Fibromyalgia as an Inflammatory Disease: A Look into the Increased Prevalence in Women

Thesis

Presented in Partial Fulfillment of the Requirements for the Degree Master of Science in the Graduate School of The Ohio State University

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2013

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Abstract

Fibromyalgia (FM), a syndrome characterized by chronic pain, fatigue, and psychological distress, has been shown to affect women significantly more than men. While the etiology of this condition is still actively debated, researchers have found evidence correlating trauma, activation of contractile fibroblasts, and the inflammatory immune response with the appearance of FM symptoms. Other studies observed higher circulating levels of pro-inflammatory cytokines, such as IL-6, in women when compared to their same-age male counterparts. Despite this emerging influx in FM research, no previous studies have combined these observations in order to explain why FM is more common in women. By reevaluating measurements collected by researchers over the past two decades, the hypothesis that greater circulating amounts of pro-inflammatory cytokines, such as IL-6, lead to the increased prevalence of FM in women is explored. The data suggests that there is a correlation between the amount of IL-6 and the appearance of FM in women. Additionally, this gathered information supports the development of a novel pathway explaining this relationship. In this pathway, it is proposed that injury activates fibroblasts within fascia, initiating the release of IL-6. The presence of the female hormone estrogen deregulates the normal inflammatory response, thus inhibiting the reduction of the response and leading to the persistent state of inflammation, resulting in the appearance of FM symptoms.
Dedication

For James, Bonnie, and Alexander Jing
Acknowledgments

I would like to extend my deepest gratitude to my advisor, Amanda Agnew, as well to the faculty, staff, and students of The Ohio State University Division of Anatomy, for their endless support and guidance.
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Chapter 1: Introduction

FM (FM) is a chronic pain syndrome that is characterized by widespread musculoskeletal pain, psychological distress, and the appearance of co-afflictions such as chronic fatigue syndrome and irritable bowel syndrome (1,2). Due to the vague clinical presentation and common co-morbidities associated with FM, standardized methods of diagnosis were not established until the late 21\textsuperscript{st} century. Previous accounts of FM symptoms described it as physical manifestation of a mental disease and as a “side effect” of depression (3). In 1990, the American College of Rheumatology (ACR) released defining characteristics and formally recognized FM as an independent disease calling for treatment. According to their analysis, diffuse pain must be felt in all quadrants of the body—right and left sides as well as above and below the waist—for more than three months, and increased tenderness must be felt after applied pressure in 11 out of 18 predetermined myofascial trigger points (4). The ACR updated the FM criteria in 2010 and introduced the somatic symptoms (SS) scale, which addresses the severity of additional, and often more debilitating, afflictions such as fatigue, unrefreshing sleep, headaches, and cognitive symptoms (5).

While these established guidelines encouraged clinicians to identify and diagnose affected patients, the etiology of FM is still actively debated. Due to the pervasive nature of FM pain, several authors have explored the role of the superficial and deep fascia as a
body-wide nociception and communication network. Previously dismissed as “packing material” for “more important body parts,” fascia is beginning to be appreciated as a highly proprioceptive organ that actively responds to and transmits mechanical forces (6,7). Additional studies have examined the histological features of fascia and furthered its role to include initiation of the inflammatory response by fibroblasts. Now emerging as a possible proponent for diseases such as plantar fasciitis and chronic low back pain, fascia may hold many of the keys for understanding muscle action and musculoskeletal pain (6,8).

Examining the cellular components of fascia may begin to explain the physical manifestations of FM pain, however, no previous studies have addressed why FM is nearly six times more likely to affect women--with a prevalence of 3.4% in women and only 0.5% in men—in terms of fascial components (9,10). Researchers and physicians are struggling to attribute this noticeable dichotomy to a single cause, and many studies are currently looking into FM as an inflammatory disease heightened by the female immune response. One such study by Asai et al. found that male peripheral blood mononuclear cells (PBMCs) produce lower levels of the pro-inflammatory cytokine, interleukin-6 (IL-6), than women. After introducing estrogen to the male PBMCs, IL-6 production greatly increased (11). Other studies have credited the appearance of FM to a single traumatic event. After injury, estrogen and testosterone can either positively or negatively regulate the cellular immune response; the disparity between immune cell activation and cytokine production results in the dichotomy in immune function between the sexes (12). Abnormal immune responses can lead to improper healing after trauma
due to mast cell degranulation and separation of myofibers in the extracellular matrix. This disruption can cause inflammation and the “deep” sensations of pain associated with FM (13).

