MTN-027

Phase 1 Safety and Pharmacokinetics Study of MK-2048/Vicriviroc (MK-4176)/MK-2048A Intravaginal Rings

Microbicide Trials Network

Funding Agencies:
Division of AIDS, US National Institute of Allergy and Infectious Diseases
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Protocol Chair:
Craig Hoesley, MD

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LIST OF ABBREVIATIONS AND ACRONYMS

AIDS	Acquired Immunodeficiency Syndrome
ALT	alanine transaminase
API	Active pharmaceutical agent
ART	antiretroviral therapy
ARV	antiretroviral
ASM	American Society for Microbiology
AST	aspartate aminotransferase
ASTM	American Society for Testing and Materials
AUC	area under the curve
BID	bis in die, twice a day
BRWG	Behavioral Research Working Group
CASI	computer assisted self-interview
CBC	complete blood count
CDC	Centers for Disease Control and Prevention
CFR	Code of Federal Regulations
CHC	combination hormonal contraceptive
C_{max}	maximum concentration
CMRB	Clinical Microbicide Research Branch
CNS	Central nerve system
CPX	Ciprofloxacin
CRF	case report form
CTA	Clinical Trial Agreement
CVF	cervical vaginal fluid
CWG	Community Working Group
DAERS	DAIDS Adverse Event Reporting System
DAIDS	Division of AIDS
DNA	deoxyribonucleic acid
EAE	expedited adverse event
EEG	Electroencephalography
ENR	Enrollment
EVA	ethylene vinyl acetate
FDA	(US) Food and Drug Administration
FHCRC	Fred Hutchinson Cancer Research Center
FRCP	Federal Rules of Civil Procedure
GCP	Good Clinical Practices
GEE	generalized estimating equations
GLP	Good laboratory practice
GMP	Good Manufacturing Practices
HBV	Hepatitis B virus
HCV	Hepatitis C virus
HEENT	Head, Eye, Ear, Nose and Throat

HHS	Health and Human Services
HIV	Human Immunodeficiency Virus
HSV	Herpes simplex virus
hu-PBL	human peripheral blood lymphocytes
IATA	International Air Transport Association
ICF	informed consent forms
ICRC	International Committee of the Red Cross
IDI	in-depth interviews
IND	Investigational New Drug
INR	International normalized ratio
IRB	Institutional Review Board
IUD	intrauterine device
IVR	Intravaginal Ring
KOH	potassium hydroxide
LDMS	Laboratory Data Management System
LLOQ	lower limit of quantification
LOC	Leadership and Coordinating Center
MPI	Maximum calculated percent inhibition
MSD	Merck Sharp & Dohme
MTN	Microbicide Trials Network
NAAT	nucleic acid amplification test
NGR	next generation rings
NIAID	National Institute of Allergy and Infectious Diseases
NICHD	*Eunice Kennedy Shriver* National Institute of Child Health and Human Development
NIH	National Institutes of Health
NIMH	National Institute of Mental Health
NOAEL	no-observed-adverse-effect-level
OBT	Optimized Background Therapy
OHRP	Office for Human Research Protections
OSS	Overall susceptibility score
PBMC	Peripheral blood mononuclear cells
PEP	post-exposure prophylaxis
PK	pharmacokinetics
PPD	Pharmaceutical Product Development
PrEP	Pre-Exposure Prophylaxis
PRO	Protocol Registration Office
PSRT	Protocol Safety Review Team
PTID	participant identification
RE	Regulatory Entity
RNA	Ribonucleic acid
RSC	Regulatory Support Center
RTI	reproductive tract infection
SAE	serious adverse event
SCR	Screening
SDMC	Statistical Data Management Center
SMC	Study Monitoring Committee
SSP	study specific procedures

STI	sexually transmitted infection
UPMC	University of Pittsburgh Medical Center
USA	United States of America
UTI	urinary tract infection
VCV	Vicriviroc
WHO	World Health Organization

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PROTOCOL TEAM ROSTER

Protocol Chair

Craig Hoesley, MD
Protocol Chair
University of Alabama at Birmingham
1530 3rd Avenue South, VH 201
Birmingham, AL 35294 USA
Phone: 205-934-3365
Fax: 205-975-3232
Email: choesley@uab.edu

Site Investigators

Beatrice A. Chen, MD, MPH
Site Investigator
Magee-Womens Hospital of UPMC
300 Halket Street
Pittsburgh, PA 15213 USA
Phone: 412-641-1403
Fax: 412-641-1133
Email: chenba@upmc.edu

Craig Hoesley, MD
Protocol Chair/Site Investigator
University of Alabama at Birmingham
1530 3rd Avenue South, VH 201
Birmingham, AL 35294 USA
Phone: 205-934-3365
Fax: 205-975-3232
Email: choesley@uab.edu

