The Use of Specialized Pro-resolving Mediators as Anti-inflammatory Agents in the Treatment of Chronic Pelvic Pain Among Patients with Endometriosis: A Systematic Review

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Abstract

Background: Endometriosis is an inflammation-dependent condition characterized by the uncontrolled development of lesions resembling the endometrium. For the host tissue to maintain homeostasis throughout the active process of inflammation resolution, endogenous factors must be activated.
Objective: To determine if using specialized pro-resolving mediators (SPM) to treat endometriosis patients' persistent pelvic pain is safe and effective.
Methods: A systematic review was performed to determine studies that determined the effect of Specialized Pro-resolving Mediators (SPM) on endometriosis control. A descriptive narrative approach was applied to synthesize the findings of this systematic review.
Results: Three prospective case-control studies and three in vitro studies were included. All studies involved Lipoxin A4 as the specialized pro-resolving mediator assessed for benefit on endometriosis control. All studies consistently reported that SPM can be potentially effective in treating endometriosis. The search terms used were: "Specialized Pro-resolving Mediators," "SPM," "Chronic inflammatory agent," pelvic pain, and endometriosis.
Conclusion: Despite the lack of trials on the use of Specialized Pro-resolving Mediators to manage chronic pelvic pain, case-control and in vitro studies are consistent in detailing the potential benefits of SPM for endometriosis control. Given the concrete pathophysiologic basis for the mechanism of action for SPM, it is highly recommended that future trials be made to determine its efficacy and safety.

Key words: endometriosis, inflammation, Specialized Pro-resolving Mediators

Introduction

The presence of endometrium, or the tissue lining the interior chamber of the uterus outside the uterine cavity, such as the fallopian tubes, ovaries, and pelvic peritoneum, is a characteristic of endometriosis, an estrogen-dependent inflammatory condition.[1] Up to 10% to 15% of all women who are of reproductive age have endometriosis. Symptoms of endometriosis commonly include excessive menstrual bleeding, chronic lower back and pelvic pain, and pain related to periods (dysmenorrhea), sex (dyspareunia), defecation (dyschezia), and urination (dysuria). But 20–25% of people might not have any symptoms.[2]

It is still unclear how endometriosis develops, even though the inflammatory immune response is known to be a key player in this process. For the host tissue to maintain homeostasis during the active process of inflammation resolution, endogenous factors must be activated.[3] In humans, a self-limited inflammatory

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response that terminates in total resolution is the optimal response to an initial stimulus. Specialized pro-resolving mediators (SPMs), a superfamily of endogenous chemical mediators that promote the resolution of inflammatory responses, are known to control the resolution phase, which is now generally acknowledged to be a biosynthetically active process.[4] Resolvins, protectins, and maresins are examples of SPM produced from omega-3 polyunsaturated fatty acids during inflammation's acute or resolution phases. SPMs work by activating various G protein-coupled receptors expressed by neurons, glial cells, and immune cells. Notably, the resolution of pain is hampered by the depletion of SPM receptors.[5] The association of SPM and endometriosis is currently expanding. This systematic review aims to integrate different studies on the association of SPM with endometriosis.

Methods

The preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P 2020) declaration were used to create this systematic review.

Inclusion Criteria

A. Types of studies. Randomized trials, observational studies, and in-vitro studies conducted among human patients published until 2024 were eligible for inclusion. Only studies written in the English language were used.
B. Population. The study population included reproductive women aged 18 years and above with endometriosis. No restrictions were applied in terms of race/ethnicity or socioeconomic status. Excluded were studies that have other comorbid gynecologic disorders.
C. Intervention. The study included specialized pro-resolving mediators (SPM), which were used in any treatment regimen protocol.
D. Comparison. Placebo, any control, or no comparison group (single arm)

Exclusion Criteria

Case reports, case series, conferences, posters, abstracts, reviews, and commentaries were excluded. Reviews, animal studies, abstracts, posters only, protocol, letters, and editorials were likewise excluded.

Information Sources

A search was performed in PubMed/MEDLINE, EMBASE, Google Scholar, Biosis, and the Cochrane Library. Print journals were not hand-searched.

