

## ORIGINAL STUDY

# Physical characteristics and properties of estradiol softgel vaginal inserts

James A. Simon, MD,<sup>1</sup> James H. Pickar, MD,<sup>2</sup> Annette M. Shadiack, PhD,<sup>3</sup> Bharat Warriar, MS,<sup>3</sup> Shelli Graham, PhD,<sup>3</sup> Brian Bernick, MD,<sup>3</sup> and Sebastian Mirkin, MD<sup>3</sup>

### Abstract

**Objective:** TX-004HR is a low-dose estradiol (E2) softgel vaginal insert designed to be rapidly dissolving and mucoadhesive. This report describes the physical attributes and pharmacokinetic parameters of the softgel vaginal insert evaluated for the treatment of moderate to severe dyspareunia due to menopausal vulvar and vaginal atrophy.

**Methods:** In vitro dissolution studies with 25- $\mu$ g E2 inserts were performed and media samples were analyzed for E2 by high-performance liquid chromatography. Effects of body position on E2 bioavailability were assessed in a phase 1, randomized trial of the 25- $\mu$ g softgel capsule versus a reference product in which women remained supine after dosing ( $n = 16$ ), and in a substudy ( $n = 16$ ) in which women were ambulatory or seated after dosing. Estradiol  $C_{\max}$ ,  $AUC_{0-24}$ , and  $t_{\max}$  were measured by high-performance liquid chromatography-tandem mass spectroscopy. A phase 2, randomized study ( $n = 50$ ) of 10- $\mu$ g E2 versus placebo inserts assessed timing of capsule disintegration at days 1 and 15.

**Results:** In vitro testing detected more than 80% of E2 in the dissolution medium by 15 minutes (first time point measured). In the phase 1 studies, baseline-corrected E2 plasma levels were not significantly different regardless of supine versus ambulatory/seated position after dosing:  $C_{\max}$ , 24.1 versus 34.3 pg/mL;  $AUC_{0-24}$ , 77.6 versus 93.7 h · pg/mL; and  $t_{\max}$ , 2.1 versus 1.9 hours, respectively. In the phase 2 study, no remnants of the softgel capsule were found at day 1 (6 hours) after dosing and day 15. Vaginal discharge was minimal (1/48 women; 2.1%).

**Conclusions:** The presented data support rapid dissolution of the softgel capsule and similar E2 pharmacokinetic parameters regardless of body position after dosing.

**Key Words:** Estradiol – Menopause – Pharmacokinetics – TX-004HR – Vaginal estrogen therapy – Vulvar and vaginal atrophy.

During menopause, decline in endogenous estrogen production can result in the thinning, drying, and loss of elasticity of the vaginal epithelium, resulting in vulvar and vaginal atrophy (VVA).<sup>1</sup> Up to 69% of postmenopausal women have clinical signs of VVA,<sup>2</sup> and nearly half of women with VVA suffer from dyspareunia, vaginal dryness, irritation, and itching.<sup>3</sup> Although VVA may be

progressive without treatment<sup>4</sup> and can significantly impair a woman's quality of life,<sup>5</sup> up to 50% of women are not being treated for their VVA symptoms.<sup>6</sup> Many women are dissatisfied with their current vaginal prescriptions because of insufficient efficacy, inconvenient application procedures, disruption and interference with sexual spontaneity, and excessive vaginal discharge.<sup>7-9</sup>

Received June 17, 2019; revised and accepted August 14, 2019.

From the <sup>1</sup>George Washington University, School of Medicine, Intim-Medicine Specialists, Washington, DC; <sup>2</sup>Columbia University Medical Center, New York, NY; and <sup>3</sup>TherapeuticsMD, Boca Raton, FL.

Funding/support: TherapeuticsMD (Boca Raton, Florida) sponsored the studies and supported the medical writing assistance of Laura J. Ninger, ELS and Dominique J. Verlaan, PhD of Precise Publications, LLC (Bedminster, New Jersey).

