Commentary

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Beyond lidocaine: selective voltage-gated sodium channel blockade for vaginal pain

Ruby A. Holland[a], Sarah E. Ross[a,b]

Female reproductive tract pain encompasses a variety of common and debilitating conditions such as vulvodynia, vaginal hyperalgesia secondary to endometriosis, pudendal neuralgia, and many others. These conditions lack effective pharmacotherapies due to a poor understanding of the mechanisms underlying sensory transmission from female reproductive organs. Studies investigating bladder and colon nociception have shown that different voltage-gated sodium (Na_V) channels play a critical and complex role in pain transmission, suggesting selective sodium channel blockade may be a more potent therapeutic strategy for pelvic pain.[8,9] However, no study has specifically investigated the role of Na_V channels in pain signaling from the reproductive tract.

In the present issue of *PAIN,* Castro et al.[3] address this gap in knowledge. Equipped with a novel ex vivo vaginal preparation, the authors investigated the mechanosensory properties and receptive fields of pelvic nerve afferents innervating the mouse vagina (**Figs. 1A and B**). Using this approach, these afferents were found to all be polymodal, with punctate receptive fields distributed along the entire length of the vagina. Interestingly, these mechanosensory responses were globally augmented through the pan-Na_V channel activator veratridine but only partly attenuated by the partially selective Na_V channel blocker tetrodotoxin (TTX), suggesting that TTX-resistant Na_V channels contribute significantly to afferent transmission from the vagina. The authors complement these results by performing single-cell polymerase chain reaction (PCR) on dorsal root ganglion (DRG) neurons retrogradely traced from the vagina and found abundant expression of Na_V channels, with distinctive expression patterns across each subtype (**Fig. 1C**). Finally, the authors measured vaginal pain sensitivity by performing vaginal balloon distention and measuring visceromotor responses in live mice. They found that TTX attenuated and veratridine enhanced pain responses in vivo and similarly altered neuronal activity in the spinal cord in vitro (**Fig. 1D**).

An important premise of the authors' experimental design is in the differential effects of TTX on Na_V channels. There are no selective pharmacological modulators of Na_V channels; however, Na_V channels can be distinguished by their sensitivity to TTX, with both TTX-resistant (TTX-R; $Na_V1.5$, $Na_V1.8$, and $Na_V1.9$) and TTX-sensitive (TTX-S; $Na_V1.1$-1.4, $Na_V1.6$, and $Na_V1.7$) subtypes.[4,5] The authors showed that TTX partially attenuated vaginal afferent excitability and concluded that TTX-R Na_V channels contribute appreciably to afferent transmission from the mouse vagina. Although this pharmacological manipulation does not within itself identify specific Na_V channel subtypes integral to vaginal pain transmission, the combination of these experiments with PCR allows for the identification of a few candidates, namely the abundantly expressed TTX-R $Na_V1.8$ and $Na_V1.9$ and TTX-S $Na_V1.1$, $Na_V1.2$, $Na_V1.6$, and $Na_V1.7$ (**Fig. 1C**). By contrast, bladder afferent excitability is almost completely dependent on TTX-S Na_V channels[8] and colon nociception is largely independent of $Na_V1.7$.[9] These findings highlight the vagina's unique neurophysiology in comparison with other pelvic organs and point to potential targets which may be specific to the treatment of vaginal pain.

The major technical advance presented in this article is the novel ex vivo single-unit extracellular vaginal afferent recording preparation (**Fig. 1B**). With their flat sheet approach, they identified mechanosensory properties of the mouse vagina, which agree with those identified by the in vivo rat studies by Berkley's group.[1,2] Furthermore, although this study solely involved pelvic nerve recordings, this preparation can also be used to record from the pudendal nerve, allowing for the study of Na_V channels in vulvar and perineal pain.

Considering the novel genetic, physiological, and behavioral findings in this study, it is likely that vaginal afferents make up a genetically distinct population or populations. Single-cell transcriptomics have revealed the genetic diversity of sensory neurons arising from a variety of sites.[7] Unfortunately, most of these studies either do not treat spinal cord level as a variable or exclude the sacral DRG entirely, where a proportion of afferents from the vulva and vagina originate. New transcriptomic evidence shows that a collection of genes within nociceptors are differentially expressed in sacral DRG compared with lumbar DRG[14]; however, no study has applied high-throughput gene expression approaches to the study of sensory neurons arising from the female reproductive tract. By performing single-cell PCR on DRG neurons retrogradely traced from the mouse vagina, the authors took a critical first step in the genetic phenotyping of vaginal sensory neurons. They characterize a highly overlapping expression profile of all Na_V channel subtypes within vagina-innervating neurons and observed significant heterogeneity between the cells they sampled. Whether or not Na_V channels themselves prove to be viable targets in vaginal pain, the Na_V channel expression patterns identified in this formative study could potentially be correlated with expression of other cell type–specific genes. Single-cell RNA sequencing analyses from neurons retrogradely traced from the vagina, as has been similarly

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[a] *Department of Neurobiology, Pittsburgh Center for Pain Research, Pittsburgh, PA, United States,* [b] *Department of Anesthesiology, University of Pittsburgh School of Medicine, Pittsburgh, PA, United States*