Search Strategy

The search terms used were: "Specialized Pro-resolving Mediators," "SPM," "Chronic inflammatory agent," pelvic pain, and endometriosis. Duplicate articles were removed, and additional relevant articles were identified by scanning the original search reference lists. No filters were used for years and countries. Search filters for English language and full-text only (free or paid) were applied.

Measurement of Outcomes

The outcome assessed was the suppressive activity on the inflammation of endometriotic epithelial cells.

Data Management

Full-text articles for potential inclusion were saved in Google Sheets. Extracted data were managed using Microsoft Excel and Microsoft Word.

Selection Process

Three authors (IJCT, MCP, and LRN) independently reviewed the titles and abstracts identified using the above search strategy. There was a comparison of papers by the same author to reduce data redundancy caused by duplicate reporting. The full-text articles of reports deemed eligible based on their titles or abstracts were obtained. At least one author selected potentially relevant papers whose full texts were retrieved and evaluated. Three authors examined articles that fit the inclusion criteria independently, and any discrepancies were resolved through discussion. Following the PRISMA 2020 criteria, a search and selection process flowchart was developed.

Data Collection Process

Study name (along with first author's name and year of publication), country where the study was conducted, source from which patients or study participants were selected, study design, treatment regimen, outcomes, effect measures, study strengths, and limitations were extracted independently by three authors using a standardized extraction form. Data were extracted independently by three authors (IJC, MCP and LRN). To ensure the correctness and consistency of the extracted data, the data extraction forms were cross-checked.

Risk of Bias Assessment in Individual Studies

Three authors (IJCT, MCP and LRN) independently assessed the methodology of the studies that were included. The methodological index for non-randomized studies (MINORS) was the tool used for quality assessment. The MINORS tool can be applied to evaluate both single- arm and comparative studies.

Data Analysis

Information from the studies included in this systematic review was consolidated by detailing individual study characteristics and conclusions and analyzing possible hypotheses to explain the mechanism of SPM in managing pain for patients with endometriosis. A meta-analysis was initially planned, but the systematic literature search did not yield sufficient articles to pool individual results quantitatively. A descriptive narrative approach was applied to this systematic review.

Results

Two hundred six articles were identified during the database search. Non-duplicate titles and abstracts were screened, and 7 articles were identified for potential inclusion. After full-text selection, 1 study was excluded because it was an animal study. A total of 6 studies were finally included in this systematic review.

Table 1 shows that the studies were of good methodologic quality. A limitation, however, is that

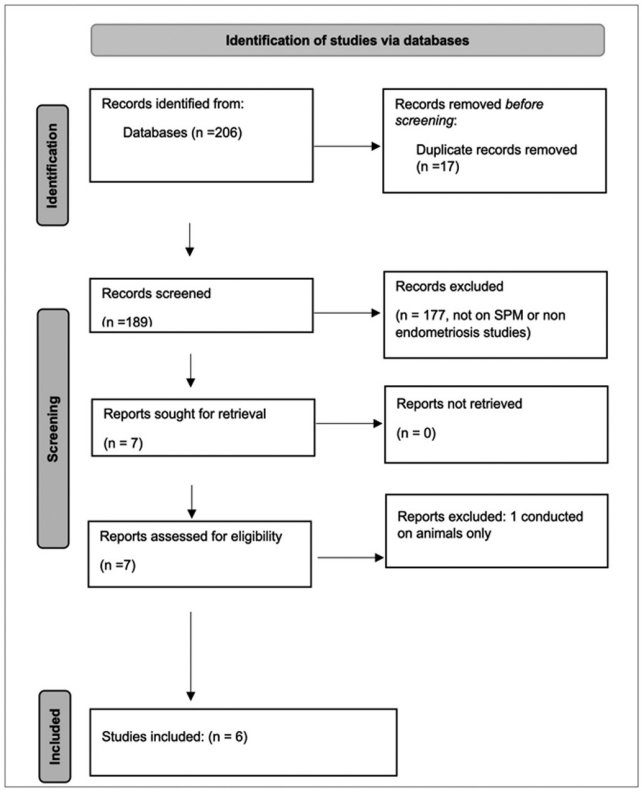

Figure 1. PRISMA flow diagram of the research study

three studies were in vitro or cellular studies only, without a control or comparative group.

