

## ORIGINAL STUDY

# Treating where it hurts—a randomized comparative trial of vestibule estradiol for postmenopausal dyspareunia

Martha F. Goetsch, MD, MPH,<sup>1</sup> Bharti Garg, MBBS, MPH,<sup>1</sup> Jen Lillemon, MD,<sup>2</sup> and Amanda L. Clark, MD, MCR, NCMP<sup>3</sup>

### Abstract

**Objective:** To compare efficacies of two strengths of estradiol cream applied to the vulvar vestibule and use of silicone lubricant to reduce intercourse pain scores in postmenopausal women with moderate/severe dyspareunia.

**Methods:** This pilot randomized comparative trial assigned 50 women to nightly applications of estradiol cream, 50 or 100 µg, for 12 weeks. We asked women to have lubricated penetration twice weekly, with intercourse or performing a tampon test. Pain, recorded in dairies, was rated using the 0-10 Numerical Rating Scale. We assessed biopsychosocial outcomes, urinary symptoms, and measured serum estradiol levels and endometrial stripe thicknesses. We performed physical examinations to determine tenderness levels of the vestibule, vagina, pelvic floor muscles, bladder, uterus, and adnexa. Comparisons were made using two-sample *t* test, Wilcoxon rank-sum test, or  $\chi^2$ /Fisher's exact test.

**Results:** Forty-seven women (94%), with a mean age of 59.7 years, completed the trial. The baseline median intercourse pain score was 8/10 (interquartile range, 6, 8). After 12 weeks, we measured no statistically significant difference between groups in the primary outcome, intercourse pain score, or any secondary outcome measure. For both groups together, the median intercourse pain score diminished by 50% after 4 weeks and 75% after 12 weeks ( $P < 0.001$ ). The most tender anatomic area, the vulvar vestibule, improved by 82% to 100% ( $P < 0.001$ ) with therapy. We did not measure a statistically significant difference in serum estradiol levels or endometrial stripe thickness between groups.

**Conclusion:** Estradiol cream applied to the vulvar vestibule, paired with precoital silicone lubricant, is a promising alternative to vaginal therapy for dyspareunia.

**Key Words:** Dyspareunia – Estradiol – Genitourinary syndrome of menopause – Lower urinary tract symptoms – Vulvar vestibule – Vulvovaginal atrophy.

The prevalence of dyspareunia associated with genitourinary syndrome of menopause (GSM) is as high as 78%.<sup>1</sup> Genitourinary syndrome of menopause is known to progress with time after menopause<sup>2-4</sup> and to reduce quality of life (QoL) for both women and their partners.<sup>5-9</sup> Intravaginal application of estradiol therapy (ET) is the standard for treating GSM. In addition to the often-expressed concerns about estrogen exposure, many women find it to be inadequate for relief.<sup>10-12</sup>

In a study of dyspareunia in hypoestrogenic breast cancer survivors, the vulvar vestibule, not the vagina, was found to be the primary site of pain associated with dyspareunia.<sup>13</sup> Further, dyspareunia in these breast cancer survivors was largely prevented by topical lidocaine applied to the vestibule, along with use of silicone lubricant during intercourse, without treating underlying atrophy.<sup>14</sup> These findings suggest that the vestibule might be the optimal target for dyspareunia therapies. Further

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From the<sup>1</sup>Department of Obstetrics and Gynecology, Oregon Health and Science University, Portland, OR; <sup>2</sup>St Luke's Clinic—Urogynecology, Boise, ID; and <sup>3</sup>Division of Urogynecology, Department of Obstetrics and Gynecology, Oregon Health and Science University, and Kaiser Permanente Center for Health Research, Portland, OR.

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Address correspondence to: Martha F. Goetsch, MD, MPH, Department of Obstetrics and Gynecology, OHSU UHN50, 3181 SW Sam Jackson Park Road, Portland, OR, 97239. E-mail: [mfgoetsch@yahoo.com](mailto:mfgoetsch@yahoo.com)

support comes from an observational study using vestibular estradiol cream to successfully treat severe vulvar pain in postmenopausal women who had failed prior therapies.<sup>15</sup> Pain slowly resolved after weeks of nightly therapy, suggesting that more intensive and prolonged therapy may be required for severe cases.

The primary objective of this pilot randomized trial was to compare the efficacies of two strengths of estradiol cream applied to the vestibule, along with use of a silicone lubricant, to reduce intercourse pain scores. The primary and most severe site of pain elicited during the baseline physical examination was the vulvar vestibule in this cohort.<sup>16</sup> In addition, we assessed secondary outcomes of QoL, physical examination findings, and parameters of safety to study this route of ET.

## METHODS

### Study design

We conducted a randomized, double-blind, comparative, pilot trial of two doses of vestibular ET. After completion of the qualifying examination and consent process, participants were sent to the Oregon Health & Science University (OHSU) research pharmacy, which assigned participants 1:1 in random permuted blocks using randomization software ([www.randomization.com](http://www.randomization.com)), dispensing their allocated medication pump directly to them. Allocation was concealed from participants and the enrolling/evaluating research team until all participants completed the study, after which time investigators and analysts were unblinded.

### Setting

“Treating Where It Hurts” was conducted at the Women's Health Research Unit of the Department of Obstetrics and Gynecology at OHSU in Portland, OR.

### Participants

We recruited a convenience sample of women by asking: “Are you postmenopausal and experiencing pain with intercourse?” using flyers and social media advertisements. Initial screening for eligibility occurred by telephone; final eligibility was determined at a clinic evaluation. Inclusion criteria were as follows: aged 40 to 75, postmenopausal status, having a stable heterosexual partnership of more than 2 years, a report of consistent dyspareunia for at least 6 months severe enough to reduce frequency of intercourse (moderate dyspareunia) or avoid intercourse (severe dyspareunia). Criteria for menopause status included amenorrhea more than 1 year for those older than 50 years, absence of ovaries; age older than 51 years, and peak climacteric symptoms more than 2 years previously for women with prior ablation or hysterectomy. Systemic ET within 6 months and local ET within 4 weeks were not permitted. Exclusion criteria were as follows: moderate/severe dyspareunia before menopause, chronic vulvar or pelvic pain, a history of estrogen-sensitive cancer, or a history of uncorrected erectile dysfunction in the partner.

### Intervention

Estradiol creams containing either 50 or 100 µg were compounded using a propylene glycol-free, fragrance-free base

cream by the Lloyd Center Pharmacy in Oregon, in collaboration with the OHSU Research Pharmacy. This compounding pharmacy adheres to the Chapter 795 (nonsterile medications) standards of the United States Pharmacopeia Convention. Participants received their allocated study drug in a 50-g pump dispenser that delivered 0.5 g with each pump, the intervention creams being identical in appearance and consistency. They also received a 100-g pump dispenser that similarly delivered 0.5 g of Pjur silicone lubricant (Pjur Group, Wasserbillig, Luxembourg). During the initial gynecologic examination, each participant was shown her vulvar vestibule with aid of a mirror and instructed where and how to digitally apply her study cream nightly. Women were instructed to apply lubricant to the vestibule before penetration.