This thesis will re-examine individually-examined factors, such as fascial components, the immune response and female hormones, in an effort to better understand the emergence of FM symptoms. By re-evaluating available studies addressing the independent roles of fibroblasts within fascia, pro-inflammatory cytokine IL-6, and estrogen on the appearance of FM symptoms, this thesis aims to suggest a novel pathway that explains why FM is more common in women. Data collected in past studies will be used in order to test the hypothesis that greater amounts of proinflammatory cytokines, specifically IL-6, lead to the increased prevalence of FM in women.
Chapter 2: Background

Fascia

Fascia is used as an enveloping term to describe the fatty loose connective tissue underlying the skin and the dense connective tissue enveloping all gross muscles, bundles of muscle fibers, and each individual muscle cell (14). Respectively referred to as superficial and deep fascia, these layers of connective tissue are intimately associated with the muscles, and are continuous with tendons and periosteum throughout the body. Due to its unspecialized and pervasive nature, it was previously assumed that the role of fascia was as a passive contributor to the musculoskeletal system and mere support of biomechanical and postural behavior (15,16). However, fascia, most notably superficial fascia, has been grossly observed to contain an abundance of blood vessels, lymphatics, and nerves that promote continuous communication between the skin and internal structures (7). Recent studies examined the internal functional components, and deemed that fascia may additionally serve as a body-wide mechanosensitive signaling system with the ability to detect and respond to biochemical changes (6,17).

Microscopically, fascia is most simply described as cells in an extracellular matrix. Macrophages, mast cells, and fibroblasts are suspended in a medium composed of ground substance, collagen, and elastic fibers that give fascia its colloidal appearance (14). In superficial fascia, 2-3 sheets of collagen with penetrating elastin fibers are
separated by a thin layer of loose connective tissue (18,19). The collagen fibers of
adjacent layers are oriented perpendicular to one another, ensuring that each layer is
mechanically independent and is able to dissipate pressure loads along different planes
(6,19). In the fascia of the upper limb, there is a greater ratio of elastic to collagen fibers.
This inherent flexibility allows for the completion of complex and fine movements
characteristic of the digits, hands, and wrists (20). In contrast, the major functions of the
lower limb are anti-gravitational and postural, and the limb itself is generally confined to
simpler movements in a few directions. Here, the elastic fibers are widely dispersed
throughout the collagen fiber bundles (20). Fat and fibrous bands tether the superficial
fascia to the deep fascia, in which collagen fibers are more tightly wound and allow for
an accumulation and dispersion of mechanical loading (6,21). In a cadaveric study, Hirata
and colleagues found intergender differences in the histological architecture of fascia.
Females displayed fewer collagen sheets than men, occasionally exhibiting only a thick
monolayer, which was reinforced anteroposteriorly by an abundance of elastic fibers.
Additionally, there was little to no smooth muscle present. The fascia in males generally
contained a greater number of collagen fiber layers, as well as an abundance of smooth
muscle suspended in the extracellular matrix (22).

Free nerve endings are perpendicularly attached to the collagen fibers throughout
the fascia and, along with encapsulated mechanoreceptors, form an extensive intrafascial
signaling network (23–25). Pacini and Ruffini corpuscles are concentrated around blood
vessels and are activated by varying amounts of sensation (24,26). Pacini bodies respond
to vibrations, fast stimulation, and rapid changes in pressure, while Ruffini corpuscles react to long term, slow, and constant manipulation (23,27).