Financial disclosure/conflicts of interest: Dr. Simon has served (within the past year, or current) as a consultant/advisor to AbbVie, Allergan plc, AMAG, Amgen, Ascend Therapeutics, Bayer Healthcare, CEEK Enterprises, Covance, Dare Bioscience, Duchesnay, Hologic, KaNDy/NeRRe Therapeutics, Mitsubishi Tanabe, ObsEva SA, Palatin Technologies, Sanofi SA, Shionogi, Sprout, and TherapeuticsMD; has received (within the past year, or current) grant/research support from AbbVie, Agile Therapeutics, Allergan plc, Bayer Healthcare, Endocuetics, GTx, Ipsen, Myovant Sciences, New England Research Institute, ObsEva SA, Palatin

Technologies, Symbio Research, TherapeuticsMD, and Viveve Medical; has also served (within the past year, or current) on the Speakers' Bureaus of AbbVie, AMAG, Duchesnay, Novo Nordisk, Shionogi, and TherapeuticsMD; and is a stockholder (direct purchase) in Sermonix Pharmaceuticals. Dr. Pickar consults for Pfizer, Shionogi, and TherapeuticsMD; and has stock options with TherapeuticsMD. Drs. Shadiack, Graham, Bernick, and Mirkin, and Mr. Warriar are employees of TherapeuticsMD with stock/stock options. Dr. Bernick is also a Board member of TherapeuticsMD.

Address correspondence to: James A. Simon, MD, George Washington University, School of Medicine, IntimMedicine Specialists, 1850 M Street, NW 450, Washington, DC 20036. E-mail: jsimon@intimmedicine.com  
This is an open access article distributed under the terms of the Creative Commons Attribution-Non Commercial-No Derivatives License 4.0 (CCBY-NC-ND), where it is permissible to download and share the work provided it is properly cited. The work cannot be changed in any way or used commercially without permission from the journal.

TX-004HR (TherapeuticsMD, Boca Raton, FL) are 17 $\beta$ -estradiol (E2) vaginal inserts containing 4-, 10-, and 25- $\mu$ g E2 doses; the 4- and 10- $\mu$ g E2 doses were approved as Imvexxy by the US Food and Drug Administration in May 2018 for the treatment of moderate to severe dyspareunia, a symptom of VVA, due to menopause.<sup>10</sup> The E2 vaginal inserts are small, light-pink, tear-shaped softgel capsules inserted manually, without the use of an applicator. One insert is administered daily for 2 weeks and then twice weekly thereafter; it can be inserted at any time of the day. Inserts of TX-004HR (4-, 10-, 25- $\mu$ g E2; identical except for E2 content) were clinically evaluated in the 12-week REJOICE trial. Each dose significantly improved the percentage of superficial and parabasal cells, vaginal pH, and the most bothersome symptom of dyspareunia (4 coprimary efficacy endpoint) compared with placebo, with an onset of effect observed as early as 2 weeks and maintained throughout the 12-week study.<sup>11</sup> Vaginal dryness (secondary endpoint) also statistically significantly improved as early as 2 weeks for the 10- and 25- $\mu$ g doses and 6 weeks for the 4- $\mu$ g dose compared with placebo.<sup>11</sup> Furthermore, a pharmacokinetics (PK) substudy demonstrated negligible to very low systemic absorption of E2 with vaginal TX-004HR, with no statistical differences observed on day 14 with the 4- and 10- $\mu$ g doses, and minimal differences with the 25- $\mu$ g dose in E2 PK parameters versus placebo.<sup>12</sup>

The softgel vaginal insert was designed with mucoadhesive properties to facilitate retention of the product in the vagina and availability of the active ingredient in a site-specific manner. Gelatin was selected as the capsule shell based on its mucoadhesive properties from its polymeric structure and for its fast-dissolving properties. Fast dissolution of the capsule is important to release the contents, allow rapid local absorption, and may potentially minimize messiness following administration. In addition, the included ingredients of the capsule were selected to be well tolerated by mucosal tissues and were developed to be viscous at body temperature to resist flow. This product was designed in part to eliminate the need for using an applicator for insertion as required with other vaginal products<sup>13-16</sup> and to potentially improve patient satisfaction, which has been considered low with currently available treatments,<sup>7</sup> in part based on low rates of continuation.<sup>6</sup>

Although data on the safety and efficacy of TX-004HR have been published,<sup>11,12,17-19</sup> how fast the softgel capsule dissolves in vitro and in vivo and whether a difference in position (supine vs ambulatory/seated) after insertion affect estradiol absorption have never been reported. Therefore, the objective of this report was to describe the characteristics and properties of the gelatin coat and the softgel capsule fill as optimized for vaginal delivery, and to summarize in vitro and in vivo evidence on the rapid release of E2.