### Symptom outcomes

The primary outcome was the change in intercourse pain score as rated by the Numerical Rating Scale (NRS, 0-10) after 4 weeks of therapy. Secondary outcomes were measured after 4 and 12 weeks of therapy and included tampon test scores, assessment of condition-specific QoL and urinary symptoms and physical examination measures. Safety assessments included serum estradiol levels and measurements of endometrial stripe thickness.

Demographic and clinical characteristics of participants were obtained by questionnaire and included medical and gynecologic histories and prior estrogen use. Condition-specific QoL was assessed using a multidimensional instrument, the Vulvar Pain Assessment Questionnaire (VPAQ), which has been validated for use in the diagnosis and treatment of chronic vulvar pain.<sup>17</sup> Its development was guided by the IMMPACT (Initiative on Methods, Measurement, and Pain Assessment in Clinical Trials) framework.<sup>18,19</sup> With permission from the instrument's creators, we adapted the VPAQ slightly for GSM, for example, changing “vulvar pain” to “penetration pain.” (See Supplemental Digital Content 1, <http://links.lww.com/MENO/B82>, which details the questionnaire we used.) The VPAQ screen begins with an illustration of the vulva that allows women to identify the locations of pain and is followed by 40 Likert items that assess the domains of pain severity, cognitive/emotional difficulties, interference with sexual function, life functions, and self-stimulation/penetration. The VPAQ desc subscale assesses descriptors of pain in three domains: burning pain, incisive pain, and sensitivity. The descriptor “dry” is not part of the VPAQ, and we added “dry” as a descriptor using the VPAQ format because the symptom of dryness is so frequent in GSM. The scoring for VPAQ subscales (range, 0-4) was not modified; there is not a total VPAQ score.

To assess the urinary parameters of GSM, we collected data on the lower urinary tract using a 13-item questionnaire created from several validated instruments. We used the Urinary Distress Inventory-6 (UDI-6),<sup>20</sup> a question from the UDI long form,<sup>21</sup> the Sandvik Severity Index,<sup>22</sup> and selected questions from the O'Leary/Sant Interstitial Cystitis Symptom Index<sup>23</sup> and Urinary tract Infection Symptom Assessment questionnaire.<sup>24</sup> To assess the potential adverse effects of ET, participants were regularly queried about breast tenderness, vaginal

bleeding, and application irritation. All questionnaires were completed at baseline and, except for demographics, repeated at 4 and 12 weeks.

### Physical examination outcomes

First, a standardized, validated tampon test was conducted, assessing insertion/removal pain using an unlubricated medium Tampax (Cincinnati, OH) and scoring the sensation using the NRS with anchors of 0 (no pain) and 10 (“the worst possible pain”).<sup>25</sup> Participants underwent gynecologic evaluations by a vulvar specialist/gynecologist (first author) or by one of two trained research women's health nurse practitioners. “Tenderness” was pain elicited by touch or pressure during an examination, with 3 defining the upper limit of “mild.”<sup>26</sup> Vestibule tenderness was assessed using the cotton-swab test at eight locations (Fig. 1), beginning in the locations least likely to be tender. A cotton-tipped swab was lightly rolled over the surfaces of the vestibule in a systematic order using no probing motions. This rolling technique has been shown to generate a pressure of 15.8 g/cm<sup>2</sup>, which is sufficient to discriminate 0-10 pain in the condition of vestibulodynia.<sup>27</sup> At each location, participants scored the touch using the 0-10 point NRS. Using a modestly lubricated Pederson speculum, the midvault vaginal mucosa, left and right, was evaluated for sensitivity with light strokes of a cotton swab and scored by participants using the NRS. Vaginal testing in this manner is an extension of the cotton-swab test for the vestibule but has not been validated. Vaginal pH was measured with a colorimetric indicator strip. After speculum removal, the pelvic floor muscles were examined digitally for ten-

derness (absent, mild, moderate, severe), the optimal technique that has shown good interrater and intrarater reliability.<sup>28</sup> The bladder, the uterus, and adnexa were palpated for tenderness (present, absent) as during bimanual examination.

### Ultrasound and laboratory assessments

Measurements of endometrial stripe thickness were obtained by transvaginal ultrasound at baseline and after 12 weeks in women with a uterus. Blood for estradiol levels was drawn at baseline, at 12 hours after the first dose of cream, and at 4 and at 12 weeks. Estradiol levels were measured by the Endocrine Technologies Core at the Oregon National Primate Research Center using a Roche cobas e411 automatic clinical immunoassay platform (Roche Diagnostics, Indianapolis, IN) with a range of 5 to 3000 pg/mL.

### Implementation

Participants were instructed to attempt penetration twice per week, either with their sexual partner or using a tampon after applying lubricant to their vestibule, and to record a pain score by NRS. The tampon test served as a proxy for penile penetration until they felt comfortable enough to attempt intercourse. Women were provided tampons, daily diary forms, the study drug, and lubricant and asked to record applications of the study drug, lubricant, and insertional activity. At 4 and 12 weeks, participants returned to clinic for repeat administration of questionnaires, gynecologic examinations, and laboratory and ultrasound assessments.

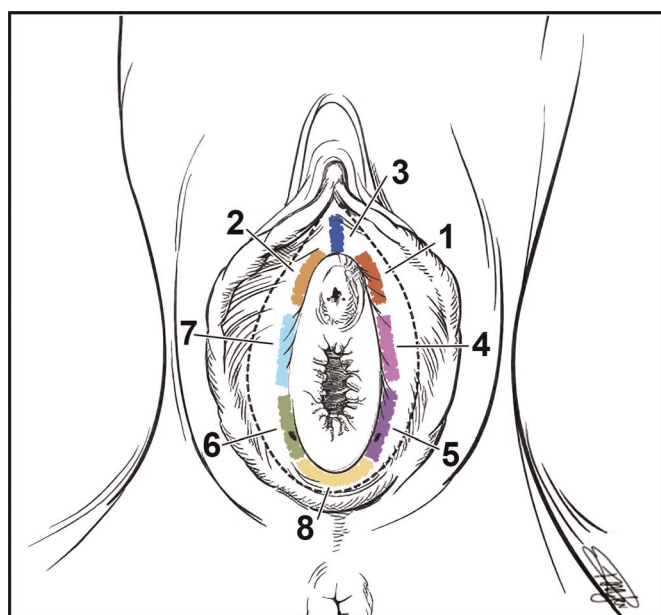
### Sample size analysis

We assumed a baseline NRS pain score of  $7 \pm 2.2$  from a previously reported study of dyspareunia,<sup>29</sup> and we assumed a 30% treatment effect after 4 weeks with the lower-dose intervention drug. A sample size of 50, divided into two arms, allowing for a 10% dropout rate in each arm, would achieve greater than 85% power to detect at least 2-point mean difference in dyspareunia pain scores between the two intervention drug strengths at 4 weeks, with an  $\alpha$  level of 0.05 for two-sample *t* test.

