pathophysiological framework for FM syndrome by describing a deep reflex hyperalgesia condition along with referred pain patterns, differentiating them from cutaneous hyperalgesia (Smythe, 1972). These findings were considered a triumph in family clinical medicine, both authors being known colloquially as the "grandparents" of FM (Inanici & Yunus, 2004). However, and in the absence of controlled studies in the field of FM, the concept and description of this disease remained doubtful. Thus, Yunus et al. (1981) they carried out the first controlled trial of clinical manifestations of FM in 50 FM patients, comparing them with 50 healthy subjects matched by age and sex. The trial confirmed the previous characteristics already recorded in prior studies in the FM population such as generalised pain, greater number of tender points, fatigue and sleep problems compared to healthy subjects. Furthermore, a set of new symptoms were described for the first time in these patients, including tension-type headaches, migraines, paresthesias, and subjective swelling, as well as the association with other functional syndromes such as irritable bowel syndrome, or restless legs syndrome(M. Yunus et al., 1981). Therefore, the documented record of these multiple symptoms raised the conception

of FM to the level of a syndrome, which is currently accepted by scientific literature (Inanici & Yunus, 2004).

Finally, in 1990 and under the criteria recommended by the American College of Rheumatology (ACR), the existing association of FM with fibrositis syndrome was definitively abandoned, to provide an official definition of FM syndrome (Wolfe & Walitt, 2013). The ACR published the first criteria for the classification and diagnosis of FM consisting of: a) generalised pain in the right and left side of the upper human body lasting for over 3 months; and b) presence of pain on digital palpation of 11 or more tender points in 18 specific points (Wolfe et al., 1990). Subsequently, these criteria have been updated over the years (2010, 2011, 2016, 2019), thus improving the knowledge and description of FM syndrome (Arnold et al., 2019; Wolfe et al., 2010, 2016).

1.1.2. Prevalence

Numerous studies in scientific literature have attempted to establish the prevalence of FM syndrome in different populations worldwide. The estimated prevalence in the general population differs between studies, which expresses the difficulty in determining the exact prevalence of this syndrome (Branco et al., 2010; Cabo-Meseguer et al., 2017; Heidari et al., 2017; Queiroz, 2013). This variation in prevalence figures can be explained by the heterogeneity observed in scientific literature of different epidemiological studies in relation to the design, recruitment criteria, patient cohorts, and their sample size (Fitzcharles et al., 2018).

In general terms, scientific literature indicates that between 0.4% and 9.3% of the general world population suffers from FM, setting the average prevalence rate at an approximate percentage of 2.7% (Queiroz, 2013). In a recent systematic review and meta-analysis on the study of FM prevalence in the general population and based on the different regions established by the World Health Organisation (WHO), the prevalence of the disease stands at 2.64% in the Europe region, 2.41% in the Americas region, 4.43% in the Eastern Mediterranean region and 1.62% in the Western Pacific region (Heidari et al., 2017). Epidemiological studies on the prevalence of FM in the European continent show its variations between different countries. For example, the prevalence observed in France stands at 2.2%, in Portugal at 3.7%, in Germany at 5.8%, and in Italy by 6.6% (Cabo-Meseguer et al., 2017). In the Spanish population, the average prevalence rate is estimated

at 2.4%, with a greater incidence in women (4.2%) than in men (0.2%) in a proportion of 21 to 1 (Cabo-Meseguer et al., 2017). Likewise, and in line with this previous review, a recent study conducted by Gayà et al. (2020) sets the prevalence rate of FM in the general Spanish population at 2.45%, with a higher percentage of affectation in women (4.49%) than in men (0.29%) (Gayà et al., 2020). In addition, medical consultations regarding FM in Spain at non-specialised health care services reflect a percentage between 2.1% and 5.7% of total consultations, compared to consultations registered at specialised rheumatology care, which represent between 10% and 20% of the total (Cabo-Meseguer et al., 2017).

Age is a determining factor to bear in mind in the development of FM syndrome. The age range between 30 and 50 years of age is the most frequent range of manifestation of the FM syndrome, although it can also appear after 50 years of age (Queiroz, 2013). On the European continent, the prevalence of FM is low in young adults, increasing, on the contrary, in the age range between 35 and 44 years of age and up to 74 and 85 years of age (Branco et al., 2010). In the Spanish population, FM usually manifests between 40 and 49 years of age, reaching its maximum prevalence around 60 and 69 years of age (Cabo-Meseguer et al., 2017; Gayà et al., 2020).

Gender is another of the most relevant issues to consider when assessing FM prevalence. FM occurs in a higher proportion in women - with an average general prevalence worldwide at 4.2% - compared to the male gender, whose prevalence is estimated at 1.4%. Accordingly, worldwide, women are more susceptible to FM than men by a 3 to 1 ratio(Queiroz, 2013). The prevalence that women present in manifesting the FM syndrome stands at 5.2% at a European level, with a general ratio of women to men under 2(Branco et al., 2010). Studies carried out in the United States indicate a much higher prevalence of FM in women (3.28%) than in men (1.06%) (Walitt et al., 2015). Furthermore, the ratio of women to men with FM differs between continents. South America is the continent with the highest ratio of women to men with FM at 12 to 1, followed by the Asian continent with a ratio of 5 to 1, the American continent with a ratio of 4 to 1, and by the European continent with a ratio of 3 to 1 (Cabo-Meseguer et al., 2017). In Spain, the prevalence of FM for females is 15.8% and 2.2% for males (Cabo-Meseguer et al., 2017). Furthermore, the female gender in the Spanish population is associated in a very significant way with the development of the FM syndrome with an

Odd Ratio (OR) of 10,156, and a 95% Confidence Interval (CI) of 5,066 to 20,352 (Gayà et al., 2020).

Among the main FM risk factors we find a series of comorbidities such as obesity, diabetes mellitus, irritable bowel syndrome, painful bladder syndrome, interstitial cystitis, celiac diseases, alterations of the temporo-mandibular joint, headache, and migraine (Fitzcharles et al., 2018; Heidari et al., 2017). In this regard, obesity has been identified as an important risk factor for FM development with a prevalence rate of 45% (Dias et al., 2017), observing a strong association with this syndrome in Spain, with an OR of 1,689 and a CI at 95% from 1,036 to 2,755 (Gayà et al., 2020). In relation to diabetes mellitus, the total prevalence of the FM syndrome is estimated to be around 14.80% with a 95% CI of 11.10 to 18.40 (Heidari et al., 2017). With respect to irritable bowel syndrome, the overall prevalence rate of FM in these patients has been estimated at 12.90% with a 95% CI of 12.70 to 13.10 (Heidari et al., 2017). Headaches, migraine and tension headaches have also shown high prevalence rates in FM patients with a percentage of 36.4%, 28.5% and 59%, respectively (M. De Tommaso et al., 2009; Marina De Tommaso et al., 2011). Finally, in other types of chronic pain conditions such as alterations in the temporo-mandibular joint, chronic low-back pain, interstitial cystitis, painful bladder syndrome, endometriosis and encephalomyelitis, high FM prevalence rates have been reported, ranging between 20 % and 65% (Fitzcharles et al., 2018).

As regards socioeconomic, sociocultural and educational factors, there is a broad consensus in the scientific community about the development of FM syndrome in people with low economic, sociocultural, and educational levels(Branco et al., 2010; Cabo-Meseguer et al., 2017; Queiroz, 2013).

Lastly, it should be noted that there seems to be discrepancies in scientific literature with respect to FM prevalence rates and their association with the person's marital status or residence in urban centres or rural areas. While certain studies report that divorce or living in rural areas are significantly associated with the development of FM(Branco et al., 2010; Cabo-Meseguer et al., 2017), other studies show high rates of FM prevalence regardless of the person's marital status (married, widowed, divorced), or whether they reside in the urban centres of large cities (Gayà et al., 2020; Queiroz, 2013).

1.1.3 Aetiopathogenesis

Despite the fact that more than 110 years have passed since William Gowers described the first characteristics of the FM syndrome through the concept of "fibrositis", its pathophysiology and underlying mechanisms remain unknown(Chinn et al., 2016; Coskun Benlidayi, 2019; Häuser et al., 2015). Central Nervous System (CNS) alterations have been considered the main key factor in the development of FM syndrome (Chinn et al., 2016; Russell & Larson, 2009). However, the current scientific literature proposes a series of factors that play an important role in the pathophysiology of this chronic condition such as vascular alterations, neurogenic dysfunctions, inflammatory factors, infectious factors, nutritional factors, and genetic factors, among others (Coskun Benlidayi, 2019; Häuser et al., 2015; Peck et al., 2020; Russell & Larson, 2009).

The main pathophysiological findings are described below, according to the affected structural or functional component:

1. **Central pain sensitisation.** Central sensitisation (CS) is defined as a physiological phenomenon produced by processing changes in the CNS that cause alterations in neuronal function, thus producing a state of hyperexcitability (hypersensitivity) to harmful and non-harmful stimuli (Neblett et al., 2017; Nijs et al., 2017). Clinically, CS manifests in the presence of allodynia (painful sensation when faced with a non-painful stimulus, such as touch), hyperalgesia (excessive sensitivity towards a stimulus that is painful under normal conditions, such as, for example, pressure), pain field expansion (pain that expands beyond the peripheral nerve area), and prolonged pain over time after a particular pain stimulus (mainly throbbing, burning, tingling, or numbness) has ceased(Nijs et al., 2010). The key element of the theory that explains the CS phenomenon resides in the fact that the sustained pain impulses will cause an alteration in ascending and descending pathways of the CNS, so that persistent pain can cause changes in the Central (CNS) and Peripheral Nervous System (PNS) (Kindler et al., 2011; Nijs et al., 2010).

   Scientific investigations have reported a decompensation in the two main pathways of CNS nociception in the FM population(Chinn et al., 2016; Russell & Larson, 2009). Given the prolonged state of pain so characteristic of patients with FM, nociceptive receptors of the peripheral tissue are overstimulated, constantly sending pain signals to the A-delta nerve fibres and C nerve fibres, which are responsible for

transmitting nociceptive impulses to the dorsal horn neurons of the spinal cord. Continued activation of these nociceptive nerve fibres produces the release of neurotransmitters and neuropeptides such as substance P, glutamate, calcitonin gene-related peptide (CGRP), nerve growth factor, and aspartate, which modulate post-synaptic electrical discharges in the dorsal horn of the spinal cord. Increased neurotransmitters and neuropeptides trigger the corresponding postsynaptic responses causing hyperstimulation of the N-methyl-D-aspartate (NMDA) receptors of second-order neurons in the dorsal horn of the spinal cord. The activation of NMDA receptors markedly disturbs the normal role that postsynaptic neurons must play, producing changes in cell membranes, allowing calcium influx, and activating protein kinase. These neurochemicals sensitise neurons with a wide dynamic range, which become hyperexcitable, leading to phenomena previously described as allodynia and hyperalgesia (Nijs & Van Houdenhove, 2009; M. B. Yunus, 2007). As a result of this process, second-order neurons are activated to stimulate a series of cerebral cortex areas and structures (thalamus, hypothalamus, pre-frontal cortex, anterior cingulate cortex, insular cortex and somatosensory cortex), which play a major role in pain processing in all its possible dimensions (sensory, affective and evaluative) (Nijs & Van Houdenhove, 2009; M. B. Yunus, 2007). This mechanism, which has been observed in the FM population, is defined as "pro-nociception" (Bellato et al., 2012; Chinn et al., 2016; Russell & Larson, 2009).

Furthermore, the CS phenomenon in FM patients does not only involve the nociceptive nerve fibres responsible for transmitting painful information to the dorsal horn of the spinal cord, but also involves and disturbs the descending inhibitory mechanisms of pain. The limitation of the activity of descending pain pathways (reticular system, brainstem, and hypothalamus) leads to insufficient release of neurotransmitters such as serotonin, norepinephrine, enkephalins and g-aminobutyric acid (GABA), which disturb the pain response modulation to the periphery. This mechanism, which has been observed in the FM population, is defined as "pro-nociception" .(Bellato et al., 2012; Chinn et al., 2016; Nijs et al., 2011; Russell & Larson, 2009; M. B. Yunus, 2007)

Finally, scientific studies have reported, both in different populations with chronic pain and in populations with FM, that the characteristic CS phenomenon in patients with this condition has increased their response capacity to a wide range of stimuli such as cold, heat, light , sound, electrical stimuli, chemicals, anxiety, panic,

depression, and emotions (Häuser et al., 2015; Nijs et al., 2011; Sluka & Clauw, 2016). Therefore, the presence of these factors and their interconnection with the physiological processes previously described indicate the need to keep the biopsychosocial model of pain in mind at all times (Nijs & Van Houdenhove, 2009).

2. **Vascular factors.** Other findings reported in patients with FM are the presence of morphological abnormalities in the diameter and density of small blood vessels (capillaries) and the presence of blood microcirculation disorders. Patients with FM syndrome present vasospastic symptoms similar to those that occurred in Raynaud's Phenomenon, increasing the sympathetic tone of blood vessels and stimulating the vasoconstrictor system (Morf et al., 2005). Continued activation of the alpha-2-adrenergic vasoconstrictor systems, endothelin-1, tyrosine kinase, serotonin, and angiotensin II produce endothelial tissue dysfunction, accompanied by a decrease in capillary density and microcirculatory blood flow. In turn, this reduces the activity of the primary muscular blood vessels and the nutritional and oxygen supply to the tissues (Choi & Kim, 2015; Morf et al., 2005).

3. **Neurogenic and neurovascular factors.** Neurogenic disorders may also lead to FM syndrome. The Autonomic Nervous System (ANS) plays an essential role in controlling the core body temperature. In this context, scientific studies show dysfunctions of the sympathetic and parasympathetic nervous systems of the ANS, of the afferent sensory nerve fibres in the area of neurovascular connection and of the arteriovenous anastomoses (AVAs) of patients with FM (Albrecht et al., 2013). Specifically, the affected AVAs could generate an alteration of the sensory innervation of the small-calibre blood vessels of the skin tissue, causing changes in blood flow and ischemia in the musculoskeletal tissue (Albrecht et al., 2013).

4. **Immunological and inflammatory factors.** Another hypothesis underlying the FM pathology is the immune system function and its interaction with inflammatory processes (Peck et al., 2020; Sluka & Clauw, 2016). Immune system cells of FM patients are highly plastic, which could alter the concentration levels of pro-inflammatory and anti-inflammatory cytokines at the systemic level or locally at the tissue level. The release of pro-inflammatory cytokines into the bloodstream, such as interleukin IL-1β, interleukin IL-6, interleukin IL-8 or tumour necrosis factor alpha,

would affect neural networks during the interaction of the nervous system with immune system cells, which would stimulate nociceptors and lead to an increase in SC, peripheral sensitisation and neuroinflammation (Peck et al., 2020; Sluka & Clauw, 2016)

5. **Genetic factors.** Finally, different genetic variants may also underlie the development of FM (Häuser et al., 2015; Peck et al., 2020). Scientific literature has amply documented with multiple studies that the inheritance of pain-related genes and family predisposition and aggregation contribute up to 50% in the development of conditions associated with chronic pain such as FM syndrome (Peck et al., 2020). Accordingly, it has been observed that first-degree relatives of patients with FM have an 8.5 times greater risk of developing this disease with a 95% CI of 2.8 to 26 (Clauw, 2014). Likewise, there are numerous genes and gene polymorphisms associated with pain hypersensitivity (for example, GRM6, GABRB3, SNPs, among others) due to changes produced in the expression and function of multiple proteins responsible for pain modulation (Peck et al., 2020). In addition, there are genes associated with the development of fatigue (polymorphism of the IL-6 gene rs1800795), joint stiffness (MTHFR gene), with sleep (polymorphism of the DAT1 gene), anxiety (polymorphism of the T102C gene) or with cognitive disorders (MYT1L gene) (D'Agnelli et al., 2019; Peck et al., 2020).

In summary, FM is a syndrome of complex and uncertain aetiology, although the CNS involvement constitutes a key element in the clinical manifestations of these patients. However, the influence of various vascular, neurogenic, immunological, inflammatory and genetic factors is currently generating great interest in the field of FM research, in an attempt to elucidating which of them constitutes the real cause of this condition(Häuser et al., 2015).

1.1.4 Classification

Generalised pain throughout the body is a condition very present in people diagnosed with FM, and it is also considered one of the most important criteria for its classification, both for patients with primary FM syndrome and secondary FM syndrome (Wolfe et al., 1990). Chronic pain is defined as persistent or recurrent pain lasting longer than 3 months, so that it loses the physiological warning function of acute

nociception.(Treede et al., 2019) People suffering from chronic pain experience anguish, pain and disability, resulting in reduced quality of life and life expectancy (Gary J. Macfarlane et al., 2017). In addition, it is one of the most frequent public health problems worldwide, requiring a major medical care effort. (Goldberg & McGee, 2011; Gary J. Macfarlane et al., 2017)

In collaboration with the WHO, the International Association for the Study of Pain (IASP) working group developed in 2019 a new classification system for chronic pain that included new taxonomic concepts related to the main diseases connected to this condition, and that have also been incorporated into the International Classification of Diseases (ICD-11) (Treede et al., 2019). Under the ICD-11, chronic pain has been assigned a specific code (code G89.4), given that it has been valued as a pathological entity in itself (Treede et al., 2019). The IASP working group, on the one hand, defined chronic primary musculoskeletal pain as recurrent pain in muscles, tendons, bones, and joints characterised by emotional distress and functional disability, and not directly attributable to a previous disease. On the other hand, it defined chronic secondary musculoskeletal pain as any persistent pain in the musculoskeletal system derived from an underlying disease such as, for example, chronic pain related to cancer processes, chronic post-traumatic pain, chronic post-surgical pain, chronic neuropathic pain, or chronic visceral (Perrot et al., 2019; Treede et al., 2019)pain. Under the established classification, FM is considered a chronic secondary musculoskeletal pain due to the presence of numerous neuropathic and rheumatic pathologies and autoimmune disorders, thus excluding the approach whereby generalised chronic pain is deemed to be the only defining factor of this syndrome (Häuser & Fitzcharles, 2018). Likewise, the diagnostic code for FM syndrome under the ICD-11 chronic pain classification is M79.7 (Perrot et al., 2019).

In summary, FM syndrome is classified into two forms: primary FM syndrome and secondary FM (Häuser et al., 2015)syndrome:

1. **Primary FM syndrome** is defined as the population who, lacking an identifiable continuous nociceptive input, experience a painful process such as damage or inflammation. In turn, these patients may develop local painful conditions such as headaches, temporomandibular disorders, dysmenorrhea, and interstitial cystitis, as well as psychological symptoms including anxiety and depression (Clauw, 2014).

2. **Secondary FM syndrome** is defined as the population who, lacking an identifiable continuous nociceptive input, experience a centralised pain process. That is, these patients present pain concomitant to a secondary disease such as rheumatic pathology (osteoarthritis, rheumatoid arthritis, lupus), autoimmune disorders and sickle cell anaemia (Clauw, 2014; Häuser et al., 2015).

1.1.5 Clinical manifestations

In relation to the clinical manifestations of FM syndrome, the most characteristic symptom is a generalised and diffuse chronic pain throughout the body, focused on the axial region, right upper body region, left upper body region, right lower body region and left lower body region, although, sometimes, the pain can start in specific places such as the lower back or cervical area(Bair & Krebs, 2020; Wolfe et al., 1990). Generally, the chronic generalised musculoskeletal pain that people with FM syndrome experience is usually accompanied by neuropathic symptoms such as dysesthesia, paraesthesia, tingling and numbness in the upper and lower extremities and a burning or stinging sensation in one or more joints (Clauw, 2014). Moreover, symptoms related to centralised pain conditions appear very frequently including allodynia (pain when faced with a normally harmless stimulus) or hyperalgesia (excessive sensitivity when faced with a normally painful stimulus) and comorbid symptoms also arising from the CNS such as fatigue, which is often reported as severe, and sleep disturbances (Clauw, 2014).

Somatic symptoms are also frequently reported by FM patients and included in current diagnostic frameworks for this disease. Among them we may highlight headaches, abdominal pain, abdominal distensions, pain in the temporo-mandibular joint, nausea, dizziness and diarrhoea (Bair & Krebs, 2020; Wolfe et al., 2016).

Other symptoms that also present clinically with the FM syndrome are those of a cognitive and psychological nature. Among this variety of symptoms, FM patients usually report memory impairment, difficulty in attention, difficulty in focusing on a specific goal or concentrating on performing a task or activity of daily living (informally known as "fibro fog"), altered moods, anxiety and depression (Bair & Krebs, 2020; Clauw, 2014).

Finally, in clinical practice, symptoms associated with sensory hyperresponsiveness may appear such as sensitivity to bright lights, sensitivity to loud noises or sounds, and sensitivity to strong odours (Bair & Krebs, 2020).

1.1.6 Socioeconomic cost.

The set of clinical manifestations previously described in the FM syndrome, together with all disorders associated therewith, expose patients to considerable personal suffering and a reduced ability to perform daily living and work activities, along with significant direct and indirect socioeconomic and health costs (Schaefer et al., 2016).

Regarding costs arising from chronic pain pathologies such as FM syndrome, studies carried out in the United States report that the average annual expenditure per patient diagnosed with FM syndrome ranges from USD 9,575 to USD 18,671, almost 5 times more (approximately USD 3,291) than people suffering from another type of pain syndrome (Berger et al., 2007; Knight et al., 2013). Likewise, it has been observed that the social and health expenditure is increased in patients with more severe FM symptoms. In this context, Chandran et al. (2012) a study conducted in the United States based on the symptom severity of patients diagnosed with FM reported that healthcare costs were USD 10,219 for patients with mild symptoms, USD 26,217 for patients with moderate symptoms, and USD 42,456 for patients with severe symptoms(Chandran et al., 2012).

Scientific literature also records various socioeconomic studies on FM syndrome carried out on the European continent (Knight et al., 2013; Pérez-Aranda et al., 2019; Perrot et al., 2012; Rivera et al., 2009). In France, like in the United States, the association between financial healthcare costs and FM symptom severity was also evaluated. Specifically, Perrot et al. (2012) the report showed an annual expenditure of EUR 6,436 for FM patients with mild symptoms, EUR 7800 for patients with moderate symptoms, and EUR 11,862 for patients with severe symptoms. (Perrot et al., 2012). In Spain, in a multi-centre study conducted by Rivera et al. (2009), where direct and indirect socioeconomic and healthcare costs in FM syndrome were analysed, it was reported that the total annual expenditure per FM patient was EUR 9,982, of which EUR 3,245.8 corresponded to direct costs and EUR 6,736.2 to indirect costs. As regards direct costs, medical consultation costs were EUR 847, diagnostic testing costs were EUR 473.5, pharmacological treatment costs were EUR 439.2, non-pharmacological treatments were EUR 1,368.1, and costs for other

health interventions were EUR 117.9. Likewise, as regards indirect costs, that is, labour costs, costs connected to the workday reduction were EUR 913.1, leave of absence costs were EUR 3,556.2, and the permanent disability pension amounted to EUR 2,266.9(Rivera et al., 2009). Therefore, the socioeconomic burden related to the FM syndrome is a very important factor to be considered. Finally, and in line with these results, another study conducted between 2012 and 2014 in Spain confirmed these high direct (consultations in primary care, specialised care and specific diagnostic tests) and indirect costs (Pérez-Aranda et al., 2019).

In summary, and given the chronic pain nature and the negative effects that FM syndrome produces at a personal or work level, this pathology has a major impact on healthcare operations and its treatment involves a high economic cost(Schaefer et al., 2016).

1.2 Neurovascular disorders and their relationship with Fibromyalgia syndrome symptoms.

Since the end of the 19th century and to this day, the study of the blood flow activity and control aroused great interest in all physiological researchers. Numerous factors such as disorders of the Autonomous Nervous System, the excessive presence of circulating substances in the bloodstream, mechanical stimulation of blood vessels, the myogenic response of arterioles to variations in intraluminal pressure, the concentration of metabolites in the vessels and tissue, and shear forces can influence the optimal perfusion of blood flow to tissues when they demand oxygen (Secomb, 2008). At the microvascular level, the greatest resistance to blood flow occurs between the arterioles originating from a main artery and the drainage venules. The regulation of the microvascular network system is controlled by the activity of a set of hormones but, above all, by the ANS, which regulates vascular tone (Bagher & Segal, 2011). In this regard, several scientific studies have confirmed that FM patients show disorders in the morphology of their capillaries as regards diameter and density, accompanied by less microcirculatory activity (Choi & Kim, 2015; Morf et al., 2005). These anomalies are due to the fact that FM patients present, like patients with Raynaud's Phenomenon, symptoms of persistent peripheral coldness that increase sympathetic vascular tone mediated by the ANS, and which cause vascular malfunction (spasm) (Morf et al., 2005). This sustained vasoconstriction activates the alpha-2-adrenergic receptors of the Sympathetic Nervous System (SNS), which are

powerful vasoconstrictors and interfere, on the one hand, in the dilations and formations of capillaries and, on the other hand, in the vasomotor tone of skeletal muscle tissue, thus disturbing the mechanism that ensures a correct supply of blood and oxygen to said tissue (Carlson et al., 2008; Choi & Kim, 2015).

On the other hand, new scientific findings indicate that this blood microcirculation disorder in FM patients can be explained by changes in the innervation of arteriovenous anastomoses (AVAs) in the skin tissue of the palms of the hands (Albrecht et al., 2013). AVAs are direct connections between small arterioles and venules, whose main function is the transport of heat from the human body's core to superficial areas. They are located in the mucosa and in glabrous skin (without hair) area of the fingers nail bed, nose, ears, feet and, above all, in the deep dermis of the hypothenar regions (100 AVAs/cm2)of the palms of the hands (Walløe, 2016). AVAs, together with cutaneous arterioles, are innervated by small fibres of vasoconstrictor sympathetic innervation and vasodilator sensory innervation, although thermoregulatory activity in this area is governed by the adrenergic vasoconstrictor system. Likewise, they regulate core body temperature, keeping the organism in a suitable thermoneutral zone, which oscillates around 26ºC and 36ºC for the naked human being at rest(Walløe, 2016). The concept of thermoneutral zone was introduced between the years 1940 and 1950 by Scholander et al. (1950) as: “the constant basal metabolism of a mammal at rest when ambient temperature ranges between 37ºC of the core body temperature and a lower temperature dependent on the insulation of the body of the mammal itself.” (Scholander et al., 1950). The metabolic rate increases via sweat production above the upper end of the thermoneutral zone. Below the lower end of the thermoneutral zone, the metabolic rate increases proportionally to the difference between ambient temperature and the core body temperature as a result of the production of heat generated by tremors and active muscle work due to cold (Walløe, 2016). AVAs act as sphincters and open or close respectively producing a decrease or an increase in blood flow to the area, allowing blood to flow straight to the venous plexuses in the extremities. Faced with a drop in temperature (exposure to cold), the hypothalamus, as the main centre for controlling the temperature of the human body, collects this information and sends nerve impulses to all the adrenergic sympathetic axons of the AVA, compromising their light to provide blood flow to the skin tissue to meet the basic metabolic demands upon this stressful event (Daanen, 2003; Walløe, 2016). In relation to AVAs, a study conducted by Albrecht et al. (2013) in women with FM syndrome reported the presence of disorders in

the peripheral vascular response. Through biopsies performed on the skin of the hypothenar eminence of the palms, researchers discovered an excessive peptidergic sensory innervation and a noradrenergic sympathetic underrepresentation of AVAs in female FM patients compared to healthy subjects. Likewise, they found that this peptidergic sensory innervation of AVAs expressed hyperresponsiveness of the alpha-2-adrenergic receptor, which produces a sympathetic release of noradrenaline/norepinephrine that constricts the smooth muscles of the tunica media of blood vessels. We shall recall here that AVAs regulate blood flow in the human body to ensure optimal thermoregulation(Walløe, 2016), so that a nervous system control disorder would lead to a deregulation of the core body temperature and a decrease in peripheral blood microcirculation (due to the closure of arterioles and venules), restricting the musculoskeletal tissue from optimal oxygenation and nutrition, favouring the deposit of metabolites and the accumulation of lactic acid that would explain the deep pain and generalised fatigue characteristic of patients with FM syndrome (Albrecht et al., 2013). Finally, these authors also indicated that this neurovascular disorder in AVAs would not only explain the symptoms in FM patients, but would also explain their exacerbation in the face of temperature changes (Albrecht et al., 2013)

Lastly, the Peripheral Nervous System through disorders of its small-calibre nerve fibres also appears to be involved in the development of the FM syndrome symptoms (Grayston et al., 2019). Currently, there is no concept or standard definition for small-calibre nerve fibre neuropathy; however, neuropathy is suspected when at least two of the following three diagnostic tests show deterioration of small nerve fibres: a) neurological examination test, b) quantitative sensory test, and c) skin biopsy always accompanied by patient's pain and dysesthesia (Devigili et al., 2008). In small-calibre neuropathy of small nerve fibres, there is much more predominant impairment in myelinated peripheral afferent nerve fibres (A-delta) and unmyelinated peripheral afferent nerve fibres (C-fibres). Both afferent fibres are involved in pain perception (allodynia and hyperalgesia), in sensory perception (paraesthesia) and in thermal perception (cold and heat) (Pickering et al., 2020). In addition, they are responsible for innervating the sweat glands that establish a bond with the ANS (Lefaucheur et al., 2015; Oaklander et al., 2013). The prevalence of small nerve fibre neuropathy in the population with FM syndrome is 49%, underlining the negative impact that the SNP has on pain and symptoms in these patients (Grayston et al., 2019). This alteration in the fibres alters the function of small blood vessels through the

production of an altered neuropeptide response and the dysregulation in the activation of α-adrenergic receptors (Grayston et al., 2019). In this way, this neurogenic microvasculopathy could explain, at least partially, the perfusion deficits at the musculoskeletal level, deep pain, exercise intolerance and a common symptom such as "mental fog" or "brain fog" characteristic of fibromyalgia. Along these lines, previous studies using microneurography have shown changes in the conductivity velocity of peripheral C-nerve fibres in subjects with FM and hyper-reactivity of C nociceptors and type 1 mechanosensitive nerve fibres (Evdokimov et al., 2019). On the other hand, and in line with the connection that the SNP presents with the ANS through sweat glands, scientific literature shows dysfunctions in the ANS of patients diagnosed with FM, which can affect sweating functions and regulation of body heat loss, altering the processes of global body thermoregulation (Elmas et al., 2016).

1.3 Pro-inflammatory and nociceptive biomarkers: the role of Nitric Oxide and Catecholamines in the general symptoms of hypersensitivity to pain in patients with Fibromyalgia syndrome.

Inflammation is a defence mechanism of the human body that protects our body and, therefore, is vital for survival. Without it, it would not be possible to react to endogenous and / or exogenous aggression and it would not be possible to provide the necessary mechanisms for tissue repair when this is damaged or injured. (Porth CM, 2011) Correct work between the immune system, the nervous system and coagulation pathways is essential for an adequate inflammatory response, which is modulated by a series of messengers called inflammatory mediators (Porth CM, 2011). Inflammatory mediators can be released into the bloodstream by immune system cells such as macrophages, lymphocytes, mast cells, and leukocytes, or by plasma cells. The most characteristic inflammatory mediators released by immune cells are pro-inflammatory and anti-inflammatory cytokines, nerve growth factor, neuropeptides, oxygen free radicals, arachidonic acid derivatives, and vasoactive amines. What is more, inflammatory mediators held in plasma include coagulation factors, acute phase proteins and complement (Porth CM, 2011)proteins.

Cytokines are small polypeptides that play a very important role in the inflammatory response. They are mainly released into the bloodstream by immune system cells (macrophages, monocytes, and T cells), although they can also be released by cells

not belonging to the immune system (Schwann cells, fibroblasts, microglia, and astrocytes). In turn, they are divided into pro-inflammatory and anti-inflammatory cytokines with immunological and physiological bodily functions (Bjurstrom et al., 2016). Cytokines can act as mediators between glial cells and neurons through central and peripheral pathways. Under normal conditions, the glia has a protective role over the CNS; in pathological conditions, however, the glia is excited and releases pro-inflammatory cytokines into the bloodstream, which sensitize nociceptors and neurons in the tissue nervous system, allowing a greater entry of painful stimuli at the central level directly and indirectly, thus facilitating the CS phenomenon (Graeber, 2010; Watkins et al., 2007).

A wide variety of scientific studies in literature has reported high levels in blood concentrations of pro-inflammatory cytokines in different populations with chronic pain. In patients with knee osteoarthritis, a pro-inflammatory basal state has been observed with an increase in the blood plasma of interleukins IL-1β, IL-6 and IL-8 (Lundborg et al., 2010). In pathologies such as complex regional pain syndrome, a significant increase in the concentrations of pro-inflammatory cytokines IL-1β and IL-6 has been shown (Alexander et al., 2005). Cross-sectional and longitudinal scientific studies in populations with chronic low-back pain and lumbar radiculopathy have indicated high concentrations of interleukins IL-6 in the patients' cerebrospinal fluid (Nagashima et al., 2009; Zin et al., 2010). Finally, in postherpetic neuralgia patients, a baseline pro-inflammatory status has been evidenced due to the high levels of interleukins IL-8 in the bloodstream compared to healthy controls (Kotani et al., 2004).

Very recent studies in the population with FM syndrome have suggested a chronic systemic inflammation underlying the pathology (Coskun Benlidayi, 2019). Accordingly, FM patients show high levels of pro-inflammatory cytokines in the bloodstream, among which interleukin IL-1β, IL-6, IL-8 and tumour necrosis factor alpha stand out (Mendieta et al., 2016; Peck et al., 2020). These concentrations of pro-inflammatory cytokines favour the development of symptoms such as generalised pain, fatigue, hyperalgesia, allodynia, and loss of sleep due to the sensitisation they induce on nociceptors (Andrés-Rodríguez et al., 2020; Rodriguez-Pintó et al., 2014; Üçeyler et al., 2011). Therefore, and given that pro-inflammatory cytokines can contribute to the initiation and / or progression of the different clinical manifestations, the regulation of concentration levels in blood plasma has been

considered as one of the main objectives in the treatment of FM syndrome (Coskun Benlidayi, 2019).

The study of the management of chronic pain in all its dimensions has generated great scientific advances in its pathophysiological knowledge, focusing most of the research on the identification and analysis of the most relevant blood biomarkers (Gunn et al., 2020). A biomarker is defined as any substance that objectively measures both the normal biological state and any pathogenic process, as well as the response to a certain method or treatment (Atkinson et al., 2001). In short, the use of biomarkers in scientific research and in clinical practice focuses on identifying patients at risk of developing a pathology, on diagnosing a disease and evaluating the response generated to an intervention and the early-stage drug development. However, successfully finding mechanistic biomarkers associated with pain proves extremely complex due to the subjective experience that lies in all pain processes and the biopsychosocial model involving chronic pain (Kalso, 2004).

One of the most important circulating biomarkers in inflammatory processes is nitric oxide (NO). NO is a free radical derived from L-arginine that acts as an intercellular messenger in inflammatory processes, being also involved in immune response mechanisms, pain modulation and regulation of the circulatory system (vasodilation) (Cury et al., 2011; Lewis et al., 1993; Schulman, 1997). NO is concentrated in the dorsal horn of the spinal cord with a very well-defined function on the nerve transmission circuits (neurotransmitter). This biomarker can be released into the bloodstream by the stimulation of multiple humoral factors (increased platelet activity, hypofibrinolysis, hyperviscosity, deformity of red blood cells, the activation of leukocytes and oxidative stress), by *"shear stress"* (tension that exerts the blood in a tangential direction on the surface of the endothelium, generating a deformity thereof), and by the prolonged activity of the C-nerve fibres in the dorsal horn of the spinal cord. The overactivity of some of these factors and processes produces the release and diffusion of NO through the synaptic cleft or outside the neurons, so it may end up exciting astrocytes and microglia (Cury et al., 2011; Gratt & Anbar, 2005). The stimulation of the normal activity of the glia causes the systemic release of pro-inflammatory cytokines, reactive oxygen species and prostaglandins that sensitize the nociceptors and neurons of the nervous tissue, prolonging the hyperexcitability of the spinal cord and favouring central sensitisation and chronic pain (Nijs et al., 2017).