Summary of Studies Included

The study of Gori and colleagues (2013)6 comprised 32 age- and BMI-matched control volunteers having hysterectomy or laparoscopy from a private clinic and two university hospitals, along with 59 endometriosis patients. During the proliferative phase of the menstrual cycle, peritoneal fluid and normal, eutopic, and ectopic endometrial biopsies were taken during surgery. In vitro mechanistic investigations were conducted using 12Z endometriotic epithelial cells. The findings demonstrated that MRP4 was expressed in both ectopic and eutopic endometrium, where it was found in the cytoplasm of glandular epithelial cells and overexpressed in peritoneal lesions. In endometriotic epithelial cells, LXA4 dose-dependently reduced MRP4 mRNA and protein levels, but did not change the expression of PGE2 metabolism-related enzymes. This happened via

Table 1. Risk of bias assessment using the MINORS tool.

	Clear study objectives	*Inclusion of consecutive samples*	*Prospective data collection*	*Appropriate endpoint*	*Appropriate follow-up*	*Drop- out <5%*	*Adequate statistical analysis*
Gori 2013[6] Prospective Case-control	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*
Chen 2014[7] Prospective Case-control	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*
Sobel 2016[8] In vitro	*Yes*	*No*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*
Wu 2017[9] In vitro	*Yes*	*No*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*
Dai 2019[10] Prospective Case-control	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*
Ghanavatineja 2021[11] *In vitro*	*Yes*	*No*	*Yes*	*Yes*	*Yes*	*Yes*	*Yes*

the estrogen receptor, according to studies using small interfering RNA and receptor antagonists. Consequently, extracellular PGE2 release was reduced by LXA4 therapy. Thus, it can be said that MRP4 is expressed in the human endometrium, increased in endometriosis of the peritoneum, and influenced by LXA4 in endometriotic epithelial cells.[26]

A study by Chen et al. (2014)[7] comprising 49 premenopausal women (of whom 30 patients had endometriosis and 19 were controls) from a university hospital was conducted using genetic analysis on human samples and primary human endometriotic stromal cells (ESCs). For in vitro research, ESCs and normal and ectopic endometrial samples from surgeries done during the proliferative phase of the menstrual cycle were used. According to the results, ectopic tissue had lower levels of ERa, PR, and LXA4 expression than the controls, whereas ectopic endometrial had higher levels of ERb expression. association analysis investigations showed that LXA4 expression had a negative association with ERb and a positive correlation with ERa in vivo. Furthermore, giving LXA4 may increase ERb expression in ESCs and, most likely via ERb, prevent the activation of p38 MAPK caused by 17b-estradiol. The results showed that, most likely via ERb in ESCs, LXA4 controls ERb expression and prevents 17bestradiol-induced activation of p38 MAPK.[7]

Sobel et al. 20168 demonstrated the effect of 17-β-Estradiol (E2) and Lipoxin A4 (LXA4) as an anti-inflammatory and pro-resolving lipid mediator against 12Z endometriotic epithelial cells. An in-vitro proteomic analysis was performed which showed that E2 affected more proteins in endometriotic epithelial cells than LXA4. The combination of E2 and LXA4 led to a decrease in the amount of controlled proteins, while LXA4 had an inhibitory impact on E2-induced signaling. Also, LXA44 modified gene expression and functional characteristics in human endometrial epithelial cells, affecting proliferation and displaying antiestrogenic properties similar to estriol. The regulated proteins play a role in several processes related to endometriosis pathophysiology and metabolism, including mRNA translation, growth, proliferation, proteolysis, and immunological responses.[8]