### Data management and statistical analyses

Study data were double entered into a REDCap electronic database.<sup>30,31</sup> The primary outcome was analyzed both as “intention to treat” (ITT) and “per protocol/as treated” (PP/AT). Missing values for the ITT analysis were estimated using baseline or last observation carried forward. Continuous variables were analyzed using independent two-sample *t* tests and categorical variables using  $\chi^2$ /Fisher's exact test. Nonparametric data were analyzed using Wilcoxon rank-sum test. We used repeated-measures analysis of variance (Friedman) to compare baseline, 4- and 12-week outcomes using the single closest score to 4 weeks ( $28 \pm 7$  days) or 12 weeks ( $84 \pm 7$  days). Statistical calculations were made using Stata (version 17; Stata Corp, College Station, TX).  $P < 0.05$  was considered statistically significant.

The OHSU Institutional Review Board approved the protocol (#16770).



**FIG. 1.** In this vulvar illustration, the vestibule's outer border is denoted with a dotted line (Hart's line), which demarcates the vestibule from the inner labia minora. The medial border is the recess immediately next to the hymen. The colored swaths indicate locations of cotton-swab touch testing corresponding to Table 3 where pain levels are reported. Black numbers indicate the order of examination. Artist: Robin M. Jensen. ©2014 Robin M. Jensen. Used with permission.

## RESULTS

Between June 2017 and August 2019, we screened 60 women and enrolled 50 eligible women who were included in the ITT analysis (Fig. 2). The PP/AT analysis included 47 women (96%) who completed 4 weeks of protocol and 46 (92%) who completed 12 weeks. One participant was noted to have lichen planus at the 4-week visit (demonstrating Wickham's striae that were not initially present); she began vulvovaginal steroid therapy and was dropped from PP/AT analyses. At 4 weeks, 45 (91%) had missed fewer than five doses of estradiol, and there was not a significant difference between groups in treatment adherence at 4 or 12 weeks. Women recorded 337 intercourse and 699 tampon pain scores, 1,036 overall, achieving overall reporting compliance of 92%. Of 556 diary-reported weeks, 0.3% lacked a single entry within a week.

The characteristics of the women at baseline are shown in Table 1, with no differences noted between the randomized groups. The mean age of participants was 60 years, with a mean duration of sexual partnership of 29 years, and mean dyspareunia duration of 6 years. Thirty-eight (76% of 50) reported prior use of systemic and/or vaginal hormone therapy, with 26 (52%) treating for dyspareunia. Eight (16%) required a washout period before enrolling. Median severity of recalled pain with intercourse by NRS was 8 (interquartile range, 6, 8). Median (interquartile range) time since last intercourse was 60 (8, 365) days, with 20 (41%) reporting intercourse in the previous month. Sexual lubricant use by 44 (88%) and vaginal moisturizer use by 5 (10%) had not prevented pain. The most consistent and severe locations of tenderness were in the vulvar vestibule. Tenderness of the pelvic floor muscles was noted in 18 women (36%). Expanded details of the baseline characteristics were reported previously.<sup>16</sup>

In both ITT and PP/AT analyses, no significant difference was measured when comparing the 50- and 100- $\mu$ g doses of estradiol (Table 2). In the PP/AT analysis at 4 and 12 weeks, no significant difference was measured between groups for intercourse and tampon test scores. During the entire study period, 42 of 47 women (89%) attempted intercourse, and no difference was detected between groups in the number of women who attempted intercourse at 4 and 12 weeks. We measured no difference in VPAQ scores, prevalence of urinary symptoms, or vestibular tenderness between groups at 12 weeks (data not shown).

With no measured differences between the groups, we combined the data to report exploratory data of outcome changes during the 12-week trial (Table 3). Intercourse pain scores decreased by 50% after 4 weeks and by 75% after 12 weeks. During the 12-week study, 12 (28% of 42 who resumed intercourse) achieved a score of zero at least once. Mild dyspareunia as a best score (NRS = 1-3)<sup>26</sup> was reached by 48%, and 24% had persistent moderate/severe dyspareunia as of 12 weeks. Of the five women who did not resume intercourse, reasons given included persistent dread of pain, introital stenosis, or other health or partner issues. Tampon test scores decreased progressively over 12 weeks. Vestibular tenderness diminished in all eight locations, achieving median values of 0 or 1. Resolution to an NRS of 0 to 3 at Bartholin's (greater vestibular gland) duct areas did not occur by 12 weeks for seven participants.

The VPAQ subscale of pain descriptors showed progressive decreases in the three domains of burning pain, incisive pain, sensitivity, and in our addition, dryness. Parameters of psychosocial and sexual function improved by 4 or 12 weeks. Women often reported urinary symptoms, but neither the proportion reporting symptoms nor the associated bother (data not shown) was statistically significantly improved by 12 weeks.

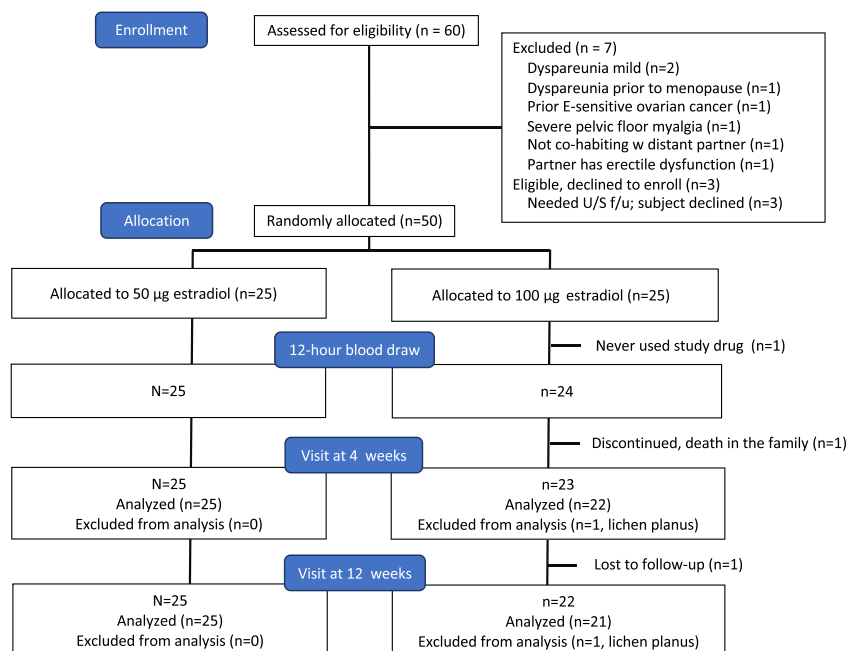


FIG. 2. CONSORT (Consolidated Standards of Reporting Trials) diagram.



