Development of an In Vitro Release Testing Method for Vaginal Creams

Weizhou Yue[1], Lingxiao Xie[1], Megan Kelchen[2], Priyanka Ghosh[2], Mengmeng Niu[2], Sam G. Raney[2], Jie Shen[1,3]

[1]University of Rhode Island, College of Pharmacy, Kingston, Rhode Island 02881
[2]Division of Therapeutic Performance I, Office of Research and Standards, Office of Generic Drugs, Center for Drug Evaluation and Research, U.S. Food and Drug Administration, Silver Spring, Maryland 20993
[3]University of Rhode Island, College of Engineering, Kingston, Rhode Island 02881

PURPOSE

Vaginal cream products account for more than 50% of the FDA-approved semi-solid dosage forms that are administered through the vaginal route. Currently, bioequivalence (BE) of vaginal creams is often established based on in vivo BE studies with comparative clinical and/or pharmacokinetic endpoints. The development of characterization-based BE approaches for vaginal creams that include formulation sameness, comparative physicochemical and structural characterization, as well as performance for the test and reference products, may mitigate the risks associated with potential failure modes for BE, ultimately facilitating patient access to generic vaginal cream products. The main objective of the present study was to develop a reproducible, validated, and discriminatory in vitro release test (IVRT) method for clindamycin phosphate (CP) vaginal creams that can facilitate a comparison of release rates between a reference product and a prospective generic drug product, as part of a characterization-based BE approach.

METHODS

CP vaginal creams were chosen as model drug products in the present research. The marketed CP vaginal cream (2% w/w, M1) was selected as the reference product. Laboratory-made CP vaginal creams composed of the same components as those used in M1 were prepared using a sweep blade mixer followed by homogenization. The prepared creams were sealed in an aluminum container after a cooling process. The physicochemical properties (e.g., pH, droplet size, and rheological properties) of the CP creams were characterized. IVRT parameters such as selection of receptor solution and artificial membrane were investigated. Following the initial IVRT method development, simulated vaginal fluid (pH 4.2) containing 3% (w/w) Brij® O20 was selected as the receptor solution and a polyethersulfone (PES) membrane (0.45 µm) was chosen as the membrane. IVRT studies of CP creams were conducted using a vertical diffusion cell (VDC) apparatus maintained at 37°C (membrane surface temperature) following application of 750 mg of the cream to the membrane. Receptor solution samples were taken at pre-determined time points and analyzed using a validated high performance liquid chromatography (HPLC) method. Reproducibility of the IVRT method was evaluated using the reference product M1 (n=6 cells/run, 3 independent runs). The discriminatory ability of the method was evaluated using laboratory-prepared CP vaginal creams at different strengths (50%, 100%, and 150% nominal strength; n=3 cells/strength).

RESULTS

- *Droplet size analysis: Brightfield upright microscopy*

Table 1. Droplet size analysis results.

	Count	Droplet Diameter (µm)			Span
		D10	D50	D90	
M1	1522	1.96	3.23	4.79	0.87
Lab-made (100% strength)	2635	2.16	4.04	6.23	1.01

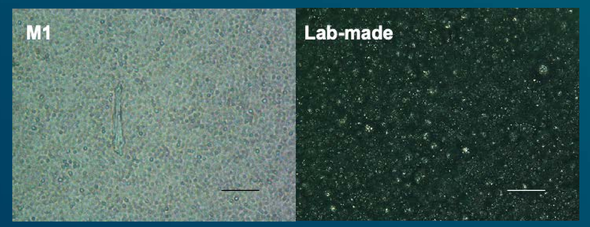

Figure 1. Representative microscopy images of CP vaginal cream. Scale bar=25 µm.

- *IVRT study conditions*
  - Apparatus: Vertical Franz diffusion cell (VDC)
  - Contact area: 1.77cm²
  - PES membrane (0.45 µm)
  - Membrane temperature: 37°C
  - Receptor solution: Simulated vaginal fluid containing 3% BrijO20 (pH 4.2)
  - Dose : 750 mg CP vaginal creams

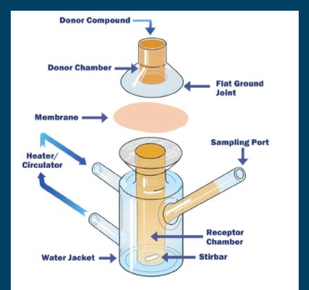

Image from PermeGear website (permegear.com/franz-cells/)

➢ *Physicochemical properties of CP vaginal creams*

- *Drug content and pH value of CP vaginal creams*

Table 2. Drug content and pH values of the marketed (M1) and laboratory (Lab)-made CP vaginal creams (mean ± SD, *n*=3).