Likewise, NO is the main relaxing factor of vascular smooth muscle due to the action it exerts on the cyclic monophosphate guanosine radical. If NO spreads excessively to a nearby area of the skin, local hyperthermia originates, which can influence the heat dissipation process by convection, and cause a deregulation of general body temperature, favouring its increase and that of the body's basal metabolism(Cheung, 2015; Gratt & Anbar, 2005). Furthermore, Albrecht et al. (2013) in their study on AVAs in the glabrous skin of the palms of FM patients, they reported that the release of compounds such as NO can lead to passive dilation of arterioles and AVAs, which would thermally stimulate the peptidergic sensory branches of the skin and trigger the activation of the "local axon reflex". In turn, this would release into the bloodstream of a large amount of substance P and CGRP receptors (powerful vasodilators), with important pro-nociceptive effects and involved in pain inflammatory type(Albrecht et al., 2013).

Scientific literature has not shown a clear consensus regarding the association between NO levels in blood plasma and the characteristic pain symptoms in subjects with FM syndrome. Although some scientific studies found that high concentrations of NO in blood serum were positively correlated with different clinical variables of FM such as headache, pressure pain thresholds, subjective perception of pain evaluated through the Visual Analogue Scale of Pain and functional capacity (Çimen et al., 2009; Rus et al., 2016; Sendur et al., 2009), other scientific studies did not find significant correlations between the concentration levels of NO and clinical characteristics such as subjective self-perceived pain and the global impact of FM symptoms(Koca et al., 2018; Ozgocmen et al., 2006).

Other circulating biomarkers and neurotransmitters to consider in inflammatory and nociceptive processes are catecholamines and aminopeptidases. On the one hand, catecholamines are a set of monoamine neurotransmitters derived from aromatic amino acids and produced by the adrenal medulla and the postganglionic fibres of the SNS. Within this group, we may highlight dopamine, norepinephrine, and epinephrine. Catecholamines are secreted into the bloodstream in response to a stress response mediated by the SNS with a fundamental role on the descending pain pathways, since they modulate the pain response towards the periphery (Rus et al., 2018). On the other hand, aminopeptidases are zinc metalloenzymes that favour the division of amino acids at the distal end of proteins and peptides. The main functions of aminopeptidases include the

regulation of oxytocin, gonadal secretion and thyroid hormones and the regulation of enkephalin metabolism (Ortega et al., 2013).

Thus, enkephalins are endogenous opioids with important analgesic effects that regulate the processes of nociception and cognitive and psychological functions such as anxiety, depression, stress, and mood. Enkephalins are synthesised in the dorsal horn of the spinal cord by different types of neurons in order to regulate the painful processes produced after peripheral injury (Henry et al., 2017). They are metabolised by enzymes called enkephalinases, among which are enkephalin-degrading aminopeptidase (EDA) and oxytocinase that metabolizes endogenous Met-enkephalin and oxytocin, with analgesic, anxiolytic and anti-stress effects (Neumann & Slattery, 2016; Xin et al., 2017). In other words, both EDA and oxytocinase are associated with the regulatory mechanisms of nociception and with cognitive and psychological processes since they participate in the enkephalin metabolism(Martínez-Martos et al., 2019).

To our knowledge, some scientific studies have determined an association between the metabolism of catecholamines and the FM syndrome, but with contradictory results, since the findings of dopamine, norepinephrine and epinephrine in plasma vary between studies, showing their increase, decrease, or even remaining steady in FM patients compared to healthy subjects(Bote et al., 2012; Giske et al., 2008; Kadetoff & Kosek, 2010; Rus et al., 2018; Torpy et al., 2000). Also, only one study has investigated the associations between enkephalinase activity in blood plasma and FM symptoms (Martínez-Martos et al., 2019). In this case-control study, the serum activity of aminopeptidases was analysed in a total of 75 women diagnosed with this pathology. Reported results indicated that there are no significant associations between the levels of enkephalinase activity in the blood serum and the global pain self-perceived by the patients evaluated through the Visual Analogue Pain Scale, as well as with respect to Fibromyalgia symptom severity analysed through the Fibromyalgia Impact Questionnaire (validated in Spanish) (Martínez-Martos et al., 2019).

As discussed in previous sections, CS is a characteristic phenomenon in subjects with FM syndrome (Chinn et al., 2016; Clauw, 2014; Stisi et al., 2007).This phenomenon can be mediated by a series of biomarkers, neurotransmitters, and neuropeptides such as NO, serotonin, norepinephrine, dopamine, epinephrine, enkephalinases, substance P or

CGRP, among others (Bellato et al., 2012; Martínez-Martos et al., 2019; Rus et al., 2018; Russell & Larson, 2009). Despite the contradictory results reflected in scientific literature, future research is recommended as changes in the activity levels of enkephalinases and catecholamines in the blood plasma can modify the regulatory events in which they are involved, ultimately leading to a pathological state in FM patients (Martínez-Martos et al., 2019).

1.4 Pro-inflammatory baseline status in populations with chronic pain and dietary intake.

Recently, evidence has arisen of the possible determining role of lifestyle, specifically that of diet quality, in patients with pathologies associated with chronic pain states(Tick, 2015; Witkamp & van Norren, 2018). Previous research postulates that a healthy dietary pattern could play an important role in pain modulation and, therefore, be considered as a symptom control strategy in clinical practice (Tick, 2015; Witkamp & van Norren, 2018). However, available evidence regarding the association between dietary intake and changes in nociception due to a decrease in the inflammatory state is very limited (Tick, 2015; Witkamp & van Norren, 2018).

Dietary interventions focused on low carbohydrate intake and high adherence to the Mediterranean diet in reducing chronic pain, inflammatory status, and oxidative stress have reported promising results in various populations with chronic pain(Kaushik et al., 2020). In adults with knee osteoarthritis, inclusion in a 12-week diet programme involving low carbohydrate consumption showed a reduction in evoked pain and oxidative stress (Strath et al., 2020). In another study, also carried out in a population with knee osteoarthritis, a reduction in pain, stiffness, an improvement in the range of joint movement and in the performance of daily living tasks was observed in patients who received an antioxidant diet programme, compared to those who consumed a placebo. The programme involved consumption of 40 grams of dehydrated blueberry powder each day over 4 months (Du et al., 2019).

Recent systematic reviews have reported that different dietary interventions can help to improve symptoms including pain in FM patients(Bair & Krebs, 2020; Rossi et al., 2015; Silva et al., 2019). However, existing studies in this field show contradictory results that may be due to their low sample size, the use of different stimulation methods to trigger

the cytokines release into the bloodstream, and the use of different analysis methods that hinder the drawing of generalisable conclusions and results(Bair & Krebs, 2020; Rossi et al., 2015; Silva et al., 2019). On the one hand, scientific studies have indicated that a vegan diet rich in lactobacteria reduces pain levels and improves sleep quality in FM patients (Kaartinen et al., 2000). Similarly, another study in patients who followed a vegetarian diet plan showed an improvement in the severity and impact of FM symptoms (Donaldson et al., 2001). On the other hand, and in contrast to these previous findings, a study conducted in FM patients linking a vegetarian diet with the possibility of improving levels of fatigue, insomnia, sleep and pain did not find significant associations between this intervention and cynical FM variables(Azad et al., 2000).

The Dietary Inflammatory Index (DII) is a recently validated dietary index that has been developed to estimate the patient's inflammation levels.(Shivappa et al., 2014). DII scores are associated with several inflammatory markers, including C-reactive protein, IL-1, IL-2, IL-6, homocysteine, and fibrinogen (Shivappa et al., 2014, 2015, 2017; Wirth et al., 2014). However, the potential of diet in patients with FM syndrome has not been previously examined. Given the baseline pro-inflammatory state of FM subjects due to the high concentrations of pro-inflammatory cytokines in the blood serum, the use of anti-inflammatory dietary interventions characterised by an abundant consumption of vegetables and fruits, of dietary antioxidants, a moderate intake of low-fat proteins as well as monounsaturated fatty acids and a limited consumption of bread and cereals (especially refined cereals), red meat and dairy, could be a promising strategy for the control of pro-inflammatory basal state and, consequently, for the improvement of symptoms, mainly chronic pain, in patients with this pathology (Bjørklund et al., 2018; Kaushik et al., 2020; Rus et al., 2017; Sears, 2015; Sears & Ricordi, 2011). The lack of results highlights the need for future research exploring the possible links between the inflammatory potential of diet and the reduction of symptoms related to FM syndrome.

1.5 Diagnosis and therapeutic approach to Fibromyalgia syndrome.

1.5.1 Diagnosis.

In 1990, the ACR established the first diagnostic criteria for FM syndrome (Wolfe et al., 1990). These criteria were:

1. Clinical history of generalised pain for at least 3 months on the left and right side of the human body, above and below the waist and in the axial skeleton (cervical spine, dorsal spine, or lumbar spine).

2. Pain upon digital palpation with an approximate force of 4 kg in at least 11 of the 18 bilateral tender points shown below:

    I. Occipital: located in the insertions of the suboccipital muscle.
    II. Cervical: located on the anterior faces of the C5-C7 intertransverse spaces.
    III. Trapezius: located at the midpoint of the upper border of the trapezius muscle.
    IV. Supraspinatus: located above the spine of the scapula near the medial border.
    V. Second rib: located at the second costochondral junction.
    VI. Lateral epicondyle: located 2 cm distal to the epicondyle.
    VII. Gluteus: located in the outer quadrant of the gluteus superior muscle.
    VIII. Greater trochanter: located posterior to the bony prominence of the trochanter.
    IX. Knee: located on the fat pad close to the joint line.

Over the years, criteria initially proposed by the ACR did not cover certain realities observed in the categorisation and evaluation of FM patients. In the first place, because key clinical characteristics such as fatigue, somatic and cognitive symptoms had not been considered and, secondly, because there was no symptom severity scale to help record symptom levels among different FM patients. For this reason, in 2010 the ACR updated the diagnostic criteria and included two main scales: the Widespread Pain Index (WPI) and the FM Symptom Severity Scale (SSS) (Wolfe et al., 2010). On the one hand, the WPI consists of a scale accompanied by a drawing where the patient notes the painful areas during the last week. The total score of the index ranges from 0 points to 19 points. On the other hand, the SSS assesses the symptom severity through a questionnaire broken down in two sections: a) a first section that encompasses symptoms of fatigue, non-restorative sleep and cognitive symptoms and in which the patient must indicate the severity of symptoms during the last week using a Likert scale with a score range from 0 points (no problem) to 3

points (serious and persistent problems); and b) a second section where somatic symptoms are assessed through a Likert scale with a score range from 0 points (no symptoms) to 3 points (large number of symptoms). The total score of the SSS ranges from 0 points to 12 points and is obtained by adding the points in the first section to the points in the second one (Wolfe et al., 2010).

In summary, this update of the ACR diagnostic criteria in 2010 established that a patient was diagnosed with FM if they met the following 3 conditions (Wolfe et al., 2010):

1. WPI score ≥7 and SSS score ≥5 or WPI score between 3-6 points and SSS score ≥9.
2. Symptoms must be present at the same level for at least 3 months.
3. The patient lacks a previously diagnosed pathology that could explain his pain.

Subsequently, in 2016, the ACR updated diagnostic criteria again after reviewing and evaluating a series of scientific reports published between 2010 and 2016 to determine the validity, usefulness, reliability and possible modifications of the criteria established in 2010 (Wolfe et al., 2016). This new update is the one used today, and considers that a person can be diagnosed with FM if they meet the following conditions(Wolfe et al., 2016):

1. Widespread pain in at least 4 of the following 5 regions: right upper body region, left upper body region, right lower body region, left lower body region, and axial region.
2. Symptoms must be present at the same level for at least 3 months.
3. WPI score ≥7 and SSS score ≥5 or WPI score between 4-6 points and SSS score ≥9.
4. The FM diagnosis is completely valid regardless of other previous diseases diagnosed in the patient.

Finally, in 2018, a public-private work association called ACTTION together with the American Pain Society proposed new diagnostic criteria for FM syndrome. This was part of a large-scale project aimed at developing a useful diagnostic system and consistent for all chronic pain disorders (Arnold et al., 2019). Diagnostic criteria established by this working group were:

1. Generalised pain at multiple sites, specifically, pain in at least 6 of the following 9 regions: head, left upper limb, right upper limb, chest, abdomen, upper spine, lower spine, left lower limb, and right lower limb.
2. Symptoms must be present at the same level for at least 3 months and must be accompanied by fatigue and moderate to severe sleep problems.
3. There is a series of symptoms which support the FM diagnosis although they are not deemed necessary for it. These symptoms include soft tissue sensitivity, stiffness, environmental hypersensitivity, and hypervigilance.

Despite this range of diagnostic criteria mentioned above, scientific literature proposes the use of a series of methods for evaluative and diagnostic purposes for FM syndrome (Bair & Krebs, 2020). The initial evaluation and examination should include a complete medical history that records a list of drug use, previous medical conditions, cardiovascular history (hypertension, dyslipidaemia), family history, predisposing factors for the development of symptoms such as lifestyle (physical inactivity or sedentary lifestyle), obesity, socioeconomic status, and psychological impairment. In the same way, the presence of comorbidities such as chemical hypersensitivity, interstitial cystitis (painful bladder syndrome) and chronic fatigue should be recorded, as they can help in achieving the treatment goals. Any type of neurological, infectious, rheumatological or endocrine problem should also be noted so that they can be further evaluated later (Bair & Krebs, 2020; Häuser et al., 2015). On the other hand, in the assessment process, reports or self-administered questionnaires such as the Visual Analog Pain Scale (VAS), the Revised Fibromyalgia Impact Questionnaire (FIQ-R), the Central Sensitisation Inventory (CSI), the Pittsburgh Sleep Quality Questionnaire (PSQI), the Multidimensional Fatigue Inventory (MFI) or the Beck Anxiety Inventory (BAI) can be provided. Different non-invasive interventions such as fingertip capillaroscopy, infrared thermography or laser Doppler can be used to assess vascular disorders(Choi & Kim, 2015; Costa et al., 2019; Morf et al., 2005). Similarly, to assess global body temperature, an infrared thermometer can be used in the external auditory canal of the ear as it provides an accurate measure of the core body temperature given the relationship of the tympanic artery with the hypothalamus (Gasim et al., 2013). Finally, findings through radiographic or neuroimaging images are not particularly useful for diagnostic or prognostic purposes in FM syndrome (Bair & Krebs, 2020).

1.5.2 Therapeutic approach to Fibromyalgia syndrome.

In 2016, the European League Against Rheumatism (EULAR), through the review of numerous clinical trials of systematic reviews, proposed a series of basic recommendations for the treatment of FM syndrome based on evidence levels and the strength of sustaining recommendations(G. J. Macfarlane et al., 2017). These criteria for recommendation strength and evidence levels established in the EULAR guideline were the following: **Grade A.** Strong recommendation. Evidence that the benefit/efficacy of the intervention or procedure is significantly greater than the possible adverse effects; **Grade B.** Evidence that the benefit / efficacy of the intervention or procedure is greater than the possible adverse effects, therefore, the treatment can be carried out without any type of problem; **Grade C.** Evidence that the benefit / efficacy of the intervention or procedure is in balance with the possible adverse effects, although there is no evidence against the treatment. The intervention is not recommended in clinical practice; **Grade D.** Evidence that the possible adverse effects of the intervention or procedure outweigh the benefits / efficacy. It is not recommended to carry out the intervention or treatment; **Grade I.** The existing evidence is poor or insufficient to decide for or against the intervention or procedure. Regarding evidence levels: **Level Ia**. Data derived from randomised controlled trials of systematic reviews with meta-analyses; **Level Ib**. Data derived from randomised clinical trials of systematic reviews without meta-analysis; **Level IIa**. Data derived from comparative observational studies with statistical differences; **Level IIb**. Data derived from non-comparative observational studies; **Level IIc**. Data derived from case report studies; **Level III**. Data derived from randomised clinical trials with inconsistent results, from small studies, reports, registries, or expert consensus. **Level IV**. There are no identifying studies in scientific literature.

The main specific recommendations cited in the 2016 EULAR guide are developed below (G. J. Macfarlane et al., 2017):

1. Pharmacologic therapy:

I. Tramadol is a type of weak opioid drug that regulates the release of neurotransmitters such as substance P, acetylcholine and norepinephrine that is recommended for the treatment of chronic pain in FM with a Relative Risk (RR) of 1.77 and a CI of 95 % from 1.26 to 2.48. **Grade A-Level Ib**.

II. Amitriptyline is a tricyclic antidepressant drug. At low doses of about 25 mg per day, it can help reduce pain and fatigue and improve sleep quality with a RR of 1.60 and a 95% CI of 1.15 to 2.24. **Grade A-Level Ia**.

III. Duloxetine and milnacipran are another type of antidepressant drug that helps reduce pain and decrease fatigue. They are recommended in patients with FM syndrome who have severe pain. RR of 1.38 and a 95% CI of 1.25 to 1.51. **Grade A-Level Ia**.

IV. Pregabalin is an anticonvulsant drug that shows positive results in reducing pain and improving sleep quality. It must be administered in patients diagnosed with FM with severe pain. RR of 1.37 and a 95% CI of 1.22 to 1.53. **Grade A-Level Ia**.

V. Cyclobenzaprine, with a structure very similar to amitriptyline, is a drug that helps improve sleep efficiency. It should be recommended for those with sleep problems truly derived from FM syndrome. RR of 4.8 and 95% CI of 3 to 11. **Grade A-Level Ia**.

2. Non-drug therapy:

I. Aerobic exercise, whether on land or in water, improves pain and symptoms in patients with FM syndrome due to the secretion of beta-endorphins and anti-inflammatory cytokines IL-4 and IL-5 with analgesic effects. Likewise, muscle strength training with weights is also beneficial in these patients, even though an increase in pain may occur at the beginning of its execution with a RR of 0.65 and a 95% CI of -0.09 to 1.39. **Grade A-Level Ia**.

II. Cognitive behavioural therapy (CBT) is a treatment modality that helps FM patients manage pain and improve quality of life through pedagogical talk therapy with patients, as a way to help them cope with negative thoughts or ideas and change their behaviour when facing an adverse event. RR of -0.29 and a 95% CI of -0.49 to -0.17. **Grade A-Level Ia**.

III. Multiple component therapies are based on the combination of physical exercise, patient education, relaxation techniques or other specific treatments such as Tai Chi

or massage, with beneficial results on generalised pain and fatigue in people with FM syndrome, with a RR of -0.37 and a 95% CI of -0.62 to -0.13. **Grade A-Level Ia**.

IV. Traditional acupuncture or electrical acupuncture techniques when combined with other treatment modalities can decrease pain symptoms in FM patients. Likewise, hydrotherapy or spa sessions can improve the symptoms of these patients with a duration of up to 14 weeks with a RR of -0.78 and a 95% CI of -1.42 to -0.13. **Grade A-Level Ia**.

V. Mindfulness therapies focused on stress reduction as well as various modalities of meditation techniques such as Qigon, Yoga or Tai Chi can help improve pain, fatigue, and the quality of sleep in FM patients. RR of -0.23 and a 95% CI of -0.46 to -0.01 for pain; RR of -0.61 and a 95% CI of -0.95 to -0.27 for sleep; RR of -0.66 and a 95% CI of -0.99 to -0.34 for fatigue. **Grade A-Level Ia**.

On the other hand, this EULAR guide also established a series of general recommendations, all based on consensus and on the unanimous opinion of experts (**Level III**) (G. J. Macfarlane et al., 2017). Thus, it was indicated that a good approach to FM syndrome requires an early diagnosis. Moreover, a comprehensive assessment of pain, the function and social context is necessary. All possible information should be provided to the patient, including any type of written material to help them understand their pathology. In general, the FM syndrome management should follow a gradual approach, always bearing in mind the complexity and heterogeneity of symptoms due to the abnormal processing of pain and the concomitant characteristics of this condition. In the same way, therapeutic interventions should focus primarily on the use of non-pharmacological therapies adapted to the pain intensity, functionality, fatigue, sleep disturbances, comorbidities, and patient preferences. In other words, it is recommended that decision-making be agreed with the patient (G. J. Macfarlane et al., 2017).

1.5.2.1 Pharmacological treatment.

The use of pharmacological therapies in chronic pain conditions such as FM syndrome implies a process of constant re-evaluation and review to guarantee the continuity of these treatments, especially those aimed at cognition and fatigue (Häuser et al., 2017). Most of the drugs used for FM treatment continue to show unsatisfactory results

today, mainly due to their lack of efficacy, the high risk of side effects, and potential to generate addictions(Häuser et al., 2017). Accordingly, the 2016 EULAR revised guidelines for FM management do not recommend the use of pharmacological therapies that include non-steroidal anti-inflammatory drugs (NSAIDs), monoamine oxidase inhibitors (MAOIs), serotonin reuptake inhibitor drugs (SSRIs), growth hormone and, above all, the use of sodium oxybate (rejected by the European Medicines Agency and by the Food and Drug Administration of the United States), strong opioids and corticosteroids (G. J. Macfarlane et al., 2017).

In contrast, drugs most used for the treatment of symptoms in FM patients usually include norepinephrine reuptake inhibitor drugs (SNRIs), among which tramadol, duloxetine and milnacipran stand out, which are considered for patients with severe pain; antidepressant drugs such as amitriptyline, antiepileptic drugs such as pregabalin, and drugs for sleep problems such as cyclobenzaprine (Häuser et al., 2017; G. J. Macfarlane et al., 2017). Numerous systematic reviews and meta-analyses support the use of these aforementioned drugs in the FM population with promising results (Häuser et al., 2009, 2011, 2013; Lunn et al., 2014; Mease et al., 2009; Murakami et al., 2017; Nishishinya et al., 2008; Roskell et al., 2011; Wiffen et al., 2013). In this context, a systematic review carried out in 2014 showed that duloxetine decreased pain in FM patients by around 50% compared to the control group that received placebo. Moreover, another study prepared by Murakami et al. (2017) indicated that duloxetine is a good drug treatment option in FM patients with severe fatigue and depression. The study also showed that this drug is safe and effective due to the follow-up and review carried out one year after the start of the study (Murakami et al., 2017). In relation to the drug milnacipran, a double-blind randomised controlled trial demonstrated the efficacy of milnacipran in relieving pain, improving general well-being, and increasing physical function in patients with FM syndrome compared to the control group (Mease et al., 2009). In the same way, another scientific study showed that the use of milnacipran had positive effects on pain intensity and fatigue (Häuser et al., 2013). With respect to tramadol, a meta-analysis carried out in 2011 showed that the prescription of this drug with a dose of 37.5 mg/4 times a day for 3 months improved pain in FM patients by around 30% compared to the control group (Roskell et al., 2011). On the other hand, the scientific literature shows beneficial effects of the use of pregabalin in FM patients (Wiffen et al., 2013). Thus, a meta-analysis of 4 randomised controlled trials with pregabalin treatment in patients diagnosed with FM

showed significant improvements in pain, sleep problems and quality of life compared to control groups (Häuser et al., 2009). Finally, the use of amitriptyline as a treatment in FM has also been supported by numerous systematic reviews in scientific literature. Nishishinya et al. (2008) indicated that the prescription of 25mg of amitriptyline per day in FM patients had positive results on pain, sleep and fatigue at 6-8 weeks of treatment, but these were not maintained at the 12-week follow-up (Nishishinya et al., 2008). Furthermore, a systematic review with meta-analysis conducted in 2011 stated that the use of amitriptyline relieved pain by about 30%, although the reported effects on fatigue and sleep were medium or low (Häuser et al., 2011).

1.5.2.2 Non-pharmacological treatment.

A wide variety of treatment methods have been studied and developed over the last few years to improve the symptoms and quality of life in FM patients (Aman et al., 2018). In relation to the study provided by EULAR on FM syndrome management and under the unanimous consensus of the experts who made up the working group, recommendations were established for the use of non-pharmacological therapies based on a grading scale (strongly recommended ; weakly recommended; not recommended; against) (G. J. Macfarlane et al., 2017). First, this working group strongly recommended physical exercise, strength training with weights, and aerobic exercise in FM patients due to the beneficial effect on pain, physical function, disability, and personal well-being. Second, and in a lower range of recommendation than performing physical exercise, they indicated different modalities of therapies that included meditation techniques (Qigon, Yoga or Tai Chi) due to their positive effects on sleep, fatigue and quality of life; mindfulness therapies for the observed improvements in pain and quality of life; cognitive-behavioural therapy for the effects on pain, disability and mood in the short term, even slightly long term; and complementary therapies such as acupuncture or hydrotherapy due to positive data related to pain and fatigue reduction and improved quality of life. Third, this working group did not recommend the use of therapies such as massage, hypnotherapy, or alternative therapies such as homeopathy, due to their low or null efficacy. Finally, they showed their outright dissatisfaction, as a precautionary and safety measure for FM patients, with the use of chiropractic techniques (G. J. Macfarlane et al., 2017).

A variety of conservative therapeutic interventions applied in the FM population are detailed below based on the publications of systematic reviews and meta-analyses of scientific literature:

1. **Physical exercise**. In relation to physical exercise programmes, scientific literature indicates positive effects on FM patients' symptoms, both in aerobic exercise activities and in strength or resistance training programmes (A. Busch et al., 2002; A. J. Busch et al., 2013). On the one hand, a systematic review of 47 studies with randomised clinical trials of different aerobic exercise programs with a duration equal to or greater than 30 minutes (if the exercises were performed once a day), or a duration equal to or greater than 10 minutes (if the exercises were performed twice a day), showed significant improvements in pain intensity and physical function in patients diagnosed with FM (A. Busch et al., 2002). On the other hand, another systematic review that included 5 randomised controlled trials focused on a resistance training programme of 8 or more repetitions per exercise for 2 or 3 times a week, reported a reduction in pain of about 3.3 cm on the numerical scale of pain as well as an improvement in the functionality of FM patients compared to the control group that received placebo (A. J. Busch et al., 2013).

2. **Acupuncture**. The use of acupuncture combined with other treatment modalities has been analysed in FM populations. A high-quality systematic review consisting of 9 randomised clinical trials with a total of 395 FM patients showed that the use of traditional acupuncture on specific established points for a time of 20 to 30 minutes in a period of 3 to 13 weeks, combined with standard therapies reduced pain by around 30% compared to those patients who only received traditional acupuncture. Likewise, the use of electrical acupuncture was correlated with a decrease in pain by 22% and with an improvement of generalised fatigue by 11% (Deare et al., 2013).

3. Cognitive behavioural therapy. Regarding this treatment modality, scientific literature shows a systematic review of 23 randomised controlled trials with more than 2,000 FM patients (Bernardy et al., 2013). The sessions consisted of a series of pedagogical conversations with the patient through understandable communication skills to provide coping strategies and control over their pain and pathology. The average duration observed in all studies was 18 hours distributed in 10 sessions over

10 weeks. The use of CBT proved to be effective in the short and long term in terms of reducing pain and improving disability and mood compared to the control group of FM patients, who received regular treatment, or who were in a waiting list to participate in research projects (Bernardy et al., 2013).

4. **Mindfulness**. Mindfulness therapy based on stress reduction and focused on body mindfulness is another of the most used current therapeutic interventions in FM syndrome (Lakhan & Schofield, 2013; Marikar Bawa et al., 2015). In this context, a systematic review and meta-analysis prepared by Lauche et al. (2013) on mindfulness sessions in FM patients with sessions lasting from 2 to 3.5 hours over 8 to 10 weeks, and with individualised activities at home lasting 30 to 45 minutes, showed immediate pain reduction after mindfulness treatment compared to the control group of FM subjects who received regular health care, or active therapies consisting of educational, supportive, relaxation and stretching techniques (Lauche et al., 2013).

5. **Meditation therapies**. The use of meditation techniques in different modalities such as Yoga, Tai Chi or Qigon have experienced remarkable growth in FM treatment (Prabhakar et al., 2019). A recent meta-analysis of systematic reviews that included 7 randomised controlled trials and a total of 362 FM patients indicated that meditation treatments such as Yoga, Qigon and Tai Chi, applied over a period of 4 to 12 weeks, with a total distributed duration In 16 hours, improved sleep quality and short- and long-term fatigue in FM patients compared to patients who received regular treatment, or active therapies (aerobic exercise, health education and stretching) (Langhorst et al., 2013).

6. **Hydrotherapy and balneotherapy**. Hydrotherapy and bath therapy are other complementary therapies used in FM pathology (Lauche et al., 2015). Several systematic reviews analysed water or mud bath therapies at a temperature of 36ºC to 37ºC or slightly higher (from 40ºC to 45ºC) with an average duration of 4 hours distributed over several weeks. Results included improvements in pain and the state of calm, with long-term effects maintained until the fourth week. Furthermore, no significant differences were reported in terms of greater or lesser effectiveness of the

use of balneotherapy or hydrotherapy separately (Langhorst et al., 2009; Naumann & Sadaghiani, 2014).

7. **Massage**. Massage is a conservative therapy that has been investigated in the management of FM through literary reviews (Yuan et al., 2015). In a meta-analysis carried out in 2014 that included 9 randomised clinical trials with a total of 404 FM patients, different massage therapy techniques with a duration of 25 to 30 minutes and about 5 sessions on average were evaluated and compared to control groups, which received a variety of therapies such as transcutaneous electrical stimulation, relaxation techniques, acupuncture, and basic health care. This meta-analysis indicated that the use of massage alone was not associated with an improvement in the clinical pain characteristics. Still, there were short-term positive effects on pain, anxiety and depression; however, clinical trials supporting these results were of low methodological quality and showed no evidence on the effects of follow-up (Li et al., 2014).

8. **Chiropractic techniques**. Chiropractic techniques must be avoided to treat FM (G. J. Macfarlane et al., 2017). Scientific studies available on it are scarce, finding only a pilot study, a quasi-randomised study, and a controlled clinical trial without significant and indeed counterproductive results. Therefore, due to the lack of evidence and in order to safeguard the health condition of FM patients, the use of chiropractic techniques is not recommended in this population (Ernst, 2009).

9. **Homeopathy**. Finally, homeopathy is another therapeutic option that the EULAR guidelines do not recommend using in FM patients(G. J. Macfarlane et al., 2017). The only studies available are a total of 4 randomised clinical trials in 2 systematic reviews with a treatment that consisted of the consumption of homeopathic medicines such as arnica montana, bryonia alba and rhus toxicodendon once a day, over 3 months and with a follow-up every 4-8 weeks through clinical interviews. Said therapy did not show beneficial effects on symptoms in FM patients compared to the placebo control group (Boehm et al., 2014; Perry et al., 2010).

1.5.2.3 New therapeutic and evaluative approaches from Physiotherapy.

1.5.2.3.1 Physiotherapy through Pain Education and electrotherapy.

Despite the broad set of non-pharmacological therapeutic treatments applied in populations with FM syndrome, new Physiotherapy lines of treatment and research have been recently proposed to control and manage FM patients' symptoms. Among them, we find pain neuroeducation (PNE) or electrotherapy techniques such as monopolar dielectric radio frequency (MDR) (Barrenengoa-Cuadra et al., 2020; Ibáñez-Vera et al., 2020; Malfliet et al., 2017).

On the one hand, pain neuroeducation is a therapeutic approach focused on education and the learning process by the patient about the mechanisms of neurophysiology, neuroanatomy and neurobiology of pain, as well as processing at the CNS level (Malfliet et al., 2017). One of the primary objectives of PNE is reconceptualizing pain on the basis that every painful process need not be associated with tissue damage, but may be due to a disorder in processing of nociceptive information in the brain. In this reconceptualisation, the patient must be provided with detailed and clarifying information about the pathophysiology of pain and the processes of central and peripheral sensitisation, always supported by written information supports (brochures, easy-to-read guides, etc.). This effort must be accompanied by examples and metaphors, since they can help reduce the hyperactivity of the Sympathetic Nervous System, the immune system, the endocrine system and the musculoskeletal system (Moseley, 2003). The PNE also seeks a high patient participation in the entire neuroeducation process. To that end, patients must be aware of the origin of their symptoms, accept the diagnosis of the disease, face the pathology in different tasks or activities of daily living, identify aggravating external factors (beliefs, cultural environment, learning capacity, clinical overinformation, etc.), work on body awareness and attention, and adhere to different aerobic, sleep hygiene or diet programmes (Aman et al., 2018; Barrenengoa-Cuadra et al., 2020). Such process will help the patient in reducing pain regions, symptom severity, catastrophizing and the impact on functional disability (Barrenengoa-Cuadra et al., 2020; García-Ríos et al., 2019; Malfliet et al., 2017).

Scientific literature includes a wide variety of scientific studies and systematic reviews showing the use of PNE in different populations with chronic pain (Andias et al.,

2018; Bodes Pardo et al., 2018; Louw et al., 2016; Watson et al., 2019; Wood & Hendrick, 2019). Accordingly, a systematic review prepared by Louw et al. (2016) that included 13 randomised clinical trials with a total of 629 patients with different chronic pain conditions (non-specific chronic low-back pain, lumbar radiculopathy, neck pain, chronic fatigue syndrome and FM syndrome) indicated that the application of various PNE programmes is effective in reducing pain, improving disability and functionality, increasing joint movement, and reducing psychological symptoms (Louw et al., 2016).

In relation to the FM population, there are also numerous scientific investigations with PNE treatment(Amer-Cuenca et al., 2020; Barrenengoa-Cuadra et al., 2020; Serrat et al., 2020; van Ittersum et al., 2014). In a study carried out by Barrenengoa-Cuadra et al. (2020) at a hospital in Bilbao (Spain) with a total of 85 FM patients, it was reported that the PNE intervention accompanied by a physical exercise and mindfulness programme improved the WPI, SSS and FIQ pain score. In addition, these benefits were maintained in the visits at 6 months and at 12 months, with benefits being observed in the medium and long term (Barrenengoa-Cuadra et al., 2020). Furthermore, a recent 12-week randomised clinical trial in 169 FM patients analysed the possible effects of a PNE intervention in conjunction with CBT, mindfulness, and meditation techniques, compared to a control group that received aerobic exercise counselling, basic health education and pharmacological treatment with duloxetine and amitriptyline. The observed data indicated important effects in reducing functional impact (d = 1.13), in self-perceived pain (d = 0.66), in depression symptoms (d = 0.69), in functional activity (d = 0.53), in fatigue (d = 0.77), anxiety (d = 0.99) and pain catastrophizing (d = 1.21) (Serrat et al., 2020). Finally, another recent single-blind randomised controlled trial evaluated the dosage of an PNE intervention and its effects on pain variables and clinical characteristics in 103 patients with FM syndrome (Amer-Cuenca et al., 2020). The PNE programme consisted of different slides prepared in power point with information on the physiology of the CNS, the characteristics of acute and chronic pain, the origin of pain in the CNS and the chronification of all painful processes through the phenomena of CS, CNS neuroplasticity and pain neuromatrix. Randomisation was configured in 4 intervention groups: group 1 received 45 minutes of the PNE program, 1 time per week over 6 sessions; Group 2 received 45 minutes of PNE, 1 time per week in a total of 2 sessions; Group 3 received 6 sessions of 15 minutes of PNE, 1 time a week; and group 4 received a biomedical education program for 45 minutes over 2 sessions. Researchers observed significant

differences in all groups in relation to pain scores with a long effect size ($\eta 2p = 0.170$) at 3 months of follow-up in favour of the group that received a longer and more PNE sessions (Amer-Cuenca et al., 2020).

Moreover, various research groups are currently applying different treatment modalities with physical agents through electrotherapy techniques such as transcutaneous electrical nerve stimulation, low-intensity laser, ultrasound or electromagnetic waves (Honda et al., 2018). From a therapeutic point of view, the use of electrical stimulation through dielectric electromagnetic signals with radio frequency generates important physiological effects in the body, which arise from its high vasodilator and calorific capacity at the molecular level on organic tissues (Rodríguez-Martín, 2014). The MDR has a frequency range between 600 and 930 kHz with electrical signals transmitted as pulses and continuously modulated manually to reduce thermal shock and avoid adaptation of skin receptors, besides focusing energy in deep tissue without damaging the overlying structures (Albornoz-Cabello et al., 2016).