**TABLE 1.** Baseline characteristics of participants

	Total n = 50	50 µg <sup>a</sup> n = 25	100 µg <sup>a</sup> n = 25
Age, mean ± SD, y	59.7 ± 6.8	59.2 ± 6.2	60.2 ± 7.5
BMI, mean ± SD, kg/m <sup>2</sup>	25.1 ± 4.6	24.6 ± 4.0	25.7 ± 5.2
Race, n (%)			
White	48 (96%)	24 (96%)	24 (96%)
Asian	1 (2%)	0 (0%)	1 (4%)
More than one race	1 (2%)	1 (4%)	0 (0%)
Education, n (%)			
9th-12th grade	1 (2%)	0 (0%)	1 (4%)
>12th grade	49 (98%)	25 (100%)	24 (96%)
Menopause			
Age at final menstrual period, Mean ± SD	48.1 ± 6.6	47.2 ± 6.2	49.0 ± 7.0
Current vasomotor symptoms, n (%)	32 (64%)	14 (56%)	18 (72%)
Prior hormone therapy, n (%)	38 (76%)	18 (72%)	20 (80%)
Sexual history			
Length of partnered relationship, years	29.3 ± 13.5	30.9 ± 13.7	27.8 ± 13.4
Duration of dyspareunia, years	6.3 ± 4.4	6.3 ± 4.9	6.4 ± 4.0
History of sexual abuse, n (%)	6 (12%)	3 (12%)	3 (12%)
Severity of dyspareunia, median (IQR)			
Intercourse pain score, NRS 0-10	8 (6, 8)	8 (5, 8)	8 (7, 8)
Baseline tampon test score, NRS 0-10	4 (3, 6)	4 (3, 6)	3 (2, 5)
Tenderness upon examination, n (%)			
Labia majora or minora	1 (2%)	1 (4%)	0 (0%)
Vestibule, score >0	50 (100%)	25 (100%)	25 (100%)
Vaginal wall, score >3	9 (18%)	5 (20%)	4 (16%)
Pelvic floor muscles	17 (34%)	11 (44%)	6 (24%)
Bladder	4 (8%)	2 (8%)	2 (8%)
Uterus, cervix, adnexa	3 (6%)	1 (4%)	2 (8%)

Exact tests, and all  $P > 0.05$ .

IQR, interquartile range; NRS, Numerical Rating Scale.

<sup>a</sup>Analyses used two-sample  $t$  test, Wilcoxon rank-sum test, or  $\chi^2$ /Fisher's exact test.

We detected no statistically significant difference in short-term measures of safety between groups (Table 4). The symptom most reported was irritation from application of the cream, which improved during treatment, affecting 11 women (23%) at 4 weeks and four women (8%) at 12 weeks. The three women who reported vaginal bleeding had one or two episodes of post-coital spotting; each had a thin endometrial stripe or no uterus. There were no major adverse events.

Serum estradiol levels were measured at baseline, 12 hours, 4 weeks, and 12 weeks (Table 4). Forty-one percent of all values were below the lower limit of the immunoassay, less than 5 pg/mL, and with therapy, these numbers decreased but were similarly distributed between treatment arms. To conservatively estimate aggregate values for each treatment group, all values reported as less than 5 were assigned a value of 5. Median estradiol levels rose slightly in each treatment arm, and the difference between strengths did not meet statistical significance. The mean endometrial stripe thickness after 12 weeks of ET did not change significantly in either group.

## DISCUSSION

Comparing results of treatment with the two strengths of estradiol cream, each used with pre-coital silicone lubricant, we found no difference in reduction in dyspareunia severity and no differences in any of the secondary outcomes. We found a 75% decrease in intercourse pain scores and an 82% to 100% reduction in tenderness measured by touch testing at the most tender anatomic area,

the vulvar vestibule, when combining both treatment groups and comparing baseline to 12-week measures. Quality of life as measured by the VPAQ was improved in the domains of sexual function and cognitive/emotional function. Urinary symptoms were frequent but not significantly altered by vestibule estradiol.

Direct comparison of our results to findings in the major studies of vaginal ET for dyspareunia is limited by the lack of standardized outcome measures and varied study protocols with respect to concomitant use of lubricants and moisturizers. Recognizing this, a brief review illustrates the general magnitudes of improvement. In a 12-week randomized controlled trial of intravaginal estradiol tablets (10 µg) plus placebo vaginal moisturizer used three times per week, the subset of women with dyspareunia as the most bothersome symptom (60%) reported a 60% reduction in pain with penetration, with a similar 60% reduction in pain in the dual-placebo group.<sup>32</sup> A 12-week randomized controlled trial of vaginal estradiol inserts (4, 10, and 25 µg) in 704 postmenopausal women with self-described moderate-severe dyspareunia found reductions of 56%, 65%, and 63%, respectively, in dyspareunia pain scores and 47% for the placebo group.<sup>33</sup> The only report of vestibular ET was a 12-week uncontrolled study of 25-µg estriol gel in 63 postmenopausal women with dyspareunia.<sup>34</sup> Dyspareunia scores were reduced by 57%, and vestibule cotton-swab scores decreased by 56%. In a study of vestibular lidocaine plus silicone lubricant in breast cancer survivors with dyspareunia, there was a median reduction in dyspareunia pain scores of 88% compared with 38% reduction with use of saline (placebo) plus silicone lubricant.<sup>14</sup> The 75%

**TABLE 2.** Intercourse pain scores after treatment, comparing treatment arms

	50 µg	100 µg	$P^a$
<b>Intercourse pain scores—intention to treat,<sup>b</sup></b>			
n = 50 (NRS 0-10 median, IQR)			
(n = 50 µg count, 100 µg count)			
Baseline, n = 25, 25	8.0 (5.0, 8.0)	8 (7.0, 8.0)	0.76
4 wk, n = 25, 25	6.0 (2.0, 8.0)	6.0 (3.0, 8.0)	0.58
<b>Intercourse pain scores—per protocol/as treated<sup>c</sup></b>			
n = 24 at 4 wk, n = 31 at 12 wk (NRS 0-10 median, IQR)			
Intercourse scores at 0 and 4 wk			
Baseline, n = 15, 9	8.0 (5.0, 8.0)	8.0 (7.0, 8.0)	0.97
4 wk, n = 15, 9	4.0 (1.0, 8.0)	3.0 (2.0, 5.0)	0.31
Intercourse scores at 0 and 12 wk			
Baseline, n = 18, 13	7.5 (5.0, 8.0)	8.0 (7.0, 8.0)	0.52
12-wk, n = 18, 13	2.0 (0.0, 3.0)	2.0 (1.0, 3.0)	0.58
<b>Tampon test scores—per protocol/as treated<sup>c</sup></b>			
n = 42 at 4 wk, 35 at 12 wk (NRS 0-10 median, IQR)			
Baseline, n = 25, 25	4.0 (3.0, 6.0)	3.0 (2.0, 5.0)	0.27
4-wk, n = 22, 20	1.0 (0, 2.0)	0 (0, 1.5)	0.15
12-wk, n = 19, 16	0 (0, 2.0)	0 (0, 1.0)	0.73

In all analyses, the diary pain score closest to 28 days (4 weeks) or 84 days (12 weeks) within a range of  $\pm 7$  days was compared with the baseline score. Participants had been instructed to record two penetration pain scores per week, either intercourse or tampon test scores.