US National Institutes of Health (NIH)

Roberta Black, PhD
Chief, Clinical Microbicide Research Branch
Division of AIDS (DAIDS), National Institute of Allergy and Infectious Diseases (NIAID),
National Institutes of Health (NIH) - U.S. Department of Health and Human Services (HHS)
5601 Fishers Lane, Room 8B62, MSC 9831
Rockville, MD 20892-9831 USA
Phone: 301-496-8199
Email: rblack@niaid.nih.gov

Naana Cleland, MHCA
Health Specialist, Clinical Microbicide Research Branch (CMRB)
Prevention Sciences Program (PSP) DAIDS, NIAID
NIH- U.S. Department of Health and Human Services (HHS)
5601 Fishers Lane, Room 8B27
Bethesda, MD 20892-9830 USA
Phone: 240-292-4779
Email: clelandn@niaid.nih.gov

Cynthia Grossman, PhD
Chief, HIV Care Engagement and Secondary Prevention Program,
National Institute of Mental Health (NIMH)
5601 Fishers Lane, Room 9G19, MSC 9831
Bethesda, MD 20892 USA
Phone: 240-627-3868
Email: grossmanc@mail.nih.gov

Jeanna Piper, MD
DAIDS Senior Medical Officer
DAIDS/NIAID
5601 Fishers Lane
Rockville, MD 20892-9831 USA
Phone: 240-292-4798
Email: piperj@niaid.nih.gov

Dianne M. Rausch, PhD
Director
DAIDS Research, NIMH
5601 Fishers Lane, Room 8D20, MSC 9831
Bethesda, MD 20892 USA
Phone: 240-627-3874
Fax: 240-627-3467
Email: drausch@mail.nih.gov

MTN Leadership and Coordinating Center (LOC) - Pitt

Katherine Bunge, MD
Protocol Safety Physician
Magee-Womens Hospital of UPMC
300 Halket Street
Pittsburgh, PA 15213 USA
Phone: 412-641-3464
Fax: 412-641-1133
Email: kbunge@mail.magee.edu

Cindy Jacobson, PharmD
Director of Pharmacy Affairs
Microbicide Trials Network
204 Craft Avenue
Pittsburgh, PA 15213 USA
Phone: 412-641-8913
Fax: 412-641-6170
Email: cjacobson@mail.magee.edu

Beth Galaska Burzuk, MID
Protocol Development Manager
Microbicide Trials Network
204 Craft Avenue
Pittsburgh, PA 15213 USA
Phone: 412-641-5579
Fax: 412-641-6170
Email: galaskaburzukb@upmc.edu

Ian McGowan, MBChB, MD, DPhil, FRCP
Co-Principal Investigator
Microbicide Trials Network
204 Craft Avenue
Pittsburgh, PA 15213 USA
Phone: 412-641-8999
Fax: 412-641-6170
Email: imcgowan@pitt.edu

Sharon Hillier, PhD
Co-Principal Investigator
Microbicide Trials Network
204 Craft Avenue
Pittsburgh, PA 15213 USA
Phone: 412-641-8933
Fax: 412-641-6170
Email:shillier@mail.magee.edu

Sharon A. Riddler, MD, MPH
Protocol Physician
UPMC, Keystone Building, Suite 510
3520 Fifth Avenue
Pittsburgh, PA 15213 USA
Phone: 412-383-1741or 412-383-1675
Fax: 412-383-2900
Email: riddler@dom.pitt.edu

Ken Ho, MD
Protocol Safety Physician
UPMC, Keystone Building, Suite 533
3520 Fifth Avenue
Pittsburgh, PA 15213 USA
Phone: 412-383-7178
Fax: 412-383-2900
Email: hok2@upmc.edu

Devika Singh, MD, MPH
Protocol Safety Physician
Box 359927, Dpt. of Global Health
ICRC, 325 Ninth Ave.
Seattle, WA 98104 USA
Phone: 206-744-8311
Fax: 206-520-3831
Email: dsingh@u.washington.edu

MTN Laboratory Center (LC)

Peter Anderson, PharmD
LC Pharmacology Core
University of Colorado School of Pharmacy
Mail Stop C238
12850 E. Montview Blvd. V20-4101
Aurora, CO 80045 USA
Phone: 303-724-6128
Email: Peter.Anderson@ucdenver.edu

Charlene S. Dezzutti, PhD
LC Director
Microbicide Trials Network
204 Craft Avenue
Pittsburgh, PA 15213 USA
Phone: 412-641-3462
Fax: 412-641-6170
Email: dezzuttics@upmc.edu