The application of MDR produces following physiological effects on our body:

a. The passage of the electromagnetic wave carries a kinetic energy that produces vasodilation, a rise in temperature, and a local increase in blood flow, allowing the elimination of harmful substances (Albornoz-Cabello et al., 2016; Rodríguez-Martín, 2014).

b. Inhibition of peripheral afferent nerve fibres that are involved in the perception of pain, and that according to the theory of the "Control Gate", would lead to pain inhibition since the electrical sensory stimulus provided by the MDR prevents the nociceptive stimuli from progressing towards the higher nervous system centres (Albornoz-Cabello et al., 2016; Rodríguez-Martín, 2014).

c. Greater permeability of the cell membrane that helps to eliminate inflammation compromising the nerve roots (Albornoz-Cabello et al., 2016; Rodríguez-Martín, 2014).

d. Release of endogenous opioids into the bloodstream with function on the descending pathways of the nervous system, which blocks the secretion of

neuropeptides such as substance P, with important pro-nociceptive effects in the body (Albornoz-Cabello et al., 2016; Rodríguez-Martín, 2014).

Scientific studies found in scientific literature for MDR in the population with chronic pain are scarce but with encouraging results (Hochsprung et al., 2018; Ibáñez-Vera et al., 2020). In this regard, a pilot study with 24 multiple sclerosis patients to assess the impact of pain in different areas applied MDR with a frequency of 800 to 900 kHz for 20 minutes, 5 days a week for 3 weeks. The study reported positive results in the mean and maximum pain scores according to the Brief Pain Inventory questionnaire, in the affectation of pain at the workplace, in the affectation of pain in the personal sphere, and in the quality of sleep and night's rest (Hochsprung et al., 2018). To our knowledge, there is just a single recent study that has evaluated the use of MDR in the FM population, with promising pain relief results in FM patients (Ibáñez-Vera et al., 2020). This single-blind randomised controlled trial in 66 FM patients was configured in 3 groups: 1) experimental group (8 sessions of 20 minutes of MDR with a frequency of 870 kHz, intensity of 30 amps and a pulsed emission at 50% on the trapezius muscle following ACR criteria); 2) simulation group (the same radiofrequency device but without the activation of the current emitting device), and 3) control group (received regular treatments, maintenance program of physical activity and continuation of their pharmacological treatment plan). The study data indicated significant differences in local pain ($p = 0.025$) and in the impact index of FM ($p = 0.031$), although with short-term effect (Ibáñez-Vera et al., 2020). Therefore, these encouraging results obtained in FM patients open a research line to assess the real, long-term influence of MDR treatment.

1.5.2.3.2 New tools for the evaluation of pain and vascular reactivity in Physiotherapy.

In the discipline of Physiotherapy, the FM syndrome is perceived as a problematic disease because it is not adjusted to a classic biomedical model. As such, there is no specific aetiology, the pathology does not present clear markers and the diagnosis is produced by ruling out other conditions (Roitenberg & Shoshana, 2019). However, physiotherapists have a series of pain and symptom assessment tools that have demonstrated satisfactory psychometric properties in FM patients (Roitenberg & Shoshana, 2019). The following stand out among them: 1) the VAS is one of the most important instruments for pain assessment in FM patients, presenting a sensitivity and specificity

(Marques et al., 2008)of 80%; 2) pressure algometry is a widely used instrument to assess sensitivity to pain with great reliability and internal consistency ($\alpha = 0.93$) (Chesterton et al., 2007); 3) the FIQ-R reports information on physical deterioration, the ability to perform a job or task, fatigue, stiffness and pain in FM patients, demonstrating high reliability with an intraclass correlation coefficient of 0.82 (Salgueiro et al., 2013); 4) the CSI assesses characteristic symptoms of this phenomenon, also showing a high reliability of 0.91 (Cuesta-Vargas et al., 2016); 5) the PSQI is a tool that evaluates the efficiency and effectiveness of sleep in FM patients, presenting high reliability with a correlation coefficient of 0.80 (Hita-Contreras et al., 2014); 6) the MFI, which assesses fatigue, is another instrument that offers proven, good psychometric properties in FM patients (intraclass correlation coefficient between 0.65 and 0.91) (Munguía-Izquierdo et al., 2012); and 7) the BAI assesses symptoms of psychological health and anxiety, and also presents good reliability with a high internal consistency of 0.93 (Magán et al., 2008).

Furthermore, infrared thermography (IT) is an optimal method to assess vascular reactivity that provides an adequate relationship between changes in blood flow at the peripheral level, and the thermal properties of the skin tissue (Fujimasa et al., 2000.; Sagaidachnyi et al., 2014). IT is based on the theory of heat transfer. The theory proposes that high heat in a body region is related to a greater blood flow and that, conversely, cold body regions represent low blood flow regions (Sagaidachnyi et al., 2014). Furthermore, IT is a non-invasive method that has high sensitivity (90%) and high specificity (86%) (Mirbod & Sugiura, 2017).

1.6 Justification of the thesis.

Patients with FM syndrome present a complex set of symptoms characterised mainly by the presence of chronic generalised musculoskeletal pain. The available scientific evidence suggests the existence of various hypotheses that attempt to explain the etiopathogenesis of the clinical manifestations of the disease, specifically the origin of the chronic pain characteristic of FM syndrome. Numerous studies indicate that the presence of vascular factors, inflammatory factors and nociceptive biomarkers in the blood serum, as well as dietary factors could be determining factors, as they all participate in the pain perception modulation and in pain processing mechanisms at the CNS level. To date, there is no scientific evidence that has examined peripheral neurovascular disorders in the glabrous skin on the dorsal and palm of the hands of FM subjects, nor the core body

temperature and its possible relationship with abnormalities of the cutaneous microvasculature and the global body thermoregulation processes, which could explain chronic pain and the different clinical manifestations of this pathology. Furthermore, the vasodilator role that NO exerts on peripheral capillaries and the high metabolic presence of enkephalinases in blood serum can influence the mechanisms of nociception and hyperalgesia, explaining symptoms such as pain, anxiety, depression, and stress. Finally, since inflammatory cytokines may be involved in the underlying mechanism of FM syndrome, a pro-inflammatory diet would be associated with hypersensitivity to pain and other symptoms of this disease. Previous research postulates that a healthy dietary pattern could play an important role in pain modulation and, therefore, be considered as a symptom control strategy in clinical practice. However, available evidence regarding the association between dietary intake and changes in nociception due to a decrease in the inflammatory state is very limited.

The initial hypotheses for the first study of the doctoral thesis was that patients with FM syndrome could present abnormalities in the peripheral vascular response on the dorsal and palm of the hands characterised by vasodilation in the face of excessive peptidergic sensory innervation of arteriovenous anastomoses, and by passive dilation of the arterioles due to the release of compounds such as nitric oxide from endothelial cells, which, in turn, would influence the global body temperature of patients with Fibromyalgia.

The initial hypothesis for the second study of the doctoral thesis was that, given the presence in the blood serum of biomarkers and neurotransmitters such as nitric oxide and enkephalins and their role in the participation in pain modulation processes, we hypothesise that the presence of circulating nitric oxide, as well as the enkephalinase activity in blood serum, could be associated with pain variables and symptoms of patients diagnosed with Fibromyalgia.

The starting hypothesis for the third study of the doctoral thesis was that, since inflammatory cytokines may be involved in the mechanism underlying the FM syndrome, we hypothesised that a pro-inflammatory diet would be associated with hypersensitivity to pain and other FM symptoms.

OBJETIVOS

OBJECTIVES

2. OBJETIVOS

2.1 Generales:

- Evaluar el componente vascular periférico de la piel del dorso y palma de las manos y la temperatura corporal global como indicadores de la actividad adrenérgica del sistema nervioso simpático y de los procesos de termogénesis en personas diagnosticadas con síndrome de Fibromialgia.

- Evaluar la presencia en a nivel sanguíneo periférico de biomarcadores vasodilatadores (óxido nítrico) y la actividad de opioides endógenos (encefalinasas) como indicadores de la microcirculación sanguínea y del estado basal inflamatorio y nociceptivo y su relación con el dolor crónico y la sintomatología en personas diagnosticadas con síndrome de Fibromialgia.

- Evaluar el potencial inflamatorio de la dieta y su relación con el dolor crónico y la sintomatología en pacientes con Fibromialgia.

2.2 Específicos:

- Comparar el nivel global del dolor, los umbrales y magnitud del dolor eléctrico, los umbrales del dolor por presión, la sensibilización central, la fatiga, la severidad e impacto de los síntomas, las alteraciones del sueño y los síntomas de ansiedad en mujeres con síndrome de Fibromialgia y mujeres sanas.

- Evaluar el patrón termográfico de la palma y dorso de las manos como medida del deterioro de la microvasculatura a nivel cutáneo e identificar posibles variaciones termográficas en mujeres con síndrome de Fibromialgia y mujeres sanas.

- Evaluar la temperatura corporal global a nivel timpánico y axilar como indicador de una disfunción del Sistema Nervioso Autónomo y de los procesos de termorregulación central en mujeres con síndrome de Fibromialgia y mujeres sanas.

- Analizar la asociación del flujo sanguíneo vascular periférico con la presencia de óxido nítrico en suero sanguíneo en mujeres con síndrome de Fibromialgia y mujeres sanas.

- Analizar la asociación de la presencia de óxido nítrico en suero sanguíneo y la temperatura corporal central en mujeres con síndrome de Fibromialgia y mujeres sanas.

- Analizar la asociación de los niveles sanguíneos de óxido nítrico y de la actividad de la aminopeptidasa degradante de encefalina (EDA) y la oxitocinasa con el dolor global, umbrales de dolor por presión, sensibilización central, impacto de los síntomas sobre la salud física y mental y los síntomas de ansiedad en una población de mujeres diagnosticadas con síndrome de Fibromialgia.

- Analizar el potencial inflamatorio de la dieta a través del Índice Dietético Inflamatorio y examinar su relación con cambios sobre los umbrales del dolor por presión, intensidad global del dolor, impacto de los síntomas, fatiga, problemas de sueño y síntomas comunes de ansiedad en mujeres con síndrome de Fibromialgia y mujeres sanas.

2. OBJECTIVES

2.1 General:

- To assess the peripheral vascular component of the skin on the dorsal and palm of the hands and the global body temperature as indicators of the adrenergic activity of the sympathetic nervous system and of thermogenesis processes in people diagnosed with Fibromyalgia syndrome.

- To assess the presence in peripheral blood of vasodilator biomarkers (nitric oxide) and the activity of endogenous opioids (enkephalinases) as indicators of blood microcirculation and of the inflammatory and nociceptive baseline state and its relationship with chronic pain and symptoms in people diagnosed with Fibromyalgia syndrome.

- To assess the inflammatory potential of diet and its relationship with chronic pain and symptoms in patients with Fibromyalgia.

2.2 Specific:

- To compare global pain level, electrical pain thresholds and magnitude, pressure pain thresholds, central sensitisation, fatigue, symptom severity and impact, sleep disturbances, and anxiety symptoms in women with Fibromyalgia syndrome vs healthy women.

- To assess the thermographic pattern of the palm and dorsal of the hands as a measure of the deterioration of the microvasculature of the skin, and to identify possible thermographic variations in women with Fibromyalgia syndrome versus healthy women.

- To assess the global body temperature at the tympanic and axillary level as an indicator of a dysfunction of the Autonomous Nervous System and of processes of central thermoregulation in women with Fibromyalgia syndrome versus healthy women.

- To analyse the association of peripheral vascular blood flow with the presence of nitric oxide in blood serum in women with Fibromyalgia syndrome versus healthy women.

- To analyse the association of the presence of nitric oxide in blood serum and core body temperature in women with Fibromyalgia syndrome versus healthy women.

- To analyse the association of nitric oxide blood levels and enkephalin degrading aminopeptidase (EDA) and oxytocinase activity with global pain, pressure pain thresholds, central sensitisation, impact of symptoms on physical and mental health and anxiety symptoms in a population of women diagnosed with Fibromyalgia syndrome.

- To analyse the inflammatory potential of diet through the Inflammatory Dietary Index and examine its relationship with changes in pressure pain thresholds, global pain intensity, impact of symptoms, fatigue, sleep problems, and common symptoms of anxiety in women with Fibromyalgia syndrome versus healthy women.

METODOLOGÍA

METHODS

3. METODOLOGÍA / METHODS

Tables 1, 2 and 3 present a summary of the methodology employed in the studies included in the PhD thesis.

3.1 Study I: "Evaluation of sympathetic adrenergic branch of cutaneous neural control throughout thermography and its relationship to nitric oxide levels in patients with Fibromyalgia".

The methods section of the Study I, added in the PhD thesis, is summarized and described below. This study has been published online in the scientific journal *Journal of Thermal Biology* (Impact Factor JCR: 2.361; Category ZOOLOGY-SCIE (18/169); Q1/T1). Table 1 shows a summary of the methodology employed in this observational case-control study.

Table 1. Summary of Material and Methods in Study I.

PAPER	STUDY DESING	PARTICIPANTS	PROCEDURES	MAIN VARIABLES	METHODS
Evaluation of sympathetic adrenergic branch of cutaneous neural control throughout thermography and its relationship to nitric oxide levels in patients with Fibromyalgia.	Observational case-control study.	• Women diagnosed with FMS (n=42). • Healthy women (n=52).	• Screening to inclusion selection criteria. • Sociodemographic and clinical characteristics were recorded: - Age. - Sex. - Height. - Weight. - Body mass index. - Hand dominance. - Drug history. - Smoking history. • Risk cardiovascular factors: - Arterial Hypertension. - Hypercholesterolemia. - Diabetes mellitus. • Other disorders: - Skin disorders. - Neurological disorders.	• Peripheral vascular response. • Core body temperature: - Tympanic. - Axillary. • Blood samples collection. • Nitric oxide levels. • Degree of the FMS symptoms.	• Infrared thermography camera (FLIR B335, FLIR Systems, Inc., USA). • Infrared skin surface temperature scanner (Derma Temp®, Model: 104920 – DT-1001-LT [USA]). • Blood from the cephalic vein of the arm into an ethylenediaminetetraacetic acid tube (BD Vacutainer® LH PST II Advance, Ref. 367374, Becton Dickinson, New Jersey, USA). • Purge system of Sievers Instruments (model NOA 280i, GE, Analytical Instruments, Colorado, USA). • Spanish version of the Revised Fibromyalgia Impact Questionnaire (FIQ-R).

FMS: Fibromyalgia syndrome.

3.1.1 Study Design and Participants.

Forty-two women diagnosed with FMS and recruited from two Spanish Fibromyalgia Associations (the Fibromyalgia Association of Granada [AGRAFIM] and the Provincial Fibromyalgia Association of Jaén [AFIXA]) participated in this observational case-control study. Fifty-two healthy women were recruited from the patients' relatives and from volunteers through a local advertisement from the University of Granada (Spain). Patient recruitment was conducted between January 2019 and June 2019, and the participants were contacted by telephone to arrange visits throughout the 6 months. Only one study visit was required per patient, and each patient was evaluated on the same day for a total of 1 hour. The study was conducted in accordance with the 2013 Helsinki Declaration and was approved by the Research Ethics Committee of the Andalusian Heath Service of Granada (Granada, Spain) (approval number, 1797-N-17). All participants signed a written informed consent form prior to their inclusion in the study.

The inclusion criteria for the patients with FMS were 1) a diagnosis in accordance with the American College of Rheumatology criteria for classifying FMS (2016 revision) (Wolfe et al., 2016) by a rheumatologist of the public health system of Andalucía (Spain); 2) an age from 18 to 70 years; and 3) no other rheumatic diseases. The inclusion criteria for the control group were 1) an age from 18 to 70 years and 2) no rheumatic diseases. The exclusion criteria for both groups were 1) male sex; 2) the presence of cardiac, renal or hepatic failure; 3) severe physical disability; 4) fever after an infection in the past two weeks; 5) hypotension/hypertension; 6) psychiatric illness; 7) neurological disorders; 8) cancer; 9) a previous history of surgery; 10) treatment with vasoactive drugs or anticoagulants or a history of drug use; and 11) skin disorders.

3.1.2 Procedures.

To obtain the demographic data, the participants were asked to complete questionnaires on their medical history, which included information on age, sex, height, weight, body mass index, and dominant hand. We employed the Spanish version of the Revised Fibromyalgia Impact Questionnaire (FIQ-R) to assess the degree of the FMS symptoms (Salgueiro et al., 2013). The main outcome measures were then evaluated that same day. Before participating in the study, the patients were ask to wear comfortable clothing such as sport shirts and sweatpants and not to wear any accessories such as watches, bracelets or rings.

All participants were asked to avoid ingesting any vasoactive substance (alcohol, caffeine, nicotine) or food during the two hours prior to the evaluation. Finally, blood samples were obtained for nitric oxide determination.

3.1.3 Measures.

3.1.3.1 Peripheral Vascular Response Assessment.

Infrared thermography (IRT) is a non-invasive technique that measures local changes in skin temperature and is a useful method for providing physiological information regarding the microvasculature and skin surface temperature (Clauw, 2014; Gaskell, 1956; Kulshreshtha et al., 2012). IRT has demonstrated a sensitivity of 90% and a specificity of 86% (Mirbod and Sugiura, 2017).

We acquired thermographic images of the dorsal and palmar sites of both hands using a FLIR B335 infrared thermography camera (FLIR Systems, Inc., USA). The camera's ambient temperature was set to 20 ºC, and the spectral emissivity was set to 0.98, given that human skin behaves as a blackbody with an emissivity of 0.96–0.99 (Ring and Ammer, 2012; Sanchez-Marin et al., 2009). All thermal images were collected in compliance with the recommendations of the European Association of Thermology (Ring and Ammer, 2012), and all thermograms were obtained in the same conditions. The participants remained in a sitting position in a room with a constant temperature of 20 ºC, following an acclimatization period of 20 min, after which the IRT was first performed on the dorsal side and then on the palmar side of both hands, capturing images from the distal phalanges to the wrist (Lim et al., 2014). The maximum, minimum and mean temperatures from each point on both hands were calculated using the camera's software (**Fig. 1**). The IRT was performed in the afternoon for both groups to control changes in circadian rhythms (Neves, 2017).

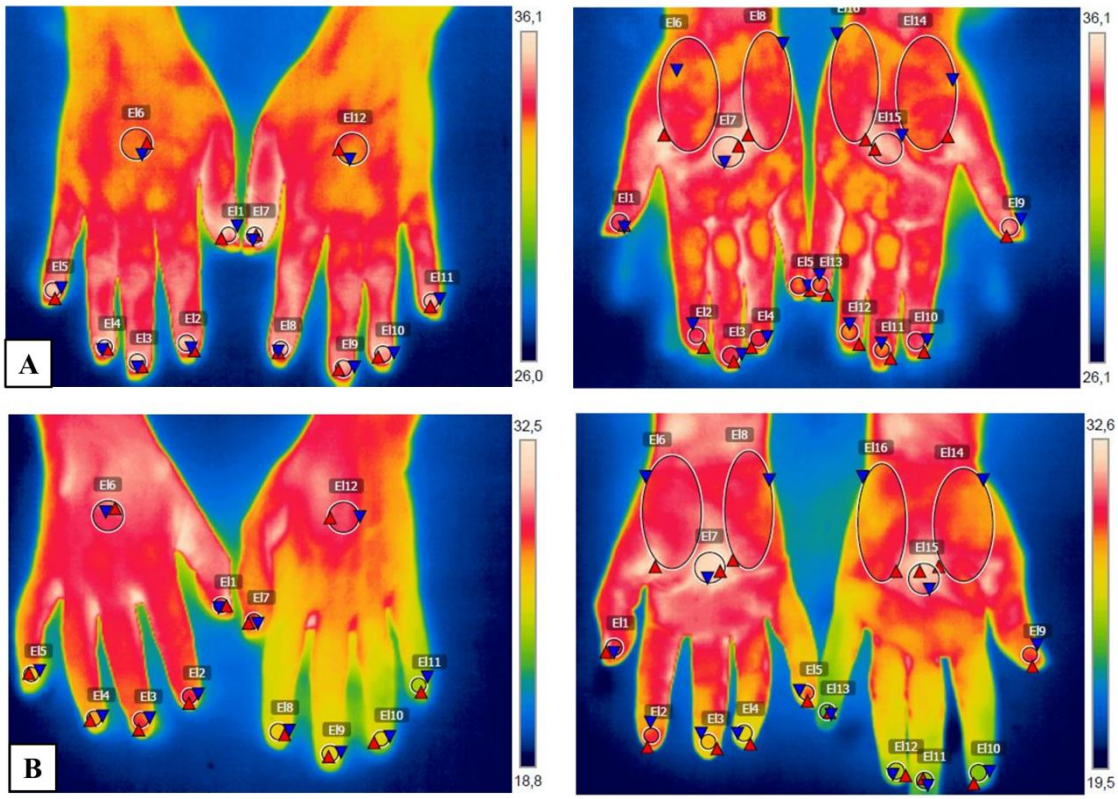

Figure 1. Thermography image of the hands of a patient diagnosed with fibromyalgia and a healthy control.

The analysis of the skin surface temperature was conducted through a circle at the centre of each dorsal and palmar fingertip (diameter 10×10 mm), at the dorsal and palmar centre of each hand (diameter 20×20 mm), at the thenar eminence of each hand, (diameter 38×72 mm), and at the hypothenar eminence of each hand (diameter 31×75 mm). (A) Image of the dorsal and palmar thermography of the hands from the same participant with fibromyalgia. (B) Image of the dorsal and palmar thermography of the hands from the same healthy control participant.

3.1.3.2 Core Body Temperature Assessment.

The core body temperature was measured in the external auditory canal with an infrared thermometer (Infrared Dermal Thermometers, Exergen). This technique provides an accurate measurement of the core temperature due to the relationship between the tympanic artery and the hypothalamus and has a sensitivity of 91% and a specificity of 90% (Bijur et al., 2016; Gasim et al., 2013). The participants' axillary temperature was also taken, given that it also reflects the body temperature (Gasim et al., 2013; Lodha et al., 2000).

3.1.3.3 Nitric Oxide Measurement.

Nitric oxide was measured in serum samples. To obtain serum, blood samples were extracted by venipuncture of the cephalic vein and collected in special tubes for serum separation (Vacutainer SST II Advance, Dickinson and Company, Franklin Lakes, NJ). Afterwards, the blood tubes stood at room temperature for 1 hour until the blood clotted. Then, the tubes were centrifuged for 10 minutes at 2000g (Avanti J-30I; Beckman Coulter, Inc, Brea, CA). Supernatant was collected, aliquoted, and kept at –80°C until used. We indirectly quantified the NO production as nitrates, nitrites and S-nitroso compounds (NOx) using an ozone chemiluminescence-based method (MacArthur et al., 2007). The total amount of NOx was determined by modifying the procedure described by Braman and Hendrix (López-Ramos et al., 2005) using the purge system of Sievers Instruments (model NOA 280i, GE, Analytical Instruments, CO). The final NOx values were referred to the total protein concentration in the initial extracts (MacArthur et al., 2007).

3.1.4 Statistical Analysis.

We performed the statistical analysis using SPSS Statistics Version 24 for Windows (IBM Corporation, Armonk, NY, USA), employing the Kolmogorov-Smirnov test to analyze the normality and distribution of the variables (P>0.05). We employed an unpaired Student's t-test with a 95% confidence interval (95% CI; α=0.05) for the continuous data to compare the differences in means between the groups for the demographic and clinical data. We performed a two-way analysis of covariance (ANCOVA) to assess the main study objective. The key variables were the maximum, minimum and mean temperatures on the dorsal and palmar sides of both hands (dominant and non-dominant) at each point, while age and body mass index (BMI) were employed as covariates. We employed linear regression to test the interactions between serum NO levels and the temperature of the skin surface of both hands and the tympanic and axillary temperatures. The results are reported as the percentage change (β) with 95% CI. Statistical significance was set at P<0.05.

We performed the sample size calculation using NCSS-PASS software. According to a previous study by Brusselmans et al. (2015), we estimated a sample size of 40 patients with FMS and 40 healthy controls would provide a 95% CI, a power of 80%, and an alpha level (α) of 0.05. The sample size was increased to 225 participants after taking into account an expected loss rate of approximately 65%.

3.2 Study II: "Associations Among Nitric Oxide and Enkephalinases With Fibromyalgia Symptoms".

The methods section of the Study II, added in the PhD thesis, is summarized and described below. This study has been accepted for publication in the scientific journal *Nursing Research* (Impact Factor JCR: 1.881; Category NURSING-SCIE (29/123); Q1/T1). Table 2 shows a summary of the methodology employed in this observational case study.

Table 2. Summary of Material and Methods in Study II.

PAPER	STUDY DESING	PARTICIPANTS	PROCEDURES	MAIN VARIABLES	METHODS
Associations Among Nitric Oxide and Enkephalinases With Fibromyalgia Symptoms.	Observational case study.	• Women diagnosed with FMS (n=58).	• Screening to inclusion selection criteria. • Sociodemographic and clinical characteristics were recorded: - Age. - Sex. - Height. - Weight. - Menopause status. - Drug history (morphine, tramadol, oxycodone, naltrexone, etc.) • Associated chronic disease: - Diabetes mellitus. - Arterial hypertension. - Cancer.	• Pain magnitude. • Pain threshold. • Global level of pain. • Pressure pain thresholds. • Central sensitization syndrome. • Severity and impact of FMS. • Anxiety. • Blood samples collection. • Nitric oxide levels. • EDA activity. • Oxytocinase activity.	• Pain Matcher® device (Cefar-Compex Scandinavia Inc, Medical AB, Lund, Sweden). • 100 mm Visual analogue scale (VAS). • Digital pressure algometer device FDIXTM (Wagner Instruments, Greenwich, CT, USA). • Central Sensitization Inventory (CSI). • Spanish version of the Revised Fibromyalgia Impact Questionnaire (FIQ-R). • Beck Anxiety Inventory (BAI). • Blood from antecubital vein into an ethylenediaminetetraacetic acid tube (BD Vacutainer® LH PST II Advance, Ref. 367374, Becton Dickinson, New Jersey, USA). • Purge system of Sievers Instruments (model NOA 280i, GE, Analytical Instruments, Colorado, USA). • Fluorometrically in triplicate in serum samples using tyrosyl-ß-naphthylamide (TyrNNap) and cystyl-ß-naphthylamide (CysNNap) as the substrates.

FMS: Fibromyalgia syndrome; EDA: enkephalin-degrading aminopeptidase.

3.2.1 Sample.

Fifty-eight patients with FM were recruited to participate in this observational case study by contacting two Associations of Fibromyalgia patients in January 2018. We selected the FM patients based on the inclusion and exclusion criteria proposed and according to the demographic and clinical data they provided in their first visit to our laboratories of the Faculty of Health Sciences of the Universities of Granada and Jaén in February 2018.

The inclusion criterion was that the patients had been previously diagnosed with FM by a professional rheumatologist of the Public Health System of Andalucía (Spain) and met the 1990 American College of Rheumatology (ACR) criteria for the classification of primary FM. The exclusion criteria were being under 18 and over 70 years old, male sex, presence of other chronic disease (diabetes mellitus, hypertension, or cancer), presence of cardiac, renal or hepatic insufficiency, severe physical disability, psychiatric illness, previous history of surgery, pregnancy or lactation, and treatment with vasoactive drugs, anticoagulants, corticosteroids, or oestrogens. Moreover, no woman was being treated with enkephalinase inhibitors or agonist / antagonist opioid receptors (morphine, tramadol, oxycodone, naltrexone, etc).

3.2.2 Procedures.

The patients provided their demographic (age, sex, height, weight, and menopause status) and clinical data by filling out questionnaires regarding their medical history. In a second visit to our laboratories between March 2018 and April 2018, blood samples were taken to the selected FM patients and then they completed several questionnaires. The same interviewer helped the FM patients to complete all the questionnaires and recorded each response.

The Ethical Committee of Research of the Andalusian Health Service of Granada approved this research (approval number: 1718-N-18). This study was conducted in accordance with the Declaration of Helsinki 2013 of the World Medical Association (WMA). The informed consent was signed by all the participants and no participant received financial incentive.

3.2.3 Measures.

3.2.3.1 Clinical Features and Pain Variables Assessment of Women with FM.

Both pain threshold and pain magnitude were evaluated using the Pain Matcher (Cefar-Compex Scandinavia Inc, Medical AB, Lund, Sweden). The device consists in the application of an electric current among the thumb and index finger with three recordings for each outcome (pain threshold and pain magnitude) with a score ranging from 0 to 60 (Alstergren & Förström, 2003). The Pain Matcher device has demonstrated to have good test-retest reliability (95% Confident Interval=0.39-0.14) (Lund et al., 2005). To assess the global level of pain, we used a Visual Analog Scale (VAS) of 100 mm, which shows a high sensibility and specificity in the FM population (Marques et al., 2008). For pain magnitude and VAS, higher values reflect worse symptomatology. For pain threshold, lower values reflect worse symptomatology.

To measure the pressure pain thresholds (PPTs) over the 11 locomotor points considered by the ACR for FM diagnosis, a digital pressure algometer device (Wagner Instruments, Greenwich, CT, USA) was used. The PPTs were obtained by increasing the pressure to a velocity of 1 kg/sec provided by the algometer of a bilaterally way over each point until the sensation of pressure turned painful (Fernández-Lao et al., 2016). We calculated the mean of three trials in each point and we used it for the main analysis. A 30-second resting period was allowed between each recording. The PPTs have reported good test-retest reliability the same day and four-day after (Intraclass Correlation Coefficients were 0.91 and 0.94-0.97, respectively) (Chesterton et al., 2007; Jones et al., 2007).

The symptoms associated with Central Sensitization Syndrome (CSS) were assessed using the Spanish version of the Central Sensitization Inventory (CSI) (Cuesta-Vargas et al., 2016), which has an internal consistency (Cronbach's alpha) of 0.88 (Neblett et al., 2013). This survey contains 25 self-assessment items and uses a five-point Likert scale, with a score ranging from 0 (means "never") to 4 (means "always") points. The total score is 100 and the cut-off score for central sensitization diagnosis is 40, with higher scores reflecting a major severity of symptoms (Neblett et al., 2017).

To measure the severity and impact of FM, we used the Spanish version of the Revised Fibromyalgia Impact Questionnaire (FIQ-R) (Salgueiro et al., 2013). This questionnaire presents three subscales (activity level [FIQ-R.1], overall impact [FIQ-R.2] and intensity of symptoms [FIQ-R.3]) with a total of 21 items, where higher scores indicate a greater impact (the score ranges from 0 to 100). The Spanish version of the FIQ-R has shown a high internal consistency value, with a Cronbach's alpha of 0.91 (Salgueiro et al., 2013).

Finally, the psychological aspects and common symptoms of anxiety were evaluated using a Spanish version of the Beck Anxiety Inventory (BAI) (Sanz & Navarro, 2003), which has a Cronbach's alpha of 0.93 (Magán et al., 2008). The BAI consists of 21 items and the score ranges from 0 to 63, with higher scores indicating higher levels of anxiety.

3.2.3.2 Preparation of Blood Samples and Measurement of Serum NO Levels, EDA Activity and Oxytocinase Activity.

The blood samples were collected in the early morning after 12 hours fast immediately before all the questionnaires were completed. We drew the blood from the antecubital vein into an ethylenediaminetetraacetic acid tube (BD Vacutainer® LH PST II Advance, Ref. 367374, Becton Dickinson, New Jersey, USA). To avoid circadian variations in the level of the biochemical parameters measured, the blood samples were collected at the same time of day and by the same practitioner (Kanabrocki et al., 2001). After blood collection, the tubes clotted for 30 min at a room temperature and then were centrifuged at 3,500 rpm (Avanti J-30I; Beckman Coulter, California (CA), USA) for 5 min at 4ºC to obtain serum samples.

NO levels were indirectly quantified as metabolites of NO (NOx) by means of an ozone chemiluminiscence-based method. We determined the total amount of NOx using the purge system of Sievers Instruments, model NOA 280i (GE, Analytical Instruments, Colorado, USA) according to a modification (Rus et al., 2010) of the procedure described by Braman and Hendrix (Braman & Hendrix, 1989). The final NOx values were referred to the total protein concentration in the initial extracts, quantified via the Bradford method (Bradford, 1976).

We measured the specific EDA (EC. 3.4.24.11) and oxytocinase (EC. 3.4.11.3) activities fluorometrically in triplicate in serum samples using tyrosyl-ß-naphthylamide (TyrNNap) and cystyl-ß-naphthylamide (CysNNap) as the substrates, respectively, and according to the methods described previously (Carrera et al., 2004; García-López et al., 2003). The amount of substrate released as a result of the EDA and oxytocinase activities was measured at 412 nm emission wavelength with an excitation wavelength of 345 nm. The final activity values were referred to the total protein concentration, quantified using the Bradford method (Bradford, 1976).

3.2.4 Statistical Analyses.

The blood samples were collected in the early morning after 12 hours fast immediately before all the questionnaires were completed. We drew the blood from the antecubital vein into an ethylenediaminetetraacetic acid tube (BD Vacutainer® LH PST II Advance, Ref. 367374, Becton Dickinson, New Jersey, USA). To avoid circadian variations in the level of the biochemical parameters measured, the blood samples were collected at the same time of day and by the same practitioner (Kanabrocki et al., 2001). After blood collection, the tubes clotted for 30 min at a room temperature and then were centrifuged at 3,500 rpm (Avanti J-30I; Beckman Coulter, California (CA), USA) for 5 min at 4ºC to obtain serum samples.

NO levels were indirectly quantified as metabolites of NO (NOx) by means of an ozone chemiluminiscence-based method. We determined the total amount of NOx using the purge system of Sievers Instruments, model NOA 280i (GE, Analytical Instruments, Colorado, USA) according to a modification (Rus et al., 2010) of the procedure described by Braman and Hendrix (Braman & Hendrix, 1989). The final NOx values were referred to the total protein concentration in the initial extracts, quantified via the Bradford method (Bradford, 1976).

We measured the specific EDA (EC. 3.4.24.11) and oxytocinase (EC. 3.4.11.3) activities fluorometrically in triplicate in serum samples using tyrosyl-ß-naphthylamide (TyrNNap) and cystyl-ß-naphthylamide (CysNNap) as the substrates, respectively, and according to the methods described previously (Carrera et al., 2004; García-López et al., 2003). The amount of substrate released as a result of the EDA and oxytocinase activities was measured at 412 nm emission wavelength with an excitation wavelength of 345 nm. The final activity values were referred to the total protein concentration, quantified using the Bradford method (Bradford, 1976).

3.3 Study III: "Dietary Inflammatory Index Scores Are Associated with Pressure Pain Hypersensitivity in Women with Fibromyalgia".

The methods section of the Study III, added in the PhD thesis, is summarized and described below. This study has been published in the scientific journal *Pain Medicine* (Impact Factor JCR: 2.513; Category MEDICINE, GENERAL & INTERNAL (51/165); Q2/T1). Table 3 shows a summary of the methodology employed in this cross-sectional study.

Table 3. Summary of Material and Methods in Study III.

PAPER	STUDY DESING	PARTICIPANTS	PROCEDURES	MAIN VARIABLES	METHODS
Dietary Inflammatory Index Scores Are Associated with Pressure Pain Hypersensitivity in Women with Fibromyalgia.	Controlled cross-sectional study. Cases and controls matched by menopause status.	• Women diagnosed with FMS (n=95). • Healthy women (n=98).	• Screening to inclusion selection criteria. • Sociodemographic and clinical characteristics were recorded: - Age. - Height. - Weight. - Menopause status - Drug history. - Smoking history. • Associated chronic disease: - Diabetes mellitus. - Arterial hypertension. - Cancer.	• Dietary habits. • Anti-inflammatory and pro-inflammatory dietary profiles. • Global level of pain. • Impact of FMS symptoms on the physical and mental health. • Severity of fatigue. • Quality of sleep. • Common anxiety symptoms. • Central sensitization. • Pressure pain thresholds (PPTs).	• Face-to-face interview to recall food preceding 24 hours. • Dietary Inflammatory Index (DII®). • 100 mm Visual analogue scale (VAS). • Spanish version of the Revised Fibromyalgia Impact Questionnaire (FIQ-R). • Multidimensional Fatigue Inventory (MFI). • Pittsburgh Sleep Quality Index (PSQI). • Beck Anxiety Inventory (BAI). • Central Sensitisation Inventory (CSI). • Digital pressure algometer device FDIXTM (Wagner Instruments, Greenwich, CT, USA).

FMS: Fibromyalgia syndrome.