## METHODS

### In vitro study

In vitro testing for dissolution of the softgel capsule (25  $\mu$ g E2) was performed using a USP Dissolution Apparatus Type 3

at 30 dips per minute with the media at 37°C. The dissolution apparatus has a reciprocating cylinder design originally developed for testing dissolution of extended-release products or poorly soluble drugs, or those for which pH/buffer changes are required in the testing procedure.<sup>20</sup> Device operation is measured in dips per minute, or the number of times per minute that the inner and outer tubes are moved up and down so that the test product is agitated within the test medium.<sup>20</sup> Samples of the media were collected for six (n = 6) softgel capsules at 15, 30, 60, 90, and 120 minutes after initiation of the dipping, and analyzed for E2 by high-performance liquid chromatography (HPLC).

### Effects of body position on E2 bioavailability

Two phase 1 studies were conducted at the Micro Therapeutic Research Labs Private Limited (Tamil Nadu, India) to assess the PK of E2 in healthy postmenopausal women following treatment with a single dose of 25- $\mu$ g E2 vaginal capsule (identical to the approved 4- and 10- $\mu$ g vaginal inserts, except for E2 content). The first study was a randomized, open-label, two-way crossover study that compared the bioavailability of a single dose of 25- $\mu$ g TX-004HR with 25- $\mu$ g Vagifem (Novo Nordisk Inc, Plainsboro, NJ)<sup>21</sup>; only TX-004HR results are reported here. Participants were randomized 1:1 to one of the possible sequences (TX-004HR followed by Vagifem or Vagifem followed by TX-004HR) in consecutive order using SAS v9.2 (SAS Institute, Cary, NC). Eligible postmenopausal women (n = 36) were 40 to 65 years of age and had a body mass index (BMI) between 18.5 and 30 kg/m<sup>2</sup>; no VVA symptoms were required. Exclusion criteria included recent use of vaginal (<1 week within enrollment), percutaneous (<4 weeks), transdermal (<8 weeks), oral (<8 weeks), or injectable (<3-6 months) hormonal therapies; recent use of any prescription medication (last 14 days); and current smoking or recreational drug use. The study protocol was approved by the Chennai Ethics Committee and informed consent was obtained from each participant before screening.

After an overnight fast of 10 hours or more, the tear-shaped, softgel capsules with the smaller end up was inserted into the vaginal canal to at least 1 to 2 inches or up to the second finger joint by a trained professional. Participants were required to remain in a supine position for 4 hours after insertion. A 14-day washout was maintained between the study periods.

The second study, which occurred approximately 7 months later, was approved by the Research Ethics Committee. Sixteen women from the first phase 1 study were randomly selected to participate only after their willingness and consent for participation in this re-dosing study was obtained. Participants were re-treated with a 25- $\mu$ g E2 vaginal softgel capsule using the same protocol as the first study, except that women were required to be ambulatory or remained seated for 4 hours after a 5-minute rest period after insertion; they were not allowed to lie down. Effect of body position (supine vs ambulatory/seated) was compared for the same 16 women enrolled in both phase 1 studies.

In both studies, participants were not blinded to treatment since PK profiles are not subjective measurements; however, bioanalytical analysts were blinded to the randomization during the course of the analysis and until the statistical analysis.