Wayne Hall, MT, ASCP
Clinical Laboratory Representative
Microbicide Trials Network
204 Craft Ave. Room A534
Pittsburgh, PA 15213 USA
Phone: 412-641-6956
Fax: 412-641-6170
Email: hallwb@mwri.magee.edu

Ratiya Pamela Kunjara Na Ayudhya, MT, ASCP
Laboratory Manager
Microbicide Trials Network
204 Craft Avenue, Room A541
Pittsburgh, PA 15213 USA
Phone: 412-641-6393
Fax: 412-641-6170
Email: pkunjara@mwri.magee.edu

MTN LOC – FHI 360

Ashley Mayo, MPH
Clinical Research Manager
359 Blackwell St., Suite 200
PO Box 21059
Durham, NC 27701 USA
Phone: 919-544-7040 Ext. 11164
Fax: 919-544-0904
Email: amayo@fhi360.org

Jonathan Paul Lucas, MPH
Senior Community Program Associate
359 Blackwell St., Suite 200
Durham, NC 27701 USA
Phone: 919.544.7040 Ext. 11458
Fax: 919.544.7261
Email: jlucas@fhi360.org

Sherri Johnson, MPH
Clinical Research Manager
FHI 360
1825 Connecticut Avenue, NW
Washington, DC 20009 USA
Phone: 202-884-8677
Fax: 202-884-8844
Email: sjohnson@fhi360.org

MTN Statistical Data Management Center (SDMC)

Jennifer Berthiaume, MSW, MPH
Project Manager
FHCRC-SCHARP
1100 Fairview Avenue North, E3-129
PO Box 19024
Seattle, WA 98109-1024 USA
Phone: 206-667-1230
Fax: 206-667-4812
Email: jberthia@scharp.org

Barbra Richardson, PhD
Faculty Statistician
FHCRC-SCHARP
1100 Fairview Avenue North
PO Box 19024
Seattle, WA 98109-1024 USA
Phone: 206-667-7788
Fax: 206-667-4812
Email: barbrar@uw.edu

MTN Behavioral Research Working Group (BRWG)

Alex Carballo-Diéguez, PhD
BRWG Representative
HIV Center for Clinical and Behavioral Studies
NYSPI & Columbia University
1051 Riverside Dr., Unit 15
New York, NY 10032 USA
Phone: 212-543-5261
Fax: 212-543-6003
Email: ac72@columbia.edu

Jose A. Bauermeister, MPH, PhD
BRWG Representative
John G. Searle Assistant Professor of Health Behavior and Health Education
Director, The Center for Sexuality & Health Disparities (SexLab)
University of Michigan School of Public Health
1415 Washington Heights, SPH I, Room 3822
Ann Arbor, MI 48109-2029
Phone: (734) 615-8414
Email: jbauerme@umich.edu

MTN Community Working Group (CWG) Representatives

Renee Weinman, BS, CCRP
MTN CWG Representative
University of Pittsburgh
Falk Medical Bldg. 7th floor
3601 Fifth Avenue
Pittsburgh, Pa 15213
Phone: 412-647-0322
Email: weinmandr@upmc.edu

MTN-027

Phase 1 Safety and Pharmacokinetics Study of MK-2048/Vicriviroc (MK-4176)/MK-2048A Intravaginal Rings

INVESTIGATOR SIGNATURE FORM

Version 1.0

December 20, 2014

A Study of the Microbicide Trials Network

Funded by:
Division of AIDS, US National Institute of Allergy and Infectious Diseases
US *Eunice Kennedy Shriver* National Institute of Child Health and Human Development
US National Institute of Mental Health
US National Institutes of Health

IND Holder:
DAIDS

I, the Investigator of Record, agree to conduct this study in full accordance with the provisions of this protocol. I will comply with all requirements regarding the obligations of investigators as outlined in the Statement of Investigator (Form FDA 1572), which I have also signed. I agree to maintain all study documentation for at least two years following the date of marketing approval for the study product for the indication in which it was studied. If no marketing application is filed, or if the application is not approved, the records will be retained for two years after the investigation is discontinued and the US Food and Drug Administration is notified. Publication of the results of this study will be governed by MTN policies. Any presentation, abstract, or manuscript will be submitted to the MTN Manuscript Review Committee, DAIDS, Merck & Co. and other entities for review prior to submission, as required by the MTN Publication Policy.

I have read and understand the information in the Investigator's Brochure(s), including the potential risks and side effects of the products under investigation, and will ensure that all associates, colleagues, and employees assisting in the conduct of the study are informed about the obligations incurred by their contribution to the study.