3.3.1 Study Design and Participants.

Between January 2018 and July 2018, 95 women diagnosed with FMS and 98 menopause-status matched healthy female controls, all aged between 30 and 70 years, were enrolled in this case-control study. Menopausal status was matched because the menopausal transition is associated with an increased risk of anxiety (Bromberger et al., 2013) and midlife women approaching and passing through the menopause are more likely to suffer sleep disturbances (Shaver & Zenk, 2000). We wrote to the two fibromyalgia associations in Spain (AGRAFIM and AFIXA) who agreed to help us to identify, approach, and recruit women with FMS to participate in this study. The controls were volunteers recruited from among the friends, relatives, and colleagues of the patients or from the Faculty of Health Sciences at the University of Granada.

The criteria for inclusion of patients with FMS in this study were: (1) a diagnosis of FMS from a rheumatology specialist; (2) the presence of symptoms consistent with the 1990 American College of Rheumatology criteria for FMS; (3) the absence of any acute or terminal illnesses. The exclusion criteria for entire sample cohort were: (1) a history of drug or alcohol abuse; (2) women who were pregnant or breastfeeding; (3) use of vasoactive drugs, contraceptives, anticoagulants, or antithrombotic therapies; (4) participants with an active tumour. A total of 225 individuals were approached to participate in the study and were screened for eligibility; after applying the inclusion and exclusion criteria, a total of 193 women were finally included.

3.3.2 Procedures.

The study was explained to each person and any questions they had were answered. We then obtained their informed consent to participation in this research, which was performed in strict compliance with the international code of medical ethics established by the World Medical Association and the Declaration of Helsinki. The study was also approved by the Ethics Committee at the University of Granada. Each participant completed a structured questionnaire which included items about their medical history and menopausal status. There were no financial incentives for participation in this study, but every participant received information promoting healthy eating habits.

3.3.3 Measures.

3.3.3.1 Pain and Clinical Manifestations Assessment.

3.3.3.1.1 Visual Analog Scale for Pain.

The patients' global level of pain was assessed using the visual analogue scale (VAS) pain score (0–100 mm, with higher scores indicating more pain). The VAS has been shown to be an important instrument in evaluating pain levels and is sensitive and specific in the assessment of pain among patients with FMS (Marques et al., 2008).

3.3.3.1.2 Fibromyalgia Impact Questionnaire.

The Spanish version of the Fibromyalgia Impact Questionnaire (FIQ-R) was used to assess the effect of FMS symptoms on the physical and mental health of patients. This self-reported questionnaire comprises 21 items which assess physical impairment, the number of days feeling good, amount of work missed, ability to work, and levels of pain, fatigue, rest, stiffness, anxiety, and depressive symptoms. The total score is the sum of all the subscales (0 to 100 points) and higher scores indicate a more negative impact (Salgueiro et al., 2013).

3.3.3.1.3 Multidimensional Fatigue Inventory.

The Multidimensional Fatigue Inventory (MFI) was used to evaluate the severity of fatigue among patients with FMS (Segura-Jiménez et al., 2016). This questionnaire contains five subscales: general fatigue, physical fatigue, mental fatigue, reduced activity, and reduced motivation. Each subscale includes four questions scored from 1 to 5 points, with higher scores indicating a higher degree of fatigue. The test-retest analysis of reliability for the MFI showed excellent correlation between the domains, ranging from 0.64 to 0.91 (Munguía-Izquierdo et al., 2012).

3.3.3.1.4 Pittsburgh Sleep Quality Index.

The Pittsburgh Sleep Quality Index (PSQI) comprises 24 items and was used to evaluate the participants' quality of sleep; they responded to 19 of these items and a person living in the same home (or hospital room) responded to the remaining 5. The PSQI evaluates 7 subdimensions: subjective quality of sleep, and sleep latency, sleep duration, sleep efficiency, sleep disturbances, sleep medication, and diurnal dysfunction. Each dimension is scored from 0 points (no problem) to 3 points (serious problem); the total score ranges from 0 to 21 points and higher scores represent poorer sleep quality (Tuba Tülay Koca et al., 2016). A previous study showed that the PSQI has good reliability and has a Cronbach alpha of 0.805 (Hita-Contreras et al., 2014).

3.3.3.1.5 Beck Anxiety Inventory.

The Beck Anxiety Inventory (BAI) was used to evaluate the psychological aspects and common symptoms of anxiety (Serdaroğlu Beyazal et al., 2018); this questionnaire contains 21 items that assess the severity of patient anxiety, with a score range from 0 points (no anxiety) to 3 points (high levels of anxiety). The total score ranges from 0 to 63 points, and higher scores indicate a higher degree of anxiety (35). The test-retest reliability analysis of the BAI showed that it has excellent internal consistency with Cronbach alpha of 0.91 (Vázquez Morejón et al., 2014).

3.3.3.1.6 Central Sensitization Inventory.

The Spanish version of the Central Sensitisation Inventory (CSI) was used to assess the frequency of health-related symptoms associated with central sensitivity syndromes (Cuesta-Vargas et al., 2016). The CSI is a 25-item survey and individuals were asked to rate their answer to each question on a 5-point Likert scale, with 0 meaning 'never' and 4 meaning 'always'. The CSI score is obtained by summing the responses, with a total possible score of 100; higher CSI scores represent greater self-reported symptomology. The test-retest reliability analysis of this questionnaire showed a correlation of 0.91 (Cuesta-Vargas et al., 2016).

3.3.3.1.7. Pressure Pain Threshold.

Pressure algometry is a quantitative method commonly used in clinical practice to assess tenderness (Chesterton et al., 2007). The pressure pain threshold (PPT) is defined as the minimum amount of pressure required for a feeling of pressure to first change to one of pain (Vanderweeën et al., 1996). A digital pressure algometer was used to bilaterally measure the PPT at the 18 tender points defined by the American College of Rheumatology for a FMS diagnosis: the occiput, trapezius, zygapophyseal joint, supraspinatus, second rib, epicondyle, gluteus, greater trochanter, and knee. The device comprises a 1 cm2 rubber disk attached to a strain gauge which displays values in kPa (Storz Medical AG, Tagerwilen, Switzerland). The PPTs of the participants were determined by gradually increasing the pressure provided by the algometer (at a rate of 1 kg/s) until the sensation reported by the individual first became painful (participants were instructed to say 'stop' at this point). The mean of three trials was calculated and used for the main analysis and a 30-second resting period was allowed between each recording. The same-day (Chesterton et al., 2007) and four-day (Jones et al., 2007) reliability of pressure algometry is high (intraclass correlation coefficients = 0.91 and 0.94 to 0.97, respectively).

3.3.3.2 Dietary Assessment.

Dietary habits were assessed using a face-to-face interview with trained investigators who asked the participants to recall all the food they had consumed in the preceding 24 hours, including nutritional supplements and beverages. When necessary, standard household measurements and pictorial food models were used to define the quantities consumed. These food records were subsequently studied using nutrient-analysis software (Nutriber 1.1.5).

3.3.3.2.1 The Dietary Inflammatory Index.

The Dietary Inflammatory Index (DII®) is based on the literature published up until 2010 linking diet to inflammatory markers (Shivappa et al., 2014). To calculate the DII® score for our study participants, we first linked their dietary data to the regionally-representative world database to obtain a robust mean and standard deviation estimate for each parameter. These then became the multipliers for expressing each individual's exposure relative to the 'standard global mean' as a z-score (by subtracting the standard global mean from the amount reported by each participant and dividing this value by the standard deviation). To minimise the effect of 'right skewing', this value was then converted to a centred percentile score by multiplying it by the respective food-parameter effect score derived from the original literature review (Shivappa et al., 2014) for each food parameter and each individual to obtain food-parameter specific DII® scores; these were then summed to obtain each participant's global DII® score. Overall, 23 food parameters including beta carotene, carbohydrates, cholesterol, energy, fibre, folate, iron, magnesium, monounsaturated fatty acid, niacin, polyunsaturated fatty acid, protein, riboflavin, saturated fatty acid, thiamine, total fat, vitamin A, vitamin B12, vitamin B6, vitamin C, vitamin D, vitamin E, and zinc were used to calculate the global DII® scores. Lower scores represent more anti-inflammatory dietary profiles and higher scores more pro-inflammatory profiles.

3.3.4 Statistical Analysis.

The data were analysed with SPSS© version 22.0 (IBM Corporation, Armonk, NY, USA). The Kolmogorov–Smirnov test was used to verify the normality of the data distribution and data were expressed as the mean ± the standard deviation (SD). To compare two groups, Mann–Whitney U tests were used, Student t-tests were employed for continuous data, and $\chi 2$ tests for categorical data. The DII® scores were analysed both as a

continuous and a categorised variable based on their quartiles. Linear regression analyses were used to determine the association between the continuous DII® scores, the PPTs of tender-point sites and, clinical symptoms adjusted for the following confounding factors: age, menopausal status, and overall energy levels. The results are reported as the percentage change (β) with 95% confidence intervals (95% CI). In the categorised DII® score analysis, generalised linear models were developed by including the mean value of each DII® quartile, the PPTs of tender-point sites, and clinical symptoms after adjustment for the same aforementioned confounding factors. Comparisons of nutrient intake across the DII® quartiles were analysed by one-way ANOVA. Probabilities exceeding 95% (alpha p-values < 0.05) were used as the threshold cut-off for statistical significance.

RESULTADOS

RESULTS

4. RESULTADOS / RESULTS

4.1 Study I: "Evaluation of sympathetic adrenergic branch of cutaneous neural control throughout thermography and its relationship to nitric oxide levels in patients with Fibromyalgia".

The results section of the Study I is showed below.

4.1.1 Demographic and Clinical Data.

Table 1 shows the participants' demographic and clinical data. A total of 42 women diagnosed with FMS (mean age, 56.45±6.58 years) and 52 healthy women (mean age, 57.15±10.52 years) met the inclusion criteria. The women diagnosed with FMS showed a significantly higher weight and BMI than the healthy women ($P \leq 0.003$). There were no significant differences in terms of age, height and serum NO levels between groups ($P \geq 0.248$). The results of the ANCOVA showed significant differences between the groups for tympanic temperature ($F=10.706$, $P=0.002$). The participants with FMS showed higher core body temperatures (Table 1) and a mean total score on the FIQ-R of 72.48±12.73.

4.1.2 Temperature of the Dorsal Fingertips and Dorsal Centre of the Hands.

The ANCOVA showed statistically significant differences between the groups for the variables minimum, maximum and mean temperature at the following dorsal sites of both hands: tip of the thumb (dominant: $F \geq 97.787$, $P \leq 0.001$; non-dominant: $F \geq 114.285$, $P \leq 0.001$); index fingertip (dominant: $F \geq 110.460$, $P \leq 0.001$; non-dominant: $F \geq 122.228$, $P \leq 0.001$); middle fingertip (dominant: $F \geq 128.550$, $P \leq 0.001$; non-dominant: $F \geq 129.516$, $P \leq 0.001$); ring fingertip (dominant: $F \geq 135.768$, $P \leq 0.001$; non-dominant: $F \geq 111.077$, $P \leq 0.001$); pinkie fingertip (dominant: $F \geq 130.445$, $P \leq 0.001$; non-dominant: $F \geq 112.445$, $P \leq 0.001$); and dorsal centre (dominant: $F \geq 64.851$, $P \leq 0.001$; non-dominant: $F \geq 67.330$, $P \leq 0.001$). The ANCOVA also revealed a significant effect for the covariate age for the minimum temperature at the dorsal side of the tip of the thumb (non-dominant: $F=4.003$, $P=0.048$). **Figure 1** shows the mean temperature of the dorsal side of the dominant and non-dominant hands of the women with fibromyalgia and those of the healthy controls.

Table 1. Demographic and clinical characteristics of the women with fibromyalgia syndrome and the healthy women.

	Women with FMS (n=42)	**Healthy women (n=52)**	***P*-value**
Age (years)	56.45±6.58	57.15±10.52	0.707
Height (cm)	159.12±5.67	157.63±6.51	0.248
Weight (kg)	72.76±12.48	65.58±10.34	0.003*
BMI (kg/m^2)	28.88±5.70	26.29±4.04	0.012*
NOx (µmol/mg protein)	27.88±21.44	29.81±18.13	0.637
Tympanic temperature ºC	36.03±0.68	35.62±0.58	0.002*
Axillary temperature ºC	35.65±0.53	35.58±0.63	0.341
FIQ-R			
FIQ-R.1	20.00±4.79	-	-
FIQ-R.2	13.79±4.46	-	-
FIQ-R.3	38.44±5.75	-	-
Total score	72.48±12.73	-	-

* Significance level $P<0.05$.

Note. Data are expressed as mean ± standard deviation (SD). FMS: Fibromyalgia Syndrome; BMI: body mass index; NOx: nitric oxide metabolites; FIQ-R= revised Fibromyalgia Impact Questionnaire; FIQ-R.1= activity level of the FIQ; FIQ-R.2= overall impact of the FIQ-R; FIQ-R.3= intensity of symptoms of the FIQ-R.

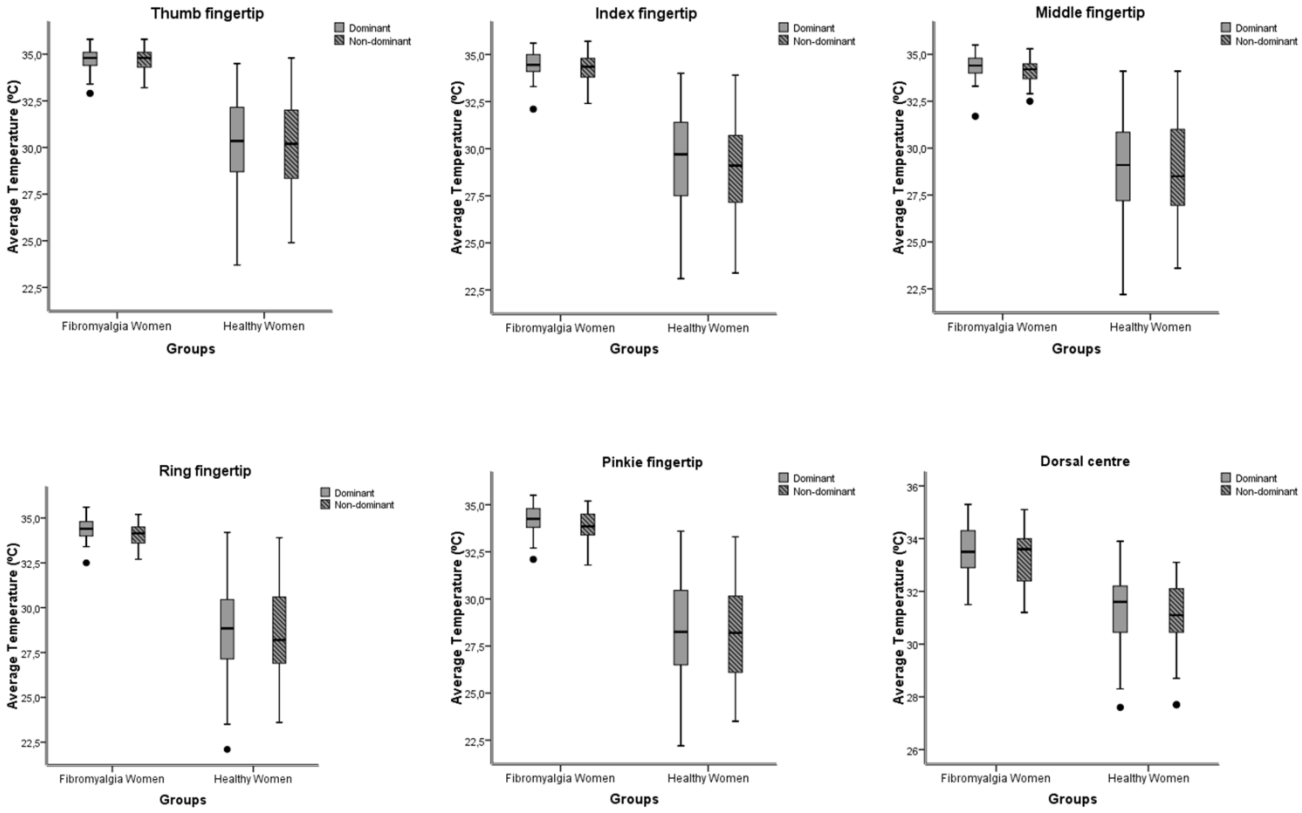

Figure 1. Box plots of mean temperature of the dorsal site of the hands for the women with fibromyalgia and the healthy controls.

The box plots show the mean temperature (ºC) from each point (tip of the thumb, index fingertip, middle fingertip, ring fingertip, pinkie fingertip, dorsal centre) of the dominant and non-dominant dorsal site of both hands between the women diagnosed with fibromyalgia and the healthy controls. In the box plots, the boundary of the box closest to zero indicates the 25th percentile, the black line within the box marks the median, and the boundary of the box farthest from zero indicates the 75th percentile. Whiskers above and below the box indicate the 10th and 90th percentiles.

4.1.3 Temperature of the Palmar Fingertips and Palmar centre of the Hands.

The ANCOVA showed statistically significant differences between the groups for the variables minimum, maximum and mean temperature at the following palmar sites of both hands: tip of the thumb (dominant: $F \geq 114.536$, $P \leq 0.001$; non-dominant: $F \geq 95.807$, $P \leq 0.001$); index fingertip (dominant: $F \geq 113.901$, $P \leq 0.001$; non-dominant: $F \geq 113.147$, $P \leq 0.001$); middle fingertip (dominant: $F \geq 150.888$, $P \leq 0.001$; non-dominant: $F \geq 122.583$, $P \leq 0.001$); ring fingertip (dominant: $F \geq 125.304$, $P \leq 0.001$; non-dominant: $F \geq 128.308$, $P \leq 0.001$); pinkie fingertip (dominant: $F \geq 78.065$, $P \leq 0.001$; non-dominant: $F \geq 96.556$, $P \leq 0.001$); centre (dominant: $F \geq 107.449$, $P \leq 0.001$; non-dominant: $F \geq 128.583$, $P \leq 0.001$); thenar eminence (dominant: $F \geq 84.179$, $P \leq 0.001$; non-dominant: $F \geq 118.286$, $P \leq 0.001$); and hypothenar eminence (dominant: $F \geq 133.310$, $P \leq 0.001$; non-dominant: $F \geq 90.660$, $P \leq 0.001$). The ANCOVA also revealed a significant effect for the covariate age for the maximum temperature at the palmar side of the tip of the thumb (dominant: $F=4.192$, $P=0.044$; non-dominant: $F=4.502$, $P=0.037$); the minimum temperature at the palmar side of the tip of the thumb (dominant: $F=5.722$, $P=0.019$; non-dominant: $F=7.024$, $P=0.009$); the mean temperature of the palmar side of the tip of the thumb (dominant: $F=4.670$, $P=0.033$; non-dominant: $F=4.674$, $P=0.033$); maximum temperature of the palmar centre (dominant: $F=5.986$, $P=0.016$; non-dominant: $F=7.007$, $P=0.010$); and mean temperature of the palmar centre (non-dominant: $F=4.771$, $P=0.032$). **Figure 2** shows the mean temperature of the palmar site of the dominant and non-dominant hands for the women with FMS and that of the healthy controls.

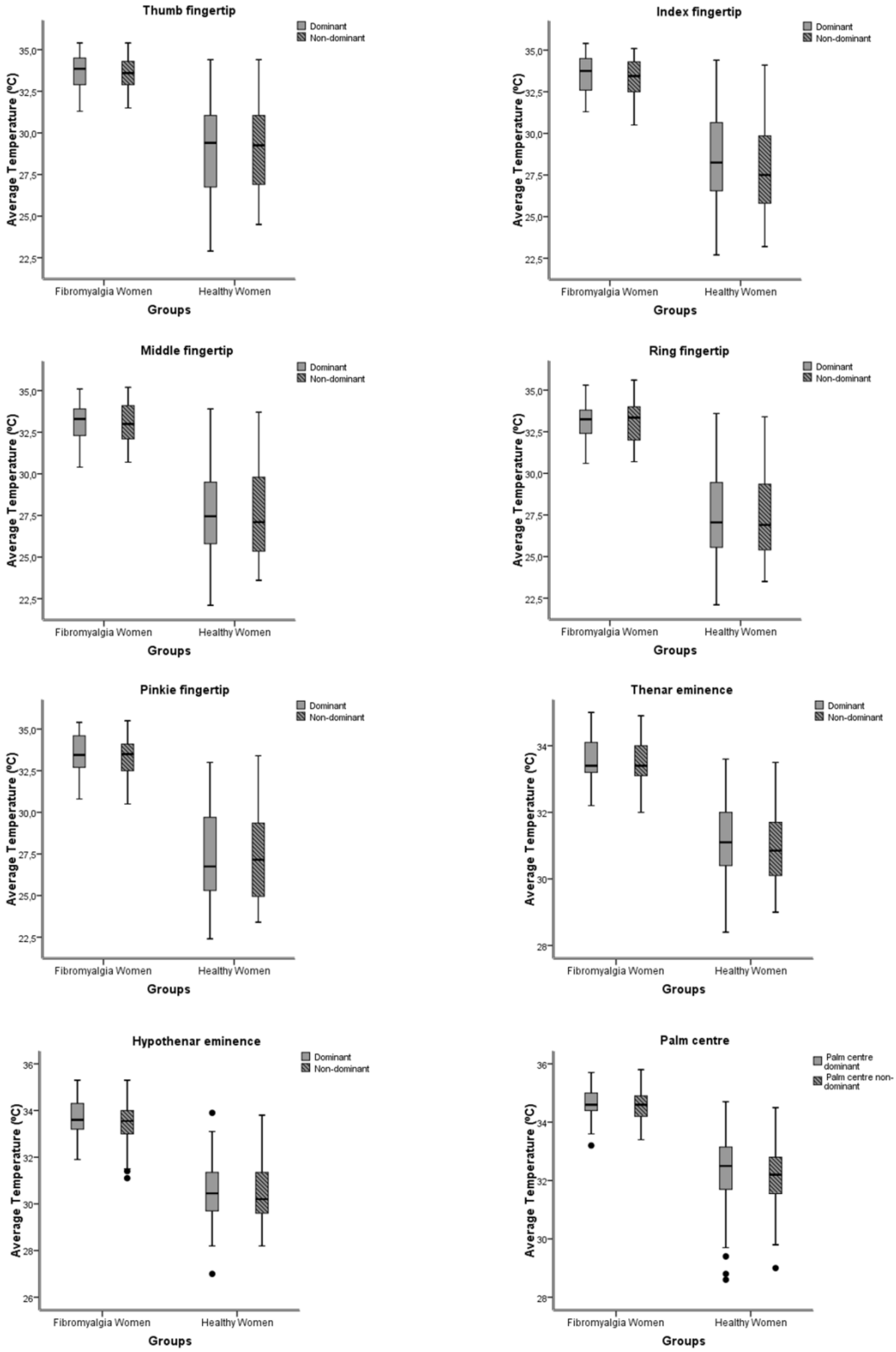

Figure 2. Box plots of mean temperature of the palmar site of the hands for the women with fibromyalgia and the healthy controls.

The box plots show the mean temperature (ºC) from each point (tip of the thumb, index fingertip, middle fingertip, ring fingertip, pinkie fingertip, thenar eminence, hypothenar eminence, palmar centre) of the dominant and non-dominant palmar site of both hands between the women diagnosed with fibromyalgia and the healthy controls. In the box plots, the boundary of the box closest to zero indicates the 25th percentile, the black line within the box marks the median, and the boundary of the box farthest from zero indicates the 75th percentile. Whiskers above and below the box indicate the 10th and 90th percentiles.

4.1.4 Association Between Thermography Image Analysis and Serum Nitric Oxide Levels.

Linear regression analysis indicated that the minimum temperature of the dorsal centre of the dominant hand (β=-3.501: 95% CI 6.805, 0.198: P=0.038); the maximum temperature of the palmar centre of the non-dominant hand (β=-5.594: 95% CI 10.106, 1.081: P=0.016); the minimum temperature of the palmar centre of the non-dominant hand (β=-4.090: 95% CI 7.905, 0.275: P=0.036); the mean temperature of the palmar centre of the non-dominant hand (β=-5.519: 95% CI 9.933, 1.106: P=0.015); and the maximum temperature of the thenar eminence of the dominant hand (β=-5.800: 95% CI 10.508, 1.092: P=0.017) were significantly associated with NO levels after adjusting for age, menopausal state and BMI in the controls but not for the women diagnosed with FMS. There were no significant differences between serum NO levels and the other temperature variables between the cases and controls **(Table 2)**.

4.1.5 Association Between Tympanic and Axillary Temperature and Serum Nitric Oxide Levels.

Lastly, the linear regression analysis for the tympanic and axillary temperature and serum NO levels showed that only tympanic temperature (β=-9.321; 95% CI 17.974, 0.669; P=0.035) was significantly associated with NO levels after adjusting for age, menopause state and BMI in the control group **(Table 3)**.

Table 2. Associations between nitric oxide levels and temperature of dorsal and palm site of both hands among the cases (women with fibromyalgia syndrome) and controls (healthy women).

			NOx Levels					
			FMS Women (n=42)			Healthy Women (n=52)		
			β	95 % CI	*P*-value	β	95 % CI	*P*-value
Dorsal site of the hand								
	Maximum ºC	**D**	7.313	(-6.641, 21.266)	0.295	-0.827	(-2.857, 1.203)	0.416
		ND	7.676	(-5.765, 21.117)	0.255	-1.350	(-3.457, 0.757)	0.204
Thumb Fingertip	**Minimum ºC**	**D**	3.479	(-3.845, 10.803)	0.342	-0.656	(-2.680, 1.367)	0.517
		ND	4.671	(-4.889, 14.232)	0.329	-1.291	(-3.518, 0.936)	0.249
	Average ºC	**D**	6.240	(-5.247, 17.728)	0.278	-0.799	(-2.802, 1.204)	0.427
		ND	6.073	(-5.584, 17.729)	0.298	-1.339	(-3.498, 0.820)	0.218
	Maximum ºC	**D**	4.653	(-5.737, 15.044)	0.370	-1.154	(-2.913, 0.605)	0.193
		ND	2.604	(-6.444, 11.652)	0.563	-1.172	(-3.110, 0.767)	0.230
Index Fingertip	**Minimum ºC**	**D**	3.271	(-5.709, 12.250)	0.465	-1.044	(-2.948, 0.860)	0.276
		ND	5.156	(-1.674, 11.985)	0.135	-0.851	(-2.890, 1.187)	0.405
	Average ºC	**D**	3.966	(-6.758, 14.690)	0.458	-1.156	(-2.960, 0.648)	0.204
		ND	2.018	(-7.133, 11.168)	0.658	-1.171	(-3.165, 0.823)	0.243
	Maximum ºC	**D**	3.690	(-7.362, 14.741)	0.503	-1.577	(-3.395, 0.240)	0.087
		ND	3.676	(-7.393, 14.745)	0.505	-1.617	(-3.500, 0.266)	0.091
Middle Fingertip	**Minimum ºC**	**D**	3.314	(-5.427, 12.054)	0.447	-1.418	(-3.272, 0.435)	0.130
		ND	1.533	(-7.991, 11.056)	0.746	-1.653	(-3.550, 0.245)	0.086
	Average ºC	**D**	3.390	(-7.686, 14.466)	0.539	-1.535	(-3.368, 0.297)	0.099
		ND	4.154	(-7.144, 15.453)	0.461	-1.654	(-3.551, 0.244)	0.086

(Continued) Table 2. Associations between nitric oxide levels and temperature of dorsal and palm site of both hands among the cases (women with fibromyalgia syndrome) and controls (healthy women).

			NOx Levels					
			FMS Women (n=42)			Healthy Women (n=52)		
			β	95 % CI	*P*-value	β	95 % CI	*P*-value
Dorsal site of the hand								
	Maximum ºC	**D**	2.912	(-8.256, 14.080)	0.600	-0.775	(-2.572, 1.021)	0.390
		ND	2.855	(-7.320, 13.030)	0.573	-1.268	(-3.179, 0.644)	0.189
Ring Fingertip	**Minimum ºC**	**D**	-2.268	(-11.418, 6882)	0.618	-0.529	(-2.413, 1.355)	0.575
		ND	-4.608	(-10.990, 1.775)	0.152	-1.154	(-3.082, 0.773)	0.234
	Average ºC	**D**	1.431	(-10.952, 13.813)	0.816	-0.724	(-2.570, 1.122)	0.434
		ND	0.102	(-11.926, 12.129)	0.986	-1.277	(-3.212, 0.658)	0.191
	Maximum ºC	**D**	3.002	(-6.970, 12.974)	0.546	-0.348	(-2.176, 1.481)	0.704
		ND	1.046	(-8.133, 10.225)	0.819	-1.181	(-3.111, 0.749)	0.224
Pinkie Fingertip	**Minimum ºC**	**D**	4.444	(-1.927, 10.815)	0.166	-0.134	(-2.026, 1.758)	0.887
		ND	-0.380	(-4.702, 3.942)	0.860	-0.930	(-3.251, 1.390)	0.424
	Average ºC	**D**	3.318	(-6.904, 13.541)	0.515	-0.318	(-2.168, 1.532)	0.731
		ND	-0.081	(-9.090, 8.928)	0.986	-1.107	(-3.073, 0.860)	0.263
	Maximum ºC	**D**	3.295	(-6.200, 12.791)	0.486	-3.301	(-7.185, 0.583)	0.094
		ND	3.559	(-5.381, 12.500)	0.425	-2.386	(-6.744, 1.972)	0.276
Dorsal Center	**Minimum ºC**	**D**	1.334	(-5.942, 8.611)	0.712	-3.501	(-6.805, -0.198)	0.038*
		ND	1.875	(-5.507, 9.257)	0.610	-2.881	(-6.561, 0.799)	0.122
	Average ºC	**D**	1.350	(-6.876, 9.577)	0.741	-3.405	(-7.028, 0.219)	0.065
		ND	2.645	(-5.327, 10.618)	0.506	-2.898	(-7.031, 1.235)	0.165

(Continued) Table 2. Associations between nitric oxide levels and temperature of dorsal and palm site of both hands among the cases (women with fibromyalgia syndrome) and controls (healthy women).

			NOx Levels					
			FMS Women (n=42)			Healthy Women (n=52)		
			β	95 % CI	*P*-value	β	95 % CI	*P*-value
Palm site of the hand								
	Maximum ºC	**D**	3.004	(-3.876, 9.883)	0.382	-0.941	(-3.007, 1.124)	0.364
		ND	0.541	(-6.986, 8.067)	0.885	-1.198	(-3.386, 0.990)	0.276
Thumb Fingertip	**Minimum ºC**	**D**	-2.727	(-8.409, 2.955)	0.337	-0.849	(-3.036, 1.338)	0.439
		ND	-0.385	(-5.782, 5.011)	0.886	-1.250	(-3.681, 1.182)	0.307
	Average ºC	**D**	1.477	(-5.288, 8.242)	0.661	-0.836	(-2.955, 1.283)	0.431
		ND	-0.053	(-7.365, 7.259)	0.988	-1.261	(-3.512, 0.991)	0.266
	Maximum ºC	**D**	3.599	(-3.178, 10.375)	0.289	-1.276	(-3.088, 0.537)	0.163
		ND	0.453	(-5.772, 6.678)	0.884	-1.046	(-2.997, 0.905)	0.286
Index Fingertip	**Minimum ºC**	**D**	2.171	(-3.889, 8.231)	0.473	-1.244	(-3.073, 0.586)	0.178
		ND	-4.070	(-10.036, 1.896)	0.175	-0.794	(-2.814, 1.226)	0.433
	Average ºC	**D**	2.629	(-4.069, 9.327)	0.432	-1.248	(-3.077, 0.581)	0.176
		ND	-0.501	(-6.577, 5.576)	0.868	-0.958	(-2.928, 1.011)	0.333
	Maximum ºC	**D**	2.027	(-5.221, 9.274)	0.574	-1.698	(-3.606, 0.210)	0.080
		ND	2.288	(-4.459, 9.034)	0.496	-1.378	(-3.303, 0.546)	0.156
Middle Fingertip	**Minimum ºC**	**D**	0.231	(-6.455, 6.917)	0.945	-1.541	(-3.494, 0.412)	0.119
		ND	1.415	(-4.549, 7.379)	0.634	-1.255	(-3.178, 0.667)	0.195
	Average ºC	**D**	0.665	(-6.287, 7.617)	0.847	-1.597	(-3.533, 0.339)	0.104
		ND	1.220	(-5.033, 7.473)	0.695	-1.361	(-3.289, 0.567)	0.162

(Continued) Table 2. Associations between nitric oxide levels and temperature of dorsal and palm site of both hands among the cases (women with fibromyalgia syndrome) and controls (healthy women).

			NOx Levels					
			FMS Women (n=42)			Healthy Women (n=52)		
			β	95 % CI	*P*-value	β	95 % CI	*P*-value
Palm site of the hand								
	Maximum ºC	**D**	1.301	(-6.179, 8.780)	0.727	-0.559	(-2.404, 1.286)	0.545
		ND	-0.625	(-6.514, 5.264)	0.831	-1.237	(-3.074, 0.599)	0.182
Ring Fingertip	**Minimum ºC**	**D**	0.207	(-4.840, 5.255)	0.934	-0.256	(-2.172, 1.660)	0.790
		ND	-0.915	(-6.393, 4.563)	0.737	-0.942	(-2.881, 0.996)	0.333
	Average ºC	**D**	-0.452	(-7.614, 6.710)	0.899	-0.483	(-2.349, 1.382)	0.605
		ND	-0.186	(-1.815, 1.218)	0.692	-1.155	(-3.052, 0.743)	0.227
	Maximum ºC	**D**	-2.857	(-9.016, 3.303)	0.353	-0.510	(-2.320, 1.301)	0.574
		ND	-1.212	(-7.525, 5.102)	0.700	-1.061	(-2.931, 0.808)	0.259
Pinkie Fingertip	**Minimum ºC**	**D**	0.057	(-3.264, 3.377)	0.973	-0.374	(-2.258, 1.510)	0.691
		ND	-2.494	(-5.885, 0.897)	0.145	-1.086	(-3.017, 0.846)	0.264
	Average ºC	**D**	-2.557	(-8.620, 3.506)	0.398	-0.432	(-2.289, 1.426)	0.642
		ND	-2.137	(-7.891, 3.617)	0.457	-1.064	(-2.930, 0.803)	0.258
	Maximum ºC	**D**	6.190	(-7.739, 20.118)	0.374	-3.627	(-7.640, 0.387)	0.075
		ND	2.826	(-11.649, 17.301)	0.695	-5.594	(-10.106, -1.081)	0.016*
Palm Center	**Minimum ºC**	**D**	-1.416	(-12.151, 9.319)	0.791	-3.603	(-7.370, 0.163)	0.060
		ND	4.888	(-5.319, 15.095)	0.338	-4.090	(-7.905, -0.275)	0.036*
	Average ºC	**D**	6.359	(-6.572, 19.290)	0.326	-3.295	(-7.282, 0.691)	0.103
		ND	3.955	(-9.300, 17.209)	0.549	-5.519	(-9.933, -1.106)	0.015*

(Continued) Table 2. Associations between nitric oxide levels and temperature of dorsal and palm site of both hands among the cases (women with fibromyalgia syndrome) and controls (healthy women).

			NOx Levels					
			FMS Women (n=42)			Healthy Women (n=52)		
			β	95 % CI	*P*-value	β	95 % CI	*P*-value
Palm site of the hand								
	Maximum ºC	**D**	4.236	(-5.853, 14.324)	0.400	-5.800	(-10.508, -1.092)	0.017*
		ND	2.842	(-7.015, 12.700)	0.563	-4.318	(-9.240, 0.605)	0.084
Thenar eminence	**Minimum ºC**	**D**	0.468	(-7.922, 8.858)	0.911	-2.490	(-6.749, 1.769)	0.245
		ND	-2.327	(-1.736, 1.212)	0.721	-4.037	(-8.512, 0.439)	0.076
	Average ºC	**D**	2.192	(-9.568, 13.953)	0.708	-4.328	(-8.937, 0.281)	0.065
		ND	-1.172	(-12.391, 10.048)	0.834	-4.336	(-9.248, 0.576)	0.082
	Maximum ºC	**D**	9.773	(-1.055, 20.602)	0.076	-3.240	(-7.575, 1.095)	0.139
		ND	6.221	(-5.281, 17.723)	0.280	-4.376	(-8.842, 0.089)	0.055
Hypothenar eminence	**Minimum ºC**	**D**	1.118	(-6.363, 8.599)	0.764	-0.719	(-4.507, 3.069)	0.704
		ND	0.075	(-4.415, 4.565)	0.973	0.024	(-3.167, 3.215)	0.988
	Average ºC	**D**	0.481	(-9.737, 10.700)	0.924	-1.579	(-5.553, 0.428)	0.428
		ND	-1.300	(-9.652, 7.053)	0.754	-4.087	(-8.443, 0.269)	0.065

*Significance level $P<0.05$

Note. a) FMS: Fibromyalgia Syndrome; b) Beta (β): regression coefficient, adjusted for age, menopause status and body mass index; c) 95% CI: 95% confidence interval; d) NOx: nitric oxide metabolites; e) ºC: Celsius degree; f) D: dominant; g) ND: non-dominant.