Primary ESC was cultivated from ovarian endometriosis tissue in a different investigation by Wu and colleagues (2017).[9] Three groups were created: the IL-1β stimulation group, the IL-1β plus LXA4 incubation group, and the control group. Proteins were evaluated by 2-D polyacrylamide gel electrophoresis (2D-PAGE), and matrix- assisted laser desorption/ionization–time of flight (MALDI-TOF) mass spectrometry (MALDI-TOF-MS) was used to further identify protein spots that were differently expressed. Transwell assays and wound

healing were used to measure the migration and invasion of ESC after therapy. Using MALDI-TOF-MS, 40 differentially expressed protein spots were successfully detected in total. The proteins that were found have connections to processes that may be implicated in the onset of endometriosis, including cell structure, metabolism, signal transduction, protein synthesis, and membrane structure. Western blot and quantitative real-time polymerase chain reaction were used for further analysis of vinculin and IL-4. Furthermore, LXA4 may inhibit ESC migration and invasion that is brought on by IL–1β. The authors concluded that LXA4 may slow down the development of endometriosis in part by altering the action of IL-1β, which is mediated in vitro by immune response-related proteins like IL-4 and inflammation-related proteins like vinculin.[9]

One study conducted by Dai et al. (2019)[10] included patients with ovarian endometriotic cysts (mean age 34 years, n=29) who had their paired eutopic endometriotic cells and ovarian endometriosis cysts removed during laparoscopic surgery. Findings showed that in contrast to the normal endometrium of controls, the ectopic endometrium of endometriosis patients showed an overexpression of COX-2. Through its receptor, formyl peptide receptor 2/lipoxin A4 receptor (FPR2/ALX), lipoxin A4 effectively inhibited IL-1β-induced COX-2 protein production in ectopic endometriotic stromal cells (ESCs). Antagonism between FPR2/ALX removed LXA4's inhibitory function. Mitogen-activated protein kinases (MAPKs), which can increase COX-2 expression, were activated by IL-1β. LXA4 pretreatment of ESCs prevented p38 MAPK phosphorylation brought on by IL-1β. It was concluded that the aberrant production of COX-2 triggers responses via the MAPKs and inflammatory pathways, which may help explain the pathophysiology of endometriosis. Moreover, by blocking the p38 MAPK signaling molecule, LXA4 reduced the production of COX-2 that was stimulated by IL- 1β.[10]

Another in vitro investigation was carried out by Ghanavatineja et al. (2021).[11] Fifteen seemingly healthy women provided their endometrial stromal cells (ESCs) and whole endometrial cells (WECs). The immunomodulatory effects of 1,25 (OH)2 D3 on lipopolysaccharide (LPS)- or lipoteichoic acid (LTA)-treated ESCs and WECs were examined. In this research, women without a history of abortion, infertility, endometriosis, or vaginal infection were recruited. Gynecologists used a Pipelle pipette to get endometrial samples during the proliferative phase of the menstrual cycle. After being gathered, WECs and ESCs were given either LPS or LTA treatment. Using the ELISA method, the amounts of TNF-α, IL-6, and IL-8 in culture supernatants were measured. RT-qPCR was used to measure the expressions of TLR2, TLR4, and MyD88. Using the Western blot method, TLR4 expression at the protein level was investigated. The findings showed that 1,25 Dihydroxycholecalciferol (1,25 (OH)2 D3) markedly decreased the generation of TNF-α in LPS-activated ESCs as well as the production of TNF-α and IL-6 by LTA-stimulated WECs. In contrast, endometrial cells challenged with LPS and LTA produced more IL-8 when 1,25 (OH)2 D3 was pretreated. Pretreatment with 1,25 (OH)2 D3 significantly inhibited the production of TLR4 protein by ESCs in response to LPS. ESCs treated with LPS showed a substantial increase in MyD88 gene expression. When 1,25 (OH)2 D3 was administered to these cells prior to LPS stimulation, the effect was reversed. The study found that 1,25 (OH)2 D3 is an immunomodulatory molecule that regulates potentially detrimental inflammatory responses linked to female reproductive tract infections, hence preserving endometrial immunological homeostasis.[11]

Discussion

The powerful endogenous substances known as pro-resolving mediators of inflammation enable the host tissue to preserve homeostasis and resist chronic inflammatory disorders. Currently, hormonal and anti-inflammatory treatments are used to treat endometriosis. However, the high cost, adverse effects, and return of the illness after therapy discontinuation place limitations on these medicines.

