Inflammatory Mechanisms in Vestibulodynia Revisited

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Although many researchers have investigated potential etiologies of vulvodynia, there is no consensus regarding its cause(s). Vulvodynia remains a diagnosis of exclusion, i.e., it is diagnosed when no precise cause, such as infection or lesions, can be identified.

Early Research

Twenty years ago, David Foster, M.D., and other researchers proposed that inflammation was one likely cause of vulvar vestibulitis, the most common subtype of vulvodynia (‘itis’ indicates inflammation). They found that inflammatory cells produced by the immune system, such as macrophages and mast cells, were present in the painful vulvar vestibule. They later determined, however, that an equal number of inflammatory cells were present in the vestibules of pain-free women (hereafter referred to as ‘controls’). The presence of these inflammatory cells in controls is unsurprising, because the vestibule surrounds the vaginal opening, a mucosal site that must defend against infection. Thus, inflammation was largely eliminated as a
cause of vulvar vestibulitis and the International Society for the Study of Vulvovaginal Disease subsequently changed the condition's name to 'vestibulodynia' ('dynia' meaning pain).

**Recent Research**

There is new evidence, however, showing that non-classical, i.e., specialized inflammation, may be a significant contributor to the development of vestibulodynia. Recent studies have found that typical inflammatory cells, e.g., macrophages, are present in roughly equivalent numbers in both women with vestibulodynia and controls, but that other inflammatory cells, such as B lymphocytes, are elevated only in women with vestibulodynia. Additionally, both the composition and arrangement of certain immune cells are different in women with vestibulodynia. Overall, these studies showed that there is more than meets the eye with regard to the presence and activation of immune cells in the painful vulvar vestibule.

Subsequent studies have supported these findings. One study demonstrated an increase in certain inflammatory mediators produced by fibroblasts (cells in connective tissue) in women's painful vulvar sites, but not in their non-painful vulvar sites. Specifically, the proinflammatory mediators, prostaglandin E$_2$ (PGE$_2$) and interleukin-6 (IL-6), were elevated only in painful vulvar sites. These mediators are also elevated in painful tissue in other chronic pain conditions and reducing their levels can help alleviate allodynia (pain with light touch). Furthermore, a connection between pain and PGE$_2$/IL-6 levels has been established in women with vestibulodynia—the higher the level of PGE$_2$ or IL-6 produced by their fibroblasts, the lower the patient's pain threshold, i.e., the amount of pressure tolerated before experiencing pain. Increased levels of other inflammatory mediators, such as tumor necrosis factor alpha (TNFα) and interleukin-1 beta (IL-1β), have also been found in fibroblasts isolated from the painful vulvar vestibule. Although there are only a few research groups currently investigating the role
of inflammation in vestibulodynia, there is growing evidence to suggest that it plays a role in the
development of vulvar pain, even though it may not be the only cause.

Hyperinnervation, or increased nerve density, in the vestibule may also contribute to
vestibulodynia, but its role is less clear, because increased nerve density typically causes
persistent itch, rather than pain. However, increased nerve density can also increase the
production of proinflammatory mediators IL-6 and PGE\textsubscript{2}. At the same time, nerve fibers express
receptors that recognize inflammatory mediators and often participate in the inflammatory
response. Although further study is warranted, it is plausible that increased nerve density in the
vestibule helps to amplify pain by potentiating inflammatory signaling.

The Origin of Inflammation
Although inflammation appears to be an important contributor to vestibular pain, we don’t know
why or how inflammatory signals are elevated in women with vestibulodynia. The majority of
these patients have experienced a period of pain-free intercourse before developing pain, and
numerous potential causes have been proposed, including genetic factors, trauma during
childbirth and vaginal infection.

Several studies point to a history of recurrent or persistent yeast infection as a precipitating
factor for vestibulodynia. More than 70 percent of these patients report having had a yeast
infection that recurred four or more times or did not resolve after several months of treatment
immediately prior to developing chronic vulvar pain. In support of this observation, researchers
demonstrated that women with vestibulodynia exhibit cutaneous hypersensitivity (an allergic
response) to \textit{Candida albicans}, a common vulvovaginal yeast pathogen. A study of mice with
recurrent \textit{Candida albicans} infections found that they eventually developed chronic vulvar
sensitivity that did not resolve, even after antifungal treatment. This finding is consistent with
what is seen in some women with vestibulodynia, i.e., they still experience vulvar pain, but no longer have an active yeast infection.