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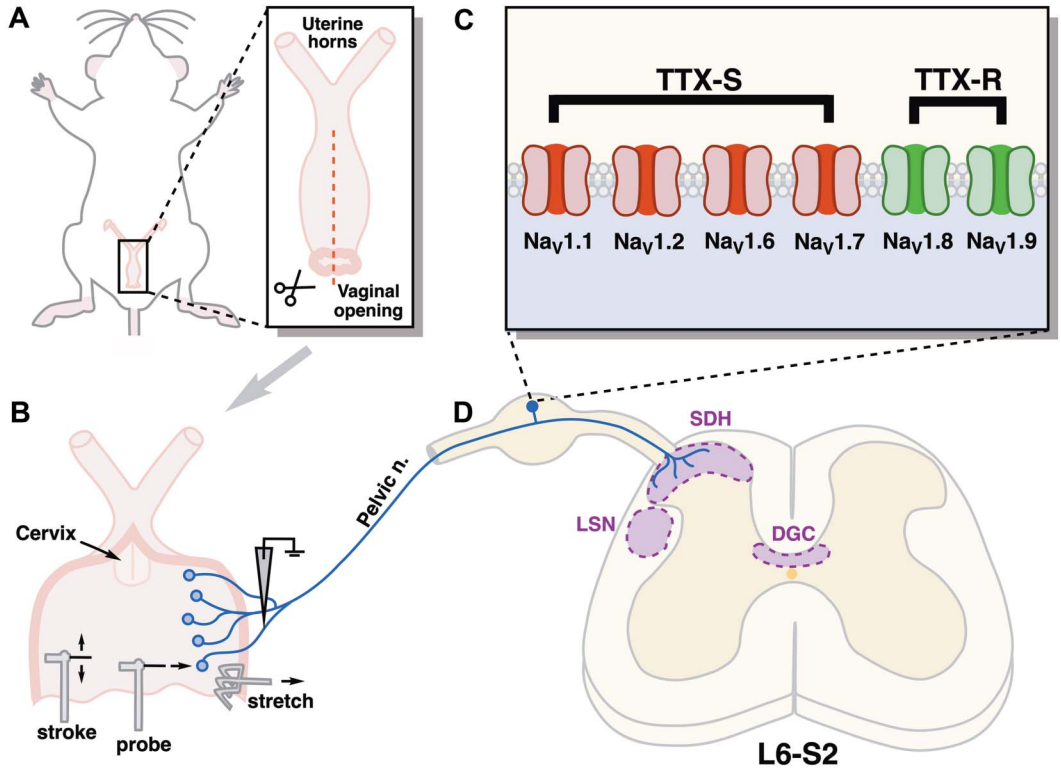

Figure 1. Voltage-gated sodium (Na_V) channels in pain transmission from the mouse vagina. (A) Schematic of the mouse female reproductive tract, with a red dashed line indicating the longitudinal cut through the vagina for the authors' flat sheet preparation. (B) The authors used a novel single-unit extracellular afferent recording preparation from pelvic nerves innervating the vagina. All vagina-innervating pelvic afferents are polymodal, responding to a combination of static probing, stroke, and circular stretch. Pelvic nerve receptive fields are small, punctate, and distributed along the entire length of the mouse vagina. (C) DRG neurons retrogradely traced from the mouse vagina abundantly express both TTX-S and TTX-R Na_V channels. (D) Pharmacological manipulation of Na_V channels in vagina-innervating sensory neurons alters signaling in the lumbosacral spinal cord at the superficial dorsal horn (SDH), lateral spinal nucleus (LSN), and dorsal gray commissure (DGC). DRG, dorsal root ganglion.

performed on colonic afferents,[10] could therefore serve to determine the cell type specificity of Na_V channels in vaginal afferents and identify additional molecular targets for the treatment of vaginal pain.

This study raises the question of whether selective Na_V channel blockade represents a feasible therapeutic strategy for reproductive tract pain. Na_V channel blockers are one of the oldest pain drugs modern medicine has had at its disposal. The ester Na_V channel blockers most commonly used for regional pain management, such as lidocaine, are nonselective because they act within the shared ion pore, rather than at unique allosteric sites.[12] In fact, lidocaine has been used to successfully treat gynecological pain in patients with dysmenorrhea due to endometriosis.[16] By contrast, its efficacy in vulvodynia has recently been discredited.[6] To better explore the complexity underlying Na_V channel-dependent mechanisms of pain transmission, genetic approaches will be essential, specifically those borne from the Cre/LoxP system. Cre or floxed lines have been developed for a few Na_V channel subtypes, including $Na_V1.8^{cre}$,[15] $Na_V1.7^{fl/fl}$,[13] and $Na_V1.6^{fl/fl}$.[11] Overall, the findings described in Castro et al. drive home the need to take this next step in the study of Na_V channel–dependent mechanisms of vaginal pain transmission and underscore the potential value of selective sodium channel blockade as a novel therapeutic strategy for the treatment of vaginal pain.

Conflict of interest statement

The authors have no conflicts of interest to declare.

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