This internal network of communication is further perpetuated by the cellular components of fascia, especially fibroblasts. Fibroblasts define tissue microenvironments, as they are responsible for producing extracellular matrix, and regulate inflammation and wound repair (14,28). These specialized cells are generally seen within muscle fibers and express the gene for alpha-smooth muscle actin, enabling them to display contractile behavior (16,29). Contractile activity is also exhibited within fascia, and several studies have observed that fascial fibroblasts display two distinct phenotypes: fusiform or stellate (30,31). Much like their role in smooth muscle, fibroblasts change shape in response to stretching, and are able to alter connective tissue tension (23). These morphological changes are also accompanied by various modifications in cell signaling, gene expression, and matrix adhesion (7,30). The detection of the associated responses suggest that a phenomenon known as mechanotransduction takes place within the fascia, as the fibroblasts are able to perceive and biochemically adapt to mechanical and applied forces at the cellular level (7,23,30,32). However, adaptation can only occur up to a certain point. Excessive stretching and contraction of fibroblasts can cause hyper-activation of the adjacent free nerve endings, which is perceived as pain (18). Additionally, studies have shown that prolonged environmental stressors activate fibroblasts to deposit excessive and disorganized extracellular matrix. This can produce abnormal adhesions and cause rigidity of the fascia (14,17).
Histological studies of subcutaneous and interstitial connective tissues have observed that fibroblasts form a reticular web with intertwining cytoplasmic processes (31). Thirty percent of these “spokes” extend through the membrane and connect adjacent cells to one another through gap junctions, forming a body-wide signaling network (15,31). This interconnected system of fibroblasts enables the effects of mechanotransduction to be communicated between cells, allowing for comprehensive movement, adaptation, lubrication, and repair of fascia (7). This network generally promotes rapid beneficial responses to external signals, but it also provides a highway for detrimental signals to overwhelm the body. Electrical and chemical reactions travel through the fascia during environmental changes, the immune response, pathological situations, and in wound healing (16,23). As observed by recent histological examination, the extracellular and cellular parts of fascia work together and form a body-wide network for proprioceptive signals, including pain, to be transmitted.

*Trigger Points*

In addition to the widespread dull pain felt by FM patients, studies have shown that up to 70% of affected individuals experience centralized and hyperirritable areas known as myofascial trigger points (MTPs) (33). These taut bands in muscle were first connected to FM in the American College of Rheumatology’s 1990 diagnostic criteria, and offered a concrete pathology on which to concentrate FM treatment. Currently, clinicians agree upon the location of 18 MTPs, which are concentrated in the muscles of the head and neck, shoulder girdle, and lower back of affected individuals (2,33).
The development of these characteristic trigger points has been attributed to acute injury or repetitive trauma that disturbs the sarcoplasmic reticulum of muscle fibers (2). To study this correlation, a group of 102 adults with neck injuries were observed for one year following their traumatic event (34). At the end of the study, 22% of the injured individuals developed FM symptoms, including nonrestorative sleep, chronic pain, appearance of MTPs, and psychological affects (34). Supplementary studies have shown that the risk of developing FM is more than 10-fold higher in adults with neck injuries than any other adults. Additionally, between 25% and 50% of patients surveyed could pinpoint a physically traumatic event within the 6 months prior to the emergence of FM symptoms (34,35).

Traumatic stress or injury involving the muscles and adjacent fascia releases free calcium ions, which cause an increase in metabolism and uncontrolled activation of the muscles’ actin-myosin contractile mechanism (2,17,33). The constant contractions form a taut band of muscle fibers, or MTPs, at the end of an extrafusal muscle spindle (2,33). While MTPs are consistently referred to as areas within muscle, several studies have observed the contractile properties of fibroblasts, which could lead to additional taut band formation within the fascia. As previously stated, some fibroblasts contain actin fibers that are able to contract in fascia as they do in muscle (14,36). This property is essential in the physical repair of fascia after injury; however, it also produces the risk of stimulated over-contraction and inadequate responses to stress or trauma (23). For example, excessive stretching of the aponeurotic fascia leads to an altered nerve arrangement and histological structure reminiscent of the taut bands of MTPs (18). The
muscle spindles in epimysial fascia can also be extended past their normal limit, activating adjacent pain fibers and muscle contraction (18). This contraction releases the neurotransmitter acetylcholine, which has been measured in significantly elevated levels around the MTPs in FM patients (18).

MTPs are highly responsive to stretching of the muscles and fascia due to their location adjacent to proprioceptive muscle spindles. While generally hypersensitive, MTP activation fluctuates in response to general stress, injury, and environmental factors (2). These stressors activate fibroblasts to produce excessive amounts of extracellular matrix and collagen fibers (14,28). Increased production of these cellular components can alter the structure of the fascia and impact pain perception and collagen metabolism (37). Histological studies show greater concentration of collagen layers around terminal nerve endings in FM patients when compared to unaffected controls (37). These hastily-produced collagen layers are also disorganized, and are believed to influence the lower pain threshold at the MTPs (37).