IQR, interquartile range; NRS, Numerical Rating Scale.

<sup>a</sup>Wilcoxon rank-sum test, median (IQR) reported.

<sup>b</sup>Absent intercourse scores at 4 weeks were estimated using baseline or last observation carried forward.

<sup>c</sup>In the per-protocol analyses, fewer participants had resumed intercourse at 4 weeks compared with 12 weeks, resulting in lower numbers for analysis; similarly, women who had intercourse twice in 1 week did not record a tampon test score for that week.

**TABLE 3.** Treatments outcomes over time for women who completed 12 weeks of therapy, combining 50- and 10- $\mu$ g groups

	Baseline	4 wk	12 wk	<i>P</i> <sup>a</sup>
Severity of dyspareunia, median (IQR)				
Intercourse pain score (n = 31)	8.0 (5.0, 8.0)	5.0 (2.0, 7.0)	2.0 (1.0, 3.0)	<0.001
Tampon pain score, (n = 32)	4.0 (3.0, 5.0)	1.0 (0.0, 2.0)	0.0 (0.0, 0.5)	<0.001
Vaginal pH, n = 46, mean $\pm$ SD	5.4 $\pm$ 0.7	4.7 $\pm$ 0.6	4.7 $\pm$ 0.6	<0.001
VPAQ scores (0-4), mean $\pm$ SD				
Pain severity				
Burning pain subscale, n = 45	2.7 $\pm$ 1.2	0.7 $\pm$ 0.9	0.7 $\pm$ 1.0	<0.001
Incisive pain subscale, n = 44	2.2 $\pm$ 1.5	0.6 $\pm$ 1.0	0.6 $\pm$ 1.1	<0.001
Sensitivity subscale, n = 45	2.6 $\pm$ 0.9	0.9 $\pm$ 0.8	0.7 $\pm$ 0.8	<0.001
Psychosocial scales				
Cognitive emotional, n = 46	1.1 $\pm$ 0.7	0.4 $\pm$ 0.5	0.3 $\pm$ 0.5	<0.001
Life interference, n = 39	0.3 $\pm$ 0.5	0.2 $\pm$ 0.3	0.1 $\pm$ 0.2	0.002
Sexual function, n = 46	2.8 $\pm$ 0.9	1.6 $\pm$ 1.2	0.8 $\pm$ 0.9	<0.001
Self-stimulation interference, n = 13	1.5 $\pm$ 1.2	1.4 $\pm$ 1.7	0.7 $\pm$ 1.3	0.11
Dryness scores, VPAQ format, n = 46	3.2 $\pm$ 1.0	2.2 $\pm$ 1.3	1.8 $\pm$ 1.2	<0.001
Vestibule sensitivity, NRS 0-10 median (IQR), n = 46				
Supraurethral midline (12:00)	2.5 (1.0, 5.0)	1.0 (0.0, 2.0)	0 (0, 1.0)	<0.001
Paraurethral (2:00, 10:00)	2.0 (0.5, 3.5)	0.5 (0.0, 1.5)	0.0 (0.0, 0.5)	<0.001
Middle (3:00, 9:00)	4.5 (3.0, 6.0)	1.5 (1.0, 3.0)	0.5 (0.0, 1.0)	<0.001
Bartholin's duct area (4:00, 8:00)	5.5 (4.0, 8.0)	2.0 (1.0, 4.0)	1.0 (0.5, 2.0)	<0.001
Posterior fourchette midline (6:00)	4.0 (3.0, 7.0)	1.5 (0.0, 3.0)	0.0 (0.0, 2.0)	<0.001
Range of scores	0-10	0-9	0-6	
Vaginal wall tenderness, NRS 0-10 median (IQR)				
Lateral walls (3:00, 9:00), n = 46	0.5 (0, 2.0)	0.0 (0.0, 0.0)	0.0 (0.0, 0.5)	<0.001
Range of scores	0-7.5	0-6	0-4	
Pelvic floor muscle tenderness, n = 46				
Perineal	6 (13.0%)	3 (6.5%)	3 (6.5%)	0.44
Levator ani	14 (30.4%)	6 (13.0%)	10 (21.7%)	0.13
Upper vaginal	7 (15.2%)	4 (8.7%)	3 (6.5%)	0.36
Urinary symptoms, yes/no n (%), n = 46				
Urinary Distress Inventory, short (UDI-6)				
Frequent urination	19 (41.3%)	Not assessed	11 (23.9%)	0.08
Urge leakage	32 (69.6%)		32 (69.6%)	1.00
Stress leakage	26 (56.5%)		24 (52.2%)	0.67
Small amounts of leakage	21 (45.6%)		20 (43.5%)	0.83
Difficulty emptying bladder	6 (13.0%)		4 (8.7%)	0.50
Pain in abdomen/genital area	5 (10.9%)		6 (13.0%)	0.75
Other instruments, n (%), n = 46				
Strong feeling of urgency <sup>b</sup>	19 (41.3%)		14 (30.4%)	0.28
Nocturia more than once <sup>c</sup>	15 (32.6%)		11 (23.9%)	0.37
Episodes of non-UTI bladder pain <sup>c</sup>	6 (13.0%)		2 (4.4%)	0.14
Episodes on non-UTI dysuria <sup>d</sup>	9 (19.6%)		3 (6.5%)	0.06
Urinary incontinence severity, mean $\pm$ SD				
Sandvik Severity Index (0-12), n = 23	4.9 $\pm$ 3.6		4.1 $\pm$ 2.7	0.10
UDI-6 score (0-100), n = 46	19.6 $\pm$ 12.4		14.8 $\pm$ 13.0	0.12

ANOVA, analysis of variance; IQR, interquartile range; NRS, Numerical Rating Scale; UDI, Urinary Distress Inventory-6; UTI, urinary tract infection; VPAQ, Vulvar Pain Assessment Questionnaire.

<sup>a</sup>Repeated-measures ANOVA or Friedman test.

<sup>b</sup>UDI, long form.