Table 3. Beta estimates and confidence intervals for the association between nitric oxide and tympanic cases (women with fibromyalgia syndrome) and controls (healthy women).

		Cases (n=42)			NOx
		β	95 % CI	p-value	β
Tympanic temperature ºC		-1.303	(-12.056, 9.450)	0.807	-9.321
Axillary temperature ºC		8.631	(-5.826, 23.087)	0.234	-7.625
Difference tympanic temperature ºC	**D**	-0.969	(-8.781, 6.842)	0.803	-0.215
	ND	0.369	(-6.726, 7.464)	0.917	1.620
Difference axillary temperature ºC	**D**	3.320	(-5.480, 12.121)	0.449	-0.608
	ND	4.381	(-3.164, 11.926)	0.247	1.609

*Significance level P<.05

Note. FMS: Fibromyalgia Syndrome; Beta (β): regression coefficient, adjusted for age, menopause status and interval; NOx: nitric oxide metabolites; ºC: Celsius degree; D: dominant; ND: non-dominant

4.2 Study II: "Associations Among Nitric Oxide and Enkephalinases With Fibromyalgia Symptoms".

The results section of the Study II is showed below.

4.2.1 Demographic and Clinical Characteristics.

The flow diagram of the selection of participants throughout the study is depicted in **Figure 3**. A total of 58 women diagnosed with FM with mean age of 56.33±7.82 years were selected and included in this study based on the inclusion and exclusion criteria proposed. **Table 4** shows the demographic and clinical data of the participants.

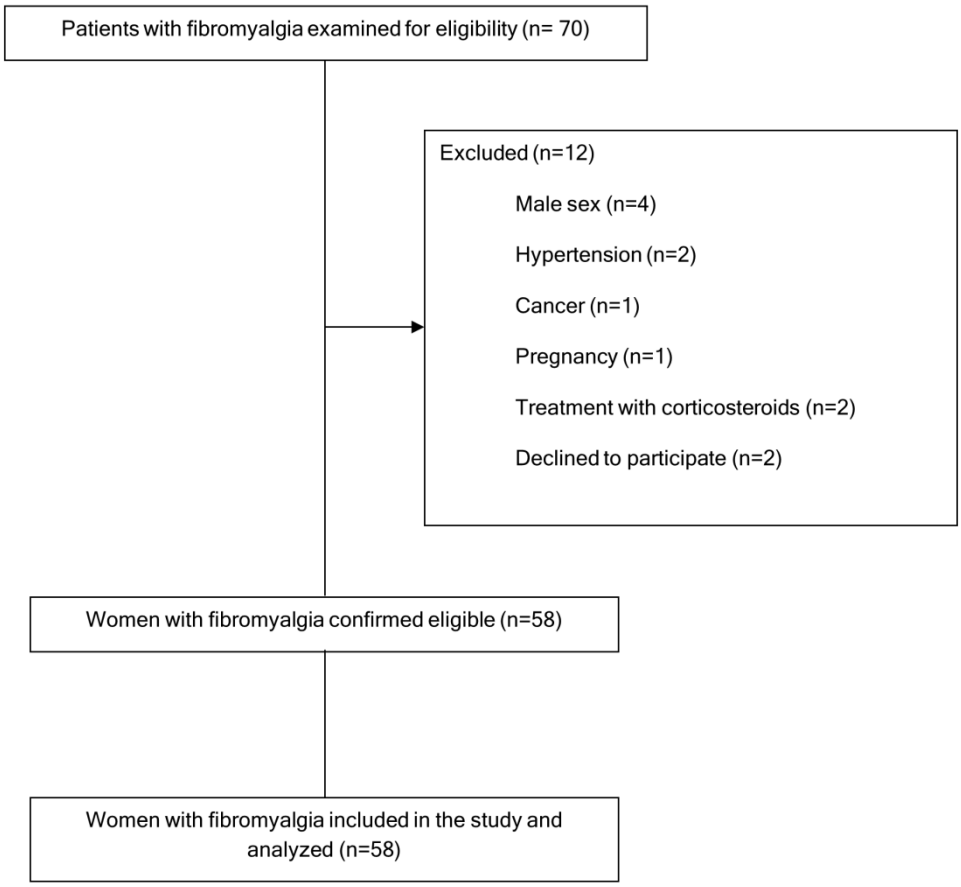

Figure 3. Flow Diagram of The Participants Throughout The Study.

Table 4. Demographic Data, Serum Parameters, and Clinical Characteristics of Women Diagnosed With Fibromyalgia.

		Women Diagnosed With Fibromyalgia (n=58)		
Variable		**Mean ± SD / Frequency (%)**	**95% CI**	**Missing data**
Age (years)		56.33±7.82	(54.26, 58.41)	
Height (cm)		158.21±5.32	(156.80, 159.62)	
Weight (kg)		72.05±12.07	(68.85, 75.26)	
BMI (kg/cm^2)		28.95±5.60	(27.46, 30.43)	
Age of menopause (years)		47.07±7.07	(44.92, 49.22)	
Menopause status				
Pre-menopausal		15 (25.86)		
Post-menopausal		43 (75.54)		
Serum NO level (µmol/mg protein)		23.91±11.98	(20.73, 27.09)	
Serum Oxytocinase activity (pmol hydrolyzed TyrNNap/min/mg protein)		53.05±11.78	(49.96, 56.15)	
Serum EDA activity (pmol hydrolysed CysNNap/min/mg protein)		76.89±50.87	(61.23, 92.55)	6
Patients with EDA activity ≤ 0.73		0.28±0.20	(0.14, 0.43)	
Patients with EDA activity > 0.73		100.10±31.74	(88.85, 111.36)	
Pain threshold electric score (mA)		6.11±3.83	(5.07, 7.14)	2
Pain electric score (mA)		12.28±8.72	(9.92, 14.64)	2
VAS (mm)		74.74±17.12	(70.19, 79.28)	
Pressure pain thresholds (kPa)				
Occiput	D	0.95±0.75	(0.75, 1.16)	
	ND	0.91±0.76	(0.71, 1.11)	
Trapezius	D	1.07±0.82	(0.86, 1.29)	
	ND	0.97±0.61	(0.81, 1.14)	
Zygapophyseal joint	D	1.07±0.85	(0.85, 1.29)	
	ND	1.08±0.88	(0.85, 1.31)	
Supraspinatus	D	1.42±1.10	(1.13, 1.71)	
	ND	1.37±0.85	(1.15, 1.60)	
Second rib	D	0.95±0.54	(0.82, 1.10)	
	ND	0.95±0.50	(0.82, 1.08)	
Epicondyle	D	1.03±0.70	(0.85, 1.22)	
	ND	1.00±0.62	(0.84, 1.17)	
Second metacarpal	D	1.25±0.80	(1.04, 1.46)	
	ND	1.16±0.65	(0.99, 1.34)	
Gluteus	D	2.03±1.71	(1.57, 2.48)	
	ND	1.95±1.40	(1.58, 2.33)	
Greater trochanter	D	2.20±1.24	(1.87, 2.52)	
	ND	2.18±1.20	(1.86, 2.50)	
Knee	D	1.68±1.22	(1.36, 2.00)	
	ND	1.95±1.26	(1.62, 2.29)	
Anterior Tibial	D	1.85±1.37	(1.48, 2.21)	
	ND	1.82±1.17	(1.51, 2.13)	
CSI		69.02±10.87	(66.13, 71.90)	

(Continued) Table 4. Demographic Data, Serum Parameters, and Clinical Characteristics of Women Diagnosed With Fibromyalgia.

	Women Diagnosed With Fibromyalgia (n=58)		
Variable	**Mean ± SD / Frequency (%)**	**95% CI**	**Missing data**
FIQ-R			
FIQ-R.1	20.49±5.03	(19.15, 21.82)	
FIQ-R.2	14.02±4.29	(12.88, 15.16)	
FIQ-R.3	38.52±6.82	(36.69, 40.35)	1
Total FIQ-R score	73.05±13.42	(69.46, 76.65)	1
BAI	32.32±9.13	(29.89, 34.74)	

Note. Data are expressed as mean ± SD (Standard Deviation) for quantitative variables, and as frequency (%) for qualitative variables. 95% CI = 95% confidence interval; BMI = body mass index; NO = nitric oxide; EDA = enkephalin-degrading aminopeptidase; mA = milliamps; VAS = visual analogue scale; kPa = kilopascals; D = dominant; ND = no dominant; CSI = Central Sensitization Inventory; FIQ-R = revised Fibromyalgia Impact Questionnaire; FIQ-R.1 = activity level of the FIQ-R; FIQ-R.2 = overall impact of the FIQ-R; FIQ-R.3 = intensity of symptoms of the FIQ-R; BAI = Beck Anxiety Inventory.

4.2.2 Associations of Serum NO Levels With Pain Threshold Electric, Pain Intensity Electric, VAS, PPTs, CSI, FIQ-R, and BAI in Patients With FM.

Table 5 shows the associations between serum NO levels and clinical symptoms of women with FM. Significant associations were found among NO levels and dominant occiput PPT (β=0.290, 95% CI [0.102, 0.478], p= .003), non-dominant occiput PPT (β=0.193, 95% CI [0.015, 0.371], p= .034), and FIQ-R.1 (β=0.031, 95% CI [0.004, 0.058], p= .027) after adjustment by age, BMI, and menopause status. Two associations approached statistical significance: NO levels with dominant epicondyle PPT (β=0.200, 95% CI [-0.005, 0.405], p= .056) and with non-dominant knee PPT (β=0.194, 95% CI [-0.021, 0.409], p= .076).

Table 5. Beta Estimates, Confidence Intervals and P-values for the Association between Nitric Oxide (NO) levels and Clinical Features in Women Diagnosed With Fibromyalgia.

Variable		Women Diagnosed With Fibromyalgia (n=58)		
		Serum NO levels		
		β	95 % CI	*p*-value
Pain threshold electric score (mA)		-0.048	(-0.376, 0.280)	0.770
Pain electric score (mA)		0.023	(-0.231, 0.278)	0.854
VAS (mm)		0.004	(-0.005, 0.012)	0.384
Pressure pain thresholds (kPa)				
Occiput	D	0.290	(0.102, 0.478)	.003*
	ND	0.193	(0.015, 0.371)	.034*
Trapezius	D	0.113	(-0.080, 0.306)	0.244
	ND	0.130	(-0.088, 0.348)	0.236
Zygapophyseal joint	D	0.122	(-0.081, 0.326)	0.234
	ND	0.108	(-0.086, 0.302)	0.269
Supraspinatus	D	0.128	(-0.071, 0.327)	0.204
	ND	0.120	(-0.095, 0.336)	0.267
Second rib	D	0.170	(-0.076, 0.415)	0.172
	ND	0.168	(-0.090, 0.425)	0.197
Epicondyle	D	0.200	(-0.005, 0.405)	.056
	ND	0.149	(-0.066, 0.365)	0.170
Second metacarpal	D	0.189	(-0.047, 0.424)	0.115
	ND	0.186	(-0.064, 0.436)	0.141
Gluteus	D	0.102	(-0.107, 0.311)	0.332
	ND	0.100	(-0.104, 0.304)	0.331
Greater trochanter	D	0.109	(-0.138, 0.355)	0.380
	ND	0.167	(-0.090, 0.423)	0.197
Knee	D	0.159	(-0.052, 0.370)	0.136
	ND	0.194	(-0.021, 0.409)	.076
Anterior Tibial	D	0.151	(-0.067, 0.368)	0.170
	ND	0.135	(-0.056, 0.327)	0.162
CSI		0.002	(-0.012, 0.016)	0.734
FIQ-R				
FIQ-R.1		0.031	(0.004, 0.058)	.027*
FIQ-R.2		0.010	(-0.026, 0.045)	0.581
FIQ-R.3		0.005	(-0.006, 0.016)	0.356
Total FIQ-R score		0.008	(-0.003, 0.019)	0.149
BAI		0.050	(-0.318, 0.419)	0.785

Note. Beta (β) represents the regression coefficient. Adjusted for age, menopause status and body mass index. 95% CI = 95% confidence interval; mA = milliamps; VAS = visual analogue scale; kPa = kilopascals; D = dominant; ND = no dominant; CSI = Central Sensitization Inventory; FIQ-R = revised Fibromyalgia Impact Questionnaire; FIQ-R.1 = activity level of the FIQ-R; FIQ-R.2 = overall impact of the FIQ-R; FIQ-R.3 = intensity of symptoms of the FIQ-R; BAI = Beck Anxiety Inventory. *Significance level: $p < .05$.

4.2.3 Associations of Serum Oxytocinase and EDA Activities With Pain Threshold Electric, Pain Intensity Electric, VAS, PPTs, CSI, FIQ-R, and BAI in Patients With FM.

The associations between serum oxytocinase and EDA activities with pain-related symptoms in FM patients are presented in **Table 6**. Linear regression analysis showed that VAS (β=0.215, 95% CI [0.031, 0.400], p= .023) and dominant knee PPT (β=2.794, 95% CI [0.147, 5.441], p= .039) were significantly associated with oxytocinase activity after adjustment for age, BMI, and menopause status. A significant association was found among EDA activity in FM patients with high EDA activity levels (> 0.73 pmol/min/mg protein) and dominant second rib PPT (β=-20.096, 95% CI [-40.080, -0.113], p= .049). A previous study identified a subpopulation of FM patients with abnormally low serum EDA activity values (Martínez-Martos et al., 2019). They established two subgroups of patients with FM based on EDA activity values according to the ROC analysis results for EDA activity in their patients (cutoff point = 0.73 pmol/min/mg protein). For this reason and based on such results (Martínez-Martos et al., 2019), in the present work we have established two subgroups of FM patients based on EDA activity values: one subgroup had values ≤ 0.73 pmol/min/mg protein and the other showed values > 0.73 pmol/min/mg protein. Our results also showed that several associations approached statistical significance. In this line, oxytocinase activity correlated with dominant zygapophyseal joint PPT (β=4.180, 95% CI [-0.336, 8.696], p= .069) and with non-dominant second metacarpal PPT (β=4.971, 95% CI [0.624, 10.566], p= .080). Similarly, EDA activity in patients with high EDA activity levels (> 0.73 pmol/min/mg protein) correlated with non-dominant second metacarpal PPT (β=-18.844, 95% CI [-38.558, 0.870], p= .060).

Table 6. Beta Estimates, Confidence Intervals and P-values for the Association between Oxytocinase and EDA in Women Diagnosed with Fibromyalgia.

		Women Diagnosed With Fibromyalgia (n=58)					
Variable		**Serum Oxytocinase activity**			**Serum EDA**		
					Patients with EDA activity ≤ 0.73 pmol/min/mg protein		
		β	**95 % CI**	***p*-value**	***β***	**95 % CI**	***p*-value**
Pain threshold electric score (mA)		4.606	(-2.720, 11.931)	0.213	-0.133	(-0.589, 0.322)	0.486
Pain electric score (mA)		1.224	(-4.526, 6.974)	0.671	0.068	(-0.239, 0.375)	0.594
VAS (mm)		0.215	(0.031, 0.400)	.023*	-0.008	(-0.025, 0.009)	0.277
Pressure pain thresholds (kPa)							
Occiput	**D**	1.482	(-3.114, 6.077)	0.520	0.062	(-0.238, 0.361)	0.620
	ND	2.696	(-1.422, 6.814)	0.195	0.042	(-0.162, 0.247)	0.616
Trapezius	**D**	2.387	(-1.971, 6.745)	0.277	-0.019	(-0.266, 0.228)	0.852
	ND	1.730	(-3.232, 6.691)	0.487	0.016	(-0.255, 0.287)	0.886
Zygapophyseal joint	**D**	4.180	(-0.336, 8.696)	.069	-0.012	(-0.223, 0.198)	0.887
	ND	3.367	(-0.965, 7.699)	0.125	0.020	(-0.207, 0.247)	0.829
Supraspinatus	**D**	1.500	(-3.043, 6.044)	0.510	0.020	(-0.206, 0.245)	0.832
	ND	2.610	(-2.250, 7.470)	0.286	-0.033	(-0.288, 0.222)	0.752
Second rib	**D**	2.190	(-3.421, 7.801)	0.437	0.005	(-0.292, 0.301)	0.970
	ND	1.633	(-4.255, 7.520)	0.580	0.019	(-0.253, 0.292)	0.863
Epicondyle	**D**	2.088	(-2.677, 6.853)	0.383	0.008	(-0.314, 0.331)	0.949
	ND	1.902	(-3.019, 6.822)	0.442	-0.038	(-0.278, 0.203)	0.704
Second metacarpal	**D**	4.411	(-0.901, 9.723)	0.102	-0.036	(-0.360, 0.288)	0.788
	ND	4.971	(0.624, 10.566)	.080	-0.058	(-0.374, 0.258)	0.657
Gluteus	**D**	1.241	(-3.509, 5.990)	0.602	-0.067	(-0.386, 0.252)	0.612
	ND	2.705	(-1.877, 7.288)	0.242	-0.016	(-0.282, 0.250)	0.881

(Continued) Table 6. Beta Estimates, Confidence Intervals and P-values for the Association between Oxyto
Clinical Features in Women Diagnosed with Fibromyalgia.

		Women Diagnosed With Fibromyalgia (n=58)					
Variable		**Serum Oxytocinase activity**			**Serum EDA**		
					Patients with EDA activity ≤ 0.73 pmol/min/mg protein		
		β	**95 % CI**	***p*-value**	***β***	**95 % CI**	***p*-value**
Pressure pain thresholds (kPa)							
Greater trochanter	**D**	3.720	(-1.788, 9.228)	0.181	-0.030	(-0.313, 0.253)	0.795
	ND	4.115	(-1.650, 9.880)	0.158	-0.021	(-0.393, 0.350)	0.888
Knee	**D**	2.794	(0.147, 5.441)	.039*	-0.002	(-0.224, 0.220)	0.985
	ND	3.091	(-1.837, 8.019)	0.214	-0.020	(-0.263, 0.222)	0.837
Anterior Tibial	**D**	3.033	(-1.897, 7.963)	0.222	-0.015	(-0.327, 0.297)	0.908
	ND	3.036	(-1.283, 7.355)	0.164	-0.036	(-0.243, 0.170)	0.669
CSI		-0.132	(-0.446, 0.183)	0.405	-0.002	(-0.021, 0.017)	0.821
FIQ-R							
FIQ-R.1		0.200	(-0.442, 0.842)	0.534	0.003	(-0.030, 0.035)	0.846
FIQ-R.2		0.017	(-0.013, 0.047)	0.266	-0.001	(-0.002, 0.002)	0.964
FIQ-R.3		0.004	(-0.003, 0.010)	0.269	-0.001	(-0.002, 0.001)	0.900
Total FIQ-R score		0.161	(-0.079, 0.402)	0.184	0.001	(-0.010, 0.012)	0.863
BAI		0.101	(-0.259, 0.462)	0.576	-0.008	(-0.021, 0.004)	0.143

Note. Beta (β) represents the regression coefficient. Adjusted for age, menopause status and body mass index. Amino
as pmol/min/mg protein. 95% CI = 95% confidence interval; EDA = enkephalin-degrading aminopeptidase; mA = m
scale; kPa = kilopascals; D = dominant; ND = no dominant; CSI = Central Sensitization Inventory; FIQ-R
Questionnaire; FIQ-R.1 = activity level of the FIQ-R; FIQ-R.2 = overall impact of the FIQ-R; FIQ-R.3 = intensity o
Beck Anxiety Inventory. *Significance level: $p < .05$.

4.3 Study III: "Dietary Inflammatory Index Scores Are Associated with Pressure Pain Hypersensitivity in Women with Fibromyalgia".

The results section of the Study III is showed below.

4.3.1 Demographic and Clinical Symptoms.

The clinical symptoms and PPTs of tender-point sites recorded for the 95 cases and 98 controls are summarised in **Table 7**. The mean DII® score was 0.28 ± 0.90 units and ranged from −2.57 (the maximum anti-inflammatory profile) to 1.95 (the maximum pro-inflammatory profile) in the FMS group. No significant differences were observed between the cases and controls for the DII® score ($p = 0.475$). Of note, FMS patients weighed more than the control group ($p = 0.005$); 76.2% of the overall cohort were postmenopausal women and 23.8% were premenopausal. As expected, patients with FMS had significantly higher levels of global pain and clinical symptoms including fatigue, sleep, anxiety, and central sensitisation than the controls ($p < 0.001$). The mean FIQ score was 72.09 ± 16.26 in women with FMS and all the PPTs of tender-point sites were significantly lower in these patients than in the controls (< 0.001).

4.3.2 Associations of DII® Scores with PPTs and Clinical Symptoms for Women Diagnosed with Fibromyalgia Syndrome and Healthy Women.

The β estimates and 95% CIs for clinical symptoms and the PPTs of tender-point sites, as well as the DII® scores for cases and controls, are presented in **Table 8**. Linear regression analysis revealed that the occiput (β = 0.350, 95% CI [0.089, 0.611], p = 0.009), trapezius (β = 0.397, 95% CI [0.134, 0.660], p = 0.004), zygapophyseal joint (β = 0.254, 95% CI [0.039, 0.469], p = 0.0021), second rib (β = 0.503, 95% CI [0.117, 0.889], p = 0.011), epicondyle (β = 0.315, 95% CI [0.010, 0.621], p = 0.043), gluteus (β = 0.147, 95% CI [0.029, 0.265], p = 0.015), greater trochanter (β = 0.193, 95% CI [0.036, 0.350], p = 0.017), and knee (β = 0.217, 95% CI [0.067, 0.366], p = 0.005) PPTs were significantly associated with the DII® after adjusting for age, menopausal status, and overall energy in patients with FMS but not in the controls. No significant differences were found between the DII® scores and the remaining clinical symptoms for cases or controls.

Table 7. Clinical symptoms and pressure pain thresholds of most tender point sites among cases (women with fibromyalgia) and controls (healthy women).

	Cases (n=95)	Controls (n=98)	p-value
Age (years)	55.76±7.96	56.08±10.33	0.808
Height (cm)	158.76±5.91	158.72±5.91	0.969
Weight (kg)	71.94±13.32	66.75±11,84	0.005
FIQ-R	72.09±16.26	-	-
VAS pain (cm)	7.38±1.80	1.40±2.22	<0.001
MFI	79.46±9.89	43.26±14.06	<0.001
PSQI	15.35±3.77	6.07±3.83	<0.001
BAI	32.53±9.88	9.70±9.25	<0.001
CSI	68.43±11.91	24.47±11.18	<0.001
PPT			
Occiput	0.87±0.68	3.33±1.35	<0.001
Trapezius	0.97±0.66	3.41±1.54	<0.001
Zygapophyseal joint	1.03±0.78	3.30±2.01	<0.001
Supraspinatus	1.25±0.82	3.94±2.04	<0.001
Second rib	0.85±0.44	2.64±1.24	<0.001
Epicondyle	0.95±0.58	3.51±1.48	<0.001
Gluteus	1.87±1.39	6.49±2.48	<0.001
Greater trochanter	2.05±1.09	6.05±2.49	<0.001
Knee	1.71±1.14	5.94±2.42	<0.001
DII®	0.32±0.89	0.23±0.91	0.475

* Significance level p<0,05

Note: Variables are shown as mean ± SD (Standard Deviation).VAS, Visual Analogue Scale; FIQ-R, Revised Fibromyalgia Impact Questionnaire; MFI, Multidimensional Fatigue Inventory; PSQI, Pittsburgh Sleep Quality Index; BAI, Beck Anxiety Inventory; CSI, Central Sensitization Inventory; Pressure pain threshold, PPT; Dietary Inflammatory Index, DII®.

Table 8. Beta estimates and confidence intervals for the association between DII® and clinical symptoms and pressure pain thresholds of most tender point sites among cases (women with fibromyalgia) and controls (healthy women).

	DII® (continuous)					
	Cases			Controls		
	β	95 % CI	p-value	β	95 % CI	p-value
VAS pain	0.012	-0.096, 0.121	0.823	0.048	-0.071, 0.166	0.422
FIQ-R	0.004	-0.007, 0.016	0.470	-	-	-
MFI	-0.001	-0.020, 0.017	0.870	0.013	-0.002, 0.029	0.089
PSQI	0.035	-0.016, 0.086	0.177	0.002	-0.064, 0.068	0.949
BAI	-0.011	-0.029, 0.007	0.220	0.017	-0.006, 0.040	0.152
CSI	-0.006	-0.022, 0.011	0.489	0.018	-0.003, 0.038	0.088
PPT						
Occiput	0.350	0.089, 0.611	0.009	-0.086	-0.259, 0.088	0.326
Trapezius	0.397	0.134, 0.660	0.004	-0.083	-0.227, 0.060	0.250
Zygapophyseal joint	0.254	0.039, 0.469	0.021	-0.075	-0.193, 0.042	0.204
Supraspinatus	0.208	-0.001, 0.417	0.051	-0.113	-0.234, 0.008	0.066
Second rib	0.503	0.117 0.889	0.011	-0.118	-0.301, 0.065	0.202
Epicondyle	0.315	0.010, 0.621	0.043	-0.134	-0.289, 0.021	0.089
Gluteus	0.147	0.029, 0.265	0.015	-0.074	-0.173, 0.026	0.143
Greater trochanter	0.193	0.036, 0.350	0.017	-0.062	-0.158, 0.035	0.205
Knee	0.217	0.067, 0.366	0.005	-0.089	-0.193, 0.015	0.090

Note: Beta represents the regression coefficient. Adjusted for age, menopause status and total energy. Dietary Inflammatory Index, DII®; VAS, Visual Analogue Scale; FIQ-R, Revised Fibromyalgia Impact Questionnaire; MFI, Multidimensional Fatigue Inventory; PSQI, Pittsburgh Sleep Quality Index; BAI, Beck Anxiety Inventory; CSI, Central Sensitization Inventory; Pressure pain threshold, PPT.

4.3.3 PPTs and Clinical Symptoms according to Quartiles of the DII® Scores in Women Diagnosed with Fibromyalgia Syndrome.

The DII® scores were divided into quartiles (Qs) according to the following cut-off points: Q1: $-2.57 \leq -0.19$; Q2: > -0.19 to ≤ 0.41; Q3: > 0.41 to ≤ 1.01; Q4: > 1.01 to 1.95. **Table 9** shows the clinical symptoms and PPTs of tender-point sites according to these quartiles in patients with FMS. After adjusting for confounding factors, the occiput ($p = 0.015$), trapezius ($p = 0.005$), zygapophyseal joint ($p = 0.012$), second rib ($p = 0.009$), greater trochanter ($p = 0.017$), and knee ($p = 0.011$) PPTs, but not the remaining clinical symptoms, were significantly associated with the DII® score quartiles in patients with FMS.

4.3.4 Nutrient Intakes according to Quartiles of the DII® Scores in Women Diagnosed with Fibromyalgia Syndrome.

Table 10 shows macro- and micronutrient intakes in patients with FMS according to the DII® quartiles. Beta carotene, energy, fibre, folate, iron, magnesium, niacin, polyunsaturated fatty acid, riboflavin, thiamine, vitamin B6, vitamin C, vitamin E, and zinc were found to be significantly higher in Q1 (the most anti-inflammatory quartile) in all cases ($p < 0.05$).

Table 9. Clinical symptoms and pressure pain thresholds among women with Fibromyalgia according to quartiles (Q) of the DII®.

	Q1 (n=20)		Q2 (n=22)		Q3 (n=23)		Q4 (n=20)		
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**P Value**
VAS pain	7.20	1.64	7.91	1.57	7.48	2.10	7.40	1.14	0.355
FIQ-R	70.50	13.33	79.90	10.45	69.17	20.09	71.90	15.37	0.056
MFI	77.75	9.46	80.55	9.83	80.17	11.04	79.90	8.92	0.669
PSQI	15.40	3.41	16.89	2.37	15.00	3.59	15.32	4.51	0.311
BAI	29.70	10.45	34.50	9.59	33.96	9.64	33.60	9.53	0.166
CSI	67.55	7.71	69.45	13.89	70.61	12.54	69.25	9.65	0.592
PPT									
Occiput	1.18	0.79	0.94	0.75	0.72	0.44	0.57	0.37	0.015
Trapezius	1.37	0.84	0.97	0.65	0.83	0.41	0.73	0.45	0.005
Zygapophyseal joint	1.50	1.12	0.97	0.72	0.90	0.53	0.80	0.53	0.012
Supraspinatus	1.61	1.14	1.31	0.89	1.08	0.52	0.99	0.51	0.066
Second rib	1.09	0.49	0.87	0.52	0.80	0.37	0.67	0.25	0.009
Epicondyle	1.24	0.74	0.95	0.59	0.85	0.43	0.78	0.35	0.059
Gluteus	2.43	1.63	2.12	1.70	1.65	1.32	1.44	0.78	0.075
Greater trochanter	2.39	0.95	2.36	1.47	1.78	0.87	1.65	0.72	0.017
Knee	2.17	1.00	1.91	1.43	1.51	1.05	1.20	0.65	0.011

Note: Adjusted for age, menopause status and total energy.

VAS, Visual Analogue Scale; FIQ-R, Revised Fibromyalgia Impact Questionnaire; MFI, Multidimensional Fatigue Inventory; PSQI, Pittsburgh Sleep Quality Index; BAI, Beck Anxiety Inventory; CSI, Central Sensitization Inventory; Pressure pain threshold, PPT.

Table 10. Nutrient intake according to the quartiles of the DII® in women with Fibromyalgia.

	Q1		Q2		Q3		Q4		
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**P Value**
DII® score	-0.88	0.65	0.15	0.17	0.72	0.19	1.31	0.26	<0.001
Beta Carotene (µg)	4481.44	3041.69	2280.25	2744.13	1746.97	2021.98	801.30	815.51	<0.001
Carbohydrate (g)	165.86	74.15	140.68	68.39	141.85	53.64	128.07	55.71	0.243
Cholesterol (mg)	224.77	155.80	268.53	215.59	207.29	129.69	208.08	137.56	0.547
Energy (kcal)	1497.09	515.45	1284.47	415.33	1255.75	352.19	1143.02	371.39	0.041
Fiber (g)	27.54	14.19	17.11	9.44	13.16	5.23	10.05	4.57	<0.001
Folate (µg)	357.99	238.73	188.59	70.93	150.99	56.62	106.94	47.70	<0.001
Iron (mg)	15.17	6.77	11.04	4.88	8.26	2.57	7.03	2.40	<0.001
Magnesium (mg)	435.64	183.54	280.33	168.62	257.84	108.41	220.58	94.11	<0.001
Mono-unsaturated fatty acid (g)	23.30	10.21	21.84	14.74	21.50	10.71	16.92	1.45	0.285
Niacin (mg)	32.19	12.92	27.50	10.87	24.76	13.22	21.82	9.41	0.025
Polyunsaturated fatty acid (g)	13.28	7.71	7.87	4.57	8.11	5.27	5.13	3.26	<0.001
Protein (g)	79.74	34.11	67.36	23.49	60.60	28.91	63.00	47.01	0.248
Riboflavin (mg)	1.51	0.42	1.47	0.95	1.25	0.49	1.06	0.32	0.042
Saturated fatty acid (g)	14.83	8.08	15.03	9.98	15.30	7.00	15.46	6.71	0.993
Thiamine (mg)	1.43	0.72	1.06	0.42	0.88	0.39	0.72	0.34	<0.001
Total fat (g)	57.08	22.65	50.26	27.99	49.57	20.66	42.07	18.75	0.179
Vitamin A (RE)	173.81	168.89	178.66	175.02	160.91	113.49	211.05	336.80	0.878
Vitamin B12 (µg)	3.89	2.62	4.93	7.12	2.83	1.97	2.29	1.66	0.126

(Continued) Table 10. Nutrient intake according to the quartiles of the DII® in women with Fibromyalgia.

	Q1		Q2		Q3		Q4		
	Mean	**SD**	**Mean**	**SD**	**Mean**	**SD**	**Mean**	**SD**	**P Value**
Vitamin B6 (mg)	1.97	0.87	1.49	0.76	1.40	0.59	0.94	0.37	<0.001
Vitamin C (mg)	183.90	104.28	127.88	59.54	104.46	53.82	62.72	52.32	<0.001
Vitamin D (µg)	3.22	4.80	3.56	4.87	2.11	2.93	0.94	1.58	0.091
Vitamin E (mg)	7.73	3.92	5.74	4.32	2.33	1.12	1.79	1.35	<0.001
Zinc (mg)	9.10	5.38	7.01	3.31	6.43	2.05	5.74	2.20	0.011

Comparisons of nutrient intake across the quartiles of the DII® were analysed using a one-way ANOVA.

Note: Dietary inflammatory index DII®.

DISCUSIÓN

DISCUSSION

5. DISCUSIÓN / DISCUSSION

5.1 Differences in hands temperature and core body temperature between women diagnosed with Fibromyalgia and healthy women.

The results of the first observational study show that the patients with FMS presented higher maximum, minimum and mean temperatures at each point of the dorsal and palmar sites of both hands than the controls. The patients with FMS also showed a higher core body temperature in the tympanic artery than the healthy women.

On the one hand, our results indicate that tympanic temperature was higher in the FMS group, suggesting increased thermogenesis. Previous studies have shown that patients with FMS have an increased core body temperature related to an imbalance of the ANS (Brusselmans et al., 2015; Elmas et al., 2016; Kulshreshtha et al., 2012). Elmas et al. (2016) measured patients' body temperature with a skin temperature probe (operating within a range of 0–50 ºC) at the inside of the right arm and observed that the temperature in patients with FMS was higher than that of the controls. Another study (Brusselmans et al., 2015) showed that patients with FMS presented increased axillary temperature compared with controls but with no changes in tympanic temperature, results that are in contrast with our findings, given that differences between the groups were only achieved at the tympanic level. The discrepancies might be explained by the different devices employed for measuring core body temperature. As Abdi et al. (2016) reported, however, the tympanic method has better accuracy and precision for detecting this variable. Supporting our results, the literature shows evidence that AVA vasodilation is related to the release of substance P and CGRP into the blood flow (Charkoudian, 2010; Holzer, 1992; Johnson & Kellogg, 2010; Minson, 2010). Substance P is delivered to the blood circulation after physical or stressful conditions (cold or heat), stimulating the mast cells that release vasoactive substances (interleukin 6) and proinflammatory mediators (tumor necrosis factor) into the blood flow (Theoharis C. Theoharides et al., 2015). Several studies have reported that levels of these compounds are elevated in patients with FMS (Paus et al., 2006; T. C. Theoharides et al., 2010; Theoharis C. Theoharides et al., 2015; Tsilioni et al., 2016). Therefore, the increase in these vasoactive markers might be related to the elevated core body temperature that we recorded in our FMS group.