Active MTPs are areas which experience spontaneous pain or pain in response to natural movements, while latent MTPs are only elicit pain under direct pressure (2,33). Shah and colleagues studied the levels of various biochemicals in the trapezius and gastrocnemius muscles in order to demonstrate the interactive molecular milieu of active MTPs. The trapezius muscle contains two bilateral MTPs, and is regularly noted as a cause of discomfort and stiffness in FM patients (38). Conversely, the gastrocnemius muscle is generally uninvolved in FM trigger point pain, and was chosen as a control muscle for this reason (38). Shah measured the levels of the inflammatory mediatory
bradykinin, substance P, pro-inflammatory cytokines IL-6 and TNF-α in both muscles of FM patients. He found elevated levels of all biochemicals in the active trapezius MTPs when compared to the gastrocnemius, indicating that active MTPs are under a chronic state of stress and inflammation (38).

Inflammatory Immune Response

There are two distinct levels of immunity in place to protect the body from further harm after injury or contact with pathogens. The innate immune response is the first active line of defense, and is a non-specific immediate defense mechanism against invading antigens (12). Innate immunity is also responsible for activating the subsequent adaptive immune response, the more robust and directed response, via cytokine release (12). Enabling communication, cytokines are proteins secreted by macrophages, monocytes, or T-cells that either enhance or suppress the activities of other immune cells (39).

Pro-inflammatory cytokines, such as interleukin-6 (IL-6), are released by immune cells in response to infection, stress, or trauma (40). In opposition to anti-inflammatory cytokines, pro-inflammatory cytokines accelerate adaptive immunity and enhance the inflammatory response (39,41–43). Under normal activation, IL-6 is a pro-inflammatory cytokine that is integral in mediating the healing process after injury (44). However, researchers have cited abnormally high levels of pro-inflammatory cytokines associated with symptoms of fatigue, unspecific pain, and depression. This myriad of symptoms is referred to by clinicians as the “sickness behavior phenomenon” and is the hallmark of relatively understudied diseases such as chronic fatigue syndrome and FM (40,42).
In addition to immune cells, several recent studies have discovered that fibroblasts also secrete IL-6 and are able to directly initiate the adaptive inflammatory response after excessive mechanical stretching of the fascia (14,28,45). After IL-6 is secreted by the stressed monocyte, macrophage, or fibroblast, it can act on adjacent or systemic cells with the IL-6 receptor, IL-6Rα. Generally, IL-6 only binds to IL-6Rα on the cell surface of hepatocytes and leukocytes. Glycoprotein 130 (gp130) then travels to the IL-6 complex, and signals a cascade of interactions that maintain cell growth and differentiation and prevent apoptosis (29,39). However, Naugler and Karin found evidence of a “trans-signaling pathway”, in which IL-6Rα binds to IL-6 in blood serum and can activate any cell in the body with gp130 on its surface, which is the majority of cells (39). This “overreaction” causes the influx of adverse effects of IL-6 seen in states of chronic inflammation (39). Other studies suggest that chronic inflammation is a result of disordered fibroblast programming, where the cells are unable to “switch off” mediators of the inflammatory response (28). Normal fibroblast secretion of IL-6 draws leukocytes and other immune cells from the circulating blood to the injured area (28). Effector cells maintain an active immune presence at the injured site until stromal cells signal for their removal (28). This reaction triggers apoptosis and phagocytosis of immune cells, and the inflammatory response gradually subsides. However, in chronic inflammatory diseases, the “survival signals” are maintained and immune cells continue to act on the area—eventually causing pain and tissue scarring (28).

Dysregulation of IL-6 signaling contributes to inflammation-associated conditions including obesity and insulin resistance, inflammatory bowel disease, arthritis, and even
the aging process (39). Supporting the “sickness behavior phenomenon,” IL-6 produces fatigue and pain in healthy people, decreases cognitive function, correlates with depression, influences pain pathways, and promotes B and T-cell proliferation (32). It is hypothesized that these “sickness behavior” symptoms are a result of IL-6 and pro-inflammatory cytokine imbalance affecting the hypothalamic-pituitary-adrenal (HPA) axis (42,46).

The HPA-axis is a neuroendocrine pathway, which controls the body’s responses to stress and immunity by reacting to circulating cytokines (40,46). In this pathway, adrenocorticotropic hormone (ACTH) is released from the anterior pituitary gland. This influx of ACTH stimulates the adrenal glands to produce glucocorticoid hormones, namely cortisol, which suppresses the immune and inflammatory pathways (47). Elevated levels of cortisol provide negative feedback on the hypothalamus and pituitary glands to suppress ACTH, and therefore additional cortisol, production (40). Gur and Oktayoglu discovered that abnormal and reduced activity along the HPA axis mirrored the “sickness behavior” symptoms and also lead to symptoms of fatigue, depression, muscle pain, and disrupted sleep (40).