<sup>c</sup>O'Leary/Sant Interstitial Cystitis Symptom Index.

<sup>d</sup>Urinary Tract Infection Symptom Assessment.

effect size in our study exceeds the typical 30% to 60% placebo effect noted in other dyspareunia studies.<sup>32,33,35-37</sup>

Whereas severity was markedly reduced, most women (71%) reported some degree of persistent dyspareunia after 12 weeks of therapy. Similarly, the studies above noted severity reduction from severe to mild. Using a scale of 0 to 3, the mean score after 12 weeks remained 1.0 for the estradiol tablets,<sup>32</sup> 0.9 for the 10- $\mu$ g estradiol inserts,<sup>33</sup> and 1.23 for introital estriol.<sup>34</sup> Whereas improvement is valuable, we do not know whether a mild level of dyspareunia is acceptable to women. In the rheumatology and urogynecology literature, there is growing use of an acceptability concept or Patient Acceptable Symptom Score (PASS),<sup>38</sup> the level of symptom severity such that the patient desires no further

treatment. Whereas a minimal clinically important difference represents one goal for women, the PASS may be more meaningful. In GSM research, neither minimal clinically important difference nor PASS levels for GSM symptom outcomes measures are known. Inadequate duration of therapy could explain remaining dyspareunia and lack of urologic effect in this study. In a study of intravaginal application of 50  $\mu$ g ET, both scores from overactive bladder were improved by 26%, but full improvement did not occur until 24 weeks of therapy.<sup>39</sup>

In each treatment arm, VPAQ descriptors of penetrative pain revealed similar severities of burning pain, incisive pain, sensitivity, and dryness. Every descriptor improved but did not resolve by 12 weeks. Women commonly report "dryness" during

TABLE 4. Adverse events and safety

	Overall n = 48	50 µg n = 25	100 µg n = 23	P <sup>a</sup>
At 4 wk, n (%)				
Breast tenderness	5 (10)	4 (16)	1 (4)	0.35
Vaginal spotting or bleeding	1 (2)	1 (4)	0 (0)	1.00
Stinging with cream application	11 (23)	5 (20)	6 (26)	0.74
At 12 wk, n (%)				
Breast tenderness	6 (13)	3 (12)	3 (14)	1.00
Vaginal spotting or bleeding	1 (2)	1 (4)	0 (0)	1.00
Stinging with cream application	4 (8)	2 (8)	2 (9)	1.00
Estradiol levels <5 pg/mL, n (%)				
Baseline	24 (54)	13 (52)	13 (57)	0.75
12 h	20 (42)	12 (48)	8 (35)	0.35
4 wk	14 (29)	9 (36)	5 (22)	0.28
12 wk	16 (33)	8 (32)	8 (35)	0.84
Estimated estradiol levels, median (IQR)				
Baseline	5.0 (5.0, 10.7)	5.0 (5.0, 10.0)	5.0 (5.0, 19.2)	0.57
12 h	7.1 (5.0, 12.5)	5.4 (5.0, 10.4)	8.6 (5.0, 17.4)	0.12
4 wk	9.5 (5.0, 13.9)	7.2 (5.0, 13.5)	10.7 (7.5, 14.1)	0.25
12 wk	9.5 (5.0, 16.1)	9.1 (5.0, 14.6)	11.1 (5.0, 18.9)	0.66
Endometrial stripe thickness, mean ± SD				
Baseline	2.8 ± 0.8	2.6 ± 0.8	3.0 ± 0.9	0.13
12 wk	2.9 ± 0.9	2.7 ± 0.8	3.0 ± 1.1	0.34

IQR, interquartile range.

<sup>a</sup>Fisher's exact test, two-sample *t* test, or Wilcoxon rank-sum test.

penetration despite use of silicone lubricant, denoting a noxious sensation rather than a literal dryness, and yet “dry” is not a listed pain descriptor in the 78-item McGill Pain Questionnaire.<sup>40</sup> The VPAQ demonstrated the detriments of dyspareunia in multiple biopsychosocial domains and improvements with ET. In addition to research use, the VPAQ could be an instrument that helps women describe their symptoms to healthcare professionals.<sup>41</sup>

Whereas the entire urogenital tract can reflect atrophy from estrogen deficiency, physical examinations demonstrated differing pain responses from the component structures. The parallel improvement in vestibule tenderness with treatment suggested that the vestibule might be the preferred target of GSM dyspareunia therapy. The tampon test was well tolerated as a surrogate measure of penetration pain, and except when baseline scores were zero, it allowed women to note therapeutic “progress,” thereby reducing dread of intercourse. Tampon test scores produced a more complete data set than intercourse scores for women naturally reluctant to be sexually active. Tampons as test devices are easily available and well known to women, but dilators could be explored as a more representative alternative.

### Safety and acceptability

The 100-µg daily estradiol cream dose used in this study represents the lowest recommended for therapy in the US Food and Drug Administration package label for vaginal estradiol cream (1–4 g of estradiol cream 0.01%).<sup>42</sup> The lower dose, 50 µg, is the currently recommended dose for clinical use in The North American Menopause Society position statement. Mean serum estradiol levels with the 50-µg dose at 12 weeks, 9.1 pg/mL, remained within the range for that of untreated postmenopausal women, which ranges from undetectable to 10.5 pg/mL by

radioimmunoassay.<sup>43,44</sup> The value for the 100-µg dose, 11.1 pg/mL, slightly exceeds that for untreated women, but both levels may be inaccurate because of our inability to quantify values less than 5 pg/mL. A 50-µg dose of estradiol cream applied to the lower vagina with a finger in women with prolapse resulted in similar levels, 9.7 pg/mL after daily application for 8 weeks and 8.7 pg/mL after twice-weekly application for 8 further weeks.<sup>45</sup> We could find no assessments using 100-µg daily.<sup>46</sup>

### Clinical and research implications

When dyspareunia is the presenting symptom in postmenopausal women, a comprehensive pelvic examination is essential to ascertain all sites of tenderness as a guide to directed therapy. Pelvic floor muscle tenderness and vaginal stenosis will need specific additional therapy. When the vestibule is the primary site of tenderness, we can be reassured that introital applications of estradiol cream, with lubricated intercourse, are a reasonable alternative to intravaginal estrogen applications for dyspareunia, but complete resolution should not be expected for all affected women by 12 weeks. The estradiol regimen required to induce full pain relief and the regimen required to maintain relief are unknown. Because GSM is a chronic condition requiring ongoing therapy, clinical follow-up is essential to optimize therapeutic benefits and balance this with potential safety concerns for each woman. Effective therapy for mild cases might be important to avoid progression to severities that require longer courses of daily therapy. Precoital, introital application of lidocaine 4% topical solution can be offered as adjunct therapy during prolonged ET. Future research should include better definition of severity, validated pain scales, and the objective physical correlates of dyspareunia used in our study. The relevance of measures of introital size, vaginal length, elasticity, and lubrication could be explored.