The primary PK variables analyzed for E2 in both studies were peak plasma concentration ( $C_{max}$ ) and area under the plasma concentration-time curve from 0 to 24 hours ( $AUC_{0-24}$ ); a secondary outcome was time to peak plasma concentration ( $t_{max}$ ). A total of 13 blood samples for each woman was collected before dosing ( $-1$ ,  $-0.5$ , and  $0$  hours) and post-dose ( $1$ ,  $2$ ,  $4$ ,  $6$ ,  $8$ ,  $10$ ,  $12$ ,  $14$ ,  $18$ , and  $24$  hours). E2 in plasma was determined using a validated HPLC-tandem mass spectrometry (HPLC-MS/MS) method (E2 concentration reference range,  $2.0$ - $703.2$  pg/mL).

### Capsule assessment in a phase 2 study

This phase 2, pilot, randomized, double-blind, placebo-controlled trial evaluated the safety and efficacy of the  $10\text{-}\mu\text{g}$  E2 softgel vaginal insert in postmenopausal women; primary results have been previously published.<sup>22</sup> Eligible women had generally good health, age 40 to 75 years, BMI of  $34\text{ kg/m}^2$  or less superficial cells of 5% or less on vaginal cytology, vaginal pH of more than 5.0, and were experiencing at least one self-assessed moderate to severe symptom of VVA (defined as vaginal dryness, vaginal pain or bleeding with sexual activity, vaginal or vulvar irritation or itching, or dysuria). Exclusion criteria were similar to those for the phase 1 studies. Women were randomized 1:1 to  $10\text{-}\mu\text{g}$  E2 or matching placebo vaginal insert, which was self-administered daily in the morning for 14 days; there were no restrictions on movement after insertion. The study was approved by an institutional review board and women gave written informed consent before participating in the study. The study was blinded to investigators, sponsor, monitors, clinical site's personnel, and participants to minimize bias.

Results of primary endpoints included changes from baseline to day 15 in vaginal cytologic parameters, vaginal pH, most bothersome VVA symptom, vaginal bleeding during sexual activity, change in the vaginal mucosa, and safety.<sup>22</sup> As part of the trial's safety assessments, the vaginal vault was visually examined by the investigators to assess for remnants of the softgel capsule on day 1 (6 hours after first insertion) and day 15 (24 hours after last insertion); these results and any Medical Dictionary for Regulatory Activities (MedDRA) reproductive system and breast disorders adverse events (AEs) are reported here.

### Statistical analyses

In the *in vitro* study, the mean percentage of estradiol dissolution was reported as an average of the six samples at each time point.

In the phase 1 studies, determination of the sample size was determined by considering the treatment ratio, intraparticipant coefficient of variation (%), significant level ( $\alpha$ ), power, and bioequivalence limits. Using the PK parameters from the 16 participants from the first phase 1 study (supine) randomly

selected for the second phase 1 study, there was approximately 78% power in determining bioequivalence when compared to the PK results in the second phase 1 study (ambulatory).

PK parameters for the two phase 1 studies are reported with descriptive statistics (arithmetic mean, geometric mean, median, minimum/maximum, standard deviation, and coefficient of variation). Analysis of variance was performed on the  $C_{max}$  and  $AUC_{0-24}$ . Significance was assessed by geometric mean ratio (%) and 90% confidence interval (CI). The CI for the geometric mean ratio was based on paired *t* tests. All concentration values below the lower limit of quantification were set to 0 for all PK and statistical calculations. Baseline concentrations were determined for each dosing period and baseline corrections were participant and period specific. The method for baseline correction was arithmetic, with the mean of the three predose concentrations (at  $-1$ ,  $-0.5$ , and  $0$  hours) being subtracted from all the postdose concentrations. If a negative concentration value resulted after baseline correction, this was set to zero. Statistical analyses were performed using SAS v9.2. Bioanalyses were conducted at Micro Therapeutic Research Labs Private Limited (Chennai, India).

In the phase 2 study, descriptive statistical analyses are presented here for the evaluation of capsule remnants and reproductive system and breast disorders adverse events.