On the other hand, our findings highlight excessive peripheral vasodilation of the microvasculature of the hands in FMS. Researchers have previously reported that ANS disorders are associated with abnormal innervation of the AVAs in the glabrous skin of the

hands of patients with FMS (Albrecht et al., 2013). The thermoregulatory activity in this area is governed by the adrenergic vasoconstrictor system, which can be activated during cold stress (Gibbins et al., 2003; Johnson & Kellogg, 2010; Kellogg, 2006). In cold conditions, such as the cold stress test, patients with FMS show similar characteristics of the Raynaud's phenomenon, including cyanosis and vasospasm on the fingers (Scolnik et al., 2016). The adrenergic axons of the AVAs are stimulated by cold conditions, can detect reduced skin surface temperature and can transmit this information to the CNS to stimulate the sympathetic vasoconstrictor system. This process impedes the blood supply to the peripheral capillaries of the superficial plexus with the objective of delivering the blood to the deep venous system, thereby maintaining tissue temperature and preserving tissue viability (Benzinger, 1963; Daanen, 2003). In line with our results regarding higher temperatures on the palmar site of the digits (33.36±1.06) and the dorsal centre temperature (33.40±0.92) of both hands in the FMS group, Scolnik et al. (2016) assessed the temperature of the digits and dorsal sides of the hands of patients with Raynaud's disease and FMS using infrared thermography. The authors showed that the patients diagnosed with FMS had significantly higher baseline digit (32.1±7.3 versus 29.0±7.3 ºC) and dorsal temperatures (31.9±4.3 versus 30.2±4.4 ºC) compared with the patients with primary Raynaud's syndrome, despite no differences in symptom characteristics between the patients with primary Raynaud's syndrome and the patients with FMS (Scolnik et al., 2016). In contrast, a recent study demonstrated a lower cold detection pain threshold in the hands of patients with FMS (29.9±0.7 ºC) compared with healthy controls (31.0±0.6 ºC) through electrochemical skin conductance (Pickering et al., 2020), a result that could be due to impaired sudomotor function in the dominant hands of the patients with FMS. More studies are needed to investigate the intertwining of the nociception-autonomic system (Pickering et al., 2020), as well as further longitudinal studies to clarify the contribution of AVAs to capillary circulation in terms of the changes in the temperature of the glabrous skin of the hands before and after an ice water test in FMS.

5.2 Nitric Oxide levels and their relationships with hands temperature and core body temperature between women with Fibromyalgia syndrome and healthy women.

We found no significant association between serum NO levels and temperature in FMS; however, a number of interactions were achieved at the dorsal and palmar sites of the hands in the controls.

The dilation of peripheral capillaries could also be caused by vasoactive compounds such as NO, which plays an important role in skin vasoactive vasodilation, eliciting hyperperfusion and local hyperthermia (Johnson et al., 1995). Gratt and Anbar (2005) reported that elevated NO levels in patients with chronic orofacial pain was associated with excessive vasodilation and hyperthermia in this region. Our data showed no differences in NO levels between the patients with FMS and the controls and no significant associations between NO levels and hand temperature. However, a number of correlations were found in the control group. Previous studies have reported that NO is not involved in reflex vasodilation in the peripheral forearm skin (Crandall & MacLean, 2001; Dietz et al., 1994). However, other studies have reported that NO plays an important role in skin vasodilation after whole body hyperthermia in healthy individuals (Dean L. Kellogg et al., 2008). Consequently, the role of NO in vasodilation and peripheral blood flow is still unclear and suggests the presence of other vasodilator mechanisms that might contribute to the effects of NO (Fujii et al., 2017; Dean L. Kellogg et al., 2008). Taking into account the contradictory results in the literature, more research is needed to validate our preliminary findings.

5.3 Relationships among Nitric Oxide levels and pain variables in women diagnosed with Fibromyalgia syndrome.

In our second study, the results have shown statistically significant associations of NO levels with PPTs of some tender points of the musculoskeletal system (dominant and non-dominant occiput) and with the daily activity level in women with FM. The associations of NO levels with dominant epicondyle PPT and with non-dominant knee PPT approached statistical significance. These results suggest that abnormalities in NO levels may be related to altered PPTs as well as to impaired activity level in patients with FM. Given its role as a vasodilator, alterations in NO levels may modify the blood microcirculation, thereby compromising the state of the musculoskeletal tissue and leading to the FM symptoms. However, previous studies have reported conflicting results on the impact of NO levels on FM clinical parameters. On one hand, several authors have observed significant correlations between NO levels and both VAS (Sendur et al., 2009) and FIQ (Rus et al., 2016) in FM patients. Other study analyzed the activity of NO synthases, the NO-producing enzymes, showing statistically significant correlations with chest pain, tender points, and migraine in patients with FM (Çimen et al., 2009). On the other hand, other studies did not find correlations between serum NO levels and FM

clinical features. Koca et al. (2018) reported no significant associations between serum NO levels and VAS and FIQ scores in patients with FM. In other case-control study, no correlations between NO levels and clinical measures such as FIQ-pain and FIQ-fatigue items were found (Ozgocmen et al., 2006).

5.4 Relationships among Enkephalin-degrading Aminopeptidase and Oxytocinase activities and pain variables in women with Fibromyalgia syndrome.

Our data have revealed significant correlations of serum oxytocinase activity with self-reported global pain, measured by VAS, and with dominant knee PPT in women with FM. The association of oxytocinase activity with dominant zygapophyseal joint PPT and with non-dominant second metacarpal PPT approached statistical significance. These results suggest that altered oxytocinase activity may be related to pain symptoms in patients diagnosed with FM. We also found a significant association between serum EDA activity in patients with high EDA activity levels and dominant second rib PPT, while the correlation with non-dominant second metacarpal PPT approached statistical significance. These results point to a relationship between altered EDA activity and pressure pain hypersensitivity in women with FM. A mechanism that could explain the widespread pain of women with FM is the attenuation on the descending pain pathways due to an alteration in the metabolism of the enkephalins. However, only one study has investigated the associations of oxytocinase and EDA activities with FM clinical characteristics (FIQ-R, VAS, Multidimensional Fatigue Inventory, BAI, and Pittsburgh Sleep Quality Index) (Martínez-Martos et al., 2019). These authors found that EDA activity significantly correlated with Pittsburgh Sleep Quality Index score in the subgroup of FM patients with low EDA activity levels, but did not find significant correlations between oxytocinase activity and any of the clinical features measured (Martínez-Martos et al., 2019). In contrast to this last result, in the present work we have observed a significant association between oxytocinase activity and VAS score in women with FM. These conflicting results may be related to the fact that in the present study we have performed a linear regression analysis adjusting by age, BMI, and menopause status to assess relationships between variables, while in the previous study, correlations were calculated by means of the Pearson's and Spearman's correlation coefficients.

5.5 Associations between Dietary Inflammatory Index® score and the pressure pain thresholds and other related symptoms in women diagnosed with Fibromyalgia syndrome.

In the third study, we found that pro-inflammatory diets were significantly associated with lower PPTs for most tender-point sites, although no significant differences in PPTs of tender-point sites, self-reported global pain, disease severity, fatigue, sleep, and anxiety with an anti-inflammatory dietary profile. These results support the hypothesis that the diet-related inflammatory potential was associated with pain hypersensitivity in individuals with FMS.

Current evidence suggests that dietary interventions may be a good therapeutic approach to reduce FMS symptoms (Arranz et al., 2010; Donaldson et al., 2001; Holton et al., 2012; Kaartine et al., 2000; Lattanzio & Imbesi, 2018; Rossi et al., 2015). Interestingly, eating a mostly raw, vegan diet (Donaldson et al., 2001) or a strict, low salt, uncooked vegan diet rich in lactobacteria (Kaartinen et al., 2000) had beneficial effects on these symptoms. Similarly, relationships between dietary habits and psychosocial outcomes such as mental health, depression, and optimism were identified in a population of women with FMS (Ruiz-Cabello et al., 2017). However, most previous work has focused on restrictive diets, isolated nutrients, or supplements, and the association between dietary inflammatory potential and the symptoms of FMS have never been investigated.

The available literature shows that sensory stimulation of healthy tissues in patients with FMS can result in pain hypersensitivity (Chinn et al., 2016). Here, we provide the first insights into the association between dietary inflammatory potential and PPTs—an accurate technique for assessing pain hypersensitivity in individuals with FMS (Chesterton et al., 2007). In a similar context, patients with rheumatoid arthritis who followed a Mediterranean diet (well-known for its anti-inflammatory properties), reported a reduction in pain levels (Sköldstam et al., 2005). Interestingly, and in agreement with our results, Tian et al. found evidence that tumour necrosis factor alpha, a widely-recognised mediator in many cytokine-dependent inflammatory events, increases in mice fed high-fat diets, resulting in fibromyalgia-like pain behaviours (Tian et al., 2018). The relationships between anti-inflammatory diets and decreased tender-point site PPTs extends the existing evidence that systemic inflammation lowers PPTs (de Goeij et al., 2013). Thus, our findings suggest that an anti-inflammatory diet might contribute to changes in pain perception in patients with FMS, reducing their pain hypersensitivity.

The mechanisms by which pro-inflammatory diets might decrease PPTs in individuals with FMS remains to be elucidated. Nonetheless, we hypothesize that anti-inflammatory diets may reduce the levels of the pro-inflammatory cytokines commonly elevated in these patients (Rodriguez-Pintó et al., 2014; Üçeyler et al., 2011b; Zhang et al., 2008), thus reducing their pain hypersensitivity. The same association was not observed in healthy women which suggests that anti-inflammatory diets only affect pain perception in this way in situations where the individual's inflammatory state is already elevated. The available literature regarding the relationship between diet and other clinical symptoms of FMS is controversial (Azad et al., 2000; Donaldson et al., 2001; Kaartinen et al., 2000). Our results indicate that DII® scores were not associated with self-assessed levels of global pain, disease severity, fatigue, sleep, anxiety, or central sensitisation as assessed using validated questionnaires. However, estimating FMS symptoms in this way is inherently limited because it is based on subjective patient-reported outcomes. Longitudinal studies will be required to better understand how the inflammatory potential of diets influence pressure-pain hypersensitivity in patients with FMS.

In contrast to these findings, a non-randomised, controlled study evaluating the effect of a strict, low-salt, uncooked vegan diet rich in lactobacteria on 18 patients with FMS, reported that VAS-assessed pain levels and sleep quality improved among these patients after a 3-month intervention period (Kaartinen et al., 2000). Moreover, Donaldson et al. showed that the FIQ but not body pain significantly improved in 30 patients with FMS after a following vegetarian diet plan (Donaldson et al., 2001). On the other hand, after assessing fatigue, insomnia, non-restorative sleep, and VAS-assessed pain levels, Azad et al. concluded that an exclusively vegetarian diet is a poor option for the treatment of FMS (Azad et al., 2000). These contradictory results may be explained by the differences in sample sizes, population characteristics, and methodology used in the assessment of FMS symptoms and dietary intakes in these studies. Given the currently limited evidence and the fact that no previous research has explored the association of these FMS outcomes with DII® scores, more research will still be needed to validate our preliminary findings and to elucidate the relationship between dietary intake and objective measures of the clinical symptoms of FMS.

5.6 Limitations of this PhD thesis.

We should recognize some limitations in the current doctoral thesis. Firstly, the first and second study presents a small sample size. So, additional researches with larger samples are needed to confirm our results. Secondly, the three studies were cross-sectional studies; therefore, no causal conclusions can be drawn. Thirdly, only women were included in the study due to the higher prevalence of Fibromyalgia syndrome among women and to avoid a possible confounder, which may limit the generalizability of the results to men. Fourth, as regards infrared thermography, there is considerable intraparticipant variability in thermographic quantification (Clark et al., 1999), which can be minimized by incorporating the protocol we employed in our study. Fifth, we did not record eye temperature, which is a reliable and reproducible method for estimating the body core temperature (Tan et al., 2009; Vardasca et al., 2019). Sixth, we did not monitor the participants' menstrual cycle phase, which could affect the reflex vascular responses (Lafferty et al., 1985). Seventh, although our findings provide new insights on the influence of serum markers on pain-related features in Fibromyalgia syndrome, future research may be needed to confirm the preliminary results in our second study. Eighth, as regards pro-inflammatory diet and its association with pain hypersensitivity, longitudinal studies are required to analyse the effect of the DII® scores and to evaluate the inclusion of anti-inflammatory diet recommendations in the guidelines for clinical management of Fibromyalgia syndrome. Ninth, although 24-hour diet recall is a reliable method to collect a variety of detailed information about the food individuals consume over a specific period, this tool has the inherent limitation that it may not accurately reflect the interviewee's normal diet at an individual level (Shim et al., 2014). To minimise the 24-hour recall bias in this third study, the recall was interviewer-driven, was administered by trained investigators, and standard household measures and pictorial food models were used to improve the accuracy of the food descriptions and quantities. Tenth, because the controls included friends and relatives of the patients, they might have had similar dietary patterns to the patient group. Finally, the DII® score was calculated based on only 23 of the 45 food parameters it originally included. However, the missing components (eugenol, flavanols, flavones, flavanones, anthocyanins, isoflavones, garlic, rosemary, saffron, ginger, turmeric, onion, tea, alcohol, and caffeine) are typically consumed in very small quantities, their reported consumption in the 24 h diet-recall was low, and previous research reported that their absence had no effect on the DII® scores (Tabung et al., 2015).

CONCLUSIONES

CONCLUSIONS

6. CONCLUSIONES

6.1 Generales:

- En general, nuestros resultados indicaron que las personas con síndrome de Fibromialgia presentan una mayor temperatura en el dorso y palma de sus manos (mayor vasodilatación) y una mayor temperatura corporal central. Estos datos sugieren una disfunción del control neural simpático a nivel cutáneo y de los procesos de termogénesis de las personas con Fibromialgia en comparación con las personas sanas sin patología.

- Del mismo modo, las personas con síndrome de Fibromialgia presentaron niveles alterados a nivel sanguíneo periférico de las concentraciones de óxido nítrico, de la actividad de la aminopeptidasa degradante de encefalina y de la actividad de la oxitocinasa, lo que puede modificar la microcirculación sanguínea y comprometer, por tanto, el estado del tejido músculo-esquelético.

- Las dietas proinflamatorias se asociaron con umbrales inferiores del dolor por presión para la mayoría de los puntos sensibles en pacientes con Fibromialgia, lo que sugiere que el potencial inflamatorio de la dieta podría desempeñar un papel relevante en la hipersensibilidad al dolor en estos pacientes.

6.2 Específicos:

- Las mujeres con síndrome de Fibromialgia presentaron mayores niveles globales de dolor, de hipersensibilidad al dolor por presión, de fatiga, de problemas de sueño, de impacto sobre la salud física y mental y de síntomas de ansiedad que las mujeres sanas.

- Las mujeres con síndrome de Fibromialgia presentaron una mayor temperatura en todos los puntos evaluados del dorso y palma de las manos a través de termografía infrarroja que las mujeres sanas. Estos datos informan de una posible alteración en la microcirculación sanguínea periférica y, por consiguiente, un daño fisiológico en la microvasculatura.

- Las mujeres con síndrome de Fibromialgia mostraron una mayor temperatura timpánica que las mujeres sanas. Estos resultados podrían indicar una disfunción del Sistema Nervioso Autónomo por la estrecha relación que presentan la arteria timpánica con el hipotálamo (principal centro del control de la temperatura del cuerpo humano).

- No se observaron asociaciones entre los niveles de concentración en suero sanguíneo del óxido nítrico y la vasodilatación periférica de la palma y el dorso de las manos de las mujeres con síndrome de Fibromialgia, aunque si se observaron algunas asociaciones negativas en mujeres sanas, lo que podría sugerir una disfunción del control neural simpático cutáneo.

- Nuestros resultados informaron de una asociación significativa entre los niveles sanguíneos de óxido nítrico y la temperatura timpánica en mujeres sanas, pero no en mujeres con síndrome de Fibromialgia, lo que indicaría una desregularización del óxido nítrico en el plasma sanguíneo que influiría en la temperatura general corporal.

- Nuestros datos revelaron asociaciones significativas de los niveles séricos de óxido nítrico y de las actividades de la aminopeptidasa degradante de encefalina y de la oxitocinasa en el plasma sanguíneo con unos mayores niveles de hipersensibilidad al dolor por presión en algunos puntos sensibles, de intensidad global del dolor y con un mayor impacto en la vida diaria en las mujeres con síndrome de Fibromialgia.

- Los perfiles dietéticos pro-inflamatorios se asociaron con unos mayores niveles de hipersensibilidad al dolor por presión para la mayoría de los puntos sensibles establecidos por el ACR en mujeres con síndrome de Fibromialgia en comparación con las mujeres sanas. Por lo tanto, se deben considerar estrategias para promover dietas antiinflamatorias para mejorar la hipersensibilidad al dolor en mujeres con Fibromialgia.

- Esta tesis doctoral ofrece un abordaje integral del síndrome de Fibromialgia que podría ayudar y mejorar las estrategias diagnósticas y de tratamiento (promoción de intervenciones dietéticas anti-inflamatorias) en estos pacientes. Además, los investigadores y clínicos podrían considerar la actividad desarrollada en la presente tesis doctoral con la finalidad de optimizar el manejo y conocimiento de esta enfermedad desde un punto de vista más global.

6. CONCLUSIONS

6.1 General:

- In general, our results indicated that people with Fibromyalgia syndrome have a higher temperature in the dorsal and palm of their hands (greater vasodilation) and a higher core body temperature. These data suggest a dysfunction of the sympathetic neural control at the cutaneous level and of the thermogenesis processes of people with Fibromyalgia compared to healthy people without pathology.

- Similarly, people with Fibromyalgia syndrome presented disorders in peripheral blood concentrations of nitric oxide, as well as in enkephalin-degrading aminopeptidase and oxytocinase activity, which can modify blood microcirculation and compromise, therefore, the musculoskeletal tissue condition.

- Pro-inflammatory diets were associated with lower pressure pain thresholds for most tender points in fibromyalgia patients, suggesting that the inflammatory potential of the diet could play a relevant role in pain hypersensitivity in such patients.

6.2 Specific:

- Women with Fibromyalgia syndrome presented higher overall levels of pain, hypersensitivity to pressure pain, fatigue, sleep problems, impact on physical and mental health, and anxiety symptoms compared to healthy women.

- Women with Fibromyalgia syndrome had a higher temperature in all the points evaluated using infrared thermography on the dorsal and palm of the hands compared to healthy women. These data indicate a possible disorder of peripheral blood microcirculation and, consequently, physiological damage to the microvasculature.

- Women with Fibromyalgia syndrome showed a higher tympanic temperature compared to healthy women. These results could indicate a dysfunction of the Autonomous Nervous System due to the close relationship that the tympanic artery presents with the hypothalamus (the main temperature control centre of the human body).

- No associations were observed between blood serum nitric oxide concentration levels and peripheral vasodilation of the palm and dorsal of the hands in women with Fibromyalgia syndrome, although some negative associations were observed in healthy women, which could suggest a dysfunction of cutaneous sympathetic neural control.

- Our results indicated a significant association between nitric oxide blood levels and tympanic temperature in healthy women, but not in women with Fibromyalgia syndrome. This could point to a dysregulation of nitric oxide in the blood plasma, which could influence the general body temperature.

- Our data revealed significant associations of serum nitric oxide levels and the activities of enkephalin-degrading aminopeptidase and oxytocinase in blood plasma, with higher levels of hypersensitivity to pressure pain in some tender points, of global pain intensity, and with a greater impact on the daily life in women with Fibromyalgia syndrome.

- Pro-inflammatory dietary profiles were associated with higher levels of pressure pain hypersensitivity for most tender points established by the American College of Rheumatology in women with Fibromyalgia syndrome compared to healthy women. Therefore, strategies to promote anti-inflammatory diets should be considered to reduce hypersensitivity to pain in women with fibromyalgia.

- This doctoral thesis offers a comprehensive approach to Fibromyalgia syndrome that could help and improve diagnostic and treatment strategies (promotion of anti-inflammatory dietary interventions) in these patients. In addition, researchers and clinicians could consider the activity developed in this doctoral thesis to

optimise the management and knowledge of this condition, from a more global point of view.

MENSAJES CLÍNICOS Y PERSPECTIVAS FUTURAS

CLINICAL MESSAGES AND FUTURE PERSPECTIVES

7. MENSAJES CLÍNICOS Y PERSPECTIVAS FUTURAS

- El síndrome de Fibromialgia no debe entenderse sólo como un dolor generalizado y difuso por todo el cuerpo humano, sino también como una patología que se encuentra acompañada de numerosos síntomas psicológicos y cognitivos. Por tanto, la exploración clínica que se ha venido realizando tradicionalmente tiempo atrás debe de progresar hacia un modelo biopsicosocial que considere la importancia de los factores psicosociales y cognitivos en la predisposición, desarrollo y cronificación de los síntomas del síndrome de Fibromialgia. En este sentido, introducir nuevos enfoques terapéuticos centrados en la neuroeducación del dolor ayudaría a los pacientes a mejorar el dolor, la sensibilización central, la catastrofización, la gravedad de los síntomas, la ansiedad, la depresión y los problemas cognitivos y psicológicos que padecen a lo largo de su vida diaria.

- Los hallazgos del presente estudio aportan un nuevo punto de vista sobre la posible influencia de las alteraciones neurovasculares a nivel cutáneo y los procesos de termogénesis en la sintomatología de los pacientes diagnosticados con síndrome de Fibromialgia. Por tanto, estudios científicos futuros deben de investigar la relación existente entre estos patrones termográficos alterados de las manos de los pacientes con Fibromialgia con las características y manifestaciones clínicas con la finalidad de obtener una mayor información en el diagnóstico y tratamiento de esta enfermedad. Además, los futuros estudios deberían también considerar el uso de la radiofrecuencia monopolar dieléctrica por los importantes efectos anti-inflamatorios y analgésicos que tienen sobre el tejido orgánico.

- La presente tesis doctoral indica también la necesidad de promover y fomentar la ingesta de alimentos anti-inflamatorios dado los resultados observados en cuanto a la asociación de perfiles dietéticos pro-inflamatorios con los niveles de hipersensibilidad al dolor en pacientes con Fibromialgia. En las medidas terapéuticas debería de estar indicada una estrategia basada en la promoción de una dieta anti-inflamatoria ya que pudiera tener una asociación beneficiosa directa con el dolor y las características clínicas de estos pacientes.

- El protocolo de evaluación y exploración de la respuesta vascular periférica así como de la temperatura corporal central puede ser utilizado por los fisioterapeutas, ofreciendo una técnica no invasiva, práctica, sin efectos secundarios y de fácil aplicación en pacientes diagnosticados con síndrome de Fibromialgia.

7. CLINICAL MESSAGES AND FUTURE PERSPECTIVES

- Fibromyalgia syndrome should not only be understood as a generalised and diffuse pain throughout the human body, but also as a pathology that is accompanied by numerous psychological and cognitive symptoms. Therefore, the clinical examination that has traditionally been conducted should progress towards a biopsychosocial model, which considers the importance of psychosocial and cognitive factors in the predisposition, development, and chronification of Fibromyalgia syndrome symptoms. In this context, introducing new therapeutic approaches focused on pain neuroeducation would help patients to improve pain, central sensitisation, catastrophizing, severity of symptoms, anxiety, depression, and cognitive and psychological issues they endure throughout their daily life.

- Findings herein provide a new point of view on the possible influence of neurovascular skin disorders and thermogenesis processes in the symptoms of patients diagnosed with Fibromyalgia syndrome. Therefore, future scientific studies should investigate the relationship between these altered hand thermographic patterns of Fibromyalgia patients with the characteristics and clinical manifestations to gain further information on the diagnosis and treatment of this condition. Furthermore, future studies should also consider the use of monopolar dielectric radiofrequency given its important anti-inflammatory and analgesic effects on organic tissue.

- This doctoral thesis also indicates the need to favour and encourage the intake of anti-inflammatory foods given the findings regarding the association of pro-inflammatory dietary profiles with pain hypersensitivity in Fibromyalgia patients. Therapeutic measures should suggest a strategy based on the promotion of anti-inflammatory diets because they may have a direct beneficial effect on pain and the clinical characteristics of these patients.

- The protocol for the assessment and exploration of the peripheral vascular response and core body temperature can be used by physiotherapists, offering a

non-invasive, practical technique, without side effects, and easy to apply in Fibromyalgia syndrome patients.

BIBLIOGRAFÍA

REFERENCES

8. BIBLIOGRAFÍA / REFERENCES

Abdi, A., Asadian, S., Khatony, A., Moradi, G. R., & Rezaei, M. (2016). Accuracy and precision of four common peripheral temperature measurement methods in intensive care patients. *Medical Devices: Evidence and Research*, *9*, 301–308. https://doi.org/10.2147/MDER.S109904

Albornoz-Cabello, M., Maya Martín, J., & Toledo Marhuenda, J. V. (2016). *Electroterapia práctica: avances en investigación clínica* (Elservier (ed.); 1ª edición).

Albrecht, P. J., Hou, Q., Argoff, C. E., Storey, J. R., Wymer, J. P., & Rice, F. L. (2013). Excessive Peptidergic Sensory Innervation of Cutaneous Arteriole-Venule Shunts (AVS) in the Palmar Glabrous Skin of Fibromyalgia Patients: Implications for Widespread Deep Tissue Pain and Fatigue. *Pain Medicine*, *14*(6), 895–915. https://doi.org/10.1111/pme.12139

Alexander, G. M., Van Rijn, M. A., Van Hilten, J. J., Perreault, M. J., & Schwartzman, R. J. (2005). Changes in cerebrospinal fluid levels of pro-inflammatory cytokines in CRPS. *Pain*, *116*(3), 213–219. https://doi.org/10.1016/j.pain.2005.04.013

Alstergren, P., & Förström, J. (2003). Acute oral pain intensity and pain threshold assessed by intensity matching to pain induced by electrical stimuli. *Journal of Orofacial Pain*, *17*(2), 151–159.

Aman, M. M., Jason Yong, R., Kaye, A. D., & Urman, R. D. (2018). Evidence-Based Non-Pharmacological Therapies for Fibromyalgia. *Current Pain and Headache Reports*, *22*(5), 33. https://doi.org/10.1007/s11916-018-0688-2

Amer-Cuenca, J. J., Pecos-Martín, D., Martínez-Merinero, P., Lluch Girbés, E., Nijs, J., Meeus, M., Ferrer Peña, R., & Fernández-Carnero, J. (2020). How Much Is Needed? Comparison of the Effectiveness of Different Pain Education Dosages in Patients with Fibromyalgia. *Pain Medicine (Malden, Mass.)*, *21*(4), 782–793. https://doi.org/10.1093/pm/pnz069

Andias, R., Neto, M., & Silva, A. G. (2018). The effects of pain neuroscience education and exercise on pain, muscle endurance, catastrophizing and anxiety in adolescents with chronic idiopathic neck pain: a school-based pilot, randomized and controlled study. *Physiotherapy Theory and Practice*, *34*(9), 682–691.

https://doi.org/10.1080/09593985.2018.1423590

Andrés-Rodríguez, L., Borràs, X., Feliu-Soler, A., Pérez-Aranda, A., Angarita-Osorio, N., Moreno-Peral, P., Montero-Marin, J., García-Campayo, J., Carvalho, A. F., Maes, M., & Luciano, J. V. (2020). Peripheral immune aberrations in fibromyalgia: A systematic review, meta-analysis and meta-regression. *Brain, Behavior, and Immunity*, *87*, 881–889. https://doi.org/10.1016/j.bbi.2019.12.020

Arnold, L. M., Bennett, R. M., Crofford, L. J., Dean, L. E., Clauw, D. J., Goldenberg, D. L., Fitzcharles, M. A., Paiva, E. S., Staud, R., Sarzi-Puttini, P., Buskila, D., & Macfarlane, G. J. (2019). AAPT Diagnostic Criteria for Fibromyalgia. *Journal of Pain*, *20*, 611–628. Churchill Livingstone Inc. https://doi.org/10.1016/j.jpain.2018.10.008

Arranz, L. I., Canela, M. Á., & Rafecas, M. (2010). Fibromyalgia and nutrition, What do we know? In *Rheumatology International*, *30*, 1417–1427. Rheumatol Int. https://doi.org/10.1007/s00296-010-1443-0

Atkinson, A. J., Colburn, W. A., DeGruttola, V. G., DeMets, D. L., Downing, G. J., Hoth, D. F., Oates, J. A., Peck, C. C., Schooley, R. T., Spilker, B. A., Woodcock, J., & Zeger, S. L. (2001). Biomarkers and surrogate endpoints: Preferred definitions and conceptual framework. In *Clinical Pharmacology and Therapeutics*, *69*, 89–95. Clin Pharmacol Ther. https://doi.org/10.1067/mcp.2001.113989

Azad, K. A., Alam, M. N., Haq, S. A., Nahar, S., Chowdhury, M. A., Ali, S. M., & Ullah, A. K. (2000). Vegetarian diet in the treatment of fibromyalgia. *Bangladesh Medical Research Council Bulletin*, *26*(2), 41–47.

Azad, K. A. K., Alam, M. N., Haq, S. A., Nahar, S., Chowdhury, M. A. J., Ali, S. M. K., & Ullah, A. K. M. A. (2000). Vegetarian diet in the treatment of fibromyalgia. *Bangladesh Medical Research Council Bulletin*, *26*(2), 41–47.

Bagher, P., & Segal, S. S. (2011). Regulation of blood flow in the microcirculation: Role of conducted vasodilation. *Acta Physiologica*, *202*(3), 271–284. https://doi.org/10.1111/j.1748-1716.2010.02244.x

Bair, M. J., & Krebs, E. E. (2020). In the clinic®: fibromyalgia. *Annals of Internal Medicine*, *172*(5), ITC33–ITC48. https://doi.org/10.7326/AITC202003030

Barrenengoa-Cuadra, M. J., Angón-Puras, L. Á., Moscosio-Cuevas, J. I., González-Lama, J., Fernández-Luco, M., & Gracia-Ballarín, R. (2021). Effectiveness of pain neuroscience education in patients with fibromyalgia: Structured group intervention in Primary Care. *Atencion Primaria*, *53*(1), 19-26 https://doi.org/10.1016/j.aprim.2019.10.007

Bellato, E., Marini, E., Castoldi, F., Barbasetti, N., Mattei, L., Bonasia, D. E., & Blonna, D. (2012). Fibromyalgia syndrome: Etiology, pathogenesis, diagnosis, and treatment. *Pain Research and Treatment*, *2012*, 426130. https://doi.org/10.1155/2012/426130

Benzinger, T. H. (1963). Peripheral cold- and central warm-reception, main origins of human thermal discomfort. *Proceedings of the National Academy of Sciences of the United States of America*, *49*(6), 832–839. https://doi.org/10.1073/pnas.49.6.832

Berger, A., Dukes, E., Martin, S., Edelsberg, J., & Oster, G. (2007). Characteristics and healthcare costs of patients with fibromyalgia syndrome. *International Journal of Clinical Practice*, *61*(9), 1498–1508. https://doi.org/10.1111/j.1742-1241.2007.01480.x

Bernardy, K., Klose, P., Busch, A. J., Choy, E. H. S., & Häuser, W. (2013). Cognitive behavioural therapies for fibromyalgia. *Cochrane Database of Systematic Reviews*, *2013*(9), CD009796. https://doi.org/10.1002/14651858.CD009796.pub2

Bijur, P. E., Shah, P. D., & Esses, D. (2016). Temperature measurement in the adult emergency department: Oral, tympanic membrane and temporal artery temperatures versus rectal temperature. *Emergency Medicine Journal*, *33*(12), 843–847. https://doi.org/10.1136/emermed-2015-205122

Bjørklund, G., Dadar, M., Chirumbolo, S., & Aaseth, J. (2018). Fibromyalgia and nutrition: Therapeutic possibilities? *Biomedicine and Pharmacotherapy*, *103*, 531–538. https://doi.org/10.1016/j.biopha.2018.04.056

Bjurstrom, M. F., Giron, S. E., & Griffis, C. A. (2016). Cerebrospinal Fluid Cytokines and Neurotrophic Factors in Human Chronic Pain Populations: A Comprehensive Review. In *Pain Practice*, *16*(2), 183–203. https://doi.org/10.1111/papr.12252

Bodes Pardo, G., Lluch Girbés, E., Roussel, N. A., Gallego Izquierdo, T., Jiménez Penick, V., & Pecos Martín, D. (2018). Pain Neurophysiology Education and Therapeutic

Exercise for Patients With Chronic Low Back Pain: A Single-Blind Randomized Controlled Trial. *Archives of Physical Medicine and Rehabilitation*, *99*(2), 338–347. https://doi.org/10.1016/j.apmr.2017.10.016

Boehm, K., Raak, C., Cramer, H., Lauche, R., & Ostermann, T. (2014). Homeopathy in the treatment of fibromyalgia-A comprehensive literature-review and meta-analysis. *Complementary Therapies in Medicine*, *22*(4), 731–742. https://doi.org/10.1016/j.ctim.2014.06.005

Bote, M. E., Garca, J. J., Hinchado, M. D., & Ortega, E. (2012). Inflammatory/stress feedback dysregulation in women with fibromyalgia. *NeuroImmunoModulation*, *19*(6), 343–351. https://doi.org/10.1159/000341664

Bradford, M. (1976). A Rapid and Sensitive Method for the Quantitation of Microgram Quantities of Protein Utilizing the Principle of Protein-Dye Binding. *Analytical Biochemistry*, *72*(1–2), 248–254. https://doi.org/10.1006/abio.1976.9999

Braman, R. S., & Hendrix, S. A. (1989). Nanogram Nitrite and Nitrate Determination in Environmental and Biological Materials by Vanadium(III) Reduction with Chemiluminescence Detection. *Analytical Chemistry*, *61*(24), 2715–2718. https://doi.org/10.1021/ac00199a007

Branco, J. C., Bannwarth, B., Failde, I., Abello Carbonell, J., Blotman, F., Spaeth, M., Saraiva, F., Nacci, F., Thomas, E., Caubère, J.-P., Le Lay, K., Taieb, C., & Matucci-Cerinic, M. (2010). Prevalence of Fibromyalgia: A Survey in Five European Countries. *Seminars in Arthritis and Rheumatism*, *39*(6), 448–453. https://doi.org/10.1016/j.semarthrit.2008.12.003

Bromberger, J. T., Kravitz, H. M., Chang, Y., Randolph, J. F., Avis, N. E., Gold, E. B., & Matthews, K. A. (2013). Does risk for anxiety increase during the menopausal transition? Study of Women's Health Across the Nation. *Menopause*, *20*(5), 488–495. https://doi.org/10.1097/gme.0b013e3182730599

Brusselmans, Griet, Nogueira, H., De Schamphelaere, E., Devulder, J., & Crombez, G. (2015). Skin Temperature during Cold Pressor Test in Fibromyalgia: an Evaluation of the Autonomic Nervous System? *Acta Anaesthesiologica Belgica*, *66*(1), 19–27. http://www.ncbi.nlm.nih.gov/pubmed/26103738

Busch, A., Barber, K., Overend, T., Peloso, P., & Schachter, C. (2002). Exercise for treating fibromyalgia syndrome. *The Cochrane Database of Systematic Reviews (Protocol), 3*, CD003786. https://doi.org/10.1002/14651858.cd003786

Busch, A. J., Webber, S. C., Richards, R. S., Bidonde, J., Schachter, C. L., Schafer, L. A., Danyliw, A., Sawant, A., Dal Bello-Haas, V., Rader, T., & Overend, T. J. (2013). Resistance exercise training for fibromyalgia. *Cochrane Database of Systematic Reviews*, *2013*(12), CD010884. https://doi.org/10.1002/14651858.CD010884

Cabo-Meseguer, A., Cerdá-Olmedo, G., & Trillo-Mata, J. L. (2017). Fibromyalgia: Prevalence, epidemiologic profiles and economic costs. In *Medicina Clinica*, *149*(10), 441–448. Ediciones Doyma, S.L. https://doi.org/10.1016/j.medcli.2017.06.008

Çakit, M. O., Çakit, B. D., Genç, H., Pervane Vural, S., Erdem, H. R., Saraçoğlu, M., & Karagöz, A. (2018). The association of skinfold anthropometric measures, body composition and disease severity in obese and non-obese fibromyalgia patients: A cross-sectional study. *Archives of Rheumatology*, *33*(1), 59–65. https://doi.org/10.5606/ArchRheumatol.2018.6180

Carlson, B. E., Arciero, J. C., & Secomb, T. W. (2008). Theoretical model of blood flow autoregulation: Roles of myogenic, shear-dependent, and metabolic responses. *American Journal of Physiology - Heart and Circulatory Physiology*, *295*(4), H1572-H1579. https://doi.org/10.1152/ajpheart.00262.2008

Carrera, M. P., Ramírez-Expósito, M. J., Valenzuela, M. T., García, M. J., Mayas, M. D., & Martínez-Martos, J. M. (2004). Serum oxytocinase activity is related to tumor growth parameters in N-methyl nitrosourea induced rat breast cancer. *Life Sciences*, *75*(11), 1369–1377. https://doi.org/10.1016/j.lfs.2004.04.009

Chandran, A., Schaefer, C., Ryan, K., Baik, R., McNett, M., & Zlateva, G. (2012). The comparative economic burden of mild, moderate, and severe fibromyalgia: Results from a retrospective chart review and cross-sectional survey of working-age U.S. adults. *Journal of Managed Care Pharmacy*, *18*(6), 415–426. https://doi.org/10.18553/jmcp.2012.18.6.415

Charkoudian, N. (2010). Mechanisms and modifiers of reflex induced cutaneous vasodilation and vasoconstriction in humans. *Journal of Applied Physiology*, *109*(4),

1221–1228. https://doi.org/10.1152/japplphysiol.00298.2010

Chesterton, L. S., Sim, J., Wright, C. C., & Foster, N. E. (2007). Interrater reliability of algometry in measuring pressure pain thresholds in healthy humans, using multiple raters. *Clinical Journal of Pain*, *23*(9), 760–766. https://doi.org/10.1097/AJP.0b013e318154b6ae

Cheung, S. S. (2015). Responses of the hands and feet to cold exposure. *Temperature (Austin, Tex.)*, *2*(1), 105–120. https://doi.org/10.1080/23328940.2015.1008890

Chinn, S., Caldwell, W., & Gritsenko, K. (2016). Fibromyalgia Pathogenesis and Treatment Options Update. *Current Pain and Headache Reports*, *20*(4), 1–10. https://doi.org/10.1007/s11916-016-0556-x

Choi, D.-H., & Kim, H.-S. (2015). Quantitative analysis of nailfold capillary morphology in patients with fibromyalgia. *The Korean Journal of Internal Medicine*, *30*(4), 531–537. https://doi.org/10.3904/kjim.2015.30.4.531

Çimen, Ö. B., Çimen, M. Y. B., Yapici, Y., & Çamdeviren, H. (2009). Arginase, NOS activities, and clinical features in fibromyalgia patients. *Pain Medicine*, *10*(5), 813–818. https://doi.org/10.1111/j.1526-4637.2009.00642.x

Clark, S., Hollis, S., Campbell, F., Moore, T., Jayson, M., & Herrick, A. (1999). The "distal-dorsal difference" as a possible predictor of secondary Raynaud's phenomenon. *The Journal of Rheumatology*, *26*(5), 1125–1128. http://www.ncbi.nlm.nih.gov/pubmed/10332978

Clauw, D. J. (2014). Fibromyalgia: A clinical review. In *JAMA - Journal of the American Medical Association*, *311*(15), 1547–1555. American Medical Association. https://doi.org/10.1001/jama.2014.3266

Coskun Benlidayi, I. (2019). Role of inflammation in the pathogenesis and treatment of fibromyalgia. *Rheumatology International*, *39*(5), 781–791. https://doi.org/10.1007/s00296-019-04251-6

Costa, A. P. C., Maia, J. M., Brioschi, M. L., & de Melo Mafra Machado, J. E. (2019). Thermography evaluation in patients with hypothyroidism and fibromyalgia by analyzing the temperatures of the palms of hands. *IFMBE Proceedings*, *68*(1), 15–19.

https://doi.org/10.1007/978-981-10-9035-6_3

Crandall, C. G., & MacLean, D. A. (2001). Cutaneous interstitial nitric oxide concentration does not increase during heat stress in humans. *Journal of Applied Physiology*, *90*(3), 1020–1024. https://doi.org/10.1152/jappl.2001.90.3.1020

Cuesta-Vargas, A. I., Roldan-Jimenez, C., Neblett, R., & Gatchel, R. J. (2016). Cross-cultural adaptation and validity of the Spanish central sensitization inventory. *SpringerPlus*, *5*(1), 1837. https://doi.org/10.1186/s40064-016-3515-4

Cury, Y., Picolo, G., Gutierrez, V. P., & Ferreira, S. H. (2011). Pain and analgesia: The dual effect of nitric oxide in the nociceptive system. *Nitric Oxide - Biology and Chemistry*, *25*(3), 243–254. https://doi.org/10.1016/j.niox.2011.06.004

D'Agnelli, S., Arendt-Nielsen, L., Gerra, M. C., Zatorri, K., Boggiani, L., Baciarello, M., & Bignami, E. (2019). Fibromyalgia: Genetics and epigenetics insights may provide the basis for the development of diagnostic biomarkers. *Molecular Pain* (Vol. 15). SAGE Publications Inc. https://doi.org/10.1177/1744806918819944

Daanen, H. A. M. (2003). Finger cold-induced vasodilation: A review. *European Journal of Applied Physiology*, *89*(5), 411–426. https://doi.org/10.1007/s00421-003-0818-2

de Goeij, M., van Eijk, L. T., Vanelderen, P., Wilder-Smith, O. H., Vissers, K. C., van der Hoeven, J. G., Kox, M., Scheffer, G. J., & Pickkers, P. (2013). Systemic Inflammation Decreases Pain Threshold in Humans In Vivo. *PLoS ONE*, *8*(12), e84159. https://doi.org/10.1371/journal.pone.0084159

De Tommaso, M., Sardaro, M., Serpino, C., Costantini, F., Vecchio, E., Pia Prudenzano, M., Lamberti, P., & Livrea, P. (2009). Fibromyalgia comorbidity in primary headaches. *Cephalalgia*, *29*(4), 453–464. https://doi.org/10.1111/j.1468-2982.2008.01754.x

De Tommaso, Marina, Federici, A., Serpino, C., Vecchio, E., Franco, G., Sardaro, M., Delussi, M., & Livrea, P. (2011). Clinical features of headache patients with fibromyalgia comorbidity. *Journal of Headache and Pain*, *12*(6), 629–638. https://doi.org/10.1007/s10194-011-0377-6

Deare, J. C., Zheng, Z., Xue, C. C. L., Liu, J. P., Shang, J., Scott, S. W., & Littlejohn, G.