**Inflammatory Response and IL-6 Production in FM**

The chronically stressed and inflammatory state exhibited in the MTPs is shown to extend to FM patient’s body as a whole. When examining the systemic inflammatory and neuroendocrine stress markers, circulating levels of pro-inflammatory cytokine, IL-6, is significantly increased in female FM patients when compared to healthy controls (40,47,48). Additionally, there is evidence claiming that FM is accompanied by
Immunosuppression as overlapping symptoms include swelling, autonomic disturbances such as irritable bowel and bladder syndromes, abnormally heightened cardiovascular responses to stress, and depression (10). Van West and Maes also measured the levels of immune cells in FM patients and in non-affected controls. In addition to higher overall levels of pro-inflammatory cytokines, patients with FM demonstrated an immune profile with fewer total lymphocytes and higher B and T-cell populations (10). In contrast to the vast majority of studies, two groups of researchers noted no significant differences in pro-inflammatory cytokine levels between those affected and unaffected by FM (49,50). However, after further analysis of the same sample populations, it was found that FM patients show decreased levels of anti-inflammatory cytokines, while the pro-inflammatory cytokine concentrations remain the same (49,50). The levels of pro- and anti-inflammatory cytokines are theorized to act as a scale: when present in the same amounts, both cytokines work together to provide protective immunity, but when one cytokine prevails, autoimmunity occurs (41,51). This theory indicates that the imbalanced cytokine levels observed in some studies will produce the same effects as when the pro-inflammatory cytokines are produced in excess.

*Estrogens*

Estrogens are the hormone complexes that regulate the menstrual and estrus cycles in women. As they are the primary female hormone, they are of great interest when studying clinical dichotomies between the sexes. In women, levels of estrogen vary greatly throughout life and exhibit significant differences throughout the pre-pubertal, pubertal and menopausal stages (52). Estrogens bind to nuclear receptors, which are
expressed by fibroblasts, myoblasts, and immunological cells that produce pro-inflammatory cytokines, including T-cells, B-cells, and macrophages (53–55). This suggested link between estrogens and immune function has been tested over the past few years, as studies have shown that estrogen *in vivo* directly increased the production of inflammatory mediators and IL-6 (12,53).

During the reproductive years, women exhibit a more efficient immune response compared to same-age males due to a more developed thymus and higher antibody concentrations (12,44). This “boost” in the female immune system has also been attributed to the increase in the circulating levels of estrogen (52). As previously stated, estrogen also initiates and influences the progression of the menstrual cycle—a relationship which is evidenced by studies that show women are more prone to symptoms of immune deficiency during menstruation (12). The decline in estrogen during menopause has also been attributed to immune effects, and can stimulate the production of pro-inflammatory cytokines by disrupting the HPA pathway (52). These studies demonstrate estrogen’s inhibitory and promotional effects throughout life and, more generally, show the ability of estrogen to impact immune system efficiency.

As discussed previously, traumatic and injurious events result in the activation of the innate immune system and the production of pro-inflammatory cytokines. However, increased levels of IL-6 leads to the enhanced systemic inflammatory response and can eventually lead to an increased risk of multiple organ dysfunction syndrome and multiple organ failure (12). In patients recovering from moderate to severe burn injuries, there is a higher mortality rate among females than males (12,56). Additionally, estrogen levels are
10-15 times greater in female patients than their uninjured female counterparts (12). This suggests that higher levels of circulating estrogen activated by trauma can actually lead to an improperly mediated immune response and decrease the body’s ability to fight off infection (12,44). Static or increased levels of IL-6 were detected in proestrus mice after induced injury. In contrast, ovariectomized mice lacking estrogen exhibited IL-6 levels that were depressed by 50% (12). Gomez et al. furthered this study and administered estrogen to elderly female mice prior to injury. After trauma, these estrogen-supplemented mice exhibited a significantly improved survival rate and “regular” levels of IL-6 when compared to their normal counterparts (52).