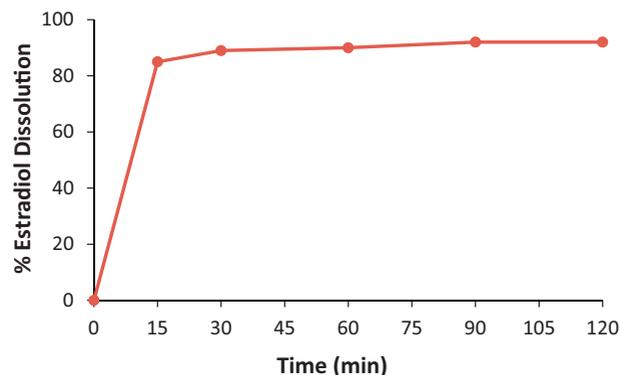
## RESULTS

### *In vitro* study

*In vitro* dissolution testing of the softgel capsules detected more than 80% (range, 81%-88%) of the E2 included in the softgel capsule in the dissolution medium within 15 minutes and more than 89% (range, 89%-94%) by 120 minutes (Fig. 1).

### Effects of body position on E2 bioavailability

From the 36 women enrolled in the first PK study, 16 were also enrolled in the second study; all 16 women completed both studies. These 16 women had a mean age of 50 years (range, 44-58 years) and BMI of  $25.8\text{ kg/m}^2$  ( $22.2$ - $29.9\text{ kg/m}^2$ ); all were of Asian race.



**FIG. 1.** *In vitro* estradiol dissolution profile of six  $25\text{-}\mu\text{g}$  softgel vaginal insert using a USP Dissolution Apparatus Type 3 at 30 dips per minute with media at  $37^\circ\text{C}$  and analyzed by high-performance liquid chromatography at different time points.

**TABLE 1.** Baseline-adjusted estradiol pharmacokinetic parameters in two phase 1 studies with the administration of a single dose of the 25-µg softgel vaginal capsule by body position

Estradiol parameters	Study 1 (supine) (mean ± SD)	Study 2 (ambulatory or seated) (mean ± SD)	GMR % (90% CI)
N	16	16	16
C <sub>max</sub> , pg/mL	24.1 ± 10.6	34.3 ± 19.5	74.6 (63.3-87.9)
AUC <sub>0-24</sub> , h · pg/mL	77.6 ± 30.4	93.7 ± 50.2	88.3 (71.9-108.4)
t <sub>max</sub> , h	2.1 ± 0.6	1.9 ± 0.7	

AUC<sub>0-24</sub>, area under the plasma concentration-time curve from 0 to 24 hours; C<sub>max</sub>, peak plasma concentration; GMR, geometric mean ratio (study 1/study 2); SD, standard deviation; t<sub>max</sub>, time to peak plasma concentration.

The 25-µg E2 softgel vaginal inserts resulted in peak E2 plasma concentrations at approximately 2 hours (Table 1); no significant differences were observed between women who remained supine compared with those who were ambulatory or remained in a seated position for baseline-corrected E2 plasma levels (Fig. 2). PK parameters were also not significantly different for the supine versus ambulatory/seated groups.

**Capsule assessment in a phase 2 study**

In the phase 2 study, 50 women were randomized to the 10-µg E2 softgel capsule (n = 24) or placebo (n = 26); 2 women receiving placebo did not complete the study. Participants who completed the study (n = 48) had a baseline mean age of 62.5 years (range, 46-75 years) and mean BMI of 27.0 kg/m<sup>2</sup> (range, 19.0-33.0 kg/m<sup>2</sup>); most women were white (92%) and fewer women were African American (6%) or Asian (2%).

Visual examination of the vaginal vault on day 1 (6 hours after first insertion) and on day 15 (24 hours after last insertion) did not detect any remnants of the softgel capsule (Table 2). One woman (1/48; 2.1%) treated with the softgel capsules (n = 1, TX-004HR) had mild vaginal discharge considered possibly related to study drug, which began on study day 14 and resolved by day 23. Other adverse events of the reproductive system and breast disorders were vaginal hemorrhage, vulvovaginal discomfort, burning sensation, or pain, and vaginal or cervical dysplasia and hot flushes (Table 3). Mild hot flushes were reported as being definitely related to treatment in one woman and not related in another woman; women neither reported having hot flushes at