(2013). Acupuncture for treating fibromyalgia. *Cochrane Database of Systematic Reviews*, *2013*(5), CD007070. https://doi.org/10.1002/14651858.CD007070.pub2

Devigili, G., Tugnoli, V., Penza, P., Camozzi, F., Lombardi, R., Melli, G., Broglio, L., Granieri, E., & Lauria, G. (2008). The diagnostic criteria for small fibre neuropathy: From symptoms to neuropathology. *Brain*, *131*(7), 1912–1925. https://doi.org/10.1093/brain/awn093

Dias, D. N. G., Marques, M. A. A., Bettini, S. C., & Paiva, E. dos S. (2017). Prevalence of fibromyalgia in patients treated at the bariatric surgery outpatient clinic of Hospital de Clínicas do Paraná - Curitiba. *Revista Brasileira de Reumatologia (English Edition)*, *57*(5), 425–430. https://doi.org/10.1016/j.rbre.2017.02.005

Dietz, N. M., Rivera, J. M., Warner, D. O., & Joyner, M. J. (1994). Is nitric oxide involved in cutaneous vasodilation during body heating in humans? *Journal of Applied Physiology*, *76*(5), 2047–2053. https://doi.org/10.1152/jappl.1994.76.5.2047

Donaldson, M S, Speight, N., & Loomis, S. (2001). Fibromyalgia syndrome improved using a mostly raw vegetarian diet: an observational study. *BMC Complementary and Alternative Medicine*, *1*, 7. https://doi.org/10.1186/1472-6882-1-7.

Du, C., Smith, A., Avalos, M., South, S., Crabtree, K., Wang, W., Kwon, Y. H., Vijayagopal, P., & Juma, S. (2019). Blueberries improve pain, gait performance, and inflammation in individuals with symptomatic knee osteoarthritis. *Nutrients*, *11*(2). https://doi.org/10.3390/nu11020290

Elmas, O., Yildiz, S., Bilgin, S., Demirci, S., Comlekci, S., Koyuncuoglu, H. R., Akkus, S., Colak, O. H., Etem Koklukaya, Arslan, E., Ozkan, O., & Bilgin, G. (2016). Physiological parameters as a tool in the diagnosis of fibromyalgia syndrome in females: A preliminary study. *Life Sciences*, *145*, 51–56. https://doi.org/10.1016/j.lfs.2015.12.029

Ernst, E. (2009). Chiropractic treatment for fibromyalgia: A systematic review. *Clinical Rheumatology*, *28*(10), 1175–1178. https://doi.org/10.1007/s10067-009-1217-9

Evdokimov, D., Frank, J., Klitsch, A., Unterecker, S., Warrings, B., Serra, J., Papagianni, A., Saffer, N., Meyer zu Altenschildesche, C., Kampik, D., Malik, R. A., Sommer, C., & Üçeyler, N. (2019). Reduction of skin innervation is associated with a severe

fibromyalgia phenotype. *Annals of Neurology*, *86*(4), 504–516. https://doi.org/10.1002/ana.25565

Fernández-Lao, C., Galiano-Castillo, N., Cantarero-Villanueva, I., Martín-Martín, L., Prados-Olleta, N., & Arroyo-Morales, M. (2016). Analysis of pressure pain hypersensitivity, ultrasound image, and quality of life in patients with chronic plantar pain: A preliminary study. *Pain Medicine (United States)*, *17*(8), 1530–1541. https://doi.org/10.1093/pm/pnv022

Fitzcharles, M. A., Perrot, S., & Häuser, W. (2018). Comorbid fibromyalgia: A qualitative review of prevalence and importance. *European Journal of Pain (United Kingdom)*, *22*(9), 1565–1576. https://doi.org/10.1002/ejp.1252

Freyberg, R. H. (1951). Non-articular rheumatism. *Bulletin of the New York Academy of Medicine*, *27*(4), 245–258.

Fujii, N., McNeely, B. D., Zhang, S. Y., Abdellaoui, Y. C., Danquah, M. O., & Kenny, G. P. (2017). Activation of protease-activated receptor 2 mediates cutaneous vasodilatation but not sweating: roles of nitric oxide synthase and cyclo-oxygenase. *Experimental Physiology*, *102*(2), 265–272. https://doi.org/10.1113/EP086092

Fujimasa, I., Chinzei, T., & Saito, I. (n.d.). Converting far infrared image information to other physiological data. *IEEE Engineering in Medicine and Biology Magazine : The Quarterly Magazine of the Engineering in Medicine & Biology Society*, *19*(3), 71–76.

García-López, M. J., Martínez-Martos, J. M., Mayas, M. D., Carrera, M. P., & Ramírez-Expósito, M. J. (2003). Effects of orchidectomy and testosterone replacement on mouse enkephalin-degrading aminopeptidase activity in the HPA axis. *General and Comparative Endocrinology*, *134*(3), 303–307. https://doi.org/10.1016/s0016-6480(03)00270-3

García-Ríos, M. C., Navarro-Ledesma, S., Tapia-Haro, R. M., Toledano-Moreno, S., Casas-Barragán, A., Correa-Rodríguez, M., & Aguilar-Ferrándiz, M. E. (2019). Effectiveness of health education in patients with fibromyalgia: A systematic review. *European Journal of Physical and Rehabilitation Medicine*, *55*(2), 301–313. https://doi.org/10.23736/S1973-9087.19.05524-2

Gasim, G. I., Musa, I. R., Abdien, M. T., & Adam, I. (2013). Accuracy of tympanic

temperature measurement using an infrared tympanic membrane thermometer. *BMC Research Notes*, *6*(1), 194. https://doi.org/10.1186/1756-0500-6-194

GASKELL, P. (1956). Are there sympathetic vasodilator nerves to the vessels of the hands. *The Journal of Physiology*, *131*(3), 647–656. https://doi.org/10.1113/jphysiol.1956.sp005489

Gayà, T. F., Ferrer, C. B., Mas, A. J., Seoane-Mato, D., Reyes, F. Á., Sánchez, M. D., Dubois, C. M., Sánchez-Fernández, S. A., Vargas, L. M. R., García Morales, P. V., Olivé, A., Muñoz, P. R., Larrosa, M., Navarro Ricós, N., Sánchez-Piedra, C., Díaz-González, F., & Bustabad-Reyes, S. (2020). Prevalence of fibromyalgia and associated factors in Spain. *Clinical and Experimental Rheumatology*, *38*(1), S47–S52.

Giacomelli, C., Sernissi, F., Sarzi-Puttini, P., Di Franco, M., Atzeni, F., & Bazzichi, L. (2013). Fibromyalgia: A critical digest of the recent literature. *Clinical and Experimental Rheumatology*, *31*(suppl.69), S1-11.

Gibbins, I. L., Jobling, P., & Morris, J. L. (2003). Functional organization of peripheral vasomotor pathways. *Acta Physiologica Scandinavica*, *177*(3), 237–245. https://doi.org/10.1046/j.1365-201X.2003.01079.x

Giske, L., Vøllestad, N. K., Mengshoel, A. M., Jensen, J., Knardahl, S., & Røe, C. (2008). Attenuated adrenergic responses to exercise in women with fibromyalgia - A controlled study. *European Journal of Pain*, *12*(3), 351–360. https://doi.org/10.1016/j.ejpain.2007.07.007

Goldberg, D. S., & McGee, S. J. (2011). Pain as a global public health priority. *BMC Public Health*, *11*(1), 770. https://doi.org/10.1186/1471-2458-11-770

Graeber, M. B. (2010). Changing face of microglia. *Science*, *330*(6005), 83–788. https://doi.org/10.1126/science.1190929

Graham, W. (1949). Fibrositis. In Hollander JL (Ed.), *Arthritis and Allied Conditions* (4 edn, pp. 622–647). Lea & Febiger.

Gratt, B. M., & Anbar, M. (2005). A pilot study of nitric oxide blood levels in patients with chronic orofacial pain. *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology*

and Endodontology, *100*(4), 441–448. https://doi.org/10.1016/j.tripleo.2004.02.081

Grayston, R., Czanner, G., Elhadd, K., Goebel, A., Frank, B., Üçeyler, N., Malik, R. A., & Alam, U. (2019). A systematic review and meta-analysis of the prevalence of small fiber pathology in fibromyalgia: Implications for a new paradigm in fibromyalgia etiopathogenesis. *Seminars in Arthritis and Rheumatism*, *48*(5), 933–940. https://doi.org/10.1016/j.semarthrit.2018.08.003

Gunn, J., Hill, M. M., Cotten, B. M., & Deer, T. R. (2020). An Analysis of Biomarkers in Patients with Chronic Pain. *Pain Physician*, *23*(1), E41–E49.

Häuser, W., Ablin, J., Fitzcharles, M. A., Littlejohn, G., Luciano, J. V., Usui, C., & Walitt, B. (2015). Fibromyalgia. *Nature Reviews Disease Primers*, *1*. https://doi.org/10.1038/nrdp.2015.22

Häuser, W., Ablin, J., Perrot, S., & Fitzcharles, M. A. (2017). Management of fibromyalgia: Key messages from recent evidence-based guidelines. *Polish Archives of Internal Medicine*, *127*(1), 47–56. https://doi.org/10.20452/pamw.3877

Häuser, W., Bernardy, K., Üçeyler, N., & Sommer, C. (2009). Treatment of fibromyalgia syndrome with gabapentin and pregabalin - A meta-analysis of randomized controlled trials. *Pain*, *145*(1–2), 69–81. https://doi.org/10.1016/j.pain.2009.05.014

Häuser, W., & Fitzcharles, M. A. (2018). Facts and myths pertaining to fibromyalgia. *Dialogues in Clinical Neuroscience*, *20*(1), 53–62. https://doi.org/10.31887/dcns.2018.20.1/whauser

Häuser, W., Petzke, F., Üeyler, N., & Sommer, C. (2011). Comparative efficacy and acceptability of amitriptyline, duloxetine and milnacipran in fibromyalgia syndrome: A systematic review with meta-analysis. *Rheumatology*, *50*(3), 532–543. https://doi.org/10.1093/rheumatology/keq354

Häuser, W., Urrútia, G., Tort, S., Üçeyler, N., & Walitt, B. (2013). Serotonin and noradrenaline reuptake inhibitors (SNRIs) for fibromyalgia syndrome. In *Cochrane Database of Systematic Reviews*, *2013*(1), CD010292. https://doi.org/10.1002/14651858.CD010292

Heidari, F., Afshari, M., & Moosazadeh, M. (2017). Prevalence of fibromyalgia in general

population and patients, a systematic review and meta-analysis. *Rheumatology International*, *37*(9), 1527–1539. https://doi.org/10.1007/s00296-017-3725-2

Henry, M. S., Gendron, L., Tremblay, M. E., & Drolet, G. (2017). Enkephalins: Endogenous Analgesics with an Emerging Role in Stress Resilience. *Neural Plasticity*, *2017*. https://doi.org/10.1155/2017/1546125

Hita-Contreras, F., Martínez-López, E., Latorre-Román, P. A., Garrido, F., Santos, M. A., & Martínez-Amat, A. (2014). Reliability and validity of the Spanish version of the Pittsburgh Sleep Quality Index (PSQI) in patients with fibromyalgia. *Rheumatology International*, *34*(7), 929–936. https://doi.org/10.1007/s00296-014-2960-z

Hochsprung, A., Escudero-Uribe, S., Ibáñez-Vera, A. J., & Izquierdo-Ayuso, G. (2018). Effectiveness of monopolar dielectric transmission of pulsed electromagnetic fields for multiple sclerosis–related pain: A pilot study. *Neurologia*, *18*, S0213-4853. https://doi.org/10.1016/j.nrl.2018.03.003

Holton, K. F., Taren, D. L., Thomson, C. A., Bennett, R. M., & Jones, K. D. (2012). The effect of dietary glutamate on fibromyalgia and irritable bowel symptoms. *Clinical and Experimental Rheumatology*, *30*(suppl.74), S10–S17.

Holzer, P. (1992). Peptidergic sensory neurons in the control of vascular functions: mechanisms and significance in the cutaneous and splanchnic vascular beds. In *Reviews of physiology, biochemistry and pharmacology*, 121, 49–146. https://doi.org/10.1007/bfb0033194

Honda, Y., Sakamoto, J., Hamaue, Y., Kataoka, H., Kondo, Y., Sasabe, R., Goto, K., Fukushima, T., Oga, S., Sasaki, R., Tanaka, N., Nakano, J., & Okita, M. (2018). Effects of Physical-Agent Pain Relief Modalities for Fibromyalgia Patients: A Systematic Review and Meta-Analysis of Randomized Controlled Trials. *Pain Research and Management*, *2018*. https://doi.org/10.1155/2018/2930632

Ibáñez-Vera, A. J., García-Romero, J. C., Alvero-Cruz, J. R., & Lomas-Vega, R. (2020). Effects of monopolar dielectric radiofrequency signals on the symptoms of fibromyalgia: A single-blind randomized controlled trial. *International Journal of Environmental Research and Public Health*, *17*(7). https://doi.org/10.3390/ijerph17072465

Inanici, F., & Yunus, M. B. (2004). History of fibromyalgia: Past to present. *Current Pain and Headache Reports*, *8*(5), 369–378. https://doi.org/10.1007/s11916-996-0010-6

Johnson, J. M., Pergola, P. E., Liao, F. K., Kellogg, D. L., & Crandall, C. G. (1995). Skin of the dorsal aspect of human hands and fingers possesses an active vasodilator system. *Journal of Applied Physiology*, *78*(3), 948–954. https://doi.org/10.1152/jappl.1995.78.3.948

Johnson, John M., & Kellogg, D. L. (2010). Local thermal control of the human cutaneous circulation. In *Journal of Applied Physiology*, *109*(4), 1229–1238. https://doi.org/10.1152/japplphysiol.00407.2010

Jones, D. H., Kilgour, R. D., & Comtois, A. S. (2007). Test-Retest Reliability of Pressure Pain Threshold Measurements of the Upper Limb and Torso in Young Healthy Women. *Journal of Pain*, *8*(8), 650–656. https://doi.org/10.1016/j.jpain.2007.04.003

Kaartinen, K., Lammi, K., Hypen, M., Nenonen, M., & Hänninen, O. (2000). Vegan diet alleviates fibromyalgia symptoms. *Scandinavian Journal of Rheumatology*, *29*(5), 308–313. https://doi.org/10.1080/030097400447697

Kaartinen, K., Lammi, K., Hypen, M., Nenonen, M., Hanninen, O., & Rauma, A. L. (2000). Vegan diet alleviates fibromyalgia symptoms. *Scandinavian Journal of Rheumatology*, *29*(5), 308–313.

Kadetoff, D., & Kosek, E. (2010). Evidence of reduced sympatho-adrenal and hypothalamic-pituitary activity during static muscular work in patients with fibromyalgia. *Journal of Rehabilitation Medicine*, *42*(8), 765–772. https://doi.org/10.2340/16501977-0597

Kalso, E. (2004). Biomarkers for pain. *Pain*, *107*(3), 199–201. https://doi.org/10.1016/j.pain.2003.12.009

Kanabrocki, E. L., Messmore, H. L., Shirazi, P., George, M., Hoppensteadt, D. A., Fareed, J., Hermida, R. C., Ayala, D. E., Ryan, M. D., Bremner, F. W., Third, J. L. H. C., & Nemchausky, B. A. (2001). Day-night variations in blood levels of nitric oxide, T-TFPI, and E-selectin. *Clinical and Applied Thrombosis/Hemostasis*, *7*(4), 339–345. https://doi.org/10.1177/107602960100700417

Kaushik, A. S., Strath, L. J., & Sorge, R. E. (2020). Dietary Interventions for Treatment of Chronic Pain: Oxidative Stress and Inflammation. *Pain and Therapy*, *9*(2). https://doi.org/10.1007/s40122-020-00200-5

Kellogg, D. L. (2006). In vivo mechanisms of cutaneous vasodilation and vasoconstriction in humans during thermoregulatory challenges. *Journal of Applied Physiology*, *100*(5), 1709–1718. https://doi.org/10.1152/japplphysiol.01071.2005

Kellogg, Dean L., Zhao, J. L., & Wu, Y. (2008). Endothelial nitric oxide synthase control mechanisms in the cutaneous vasculature of humans in vivo. *American Journal of Physiology - Heart and Circulatory Physiology*, *295*(1). https://doi.org/10.1152/ajpheart.00082.2008

Kindler, L. L., Bennett, R. M., & Jones, K. D. (2011). Central Sensitivity Syndromes: Mounting Pathophysiologic Evidence to Link Fibromyalgia with Other Common Chronic Pain Disorders. *Pain Management Nursing*, *12*(1), 15–24. https://doi.org/10.1016/j.pmn.2009.10.003

Knight, T., Schaefer, C., Chandran, A., Zlateva, G., Winkelmann, A., & Perrot, S. (2013). Health-resource use and costs associated with fibromyalgia in France, Germany, and the United States. *ClinicoEconomics and Outcomes Research*, *5*(1), 171–180. https://doi.org/10.2147/CEOR.S41111

Koca, T. T., Seyithanoglu, M., Sagiroglu, S., Berk, E., & Dagli, H. (2018). Frequency of audiological complaints in patients with fibromyalgia syndrome and its relationship with oxidative stress. *Nigerian Journal of Clinical Practice*, *21*(10), 1271–1277. https://doi.org/10.4103/njcp.njcp_95_18

Koca, Tuba Tülay, Acet, G. K., Tanrıkut, E., & Talu, B. (2016). Evaluation of sleep disorder and its effect on sexual dysfunction in patients with Fibromyalgia syndrome. *Turk Jinekoloji ve Obstetrik Dernegi Dergisi*, *13*(4), 167–171. https://doi.org/10.4274/tjod.17047

Kotani, N., Kudo, R., Sakurai, Y., Sawamura, D., Sessler, D. I., Okada, H., Nakayama, H., Yamagata, T., Yasujima, M., & Matsuki, A. (2004). Cerebrospinal fluid interleukin 8 concentrations and the subsequent development of postherpetic neuralgia. *American Journal of Medicine*, *116*(5), 318–324. https://doi.org/10.1016/j.amjmed.2003.10.027

Kulshreshtha, P., Gupta, R., Yadav, R. K., Bijlani, R. L., & Deepak, K. K. (2012). A comprehensive study of autonomic dysfunction in the fibromyalgia patients. *Clinical Autonomic Research*, *22*(3), 117–122. https://doi.org/10.1007/s10286-011-0150-6

Lafferty, K., De Trafford, J. C., Potter, C., Roberts, V. C., & Cotton, L. T. (1985). Reflex vascular responses in the finger to contralateral thermal stimuli during the normal menstrual cycle: a hormonal basis to Raynaud's phenomenon? *Clinical Science (London, England : 1979)*, *68*(6), 639–645. http://www.ncbi.nlm.nih.gov/pubmed/2485265

Lakhan, S. E., & Schofield, K. L. (2013). Mindfulness-Based Therapies in the Treatment of Somatization Disorders: A Systematic Review and Meta-Analysis. *PLoS ONE*, *8*(8), e71834. https://doi.org/10.1371/journal.pone.0071834

Langhorst, J., Klose, P., Dobos, G. J., Bernardy, K., & Häuser, W. (2013). Efficacy and safety of meditative movement therapies in fibromyalgia syndrome: A systematic review and meta-analysis of randomized controlled trials. *Rheumatology International*, *33*(1), 193–207. https://doi.org/10.1007/s00296-012-2360-1

Langhorst, J., Musial, F., Klose, P., & Häuser, W. (2009). Efficacy of hydrotherapy in fibromyalgia syndrome-a meta-analysis of randomized controlled clinical trials. *Rheumatology*, *48*(9), 1155–1159. https://doi.org/10.1093/rheumatology/kep182

Lattanzio, S. M., & Imbesi, F. (2018). Fibromyalgia syndrome: A case report on controlled remission of symptoms by a dietary strategy. *Frontiers in Medicine*, *5*(APR). https://doi.org/10.3389/fmed.2018.00094

Lauche, R., Cramer, H., Dobos, G., Langhorst, J., & Schmidt, S. (2013). A systematic review and meta-analysis of mindfulness-based stress reduction for the fibromyalgia syndrome. *Journal of Psychosomatic Research*, *75*(6), 500–510. https://doi.org/10.1016/j.jpsychores.2013.10.010

Lauche, R., Cramer, H., Haüser, W., Dobos, G., & Langhorst, J. (2015). A Systematic Overview of Reviews for Complementary and Alternative Therapies in the Treatment of the Fibromyalgia Syndrome. *Evidence-based Complementary and Alternative Medicine, 2015*, 610615. https://doi.org/10.1155/2015/610615

Lefaucheur, J. P., Wahab, A., Planté-Bordeneuve, V., Sène, D., Ménard-Lefaucheur, I.,

Rouie, D., Tebbal, D., Salhi, H., Créange, A., Zouari, H., & Ng Wing Tin, S. (2015). Diagnosis of small fiber neuropathy: A comparative study of five neurophysiological tests. *Neurophysiologie Clinique*, *45*(6), 445–455. https://doi.org/10.1016/j.neucli.2015.09.012

Lewis, D. A., Rud, K. S., & Miller, V. M. (1993). Cofactors of constitutive nitric oxide synthase and endothelium-dependent relaxations in canine femoral veins. *Journal of Cardiovascular Pharmacology*, *22*(3), 443–448. https://doi.org/10.1097/00005344-199309000-00015

Li, Y. H., Wang, F. Y., Feng, C. Q., Yang, X. F., & Sun, Y. H. (2014). Massage therapy for fibromyalgia: A systematic review and meta-analysis of randomized controlled trials. *PLoS ONE*, *9*(2), e89304. https://doi.org/10.1371/journal.pone.0089304

Lim, M. J., Kwon, S. R., Jung, K. H., Joo, K., Park, S. G., & Park, W. (2014). Digital thermography of the fingers and toes in Raynaud's phenomenon. *Journal of Korean Medical Science*, *29*(4), 502–506. https://doi.org/10.3346/jkms.2014.29.4.502

Lodha, R., Mukerji, N., Sinha, N., Pandey, R. M., & Jain, Y. (2000). Is axillary temperature an appropriate surrogate for core temperature? *Indian Journal of Pediatrics*, *67*(8), 571–574. https://doi.org/10.1007/BF02758482

López-Ramos, J. C., Martínez-Romero, R., Molina, F., Cañuelo, A., Martínez-Lara, E., Siles, E., & Peinado, M. A. (2005). Evidence of a decrease in nitric oxide-storage molecules following acute hypoxia and/or hypobaria, by means of chemiluminescence analysis. *Nitric Oxide - Biology and Chemistry*, *13*(1), 62–67. https://doi.org/10.1016/j.niox.2005.05.003

Louw, A., Zimney, K., Puentedura, E. J., & Diener, I. (2016). The efficacy of pain neuroscience education on musculoskeletal pain: A systematic review of the literature. In *Physiotherapy Theory and Practice*, *32*(5), 332–355. https://doi.org/10.1080/09593985.2016.1194646

Lund, I., Lundeberg, T., Kowalski, J., Sandberg, L., Budh, C. N., & Svensson, E. (2005). Evaluation of variations in sensory and pain threshold assessments by electrocutaneous stimulation. *Physiotherapy Theory and Practice*, *21*(2), 81–92. https://doi.org/10.1080/09593980590922307

Lundborg, C., Hahn-Zoric, M., Biber, B., & Hansson, E. (2010). Glial cell line-derived neurotrophic factor is increased in cerebrospinal fluid but decreased in blood during long-term pain. *Journal of Neuroimmunology*, *220*(1–2), 108–113. https://doi.org/10.1016/j.jneuroim.2010.01.007

Lunn, M. P. T., Hughes, R. A. C., & Wiffen, P. J. (2014). Duloxetine for treating painful neuropathy, chronic pain or fibromyalgia. *Cochrane Database of Systematic Reviews*, *2014*(1), CD007115. https://doi.org/10.1002/14651858.CD007115.pub3

MacArthur, P. H., Shiva, S., & Gladwin, M. T. (2007). Measurement of circulating nitrite and S-nitrosothiols by reductive chemiluminescence. *Journal of Chromatography B: Analytical Technologies in the Biomedical and Life Sciences*, *851*(1-2), 93–105. https://doi.org/10.1016/j.jchromb.2006.12.012

Macfarlane, G. J., Kronisch, C., Dean, L. E., Atzeni, F., Häuser, W., Flub, E., Choy, E., Kosek, E., Amris, K., Branco, J., Dincer, F., Leino-Arjas, P., Longle, K., McCarthy, G. M., Makri, S., Perrot, S., Sarzi-Puttini, P., Taylor, A., & Jones, G. T. (2017). EULAR revised recommendations for the management of fibromyalgia. *Annals of the Rheumatic Diseases*, *76*(2), 318–328. https://doi.org/10.1136/annrheumdis-2016-209724

Macfarlane, Gary J., Barnish, M. S., & Jones, G. T. (2017). Persons with chronic widespread pain experience excess mortality: Longitudinal results from UK Biobank and meta-Analysis. *Annals of the Rheumatic Diseases*, *76*(11), 1815–1822. https://doi.org/10.1136/annrheumdis-2017-211476

Magán, I., Sanz, J., & García-Vera, M. P. (2008). Psychometric properties of a Spanish version of the Beck anxiety inventory (BAI) in general population. *Spanish Journal of Psychology*, *11*(2), 626–640. https://doi.org/10.1017/s1138741600004637

Malfliet, A., Van Oosterwijck, J., Meeus, M., Cagnie, B., Danneels, L., Dolphens, M., Buyl, R., & Nijs, J. (2017). Kinesiophobia and maladaptive coping strategies prevent improvements in pain catastrophizing following pain neuroscience education in fibromyalgia/chronic fatigue syndrome: An explorative study. *Physiotherapy Theory and Practice*, *33*(8), 653–660. https://doi.org/10.1080/09593985.2017.1331481

Marikar Bawa, F. L., Mercer, S. W., Atherton, R. J., Clague, F., Keen, A., Scott, N. W., &

Bond, C. M. (2015). Does mindfulness improve outcomes in patients with chronic pain? Systematic review and meta-analysis. *British Journal of General Practice*, *65*(635), e387–e400. https://doi.org/10.3399/bjgp15X685297

Marques, A. P., Assumpção, A., Matsutani, L. A., Bragança Pereira, C. A., & Lage, L. (2008). Pain in fibromyalgia and discriminativen power of the instruments: Visual Analog Scale. Dolorimetry and the McGill Pain Questionnaire. *Acta Reumatologica Portuguesa*, *33*(3), 345–351.

Martínez-Martos, J. M., Correa-Rodríguez, M., Rus, A., Molina, F., Ramírez-Expósito, M. J., & Aguilar-Ferrandiz, M. E. (2019). Altered Serum Oxytocinase and Enkephalin-Degrading Aminopeptidase Activities in Patients With Fibromyalgia. *Biological Research for Nursing*, *21*(4), 431–439. https://doi.org/10.1177/1099800419854207

Mease, P. J., Clauw, D. J., Gendreau, R. M., Rao, S. G., Kranzler, J., Chen, W., & Palmer, R. H. (2009). The efficacy and safety of milnacipran for treatment of fibromyalgia. A randomized, double-blind, placebo-controlled trial. *Journal of Rheumatology*, *36*(2), 398–409. https://doi.org/10.3899/jrheum.080734

Mendieta, D., la Cruz-Aguilera, D. L. De, Barrera-Villalpando, M. I., Becerril-Villanueva, E., Arreola, R., Hernández-Ferreira, E., Pérez-Tapia, S. M., Pérez-Sánchez, G., Garcés-Alvarez, M. E., Aguirre-Cruz, L., Velasco-Velázquez, M. A., & Pavón, L. (2016). IL-8 and IL-6 primarily mediate the inflammatory response in fibromyalgia patients. *Journal of Neuroimmunology*, *290*, 22–25. https://doi.org/10.1016/j.jneuroim.2015.11.011

Minson, C. T. (2010). Thermal provocation to evaluate microvascular reactivity in human skin. *Journal of Applied Physiology*, *109*(4), 1239–1246. https://doi.org/10.1152/japplphysiol.00414.2010

Mirbod, S. M., & Sugiura, H. (2017). A non-invasive technique for the evaluation of peripheral circulatory functions in female subjects with Raynaud's phenomenon. *Industrial Health*, *55*(3), 275–284. https://doi.org/10.2486/indhealth.2016-0201

Moldofsky, H., Scarisbrick, P., England, R., & Smythe, H. (1975). Musculoskeletal symptoms and non REM sleep disturbance in patients with "fibrositis syndrome" and healthy subjects. *Psychosomatic Medicine*, *37*(4), 341–351.

https://doi.org/10.1097/00006842-197507000-00008

Morf, S., Amann-Vesti, B., Forster, A., Franzeck, U. K., Koppensteiner, R., Uebelhart, D., & Sprott, H. (2005). Microcirculation abnormalities in patients with fibromyalgia - measured by capillary microscopy and laser fluxmetry. *Arthritis Research & Therapy*, *7*(2), R209-216. https://doi.org/10.1186/ar1459

Moseley, G. L. (2003). A pain neuromatrix approach to patients with chronic pain. *Manual Therapy*, *8*(3), 130–140. https://doi.org/10.1016/S1356-689X(03)00051-1

Munguía-Izquierdo, D., Segura-Jiménez, V., Camiletti-Moirón, D., Pulido-Martos, M., Alvarez-Gallardo, I. C., Romero, A., Aparicio, V. A., Carbonell-Baeza, A., & Delgado-Fernández, M. (2012). Multidimensional Fatigue Inventory: Spanish adaptation and psychometric properties for fibromyalgia patients. The Al-Andalus study. *Clinical and Experimental Rheumatology*, *30*(6 Suppl 74), 94–102.

Murakami, M., Osada, K., Ichibayashi, H., Mizuno, H., Ochiai, T., Ishida, M., Alev, L., & Nishioka, K. (2017). An open-label, long-term, phase III extension trial of duloxetine in Japanese patients with fibromyalgia. *Modern Rheumatology*, *27*(4), 688–695. https://doi.org/10.1080/14397595.2016.1245237

Nagashima, H., Morio, Y., Yamane, K., Nanjo, Y., & Teshima, R. (2009). Tumor necrosis factor-α, interleukin-1β, and interleukin-6 in the cerebrospinal fluid of patients with cervical myelopathy and lumbar radiculopathy. *European Spine Journal*, *18*(12), 1946–1950. https://doi.org/10.1007/s00586-009-1069-7

Naumann, J., & Sadaghiani, C. (2014). Therapeutic benefit of balneotherapy and hydrotherapy in the management of fibromyalgia syndrome: A qualitative systematic review and meta-analysis of randomized controlled trials. *Arthritis Research and Therapy*, *16*(4), R141. https://doi.org/10.1186/ar4603

Neblett, R., Cohen, H., Choi, Y., Hartzell, M. M., Williams, M., Mayer, T. G., & Gatchel, R. J. (2013). The central sensitization inventory (CSI): Establishing clinically significant values for identifying central sensitivity syndromes in an outpatient chronic pain sample. *Journal of Pain*, *14*(5), 438–445. https://doi.org/10.1016/j.jpain.2012.11.012

Neblett, R., Hartzell, M. M., Mayer, T. G., Cohen, H., & Gatchel, R. J. (2017).