Studies have found that human vulvar fibroblasts isolated from painful vestibular sites are exquisitely sensitive to both live yeast and particles isolated from yeast. As few as 10 yeast cells (much less than what can be detected clinically) elicit a significant response, characterized by high levels of both IL-6 and PGE\textsubscript{2} production by these fibroblasts. However, fibroblasts from patients’ non-painful vulvar sites fail to respond and require up to a 1,000 times higher yeast dose to increase IL-6 and PGE\textsubscript{2} levels. These findings demonstrate that cells in the painful vulvar vestibule are hypersensitive to yeast and their response causes an increase in proinflammatory mediators associated with pain. Thus far, the connection is clearest for yeast, but it is likely that other vulvovaginal pathogens play a role. Most women self-diagnose yeast infections, which is unreliable in the absence of a diagnostic test. Since symptoms of a yeast infection are similar to those of a viral or bacterial infection (or a microbiological imbalance), some women who report a history of yeast infections may have actually had one of these other conditions instead.

Although the above findings indicate that inflammation contributes to vulvar pain, specifically via a response to yeast and possibly other vulvovaginal pathogens, they do not explain why the vestibule is inherently sensitive to inflammatory stimuli. To discover the underlying mechanism(s) for this sensitivity, our University of Rochester research group identified pattern recognition receptors (PRRs), special receptors in humans that recognize microbes by spotting conserved patterns on microbes that distinguish them from the body’s own cells. A dozen PRRs implicated in recognizing harmful vulvovaginal microbes (e.g., yeast) are slightly, but significantly, more abundant in fibroblasts taken from a patient’s painful vulvar site than those obtained from non-painful sites. When fibroblasts are treated with inhibitors that reduce the
amount of these PRRs or reduce the ability of these PRRs to function, both IL-6 and PGE\textsubscript{2} levels drop significantly. This finding tells us that these PRRs play a role in generating the inflammatory signals linked to pain in women with vestibulodynia.

PRRs are part of a larger signaling system that turns on the production of the pain-associated inflammatory mediators, IL-6 and PGE\textsubscript{2}. Activation of any one of these receptors can turn on signaling, such that turning off only one of them while the rest are working is not sufficient to stop inflammatory signaling. As an analogy, If a car is rolling downhill, removing the engine will not be enough to stop it. Each PRR turns on IL-6 and PGE\textsubscript{2} production by first turning on nuclear factor kappa B (NF\kappa B), a master inflammatory regulator. NF\kappa B turns on IL-6 and PGE\textsubscript{2} production in response to more than a dozen inflammatory activators, including yeast. Thus, impairing the function of NF\kappa B can completely turn off signaling, similar to putting a brick wall in front of the rolling car. These findings support the idea that heightened proinflammatory signaling contributes to vulvar allodynia and establishes a mechanism for this heightened signaling. Furthermore, this research identifies potential targets for the development of new vestibulodynia therapies, e.g., inhibiting NF\kappa B.

Interestingly, this inflammatory response seems to be an exaggeration of a normal response, because fibroblasts from vestibules of controls also express elevated amounts of PRRs and produce more IL-6 and PGE\textsubscript{2} than fibroblasts from external vulvar sites. A simple analogy is to think of these receptors as a net to catch fish, or in this case, a defense system to recognize invading microbes. The bigger the net, the greater likelihood you will catch a fish, even if there are few fish. For a woman without vulvar pain, this is a helpful response that allows the vestibule, an area that immediately surrounds and protects the vagina, to recognize harmful pathogens and reduce the risk of infection. However, in women with vestibulodynia, the net is too big and catches fish it is not meant to catch, e.g., non-harmful microbes that are part of a
woman’s normal vulvovaginal flora. The end result is a persistent and detrimental inflammatory response that leads to unremitting pain.

Conclusion
With the identification and improved understanding of these inflammatory mechanisms, we can unlock new avenues for the development of more effective vulvodynia therapies. Specifically, a treatment that removes unnecessarily elevated inflammatory signaling, while preserving the ability of the vestibule to detect infectious threats, would be highly desirable. Our University of Rochester group is actively working on this goal and has developed a mouse model of vestibulodynia to test promising new therapies that may alleviate both pain and inflammation without compromising microbial defenses.