LXA4 is the most researched mediator, having a favorable effect on several aspects of the development and maintenance of endometriosis.[10] In human embryonic stem cells, LXA4 down-regulated the expression of proinflammatory-associated genes (TNF-a and IL-6), altered the expression of

estrogen receptor β (Erβ), and prevented E2-induced activation of p38 MAPK.[7]

Ectopic endometrium is seen to develop into endometriosis after completing the attachment–aggression–angiogenesis three-step process. Endometriosis development may be influenced by a strong inflammatory reaction. The most significant pro- inflammatory cytokine in endometriosis is IL-1b, which intensifies the inflammatory state and encourages the release of other cytokines and growth factors, eventually facilitating the implantation and development of an ectopic lesion. Endogenous lipoxin A4 is a pro- resolving and anti-inflammatory mediator with a strong affinity for the FPR2/LXA4 receptor (FPR2/ALX). Furthermore, the initiation of pregnancy and the inflammation linked to menstruation were both regulated by LXA4 signaling via FPR2/ALX. LXA4 is the most researched mediator, having a favorable effect on several aspects of the development and maintenance of endometriosis.[10]

In local lesions, Cyclooxygenase-2 gene (COX-2) expression is greater than in ectopic endometriosis.[10] In ectopic and ectopic endometrial tissues, the methylation status of the COX-2promoter DNA was considerably lower than in controls.[12] PGE2 is produced in the peritoneal fluid as a consequence of the increased expression of COX- 2, which indicates enhanced COX-2 enzyme synthesis. The pathophysiology of endometriosis may be influenced by elevated COX-2 expression in the ectopic lesion and enhanced sensitivity of IL-1b induced COX-2 expression in ESCs.

The development of endometriosis involves the involvement of inflammatory response and mitogen-activated protein kinase (MAPK) signaling pathways.[13] The regulation of distinct biological responses in all eukaryotic cells is facilitated by the activation of MAPKs in response to a variety of stimuli, such as growth factors, cytokines, antigens, toxins, and medicines. MMAPK signal pathway included COX-2 expression in ESCs stimulated by IL-1b. In vitro, LXA4 inhibited the phosphorylated p38 MAPK and COX-2 expression. In light of this, COX-2 expression in ESCs is linked to the MAPKs pathway, and the p38 MAPK pathway is linked to COX-2 expression caused by IL-1b. Furthermore, LXA4, an endogenous lipid, has the potential to be a novel and effective pharmacologic agent for the treatment of endometriosis since it inhibits the p38 MAPK pathway, which is responsible for IL-1b-induced COX-2 production.[10] Furthermore, assays for cell invasion and wound healing similarly showed that LXA4 reduces the capacity of ESCs to migrate and invade when stimulated by IL-1β. This implies that LXA4 may further prevent the implantation of endometriotic cells in the pelvic cavity or ovary by increasing the adherence of ESC and reducing motility by upregulating vinculin expression.[9]

Furthermore, the body produces the most powerful estrogen, E2, and ERα and ERβ play a major role in mediating estrogen signaling, which promotes the growth of ectopic tissue and controls inflammatory reactions. Endometriosis is associated with decreased levels of ERα and PR and overexpression of ERβ, which is in line with previous studies.[34,35] The elevated amount of E2 is most likely related to the low level of LXA4 and abnormal ER expression in endometriosis tissue. Inhibiting inflammatory reactions and reducing estrogen signaling simultaneously may be achieved by ERβ activation, which is advantageous for endometriosis.[7]

A limitation of the current systematic review is the lack of clinical trials that compared the actual clinical use of Specialized Pro-resolving Mediators with placebo or control. Most studies included were in vitro proteomic methods which are characterized by some heterogeneous methods, and further standardization and optimization for clinical use is necessary.

In summary, endometriosis may be caused by abnormal expression of various molecules, as suggested by the possibility that Specialized Pro-resolving Mediators such as LXA4 may inhibit the progression of the disease by lowering or raising the effect of some cytokines, such as IL-1β, mediated via some inflammation-related proteins and immune response-related protein.[9] Even after going over the body of research, there is much more to learn about the Specialized Pro-resolving Mediators pathways in endometriosis patients. It is crucial to remember that the immune system and aberrant inflammatory reactions are crucial in the onset, maintenance, and progression of this chronic illness, making the therapeutic potential of these chemicals clear.

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