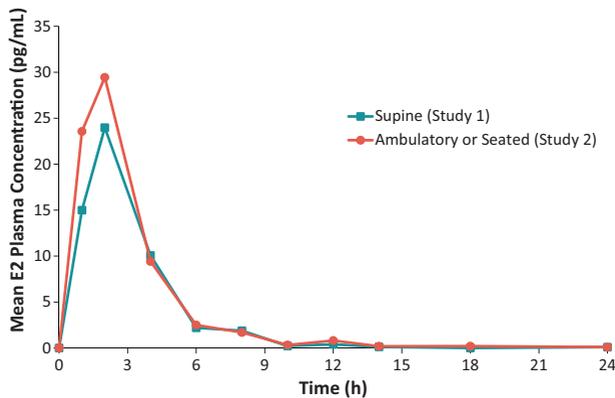
baseline nor any other adverse events during the study. No breast disorder adverse events were reported.

**DISCUSSION**

The softgel capsule vaginal insert was designed to be mucoadhesive and rapidly dissolving to release the drug quickly and absorb fully, which may minimize messiness during application. Similar PK parameters were observed in clinical studies in both the supine and seated/ambulatory positions, presumably due to the mucoadhesive gelatin polymers in the capsule shell and the viscosity of the fill material. The results suggest that the formulation allows for flexible timing and positions for dosing.<sup>10</sup> Furthermore, data from the in vitro study demonstrated that the gelatin capsule shell ruptured and significant dissolution of the soft gelatin capsule occurred within 15 minutes. Complete dissolution of the capsule within 6 hours of insertion, as observed in the phase 2 study, makes the solubilized E2 included in the capsule available for absorption to the vaginal mucosa.

While not clinically meaningful or significantly different, the C<sub>max</sub> and AUC were numerically higher in women who were ambulatory or seated compared with those who remained supine. This may be due to the small number of participants, and/or the fact that the data were not collected in the same study. It is also possible that these data are related to an increase in vaginal surface area coming into contact with the estradiol as the women moved around or an increase in blood flow with activity.

Manually placing the estradiol softgel vaginal insert into the lower third of the vagina may play a role in its rapid onset of action, providing estrogenization of the tissues in and around the vagina including the vulva and vestibule. Furthermore, capsule placement into the lower third of the vagina, may limit the transfer of estrogen to the uterus.<sup>23</sup> Placement of estrogen products into the upper third of the vagina (such as



**FIG. 2.** Baseline-adjusted mean plasma estradiol (E2) concentrations in two phase 1 studies with administration of a single 25-µg E2 softgel vaginal insert in the same 16 postmenopausal women by body position (supine or ambulatory).

**TABLE 2.** Softgel capsule disintegration state in the vagina on days 1 and 15 in the phase 2 study

	10 µg E2 (n = 24)		Placebo (n = 26)	
	Day 1 <sup>a</sup>	Day 15 <sup>b</sup>	Day 1 <sup>a</sup>	Day 15 <sup>b</sup>
No evidence of capsule present	23 (96)	24 (100)	26 (100)	24 (92)
Evidence of capsule present	0	0	0	0
Assessment not done	1 (4)	0	0	2 (8)

Data presented as n (%).

E2, estradiol.

<sup>a</sup>Assessment 6 hours after insertion.

<sup>b</sup>Assessment 24 hours after insertion.

**TABLE 3.** Adverse events coded by the MedDRA System of the reproductive system and breast disorders in the phase 2 study

Treatment	Adverse event	n	Relation to study drug	Severity
Placebo (n = 24)	Vaginal hemorrhage	2	Not related	Mild
	Vulvovaginal discomfort	1	Not related	Moderate
TX-004HR (n = 24)	Cervical dysplasia	1	Possible	Mild
	Hot flushes	2	Not related (n = 1), definite (n = 1)	Mild
	Vaginal discharge	1	Possible	Mild
	Vaginal dysplasia	3	Possible	Mild
	Vulvovaginal burning sensation	1	Possible	Mild
	Vulvovaginal pain	1	Possible	Mild
	Vulvovaginal pruritus	1	Possible	Mild

MedDRA, Medical Dictionary for Regulatory Activities.