Formulations	Drug Content (%)	pH Room Temp. (21°C)	pH 37°C
M1	2.04 ± 0.020	3.75 ± 0.02	3.66 ± 0.03
Lab-made (50% strength)	1.07 ± 0.006	4.09 ± 0.03	4.15 ± 0.02
Lab-made (100% strength)	2.02 ± 0.006	3.68 ± 0.14	3.96 ± 0.02
Lab-made (150% strength)	3.12 ± 0.013	3.53 ± 0.01	3.76 ± 0.02

- *Rheological properties*

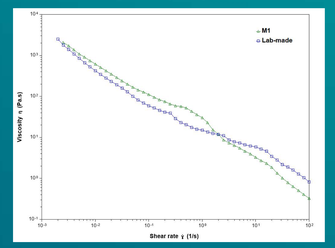

Figure 2. Flow sweep of the marketed product M1 and laboratory-made cream (100% strength) characterized by rheometer. All CP creams showed shear thinning behavior.

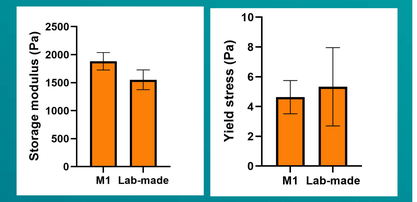

Figure 3. Average storage modulus and yield stress of marketed (M1) and laboratory made (100% strength) CP vaginal creams (mean ± SD, n=3).

Table 3. Average viscosities of CP vaginal creams (mean ± SD, *n*=3).

	Viscosity (Pa.s)		
	0.005 (1/s)	1 (1/s)	50 (1/s)
M1	890.42 ± 161.71	26.15 ± 4.62	0.62 ± 0.01
Lab-made (100 strength%)	688.39 ± 163.86	13.53 ± 1.50	1.57 ± 0.04

➢ *In vitro release test (IVRT) method development*

- *Reproducibility study (IVRT method)*

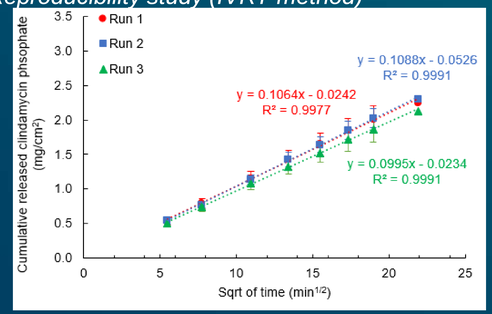

Figure 4. Mean (± SD) in vitro release profiles of M1 obtained using the developed VDC method at 37°C (three runs, *n*=6 per run).

Table 4. IVRT release rate of marketed (M1) vaginal creams analyzed using the Higuchi model (8-hr) (mean ± SD, n=6 per run). Good reproducibility with inter- and intra-day CV%<15%.

	Run 1	Run 2	Run 3	Mean
Release rate (8-hr) (mg/cm²/min^1/2)	0.106±0.007	0.109±0.011	0.0996±0.011	0.105±0.005
CV (%)	7.08	10.28	10.59	9.32
Drug depletion (%)	27.62±1.37	27.40±2.62	25.36±2.55	26.79±1.25
CV (%)	4.94	9.57	10.06	4.66

- *Discriminatory ability study of VDC method*

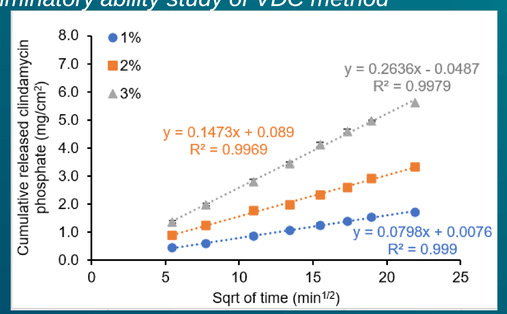

Figure 5. Mean (± SD) in vitro release profiles of three laboratory-made CP vaginal creams of different nominal strengths (*n*=3 per strength).

Table 5. IVRT release rates of laboratory-made creams with different strengths analyzed using the Higuchi model (8-hr) (mean ± SD, *n*=3 per strength). Observed differences in the release rate between the 100% strength M1 and laboratory-made creams products may be potentially attributable to differences in physicochemical properties between the two products.

Sample	Release rate (mg/cm²/min^1/2)
50% strength	0.080±0.005
100% strength	0.147±0.001
150% strength	0.264±0.003

CONCLUSIONS

An IVRT study that can demonstrate equivalent rates of release of an active ingredient from a prospective generic vaginal cream product compared to a reference product may be a relevant test that can be utilized to mitigate risks associated with potential failure modes for BE. The IVRT method using the VDC apparatus demonstrated good reproducibility in evaluating the release rates of CP from vaginal creams, and based on the preliminary data, the IVRT method appears to have good discrimination, illustrating the feasibility of using such an IVRT method for vaginal creams. Additional research is planned to evaluate the release of CP from the marketed and laboratory-prepared formulations using the developed IVRT method.

ACKNOWLEDGEMENT

This project is supported by the Food and Drug Administration (FDA) of the U.S. Department of Health and Human Services (HHS) as part of a grant (U01FD006721) totaling $500,000 with 100 percent funded by FDA/HHS. The contents are those of the author(s) and do not necessarily represent the official views of, nor an endorsement, by FDA/HHS, or the U.S. Government.

CONTACT INFORMATION: Dr. Jie Shen (jie_shen@uri.edu)
University of Rhode Island