## Strengths and limitations

Strengths of our study include using metered doses of estradiol cream and lubricant and instruments and physical examination outcome measures with strong psychometric properties from the domain of vulvar pain research.<sup>17,20-25,27,28</sup> This study is generalizable only to urban, heterosexually active, White women with similarly defined moderate/severe dyspareunia. The absence of a placebo control precludes measurement of the magnitude of a treatment benefit. Use of the more sensitive liquid or gas chromatography/mass spectroscopy would have provided more exact measurements of systemic estradiol levels.

## CONCLUSION

When a comprehensive genital examination reveals the vestibule to be the site of greatest tenderness, estradiol cream targeted to the vulvar vestibule is a reasonable alternative to intra-vaginal applications for treatment of GSM dyspareunia. With similar efficacies, the 50- $\mu$ g dose is preferable to the 100- $\mu$ g dose regarding concerns about absorption, but the need for prolonged nightly dosing to achieve meaningful correction deserves more detailed research. Use of outcome measures from vulvar pain research provides a more expansive description of dyspareunia associated with GSM and may help to refine our therapies.

## REFERENCES

- Palma F, Volpe A, Villa P, Cagnacci A; Writing Group of AGATA Study. Vaginal atrophy of women in postmenopause. Results from a multicentric observational study: the AGATA study. *Maturitas* 2016;83:40-44. doi: 10.1016/j.maturitas.2015.09.001
- The NAMS 2020 GSM Position Statement Editorial Panel. The 2020 genitourinary syndrome of menopause position statement of The North American Menopause Society. *Menopause* 2020;27:976-992. doi: 10.1097/GME.0000000000001609
- Lonn  -Hoffmann RA, Dennerstein L, Leher P, Szoek C. Sexual function in the late postmenopause: a decade of follow-up in a population-based cohort of Australian women. *J Sex Med* 2014;11:2029-2038. doi: 10.1111/jsm.12590
- Avis NE, Brockwell S, Randolph JF Jr., et al. Longitudinal changes in sexual functioning as women transition through menopause: results from the Study of Women's Health Across the Nation. *Menopause* 2009;16:442-452. doi: 10.1097/gme.0b013e3181948dd0
- Nappi RE, Kingsberg S, Maamari R, Simon J. The CLOSER (CLarifying Vaginal Atrophy's Impact On SEx and Relationships) survey: implications of vaginal discomfort in postmenopausal women and in male partners. *J Sex Med* 2013;10:2232-2241. doi: 10.1111/jsm.12235
- Simon JA, Kokot-Kierepa M, Goldstein J, Nappi RE. Vaginal health in the United States: results from the Vaginal Health: Insights, Views & Attitudes survey. *Menopause* 2013;20:1043-1048. doi: 10.1097/GME.0b013e318287342d
- Simon JA, Nappi RE, Kingsberg SA, Maamari R, Brown V. CLarifying Vaginal Atrophy's Impact On SEx and Relationships (CLOSER) survey: emotional and physical impact of vaginal discomfort on North American postmenopausal women and their partners. *Menopause* 2014;21:137-142. doi: 10.1097/GME.0b013e318295236f
- Gandhi J, Chen A, Dagur G, et al. Genitourinary syndrome of menopause: an overview of clinical manifestations, pathophysiology, etiology, evaluation, and management. *Am J Obstet Gynecol* 2016;215:704-711. doi: 10.1016/j.ajog.2016.07.045
- Shiffen JL, Zinzavage R, Cho EL, et al. Women's experience of vulvovaginal symptoms associated with menopause. *Menopause* 2019;26:341-349. doi: 10.1097/GME.0000000000001275
- Kao A, Binik YM, Kapuscinski A, Khalife S. Dyspareunia in postmenopausal women: a critical review. *Pain Res Manag* 2008;13:243-254. doi: 10.1155/2008/269571
- Kao A, Binik YM, Amsel R, Funaro D, Leroux N, Khalife S. Biopsychosocial predictors of postmenopausal dyspareunia: the role of steroid hormones, vulvovaginal atrophy, cognitive-emotional factors, and dyadic adjustment. *J Sex Med* 2012;9:2066-2076. doi: 10.1111/j.1743-6109.2012.02771.x
- Kingsberg SA, Wysocki S, Magnus L, Krychman ML. Vulvar and vaginal atrophy in postmenopausal women: findings from the REVIVE (REal Women's Views of Treatment Options for Menopausal Vaginal ChangEs) survey. *J Sex Med* 2013;10:1790-1799. doi: 10.1111/jsm.12190
- Goetsch MF, Lim JY, Caughey AB. Locating pain in breast cancer survivors experiencing dyspareunia: a randomized controlled trial. *Obstet Gynecol* 2014;123:1231-1236. doi: 10.1097/AOG.0000000000000283
- Goetsch MF, Lim JY, Caughey AB. A practical solution for dyspareunia in breast cancer survivors: a randomized controlled trial. *J Clin Oncol* 2015;33:3394-3400. doi: 10.1200/JCO.2014.60.7366
- Goetsch MF. Unprovoked vestibular burning in late estrogen-deprived menopause: a case series. *J Low Genit Tract Dis* 2012;16:442-446. doi: 10.1097/LGT.0b013e31825c2d28
- Goetsch MF, Garg B, Lillemon J, Clark AL. Where does postmenopausal dyspareunia hurt? A cross-sectional report. *Menopause* 2022;29:646-653. doi: 10.1097/GME.0000000000001956
- Dargie E, Holden RR, Pukall CF. The Vulvar Pain Assessment Questionnaire inventory. *Pain* 2016;157:2672-2686. doi: 10.1097/j.pain.0000000000000682
- Dworkin RH, Turk DC, Farrar JT, et al. Core outcome measures for chronic pain clinical trials: IMMPACT recommendations. *Pain* 2005;113(1-2):9-19. doi: 10.1016/j.pain.2004.09.012
- Pukall CF, Bergeron S, Brown C, Bachmann G, Wesselmann U, Vulvodysnia Collaborative Research Group. Recommendations for self-report outcome measures in vulvodysnia clinical trials. *Clin J Pain* 2017;33:756-765. doi: 10.1097/AJP.0000000000000453
- Barber MD, Walters MD, Bump RC. Short forms of two condition-specific quality-of-life questionnaires for women with pelvic floor disorders (PFDI-20 and PFIQ-7). *Am J Obstet Gynecol* 2005;193:103-113. doi: 10.1016/j.ajog.2004.12.025
- Uebersax JS, Wyman JF, Shumaker SA, McClish DK, Fantl JA. Short forms to assess life quality and symptom distress for urinary incontinence in women: the Incontinence Impact Questionnaire and the Urogenital Distress Inventory. Continence Program for Women Research Group. *Neurourol Urodyn* 1995;14:131-139. doi: 10.1002/nau.1930140206
- Hanley J, Capewell A, Hagen S. Validity study of the severity index, a simple measure of urinary incontinence in women. *BMJ* 2001;322:1096-1097. doi: 10.1136/bmj.322.7294.1096
- O'Leary MP, Sant GR, Fowler FJ Jr., Whitmore KE, Spolarich-Kroll J. The interstitial cystitis symptom index and problem index. *Urology* 1997;49(5A suppl):58-63. doi: 10.1016/s0090-4295(99)80333-1
- Clayson D, Wild D, Doll H, Keating K, Gondek K. Validation of a patient-administered questionnaire to measure the severity and bothersomeness of lower urinary tract symptoms in uncomplicated urinary tract infection (UTI): the UTI Symptom Assessment questionnaire. *BJU Int* 2005;96:350-359. doi: 10.1111/j.1464-410X.2005.05630.x
- Foster DC, Beth Kotok M, Huang LS, et al. The tampon test for vulvodysnia treatment outcomes research: reliability, construct validity, and responsiveness. *Obstet Gynecol* 2009;113:825-832. doi: 10.1097/AOG.0b013e31819bda7c
- Woo A, Lechner B, Fu T, et al. Cut points for mild, moderate, and severe pain among cancer and non-cancer patients: a literature review. *Ann Palliat Med* 2015;4:176-183. doi: 10.3978/j.issn.2224-5820.2015.09.04
- Stenson AL, Leclair CM, Goetsch MF. Comparing vestibule examination techniques: light touch, serial forces, and the lidocaine test. *J Low Genit Tract Dis* 2021;25:236-242. doi: 10.1097/LGT.0000000000000605
- Kapurubandara SC, Lowes B, Sansom-Daly UM, Deans R, Abbott JA. A systematic review of diagnostic tests to detect pelvic floor myofascial pain. *Int Urogynecol J* 2022;33:2379-2389. doi: 10.1007/s00192-022-05258-7
- Kao A, Binik YM, Amsel R, Funaro D, Leroux N, Khalife S. Challenging atrophied perspectives on postmenopausal dyspareunia: a systematic description and synthesis of clinical pain characteristics. *J Sex Marital Ther* 2012;38:128-150. doi: 10.1080/0092623X.2011.569641
- Harris PA, Taylor R, Thielke R, Payne J, Gonzalez N, Conde JG. Research electronic data capture (REDCap)—a metadata-driven methodology and workflow process for providing translational research informatics support. *J Biomed Inform* 2009;42:377-381. doi: 10.1016/j.jbi.2008.08.010
- Harris PA, Taylor R, Minor BL, et al. The REDCap Consortium: building an international community of software platform partners. *J Biomed Inform* 2019;95:103208. doi: 10.1016/j.jbi.2019.103208
- Mitchell CM, Reed SD, Diem S, et al. Efficacy of vaginal estradiol or vaginal moisturizer vs placebo for treating postmenopausal vulvovaginal