products placed with an applicator) may allow for the distribution of estrogen to the uterus via a “first uterine pass effect” through the close proximity of veins and arteries,<sup>23,24</sup> potentially increasing the risk of endometrial effects of estrogens such as hyperplasia or cancer.<sup>25</sup>

Using the estradiol softgel vaginal inserts may address some of the inconveniences women have reported with other vaginal estrogens. In the REVIVE (Real Women’s Views of Treatment Options for Menopausal Vaginal ChangEs) survey, US postmenopausal women with VVA and/or symptoms of VVA reported being concerned about the inconvenience and messiness of vaginal products prescribed for treating VVA and its symptoms.<sup>7</sup> Another US survey reported similar results, with women reporting messiness, not having time needed for the application, or the application process being generally unpleasant.<sup>26</sup> Most Food and Drug Administration–approved treatments for VVA, including Vagifem (E2 vaginal insert), Intrarosa (prasterone vaginal insert; AMAG Pharmaceuticals Inc, Waltham, MA), Estrace (E2 vaginal cream; Allergan USA Inc, Madison, NJ), and Premarin (conjugated estrogens cream; Wyeth Pharmaceuticals Inc, a subsidiary of Pfizer, Philadelphia, PA), require an applicator for dosing, unlike Imvexxy.<sup>13–16</sup> In contrast, a study of patient satisfaction with the softgel vaginal insert reported that more than 85% of women found it easy to use, and significantly more users of all E2 doses (4, 10, and 25 µg) were “satisfied” or “very satisfied” compared with placebo and preferred the softgel product over their previously used treatments.<sup>27</sup>

Overall, only one case (2.1%, 1/48) of vaginal discharge possibly related to study drug was reported in the phase 2 capsule assessment study. A similar vaginal discharge rate (3.7%; 28/764) was observed in the phase 3 REJOICE trial, although a higher rate for vaginal discharge was observed in the placebo group (6.8%, 13/192) than in the TX-004HR groups (4 µg, 2.6% [5/191], 10 µg, 3.1% [6/191], and 25 µg, 2.1% [4/190]).<sup>11</sup> Similar to the pilot study, most AEs in the REJOICE trial were mild to moderate, and no participants discontinued because of vaginal discharge AEs.

## CONCLUSIONS

These data show rapid *in vitro* dissolution of the study drug, comparable E2 PK parameters regardless of patient positioning after dosing, and complete disappearance of the softgel capsule by 6 hours after administration. In conjunction with phase 3

data showing significant improvements in dyspareunia, vaginal dryness, and objective measures of vaginal atrophy (vaginal pH, percentage of superficial and parabasal cells),<sup>11</sup> our results suggest that the physical characteristics of the softgel capsule reported here may help overcome some of the drawbacks of currently available vaginal estrogen products.

**Acknowledgments:** The authors acknowledge the medical writing assistance provided by Laura J. Ninger, ELS and Dominique J. Verlaan, PhD of Precise Publications, LLC (Bedminster, NJ).

## REFERENCES

- MacBride MB, Rhodes DJ, Shuster LT. Vulvovaginal atrophy. *Mayo Clin Proc* 2010;85:87-94.
- Gass ML, Cochrane BB, Larson JC, et al. Patterns and predictors of sexual activity among women in the Hormone Therapy trials of the Women’s Health Initiative. *Menopause* 2011;18:1160-1171.
- Santoro N, Komi J. Prevalence and impact of vaginal symptoms among postmenopausal women. *J Sex Med* 2009;6:2133-2142.
- Dennerstein L, Dudley EC, Hopper JL, Guthrie JR, Burger HG. A prospective population-based study of menopausal symptoms. *Obstet Gynecol* 2000;96:351-358.
- Nappi RE, Kokot-Kierepa M. Women’s voices in the menopause: results from an international survey on vaginal atrophy. *Maturitas* 2010;67:233-238.
- Krychman M, Graham S, Bernick B, Mirkin S, Kingsberg SA. The Women’s EMPOWER survey: women’s knowledge and awareness of treatment options for vulvar and vaginal atrophy remains inadequate. *J Sex Med* 2017;14:425-433.
- 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.
- Nappi RE, Palacios S, Panay N, Particco M, Krychman ML. Vulvar and vaginal atrophy in four European countries: evidence from the European REVIVE Survey. *Climacteric* 2016;19:188-197.
- Kingsberg S, Krychman M, Graham S, Bernick B, Mirkin S. The Women’s EMPOWER Survey: Identifying women’s perceptions on vulvar and vaginal atrophy (VVA) and its treatment. *J Sex Med* 2017;14:413-424.
- Imvexxy (estradiol vaginal inserts) Prescribing Information. Boca Raton, FL: TherapeuticsMD; 2018.
- Constantine G, 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.
- Archer DF, Constantine GD, Simon J, et al. TX-004HR vaginal estradiol has negligible to very low systemic absorption of estradiol. *Menopause* 2017;24:510-516.
- Vagifem (estradiol vaginal tablets) Prescribing Information. Plainsboro, NJ: Novo Nordisk Pharmaceuticals Inc; 2017.
- Intrarosa (prasterone insert) Prescribing Information. Waltham, MA: AMAG Pharmaceuticals; 2018.