In 2008, Bird and colleagues determined that estrogen results in greater production of IL-6 and therefore, in a more robust immediate immune response. However, studies have shown that IL-6 deficiency actually improves immune response following a burn injury (56). There is additional evidence that osteoarthritis and cardiovascular disease, both attributed to systemic inflammation, are more prevalent in women following menopause (55). Researchers have also shown that post-menopausal women exhibit a constant hyper-inflammatory state with higher circulating levels of IL-6 even in the absence of an immunological trigger (52). This can lead to the slower recovery and greater prevalence of complications in elderly women after injury. Estrogen’s ability to attenuate the abnormal production of IL-6 means that any alteration, increase or loss, to the normal circulating levels of estrogen activates the production of pro-inflammatory cytokine production (52,55).
Chapter 3: Data Analysis

Materials and Methods

To determine the correlation between increased IL-6 levels and the appearance of FM in women, several comparisons of this pro-inflammatory cytokine were made by examining publically available, peer-reviewed articles. For the eight papers that supplied data, IL-6 levels were compared between men and women at rest, between both sexes after injury or stress, and in women with FM versus healthy female controls.

Each study measured circulating IL-6 levels in picograms per milliliter (pg/mL). These values were converted into ratios (i.e. 400 pg/mL IL-6 concentration in females versus 200 pg/mL in males equals 2:1) in order to standardize the results of the various studies with different sample sizes. While Crisostoma et al., Bird et al., and Plackett et al. did not specify a sample size, Asai et al. compared IL-6 values in 10 males to 10 females (12,56,57,11). Prather et al. examined 28 males and 25 females; the latter of which was further separated into groups of 10 pre-menopausal women and 15 post-menopausal women (58). Ortega et al. compared 9 female FM patients to 9 age-matched healthy women (59). Geiss et al. did the same with 12 FM individuals and 15 age-matched controls, and Bote et al. compared IL-6 levels in 25 FM to 20 age-matched healthy controls (47,60).
Results

When comparing the ratios of serum IL-6 in resting females and males, it was found that females have intrinsically greater levels of IL-6, ranging from 1.5 to 25 times greater than male concentrations (Figure 1). Crisostomo et al. were the only group to use mice in their experiment; when this data is removed, the comparison of IL-6 levels remain 1.5-2 times greater in human females than males (57). This relationship is altered when taking into account a traumatic injury or stressful event. During these inflammatory triggers, the male IL-6 levels elevate to 1.67 to 4.5 times greater than female IL-6 concentrations (Figure 2). Prather et al. showed an initial male to female ratio of 1:1.06 (not statistically significant), but further differentiated the female sample to pre- and post-menopausal women. This stratification yielded statistically significant results, indicating that post-menopausal females produce twice as much IL-6 than males during a stressful event. Pre-menopausal female and male ratios effectively remained at 1:1 (Figure 2). A third comparison shows that women with FM exhibit between 1.25 and 4 times greater resting IL-6 levels than their age-matched female counterparts (Figure 3).

The results of IL-6 comparisons did not fully support my hypothesis that greater IL-6 concentrations explains the increased prevalence of FM in women, as males exhibit increased levels during a stressful event.
Figure 1: Ratios of Resting IL-6 Levels in Females and Males
Figure 2: Ratios of Stress-Activated IL-6 Levels in Females and Males

Sample size:
Crisostomo et al.: not specified
Plackett et al.: not specified
Prather et al.: 15 postmenopausal females, 10 premenopausal females, 28 males
Figure 3: Ratios of IL-6 Levels in Healthy Women and Female Fibromyalgia Patients

Sample size:
- Bote et al.: 25 FM patients, 20 HW controls
- Geiss et al.: 12 FM patients, 15 HW controls
- Ortega et al.: 9 FM patients, 9 HW controls
Chapter 4: Discussion

The complexity of my findings initiated an additional evaluation of available research, taking into account trauma or injury, the inflammatory role of fibroblasts, IL-6, and estrogen’s effects on the appearance of FM symptoms. As a result of this research, I suggest a novel pathway that explains my charted findings and the appearance of FM symptoms in women (Figure 4). This pathway is initiated by physical trauma or injury that activates fascial fibroblasts. Fibroblasts elicit excessive production of IL-6, which normally subsides after a few days and deactivates the inflammatory immune response. However, the production of estrogen interferes with the recall of IL-6, and triggers the female body to maintain a constant inflammatory state observed as FM.