- symptoms: a randomized clinical trial. *JAMA Intern Med* 2018;178:681-690. doi: 10.1001/jamainternmed.2018.0116
33. Constantine GD, Simon JA, Pickar JH, et al. The REJOICE trial: a phase 3 randomized, controlled trial evaluating the safety and efficacy of a novel vaginal estradiol soft-gel capsule for symptomatic vulvar and vaginal atrophy. *Menopause* 2017;24:409-416. doi: 10.1097/gme.0000000000000786
34. Murina F, Graziottin A, Felice R, Di Francesco S. Coital pain in the elderly: could a low dose estriol gel thrill the vulvar vestibule? *Eur J Obstet Gynecol Reprod Biol* 2016;207:121-124. doi: 10.1016/j.ejogrb.2016.10.016
35. Foster DC, Kotok MB, Huang LS, et al. Oral desipramine and topical lidocaine for vulvodynia: a randomized controlled trial. *Obstet Gynecol* 2010;116:583-593. doi: 10.1097/AOG.0b013e3181e9e0ab
36. Andrews JC. Vulvodynia interventions—systematic review and evidence grading. *Obstet Gynecol Surv* 2011;66:299-315. doi: 10.1097/OGX.0b013e3182277fb7
37. Miranda Varella Pereira G, Soriano Marcolino M, Silveira Nogueira Reis Z, Vale de Castro Monteiro M. A systematic review of drug treatment of vulvodynia: evidence of a strong placebo effect. *BJOG* 2018;125:1216-1224. doi: 10.1111/1471-0528.15223
38. Sanderson DJ, Zavez A, Meekins AR, et al. The patient acceptable symptom state in female urinary incontinence. *Female Pelvic Med Reconstr Surg* 2022;28:33-39. doi: 10.1097/SPV.0000000000001055
39. Ellington DR, Szychowski JM, Malek JM, Gerten KA, Burgio KL, Richter HE. Combined tolterodine and vaginal estradiol cream for overactive bladder symptoms after randomized single-therapy treatment. *Female Pelvic Med Reconstr Surg* 2016;22:254-260. doi: 10.1097/spv.0000000000000256
40. Melzack R. The McGill Pain Questionnaire: major properties and scoring methods. *Pain* 1975;1:277-299. doi: 10.1016/0304-3959(75)90044-5
41. Dargie E, Pukall CF, Goetsch M, Stenson A, Leclair C. The clinical utility of the Vulvar Pain Assessment Questionnaire: a pilot study. *J Low Genit Tract Dis* 2019;23:272-278. doi: 10.1097/lgt.0000000000000488
42. Food and Drug Administration. Estrace Cream [package insert]. Available at: [https://www.accessdata.fda.gov/drugsatfda\\_docs/label/2005/86069s018lbl.pdf](https://www.accessdata.fda.gov/drugsatfda_docs/label/2005/86069s018lbl.pdf). Accessed October 13, 2021.
43. Santen RJ. Vaginal administration of estradiol: effects of dose, preparation and timing on plasma estradiol levels. *Climacteric* 2015;18:121-134. doi: 10.3109/13697137.2014.947254
44. Santen RJ, Mirkin S, Bernick B, Constantine GD. Systemic estradiol levels with low-dose vaginal estrogens. *Menopause* 2020;27:361-370. doi: 10.1097/gme.0000000000001463
45. Illston JD, Wheeler TL, Parker CR, et al. Low-dose 17-β-estradiol cream for vaginal atrophy in a cohort without prolapse: serum levels and vaginal response including tissue biomarkers associated with tissue remodeling. *Maturitas* 2015;81:475-479. doi: 10.1016/j.maturitas.2015.05.010
46. Crandall CJ, Diamant A, Santoro N. Safety of vaginal estrogens: a systematic review. *Menopause* 2020;27:339-360. doi: 10.1097/GME.0000000000001468