15. Estrace (Cream (estradiol vaginal cream, USP, 0.01%) Prescribing Information. Madison, NJ: Allergan USA, Inc; 2018.
16. Premarin (conjugated estrogens) Vaginal Cream Prescribing Information. Philadelphia, PA: Wyeth Pharmaceuticals Inc; 2018.
17. Constantine GD, Bouchard C, Pickar JH, et al. Consistency of effect with a low-dose, estradiol vaginal capsule (TX-004HR): evaluating improvement in vaginal physiology and moderate-to-severe dyspareunia in subgroups of postmenopausal women. *J Womens Health (Larchmt)* 2017;26:616-623.
18. Simon JA, Archer DF, Kagan R, et al. Visual improvements in vaginal mucosa correlate with symptoms of VVA: data from a double-blind, placebo-controlled trial. *Menopause* 2017;24:1003-1010.
19. Simon JA, Kagan R, Archer DF, et al. TX-004HR clinically improves symptoms of vulvar and vaginal atrophy in postmenopausal women. *Climacteric* 2019;22:412-418.
20. Shah M. Drug Dissolution Apparatus III USP (Reciprocating Cylinder) March 2011: Available at: <http://www.pharmatips.in/Articles/Pharmaceutics/Tablet/Drug-Dissolution-Apparatus-III-USP-Reciprocating-Cylinder.aspx>.
21. Pickar JH, Amadio JM, Bernick BA, Mirkin S. Pharmacokinetic studies of solubilized estradiol given vaginally in a novel softgel capsule. *Climacteric* 2016;19:181-187.
22. Pickar JH, Amadio JM, Hill JM, Bernick BA, Mirkin S. A randomized, double-blind, placebo-controlled phase 2 pilot trial evaluating a novel, vaginal softgel capsule containing solubilized estradiol. *Menopause* 2016;23:506-510.
23. Cicinelli E, Di NE, De ZD, et al. Placement of the vaginal 17beta-estradiol tablets in the inner or outer one third of the vagina affects the preferential delivery of 17beta-estradiol toward the uterus or periurethral areas, thereby modifying efficacy and endometrial safety. *Am J Obstet Gynecol* 2003;189:55-58.
24. Cicinelli E, De Ziegler D, Morgese S, Bulletti C, Luisi D, Schonauer LM. First uterine pass effect'' is observed when estradiol is placed in the upper but not lower third of the vagina. *Fertil Steril* 2004;81:1414-1416.
25. Constantine G, Graham S, Lapane K, et al. Endometrial safety of low-dose vaginal estrogens in menopausal women: a systematic review of the evidence. *Menopause* 2019;26:800-807.
26. Minkin MJ, Maamari R, Reiter S. Postmenopausal vaginal atrophy: evaluation of treatment with local estrogen therapy. *Int J Womens Health* 2014;6:281-288.
27. Kingsberg S, Kroll R, Goldstein I, et al. Patient acceptability and satisfaction with a low-dose solubilized vaginal estradiol softgel capsule, TX-004HR. *Menopause* 2017;24:894-899.