Fibroblasts

One initiator of the increased pro-inflammatory cytokine levels in FM patients are fibroblasts, which are triggered by stress or injury to produce IL-6 (61). Within fascia, this initiated inflammatory response increases the excitability of nearby free, unmyelinated nerve endings, as well as the encapsulated Pacini and Ruffini corpuscles (62). Besson also observed the presence of “sleeping nociceptors”, which are generally inactive histological components that are only excitable under inflammatory conditions (62). These “sleeping nociceptors” and traditional nervous receptors transmit noxious
signals throughout the body, and lead the protective inflammatory response to be abnormally perceived as pain (62). Inflammation of the fascia, as a result of IL-6 production by fibroblasts, is also a source of long-term activation of the dorsal horn neurons of the spinal cord (14,40). This phenomenon is known as central sensitization, and refers to overreaction of the central nervous system to painful stimuli as a result of constant input (14,40). While attributed to chronic pain conditions such as endometriosis and peripheral arterial disease, central sensitization in FM patients may occur spontaneously or as a result of active MTPs (14). Additionally, astrocytes and microglia cells within the fascia can be sensitized by the chronic inflammatory state and produce the exaggerated pain response characteristic of FM (63).

Hypothalamic-Pituitary-Adrenal (HPA) Axis

The injury-induced increase in IL-6 and resultant fascial inflammation directly affects the regulation of the stress-induced HPA- axis (64). More specifically, it has been suggested that IL-6 production is increased in FM patients due to deficiencies in the glucocorticoid receptors (60). As part of the HPA pathway, glucocorticoid secretion of cortisol is induced by physical or biochemical stress, and protects the body by regulating the production of IL-6 (60). Geiss and colleagues found that FM patients exhibit both hyper- and hypo-cortisolemic patterns; elevated levels of cortisol are seen in MTPs, and the reduced, hypocortisolemic state can be seen within the first hour of waking (60,65). These altered states are a result of improper binding and signaling of the glucocorticoid receptors, and it was hypothesized that the hypocortisolemic state will increase the core symptoms of FM—pain and chronic fatigue—by enhancing the sensitivity of
glucocorticoid receptors and increasing IL-6 production (60,65). To test this claim, researchers administered a pressure pain threshold test on all 18 MTPs in FM patients and controls. The subjects’ pain response, cortisol and IL-6 production levels were measured after increased physical pressure on the taut band sites. Consistent with their hypothesis, Geiss et al. found that IL-6 levels were 4 times higher in FM patients, and higher pain and fatigue symptoms were reported in all affected individuals after the test (60).

The increased IL-6 concentrations characteristic of FM patients can also stimulate the hypothalamus and delay the responsiveness of ACTH, altering the function of this HPA stress pathway (66). In addition to the impacts on ACTH, Torpy and colleagues found increases in heart rate and inhibition of glucocorticoid receptor function in female patients after administering additional levels of IL-6 to FM-affected individuals (66). These factors indicate that FM correlates with disorders of the stress system, and the reduced function of the HPA axis leads to the characteristic symptoms of fatigue, depressed mood, pain, and disrupted sleep (40,66).

Estrogen Leads to Improper Cell-Mediated Immune Response

As women represent more than 90% of FM cases, the impact of female hormones are of great interest to those studying the emergence of FM symptoms (10). Additionally, the risk of developing FM increases with age, corresponding with decreases in estrogen levels (67). Researchers theorize that these effects are due to the abnormal responses in women following biopsychological stressors, which significantly impact the cell-mediated immune response and the HPA-axis.
To measure the direct inflammatory effects of such stressors, Prather and colleagues recorded IL-6 levels in men and women throughout an evaluative public speaking task (58). While men showed a measureable decrease in circulating IL-6 amounts, young women showed no change (58). This study was then extended to include post-menopausal women, and researchers discovered a significant increase in IL-6 production in estrogen-reduced women following the stressful speaking task (58). Darnall et al. turned their attention to the effects of pain catastrophizing—the cascade of negative emotions following pain—on IL-6 production between men and women (68). The negative emotions studied include ruminating and magnifying pain, and experiencing helplessness as a result of pain’s chronic appearance (68). After examining the compounding effects of pain catastrophizing, researchers concluded that women experience this cascade of negative emotions more often than men, and IL-6 levels increase in direct response to these psychological stressors (68). Additionally, while in this state of negativity, women report experiencing longer stages of physical pain (68). Emotional triggers play a more active role in women as a whole, and indicate how “real-life psychological stressors” can amplify the inflammatory response and increase the instances of chronic pain in women (68).