Establishing Clinically Relevant Severity Levels for the Central Sensitization Inventory. *Pain Practice*, *17*(2), 166–175. https://doi.org/10.1111/papr.12440

Neumann, I. D., & Slattery, D. A. (2016). Oxytocin in General Anxiety and Social Fear: A Translational Approach. *Biological Psychiatry*, *79*(3), 213–221. https://doi.org/10.1016/j.biopsych.2015.06.004

Neves, E. B. (2017). The effect of body fat percentage and body fat distribution on skin surface temperature with infrared thermography. *Journal of Thermal Biology*, *66*, 1–9. https://doi.org/10.1016/j.jtherbio.2017.03.006

Nijs, J., Loggia, M. L., Polli, A., Moens, M., Huysmans, E., Goudman, L., Meeus, M., Vanderweeën, L., Ickmans, K., & Clauw, D. (2017). Sleep disturbances and severe stress as glial activators: key targets for treating central sensitization in chronic pain patients? *Expert Opinion on Therapeutic Targets*, *21*(8), 817–826. https://doi.org/10.1080/14728222.2017.1353603

Nijs, J., Paul van Wilgen, C., Van Oosterwijck, J., van Ittersum, M., & Meeus, M. (2011). How to explain central sensitization to patients with "unexplained" chronic musculoskeletal pain: Practice guidelines. *Manual Therapy*, *16*(5), 413–418. https://doi.org/10.1016/j.math.2011.04.005

Nijs, J., & Van Houdenhove, B. (2009). From acute musculoskeletal pain to chronic widespread pain and fibromyalgia: Application of pain neurophysiology in manual therapy practice. *Manual Therapy*, *14*(1), 3–12. https://doi.org/10.1016/j.math.2008.03.001

Nijs, J., Van Houdenhove, B., & Oostendorp, R. A. B. (2010). Recognition of central sensitization in patients with musculoskeletal pain: Application of pain neurophysiology in manual therapy practice. *Manual Therapy*, *15*(2), 135–141. https://doi.org/10.1016/j.math.2009.12.001

Nishishinya, B., Urrútia, G., Walitt, B., Rodriguez, A., Bonfill, X., Alegre, C., & Darko, G. (2008). Amitriptyline in the treatment of fibromyalgia: A systematic review of its efficacy. *Rheumatology (Oxford)*, *47*(12), 1741–1746. https://doi.org/10.1093/rheumatology/ken317

Oaklander, A. L., Herzog, Z. D., Downs, H. M., & Klein, M. M. (2013). Objective

evidence that small-fiber polyneuropathy underlies some illnesses currently labeled as fibromyalgia. *Pain*, *154*(11), 2310–2316. https://doi.org/10.1016/j.pain.2013.06.001

Ortega, R., Saavedra-Alías, J., Liébana-Cañada, A., Sánchez-Muñoz, B., Martínez-Martos, J., & Ramírez-Expósito, M. (2013). Circulating Aminopeptidase Activities in Men and Women with Essential Hypertension. *Current Medicinal Chemistry*, *20*(38), 4935–4945. https://doi.org/10.2174/15672050113109990205

Ozgocmen, S., Ozyurt, H., Sogut, S., Akyol, O., Ardicoglu, O., & Yildizhan, H. (2006). Antioxidant status, lipid peroxidation and nitric oxide in fibromyalgia: Etiologic and therapeutic concerns. *Rheumatology International*, *26*(7), 598–603. https://doi.org/10.1007/s00296-005-0079-y

Paus, R., Theoharides, T. C., & Arck, P. C. (2006). Neuroimmunoendocrine circuitry of the "brain-skin connection." *Trends in Immunology*, *27*(1), 32–39. https://doi.org/10.1016/j.it.2005.10.002

Peck, M. M., Maram, R., Mohamed, A., Ochoa Crespo, D., Kaur, G., Ashraf, I., & Malik, B. H. (2020). The Influence of Pro-inflammatory Cytokines and Genetic Variants in the Development of Fibromyalgia: A Traditional Review. *Cureus*, *12*(9). https://doi.org/10.7759/cureus.10276

Pérez-Aranda, A., Andrés-Rodríguez, L., Feliu-Soler, A., Núñez, C., Stephan-Otto, C., Pastor-Mira, M. A., López-Roig, S., Peñacoba, C., Calandre, E. P., Slim, M., Salgueiro, M., Feixas, G., & Luciano, J. V. (2019). Clustering a large Spanish sample of patients with fibromyalgia using the Fibromyalgia Impact Questionnaire-Revised: Differences in clinical outcomes, economic costs, inflammatory markers, and gray matter volumes. *Pain*, *160*(4), 908–921. https://doi.org/10.1097/j.pain.0000000000001468

Perrot, S., Cohen, M., Barke, A., Korwisi, B., Rief, W., & Treede, R.-D. (2019). The IASP classification of chronic pain for ICD-11. *Pain*, *160*(1), 77–82. https://doi.org/10.1097/j.pain.0000000000001389

Perrot, S., Schaefer, C., Knight, T., Hufstader, M., Chandran, A. B., & Zlateva, G. (2012). Societal and individual burden of illness among fibromyalgia patients in France: Association between disease severity and OMERACT core domains. *BMC*

Musculoskeletal Disorders, *13*, 22. https://doi.org/10.1186/1471-2474-13-22

Perry, R., Terry, R., & Ernst, E. (2010). A systematic review of homoeopathy for the treatment of fibromyalgia. *Clinical Rheumatology*, *29*(5), 457–464. https://doi.org/10.1007/s10067-009-1361-2

Pickering, G., Achard, A., Corriger, A., Sickout-Arondo, S., Macian, N., Leray, V., Lucchini, C., Cardot, J. M., & Pereira, B. (2020). Electrochemical Skin Conductance and Quantitative Sensory Testing on Fibromyalgia. *Pain Practice*, *20*(4), 348–356. https://doi.org/10.1111/papr.12857

Porth CM. (2011). Inflammation, the inflammatory response and fever. In W. K. H. W. & Wilkins (Ed.), *Essentials of pathophysiology* (3rd edn, pp. 51–72). China.

Prabhakar, A., Kaiser, J. M., Novitch, M. B., Cornett, E. M., Urman, R. D., & Kaye, A. D. (2019). The Role of Complementary and Alternative Medicine Treatments in Fibromyalgia: a Comprehensive Review. *Current Rheumatology Reports*, 21(5), 14. https://doi.org/10.1007/s11926-019-0814-0

Queiroz, L. P. (2013). Worldwide epidemiology of fibromyalgia topical collection on fibromyalgia. *Current Pain and Headache Reports*, *17*(8), 356. https://doi.org/10.1007/s11916-013-0356-5

Ring, E. F. J., & Ammer, K. (2012). Infrared thermal imaging in medicine. *Physiological Measurement*, *33*(3), R33–R46. https://doi.org/10.1088/0967-3334/33/3/R33

Rivera, J., Rejas, J., Esteve-Vives, J., & Vallejo, M. A. (2009). Resource utilisation and health care costs in patients diagnosed with fibromyalgia in Spain. *Clinical and Experimental Rheumatology*, *27*(5 suppl. 56), S39-45.

Rodríguez-Martín, J. M. (2014). *Electroterapia en Fisioterapia* (3ª edición). Editorial Médica Panamericana.

Rodriguez-Pintó, I., Agmon-Levin, N., Howard, A., & Shoenfeld, Y. (2014). Fibromyalgia and cytokines. *Immunology Letters*, *161*(2), 200–203. https://doi.org/10.1016/j.imlet.2014.01.009

Roitenberg, N., & Shoshana, A. (2019). Physiotherapists' accounts of fibromyalgia: role-uncertainty and professional shortcomings. *Disability and Rehabilitation*.

https://doi.org/10.1080/09638288.2019.1632939

Roskell, N. S., Beard, S. M., Zhao, Y., & Le, T. K. (2011). A meta-analysis of pain response in the treatment of fibromyalgia. *Pain Practice*, *11*(6), 516–527. https://doi.org/10.1111/j.1533-2500.2010.00441.x

Rossi, A., Di Lollo, A. C., Guzzo, M. P., Giacomelli, C., Atzeni, F., Bazzichi, L., & Di Franco, M. (2015). Fibromyalgia and nutrition: What news? *Clinical and Experimental Rheumatology*, *33*, S117–S125.

Ruiz-Cabello, P., Soriano-Maldonado, A., Delgado-Fernandez, M., Alvarez-Gallardo, I. C., Segura-Jimenez, V., Estevez-Lopez, F., Camiletti-Moirón, D., & Aparicio, V. A. (2017). Association of Dietary Habits with Psychosocial Outcomes in Women with Fibromyalgia: The al-Ándalus Project. *Journal of the Academy of Nutrition and Dietetics*, *117*(3), 422-432.e1. https://doi.org/10.1016/j.jand.2016.09.023

Rus, A., Castro, L., Del Moral, M. L., & Peinado, Á. (2010). Inducible NOS inhibitor 1400W reduces hypoxia/re-oxygenation injury in rat lung. *Redox Report*, *15*(4), 169–178. https://doi.org/10.1179/174329210X12650506623609

Rus, A., Molina, F., Del Moral, M. L., Ramírez-Expósito, M. J., & Martínez-Martos, J. M. (2018). Catecholamine and Indolamine Pathway: A Case–Control Study in Fibromyalgia. *Biological Research for Nursing*, *20*(5), 577–586. https://doi.org/10.1177/1099800418787672

Rus, A., Molina, F., Gassó, M., Camacho, M. V., Peinado, M. Á., & Moral, M. L. del. (2016). Nitric Oxide, Inflammation, Lipid Profile, and Cortisol in Normal- and Overweight Women With Fibromyalgia. *Biological Research for Nursing*, *18*(2), 138–146. https://doi.org/10.1177/1099800415591035

Rus, A., Molina, F., Ramos, M. M., Martínez-Ramírez, M. J., & del Moral, M. L. (2017). Extra Virgin Olive Oil Improves Oxidative Stress, Functional Capacity, and Health-Related Psychological Status in Patients With Fibromyalgia: A Preliminary Study. *Biological Research for Nursing*, *19*(1), 106–115. https://doi.org/10.1177/1099800416659370

Russell, I. J., & Larson, A. A. (2009). Neurophysiopathogenesis of Fibromyalgia Syndrome: A Unified Hypothesis. *Rheumatic Disease Clinics of North America*,

34(2), 421–435. https://doi.org/10.1016/j.rdc.2009.06.005

Sagaidachnyi, A. A., Skripal, A. V, Fomin, A. V, & Usanov, D. A. (2014). Determination of the amplitude and phase relationships between oscillations in skin temperature and photoplethysmography-measured blood flow in fingertips. *Physiological Measurement*, *35*(2), 153–166. https://doi.org/10.1088/0967-3334/35/2/153

Salgueiro, M., García-Leiva, J. M., Ballesteros, J., Hidalgo, J., Molina, R., & Calandre, E. P. (2013). Validation of a Spanish version of the Revised Fibromyalgia Impact Questionnaire (FIQR). *Health and Quality of Life Outcomes*, *11*(1), 132. https://doi.org/10.1186/1477-7525-11-132

Sanchez-Marin, F. J., Calixto-Carrera, S., & Villaseñor-Mora, C. (2009). Novel approach to assess the emissivity of the human skin. *Journal of Biomedical Optics*, *14*(2), 024006. https://doi.org/10.1117/1.3086612

Sarzi-Puttini, P., Giorgi, V., Marotto, D., & Atzeni, F. (2020). Fibromyalgia: an update on clinical characteristics, aetiopathogenesis and treatment. *Nature Reviews Rheumatology*, *16*(11), 645–660. https://doi.org/10.1038/s41584-020-00506-w

Schaefer, C., Mann, R., Masters, E. T., Cappelleri, J. C., Daniel, S. R., Zlateva, G., McElroy, H. J., Chandran, A. B., Adams, E. H., Assaf, A. R., McNett, M., Mease, P., Silverman, S., & Staud, R. (2016). The Comparative Burden of Chronic Widespread Pain and Fibromyalgia in the United States. *Pain Practice*, *16*(5), 565–579. https://doi.org/10.1111/papr.12302

Scholander, P. F., Hock, R., Walters, V., Johnson, F., & Irving, L. (1950). Heat regulation in some arctic and tropical mammals and birds. *The Biological Bulletin*, *99*(2), 237–258. https://doi.org/10.2307/1538741

Schulman, H. (1997). Nitric oxide: A spatial second messenger. *Molecular Psychiatry*, *2*(4), 296–299. https://doi.org/10.1038/sj.mp.4000197

Scolnik, M., Vasta, B., Hart, D. J., Shipley, J. A., McHugh, N. J., & Pauling, J. D. (2016). Symptoms of Raynaud's phenomenon (RP) in fibromyalgia syndrome are similar to those reported in primary RP despite differences in objective assessment of digital microvascular function and morphology. *Rheumatology International*, *36*(10), 1371–1377. https://doi.org/10.1007/s00296-016-3483-6

Sears, B. (2015). Anti-inflammatory Diets. *Journal of the American College of Nutrition*, *34*, 14–21. https://doi.org/10.1080/07315724.2015.1080105

Sears, B., & Ricordi, C. (2011). Anti-inflammatory nutrition as a pharmacological approach to treat obesity. *Journal of Obesity*, *2011*. https://doi.org/10.1155/2011/431985

Secomb, T. W. (2008). Theoretical models for regulation of blood flow. *Microcirculation*, *15*(8), 765–775. https://doi.org/10.1080/10739680802350112

Segura-Jiménez, V., Castro-Piñero, J., Soriano-Maldonado, A., Álvarez-Gallardo, I. C., Estévez-López, F., Delgado-Fernández, M., & Carbonell-Baeza, A. (2016). The association of total and central body fat with pain, fatigue and the impact of fibromyalgia in women; Role of physical fitness. *European Journal of Pain (United Kingdom)*, *20*(5), 811–821. https://doi.org/10.1002/ejp.807

Sendur, O. F., Turan, Y., Tastaban, E., Yenisey, C., & Serter, M. (2009). Serum antioxidants and nitric oxide levels in fibromyalgia: A controlled study. *Rheumatology International*, *29*(6), 629–633. https://doi.org/10.1007/s00296-008-0738-x

Serdaroğlu Beyazal, M., Tüfekçi, A., Kirbaş, S., & Topaloğlu, M. S. (2018). The impact of fibromyalgia on disability, anxiety, depression, sleep disturbance, and quality of life in patients with migraine. *Noropsikiyatri Arsivi*, *55*(2), 140–145. https://doi.org/10.5152/npa.2016.12691

Serrat, M., Almirall, M., Musté, M., Sanabria-Mazo, J. P., Feliu-Soler, A., Méndez-Ulrich, J. L., Luciano, J. V., & Sanz, A. (2020). Effectiveness of a Multicomponent Treatment for Fibromyalgia Based on Pain Neuroscience Education, Exercise Therapy, Psychological Support, and Nature Exposure (NAT-FM): A Pragmatic Randomized Controlled Trial. *Journal of Clinical Medicine*, *9*(10), 3348. https://doi.org/10.3390/jcm9103348

Shaver, J. L. F., & Zenk, S. N. (2000). Sleep disturbance in menopause. In *Journal of Women's Health and Gender-Based Medicine*, *9*(2), 109-118. https://doi.org/10.1089/152460900318605

Shim, J.-S., Oh, K., & Kim, H. C. (2014). Dietary assessment methods in epidemiologic

studies. *Epidemiology and Health*, *36*, e2014009. https://doi.org/10.4178/epih/e2014009

Shivappa, N., Hebert, J. R., Marcos, A., Diaz, L. E., Gomez, S., Nova, E., Michels, N., Arouca, A., González-Gil, E., Frederic, G., González-Gross, M., Castillo, M. J., Manios, Y., Kersting, M., Gunter, M. J., De Henauw, S., Antonios, K., Widhalm, K., Molnar, D., … Huybrechts, I. (2017). Association between dietary inflammatory index and inflammatory markers in the HELENA study. *Molecular Nutrition and Food Research*, *61*(6). https://doi.org/10.1002/mnfr.201600707

Shivappa, N., Hébert, J. R., Rietzschel, E. R., De Buyzere, M. L., Langlois, M., Debruyne, E., Marcos, A., & Huybrechts, I. (2015). Associations between dietary inflammatory index and inflammatory markers in the Asklepios Study. *British Journal of Nutrition*, *113*(4), 665–671. https://doi.org/10.1017/S000711451400395X

Shivappa, N., Steck, S. E., Hurley, T. G., Hussey, J. R., & Hébert, J. R. (2014). Designing and developing a literature-derived, population-based dietary inflammatory index. *Public Health Nutrition*, *17*(08), 1689–1696. https://doi.org/10.1017/S1368980013002115

Silva, A. R., Bernardo, A., Costa, J., Cardoso, A., Santos, P., de Mesquita, M. F., Vaz Patto, J., Moreira, P., Silva, M. L., & Padrão, P. (2019). Dietary interventions in fibromyalgia: a systematic review. *Annals of Medicine*, *51*(1), 2–14. https://doi.org/10.1080/07853890.2018.1564360

Sköldstam, L., Brudin, L., Hagfors, L., & Johansson, G. (2005). Weight reduction is not a major reason for improvement in rheumatoid arthritis from lacto-vegetarian, vegan or Mediterranean diets. *Nutrition Journal*, *4*, 15. https://doi.org/10.1186/1475-2891-4-15

Sluka, K. A., & Clauw, D. J. (2016). Neurobiology of fibromyalgia and chronic widespread pain. *Neuroscience*, *338*, 114–129. https://doi.org/10.1016/j.neuroscience.2016.06.006

Smythe, H. A. (1972). Nonarticular rheumatism and psychogenic musculoskeletal syndromes. In McCarty DJ. (Ed.), *Arthritis and Allied Conditions* (edn, pp. 881–891). Lea & Febiger.

Stisi, S., Cazzola, M., Buskila, D., Spath, M., Giamberardino, M. A., Sarzi-Puttini, P.,

Arioli, G., Alciati, A., Leardini, G., Gorla, R., Marsico, A., Ceccherelli, F., Bazzichi, L., Carignola, R., Gracely, R. H., Salaffi, F., Marinangeli, F., Torta, R., Di Franco, M., … Atzeni, F. (2007). Meccanismi eziopatogenetici della sindrome fibromialgica. *Reumatismo*, *60*(suppl. 1), 25–35. https://doi.org/10.4081/reumatismo.2008.1s.25

Strath, L. J., Jones, C. D., George, A. P., Lukens, S. L., Morrison, S. A., Soleymani, T., Locher, J. L., Gower, B. A., & Sorge, R. E. (2020). The effect of low-carbohydrate and low-fat diets on pain in individuals with knee osteoarthritis. *Pain Medicine (United States)*, *21*(1), 150–160. https://doi.org/10.1093/pm/pnz022

Sturgeon, J. A., Darnall, B. D., Zwickey, H. L., Wood, L. J., Hanes, D. A., Zava, D. T., & Mackey, S. C. (2014). Proinflammatory cytokines and DHEA-S in women with fibromyalgia: Impact of psychological distress and menopausal status. *Journal of Pain Research*, *7*, 707–716. https://doi.org/10.2147/JPR.S71344

Tabung, F. K., Steck, S. E., Zhang, J., Ma, Y., Liese, A. D., Agalliu, I., Hingle, M., Hou, L., Hurley, T. G., Jiao, L., Martin, L. W., Millen, A. E., Park, H. L., Rosal, M. C., Shikany, J. M., Shivappa, N., Ockene, J. K., & Hebert, J. R. (2015). Construct validation of the dietary inflammatory index among postmenopausal women. *Annals of Epidemiology*, *25*(6), 398–405. https://doi.org/10.1016/j.annepidem.2015.03.009

Tan, J. H., Ng, E. Y. K., Rajendra Acharya, U., & Chee, C. (2009). Infrared thermography on ocular surface temperature: A review. *Infrared Physics and Technology*, *52*(4), 97-108. https://doi.org/10.1016/j.infrared.2009.05.002

Theoharides, T. C., Zhang, B., Kempuraj, D., Tagen, M., Vasiadi, M., Angelidou, A., Alysandratos, K.-D., Kalogeromitros, D., Asadi, S., Stavrianeas, N., Peterson, E., Leeman, S., & Conti, P. (2010). IL-33 augments substance P-induced VEGF secretion from human mast cells and is increased in psoriatic skin. *Proceedings of the National Academy of Sciences*, *107*(9), 4448–4453. https://doi.org/10.1073/pnas.1000803107

Theoharides, Theoharis C., Valent, P., & Akin, C. (2015). Mast cells, mastocytosis, and related disorders. *New England Journal of Medicine*, *373*(2), 163-172. https://doi.org/10.1056/NEJMra1409760

Tian, D., Tian, M., Zhang, L., Zhao, P., Cui, Y., & Li, J. (2018). High fat diet sensitizes fibromyalgia-like pain behaviors in mice via tumor necrosis factor alpha. *PLoS ONE*,

13(2), e0190861. https://doi.org/10.1371/journal.pone.0190861

Tick, H. (2015). Nutrition and Pain. *Physical Medicine and Rehabilitation Clinics of North America*, *26*(2), 309–320. https://doi.org/10.1016/j.pmr.2014.12.006

Torpy, D. J., Papanicolaou, D. A., Lotsikas, A. J., Wilder, R. L., Chrousos, G. P., & Pillemer, S. R. (2000). Responses of the sympathetic nervous system and the hypothalamic-pituitary-adrenal axis to interleukin-6: A pilot study in fibromyalgia. *Arthritis and Rheumatism*, *43*(4), 872–880. https://doi.org/10.1002/1529-0131(200004)43:4<872::AID-ANR19>3.0.CO;2-T

Traut, E. F. (1968). Fibrositis. *Journal of the American Geriatrics Society*, *16*(5), 531–538. https://doi.org/10.1111/j.1532-5415.1968.tb02092.x

Treede, R. D., Rief, W., Barke, A., Aziz, Q., Bennett, M. I., Benoliel, R., Cohen, M., Evers, S., Finnerup, N. B., First, M. B., Giamberardino, M. A., Kaasa, S., Korwisi, B., Kosek, E., Lavand'Homme, P., Nicholas, M., Perrot, S., Scholz, J., Schug, S., … Wang, S. J. (2019). Chronic pain as a symptom or a disease: The IASP Classification of Chronic Pain for the International Classification of Diseases (ICD-11). *Pain*, *160*(1), 19–27. https://doi.org/10.1097/j.pain.0000000000001384

Tsilioni, I., Russell, I. J., Stewart, J. M., Gleason, R. M., & Theoharides, T. C. (2016). Neuropeptides CRH, SP, HK-1, and Inflammatory Cytokines IL-6 and TNF Are Increased in Serum of Patients with Fibromyalgia Syndrome, Implicating Mast Cells. *Journal of Pharmacology and Experimental Therapeutics*, *356*(3), 664–672. https://doi.org/10.1124/jpet.115.230060

Üçeyler, N., Häuser, W., & Sommer, C. (2011). Systematic review with meta-analysis: Cytokines in fibromyalgia syndrome. *BMC Musculoskeletal Disorders*, *12*, 245. https://doi.org/10.1186/1471-2474-12-245

van Ittersum, M. W., van Wilgen, C. P., van der Schans, C. P., Lambrecht, L., Groothoff, J. W., & Nijs, J. (2014). Written Pain Neuroscience Education in Fibromyalgia: A Multicenter Randomized Controlled Trial. *Pain Practice*, *14*(8), 689–700. https://doi.org/10.1111/papr.12137

Vanderweeën, L., Oostendorp, R. A. B., Vaes, P., & Duquet, W. (1996). Pressure algometry in manual therapy. *Manual Therapy*, *1*(5), 258–265.

https://doi.org/10.1054/math.1996.0276

Vardasca, R., Magalhaes, C., Marques, D., Moreira, J., Frade, R., Seixas, A., Mendes, J., & Ring, F. (2019). Bilateral assessment of body core temperature through axillar, tympanic and inner canthi thermometers in a young population. *Physiological Measurement*, *40*(9). https://doi.org/10.1088/1361-6579/ab2af6

Vázquez Morejón, A. J., Vázquez-Morejón Jiménez, R., & Zanin, G. B. (2014). Beck anxiety inventory: Psychometric characteristics in a sample from the clinical spanish population. *Spanish Journal of Psychology*, *17*(2). https://doi.org/10.1017/sjp.2014.76

Walitt, B., Nahin, R. L., Katz, R. S., Bergman, M. J., & Wolfe, F. (2015). The Prevalence and Characteristics of Fibromyalgia in the 2012 National Health Interview Survey. *PLOS ONE*, *10*(9), e0138024. https://doi.org/10.1371/journal.pone.0138024

Walløe, L. (2016). Arterio-venous anastomoses in the human skin and their role in temperature control. *Temperature (Austin, Tex.)*, *3*(1), 92–103. https://doi.org/10.1080/23328940.2015.1088502

Watkins, L. R., Hutchinson, M. R., Ledeboer, A., Wieseler-Frank, J., Milligan, E. D., & Maier, S. F. (2007). Glia as the "bad guys": Implications for improving clinical pain control and the clinical utility of opioids. *Brain, Behavior, and Immunity*, *21*(2), 131–146. https://doi.org/10.1016/j.bbi.2006.10.011

Watson, J. A., Ryan, C. G., Cooper, L., Ellington, D., Whittle, R., Lavender, M., Dixon, J., Atkinson, G., Cooper, K., & Martin, D. J. (2019). Pain Neuroscience Education for Adults With Chronic Musculoskeletal Pain: A Mixed-Methods Systematic Review and Meta-Analysis. In *Journal of Pain*, *20*(10), 1140.e1-1140.e22. https://doi.org/10.1016/j.jpain.2019.02.011

Watt, F. E. (2018). Musculoskeletal pain and menopause. *Post Reproductive Health*, *24*(1), 34–43. https://doi.org/10.1177/2053369118757537

Wiffen, P. J., Derry, S., Moore, R. A., Aldington, D., Cole, P., Rice, A. S. C., Lunn, M. P. T., Hamunen, K., Haanpaa, M., & Kalso, E. A. (2013). Antiepileptic drugs for neuropathic pain and fibromyalgia - an overview of Cochrane reviews. *Cochrane Database of Systematic Reviews*, *2013*(11), CD010567. https://doi.org/10.1002/14651858.CD010567.pub2

Wirth, M. D., Burch, J., Shivappa, N., Violanti, J. M., Burchfiel, C. M., Fekedulegn, D., Andrew, M. E., Hartley, T. A., Miller, D. B., Mnatsakanova, A., Charles, L. E., Steck, S. E., Hurley, T. G., Vena, J. E., & Hébert, J. R. (2014). Association of a dietary inflammatory index with inflammatory indices and metabolic syndrome among police officers. *Journal of Occupational and Environmental Medicine*, *56*(9), 986–989. https://doi.org/10.1097/JOM.0000000000000213

Witkamp, R. F., & van Norren, K. (2018). Let thy food be thy medicine….when possible. *European Journal of Pharmacology*, *836*, 102–114. https://doi.org/10.1016/j.ejphar.2018.06.026

Wolfe, F., Clauw, D. J., Fitzcharles, M. A., Goldenberg, D. L., Häuser, W., Katz, R. L., Mease, P. J., Russell, A. S., Russell, I. J., & Walitt, B. (2016). 2016 Revisions to the 2010/2011 fibromyalgia diagnostic criteria. *Seminars in Arthritis and Rheumatism*, *46*(3), 319–329. https://doi.org/10.1016/j.semarthrit.2016.08.012

Wolfe, F., Clauw, D. J., Fitzcharles, M. A., Goldenberg, D. L., Katz, R. S., Mease, P., Russell, A. S., Russell, I. J., Winfield, J. B., & Yunus, M. B. (2010). The American College of Rheumatology preliminary diagnostic criteria for fibromyalgia and measurement of symptom severity. *Arthritis Care and Research*, *62*(5), 600–610. https://doi.org/10.1002/acr.20140

Wolfe, F., Smythe, H. A., Yunus, M. B., Bennett, R. M., Bombardier, C., Goldenberg, D. L., Tugwell, P., Campbell, S. M., Abeles, M., Clark, P., Fam, A. G., Farber, S. J., Fiechtner, J. J., Michael Franklin, C., Gatter, R. A., Hamaty, D., Lessard, J., Lichtbroun, A. S., Masi, A. T., … Sheon, R. P. (1990). The american college of rheumatology 1990 criteria for the classification of fibromyalgia. *Arthritis & Rheumatism*, *33*(2), 160–172. https://doi.org/10.1002/art.1780330203

Wolfe, F., & Walitt, B. (2013). Culture, science and the changing nature of fibromyalgia. *Nature Reviews Rheumatology*, *9*(12), 751-755. https://doi.org/10.1038/nrrheum.2013.96

Wood, L., & Hendrick, P. A. (2019). A systematic review and meta-analysis of pain neuroscience education for chronic low back pain: Short-and long-term outcomes of pain and disability. In *European Journal of Pain (United Kingdom)*, *23*(2), 234–249. https://doi.org/10.1002/ejp.1314

Xin, Q., Bai, B., & Liu, W. (2017). The analgesic effects of oxytocin in the peripheral and central nervous system. *Neurochemistry International*, *103*, 57–64. https://doi.org/10.1016/j.neuint.2016.12.021

Yuan, S. L. K., Matsutani, L. A., & Marques, A. P. (2015). Effectiveness of different styles of massage therapy in fibromyalgia: A systematic review and meta-analysis. *Manual Therapy*, *20*(2), 257-264. https://doi.org/10.1016/j.math.2014.09.003

Yunus, M. B. (2007). Fibromyalgia and Overlapping Disorders: The Unifying Concept of Central Sensitivity Syndromes. *Seminars in Arthritis and Rheumatism*, *36*(6), 339–356. https://doi.org/10.1016/j.semarthrit.2006.12.009

Yunus, M., Masi, A. T., Calabro, J. J., Miller, K. A., & Feigenbaum, S. L. (1981). Primary fibromyalgia (fibrositis): Clinical study of 50 patients with matched normal controls. *Seminars in Arthritis and Rheumatism*, *11*(1), 151–171. https://doi.org/10.1016/0049-0172(81)90096-2

Zhang, Z., Cherryholmes, G., Mao, A., Marek, C., Longmate, J., Kalos, M., St. Amand, R. P., & Shively, J. E. (2008). High plasma levels of MCP-1 and eotaxin provide evidence for an immunological basis of fibromyalgia. *Experimental Biology and Medicine*, *233*(9), 1171–1180. https://doi.org/10.3181/0712-RM-328

Zin, C. S., Nissen, L. M., O'Callaghan, J. P., Moore, B. J., & Smith, M. T. (2010). Preliminary study of the plasma and cerebrospinal fluid concentrations of IL-6 and IL-10 in patients with chronic pain receiving intrathecal opioid infusions by chronically implanted pump for pain management. *Pain Medicine*, *11*(4), 550–561. https://doi.org/10.1111/j.1526-4637.2010.00821.x

APPENDIX 1

Ethics Committee of the University of Granada (Spain)

JUNTA DE ANDALUCIA

CONSEJERÍA DE SALUD
Dirección General de Calidad, Investigación, Desarrollo e Innovación

DICTAMEN ÚNICO EN LA COMUNIDAD AUTÓNOMA DE ANDALUCÍA

D/Dª: CRISTINA LUCIA DAVILA FAJARDO como secretario/a del CEIM/CEI Provincial de Granada

CERTIFICA

Que este Comité ha evaluado la propuesta del promotor/investigador (No hay promotor/a asociado/a) para realizar el estudio de investigación titulado:

TÍTULO DEL ESTUDIO:	Alteraciones vasculares, calidad de la dieta y su relación con el dolor en pacientes con Fibromialgia ,(Alteraciones vasculares y nutricionales en fibromialgia)
Protocolo, Versión:	1
HIP, Versión:	1
CI, Versión:	1

Y que considera que:

Se cumplen los requisitos necesarios de idoneidad del protocolo en relación con los objetivos del estudio y se ajusta a los principios éticos aplicables a este tipo de estudios.

La capacidad del/de la investigador/a y los medios disponibles son apropiados para llevar a cabo el estudio.

Están justificados los riesgos y molestias previsibles para los participantes.

Que los aspectos económicos involucrados en el proyecto, no interfieren con respecto a los postulados éticos.

Y que este Comité considera, que dicho estudio puede ser realizado en los Centros de la Comunidad Autónoma de Andalucía que se relacionan, para lo cual corresponde a la Dirección del Centro correspondiente determinar si la capacidad y los medios disponibles son apropiados para llevar a cabo el estudio.

Lo que firmo en Granada a 31/12/2018

D/Dª. CRISTINA LUCIA DAVILA FAJARDO, como Secretario/a del CEIM/CEI Provincial de Granada

Código Seguro De Verificación:	e64d66881b1e876ea2b068519af000cd202e8ebf	Fecha	31/12/2018
Normativa	Este documento incorpora firma electrónica reconocida de acuerdo a la Ley 59/2003, de 19 de diciembre, de firma electrónica.		
Firmado Por	Cristina Lucia Davila Fajardo		
Url De Verificación	https://www.juntadeandalucia.es/salud/portaldeetica/xhtml/ayuda/verificarFirmaDocumento.iface/code/e64d66881b1e876ea2b068519af000cd202e8ebf	Página	1/2

CERTIFICA

Que este Comité ha ponderado y evaluado en sesión celebrada el 21/12/2018 y recogida en acta 12/18 la propuesta del/de la Promotor/a (No hay promotor/a asociado/a), para realizar el estudio de investigación titulado:

TÍTULO DEL ESTUDIO:	Alteraciones vasculares, calidad de la dieta y su relación con el dolor en pacientes con Fibromialgia ,(Alteraciones vasculares y nutricionales en fibromialgia)
Protocolo, Versión:	1
HIP, Versión:	1
CI, Versión:	1

Que a dicha sesión asistieron los siguientes integrantes del Comité:

Presidente/a

D/Dª. Fidel Fernández Quesada

Vicepresidente/a

D/Dª. Francisco Manuel Luque Martínez

Secretario/a

D/Dª. CRISTINA LUCIA DAVILA FAJARDO

Vocales

D/Dª. JESÚS CARDONA CONTRERAS
D/Dª. LUIS MIGUEL DOMENECH GIL
D/Dª. Jesús Martínez Tapias
D/Dª. Juan Ramón Delgado Pérez
D/Dª. Berta Gorlat Sánchez
D/Dª. José Dario Sánchez López
D/Dª. Juana María de Haro Castellano
D/Dª. José Cabeza Barrera
D/Dª. Juan Morales Arcas
D/Dª. Juan Mozas Moreno
D/Dª. José Uberos Fernández
D/Dª. José Antonio López Escámez
D/Dª. MAXIMILIANO OCETE ESPINOLA
D/Dª. Joaquina Martínez Galán
D/Dª. AURORA BUENO CAVANILLAS
D/Dª. Paloma Muñoz de Rueda
D/Dª. Manuel Gálvez Ibáñez
D/Dª. Esther Espínola García
D/Dª. FRANCISCO LUIS MANZANO MANZANO
D/Dª. MARÍA DEL PILAR GONZÁLEZ CARRIÓN
D/Dª. JUAN ROMERO COTELO
D/Dª. JUAN DIAZ GARCIA
D/Dª. Pilar Guijosa Campos
D/Dª. José Luis Martín Ruiz

Que dicho Comité, está constituido y actua de acuerdo con la normativa vigente y las directrices de la Conferencia Internacional de Buena Práctica Clínica.

Lo que firmo en Granada a 31/12/2018

Código Seguro De Verificación:	e64d66881b1e876ea2b068519af000cd202e8ebf	Fecha	31/12/2018
Normativa	Este documento incorpora firma electrónica reconocida de acuerdo a la Ley 59/2003, de 19 de diciembre, de firma electrónica.		
Firmado Por	Cristina Lucia Davila Fajardo		
Url De Verificación	https://www.juntadeandalucia.es/salud/portaldeetica/xhtml/ayuda/verificarFirmaDocumento.iface/code/e64d66881b1e876ea2b068519af000cd202e8ebf	Página	2/2