Other studies looked for the development of psychological stress reactions—the psychological adaptions to an activated HPA axis—between men and women (69). Rohleder and colleagues found glucocorticoid sensitivity increased in men and decreased in women one hour after a psychologically stressful event. Cytokine production also decreased in men and increased in women, indicating gender-specific alterations of the
The HPA pathway (46). The HPA-axis shows dysregulation in the presentation of FM, as women with FM are shown to have lower systemic concentrations of ACTH and higher concentration of cortisol than their age-matched control group (47). Estrogen’s alteration of the HPA pathway is therefore able to modify the immune responses and cytokine secretion (52).

While emotional factors may play a more detrimental role in women than in men, many studies have indicated that women physically exhibit lower thresholds to pain, and may experience greater levels of pain in general (70–72). For example, when electrocutaneous stimulation was applied to the forearm, women expressed a higher pain response (71). This may be due to the greater average thickness of subcutaneous fat and fascia, leading to more free and encapsulated nerve endings, in women (22,71). The direct involvement of estrogen in the HPA-axis and stress response, as well as the exhibited variation in psychological stress reactions increases the risk of developing FM in women.

Additionally, the constant release of IL-6, triggered by constant psychological stressors, can increase the risk for chronic inflammatory disorders (58). The chronic stress input also produces mitochondrial damage and vasoconstriction, reducing blood flow to body tissues and leading to areas of referred and constant pain, characteristic of FM (69).

Proposed Injury-Induced Fibromyalgia Pathway

The interplay of all aforementioned factors is described in my proposed injury-induced FM pathway (Figure 4). After a traumatic injury, stretching and tearing of the
fascia activates IL-6 release by fascial fibroblasts. Excessive IL-6 production enhances the systemic inflammatory response and activates the HPA-axis due to the resulting influx of pro-inflammatory cytokines (42,44). Under normal circumstances, the responses of both the inflammatory system and HPA-axis will subside once the biochemical balance is restored. However, the presence of estrogen alters these responses—resulting in the chronic inflammation and “sickness behavior” symptoms characteristic of FM (44).

While my analysis of IL-6 levels and proposed injury-induced pathway seek to explain the prevalence of FM in women, more research and testing must be completed to prove the veracity of these claims. The IL-6 measurements gathered were used as an indication of inflammation in each population; however, additional inflammatory biomarkers should be studied. In addition, the impact of IL-6 can be further examined as dysregulated IL-6 levels have been observed in diseases such as artherosclerosis, Crohn’s disease and rheumatoid arthritis (29). Research into these areas of autoimmunity and chronic inflammation may offer greater understanding and treatment developments for FM. Because of the increased prevalence of FM in women, the impact of estrogen on all areas of my proposed pathway should continue to be examined. Genetic information and resultant embryological development may also explain why women may be “predisposed” to FM, and may also explain why FM does not universally develop in women after injury.

After testing my hypothesis involving only IL-6 levels, I have determined that FM is a multifactorial condition. To further explain the increased prevalence of FM in women, each contribution to the appearance of FM symptoms outlined in my proposed
pathway should be explored. In addition, the interaction between these outlined factors should be examined to verify the interactions between each step in the pathway, and the pathway as a whole.
Figure 4: Proposed Injury-Induced Fibromyalgia Pathway
Chapter 5: Conclusion

The etiology of FM is one that continues to be studied and debated. In hopes of explaining the prevalence of FM in women, this thesis hypothesized that increased IL-6 levels would be seen in women, leading to the greater prevalence of FM in this gender. To examine this proposal, I compared concentrations of IL-6 between resting and stressed women and men, and FM and healthy age-matched women. Though comprehensive conclusions are yet to be reached, I propose that FM is an inflammatory disease and offer a novel interpretation of previous research further explaining my hypothesis. The intrinsic complexity of the immune system, with antigen non-specific and specific innate and adaptive immunities, leaves the human body prone to dysfunction exhibited as autoimmunity, allergy, and chronic inflammation (41). In FM patients, the continuous activation of the immune system is due to the abnormal interactions between pro-inflammatory cytokines released from fascial fibroblasts and estrogen hormones. Traumatic injury physically alters the composition of fascia, leading to activation of the fascial fibroblasts as part of the inflammatory response. The release of IL-6 from the fibroblasts influences the functioning of the HPA- axis, which is also adversely affected by estrogens. These interactions are verified by my specific findings, and offer a complex explanation for the unusual phenomenon of FM.
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