_____
Name of Investigator of Record

_____ _____
Signature of Investigator of Record Date

MTN-027

Phase 1 Safety and Pharmacokinetics Study of MK-2048/Vicriviroc (MK-4176)/MK-2048A Intravaginal Rings

PROTOCOL SUMMARY

Short Title:	Safety and PK of IVRs Containing VCV (MK-4176) and/or MK-2048
Clinical Phase:	Phase 1
IND Sponsor:	DAIDS
Protocol Chair:	Craig Hoesley, MD
Sample Size:	Approximately 48 women
Study Population:	Healthy, HIV-uninfected, sexually abstinent women between the ages of 18-45 (inclusive)
Study Sites:	US sites selected by the MTN Executive Committee
Study Design:	Multi-site, single-blind, four-arm, randomized, placebo-controlled trial
Study Duration:	Approximately 5 weeks per participant, with approximately 6-9 months for planned accrual at each site
Study Products:	Vicriviroc (MK-4176) Intravaginal Ring (IVR) MK-2048 IVR MK-2048A (Vicriviroc [MK-4176] + MK-2048) IVR Placebo IVR
Study Regimen:	Participants will be randomized to study IVR assignment in a 1:1:1:1 ratio. Participants will insert one IVR to be used for a period of approximately 28 days, followed by approximately 7 days of no study product use.

Primary Objectives:

- Assess and compare the safety of ethylene-vinyl acetate IVRs containing 182 mg vicriviroc (MK-4176), or 30 mg MK-2048, or 182 mg vicriviroc (MK-4176) + 30 mg MK-2048, when used continuously for 28 days by healthy, HIV-uninfected, sexually abstinent women, as compared with the placebo IVR

- Examine local and systemic pharmacokinetics of vicriviroc (MK-4176) and MK-2048 in vaginal fluid, plasma and cervical tissue during and after 28 days continuous use of an IVR containing 182 mg vicriviroc (MK-4176), or 30 mg MK-2048, or 182 mg vicriviroc (MK-4176) + 30 mg MK-2048

Primary Endpoints:

- The primary safety endpoints are the proportion of women in each of the four IVR regimens (182 mg vicriviroc (MK-4176), 30 mg MK-2048, 182 mg vicriviroc (MK-4176) + 30 mg MK-2048, or placebo) with:
  - Genitourinary events Grade 1 or higher as defined by the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Version 1.0, Dec 2004 (Clarification dated August 2009), Addendum 1, (Female Genital Grading Table for Use in Microbicide Studies) judged to be related to study product
  - Adverse events Grade 2 or higher as defined by the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Version 1.0, Dec 2004 (Clarification dated August 2009)

- The pharmacokinetic endpoints are:
  - Local and systemic concentrations of vicriviroc (MK-4176) and MK-2048 in plasma, vaginal fluids and cervical tissue during and after 28 days of continuous use of an IVR containing 182 mg vicriviroc (MK-4176), or 30 mg MK-2048, or 182 mg vicriviroc (MK-4176) + 30 mg MK-2048 (MK-2048A)

Secondary Objectives:

- Evaluate the acceptability of the study IVR in HIV-uninfected sexually abstinent women over 28 days of use

- Evaluate the adherence to the study IVR in HIV-uninfected sexually abstinent women over 28 days of use

Secondary Endpoints:

- Acceptability
  - Participant report of acceptability including genitourinary and emotional (dis)comfort, awareness/feeling during daily activities, ring insertion/removal issues, and willingness to use in the future

- Adherence
  - Participant report of frequency of study IVR removal/expulsion and duration without IVR inserted in vagina

Exploratory Objectives:

- Evaluate the HIV inhibitory activity of cervical tissue after ring use

- Assess drug levels in rectal fluid

- Describe the genital microenvironment during 28 days of study product use and follow-up

- Evaluate the potential relationship between participant self-report of adherence, vicriviroc (MK-4176) and MK-2048 remnant content in returned IVRs, and PK levels

Exploratory Endpoints:

- HIV-1 Inhibitory Activity:
  - Measures of HIV-1 inhibition within cervical tissue after ring use

- Drug Concentration in Rectal Fluid:
  - Local concentrations of vicriviroc (MK-4176) and MK-2048 in rectal fluid

- Genital Microenvironment:
  - Abnormal vaginal flora as assessed by Gram stains
  - Presence of candidate biomarkers of safety and efficacy in mucosal secretions
  - Quantitative vaginal culture

- Evaluation of the Potential Relationship between Various Measures of Adherence:
  - Participant report of duration without IVR inserted in vagina
  - Remnant content of vicriviroc (MK-4176) and MK-2048
  - Local and systemic concentrations of vicriviroc (MK-4176) and MK-2048 in plasma, vaginal fluids and cervical tissue

Figure 1: MTN-027 Study Visit Schedule

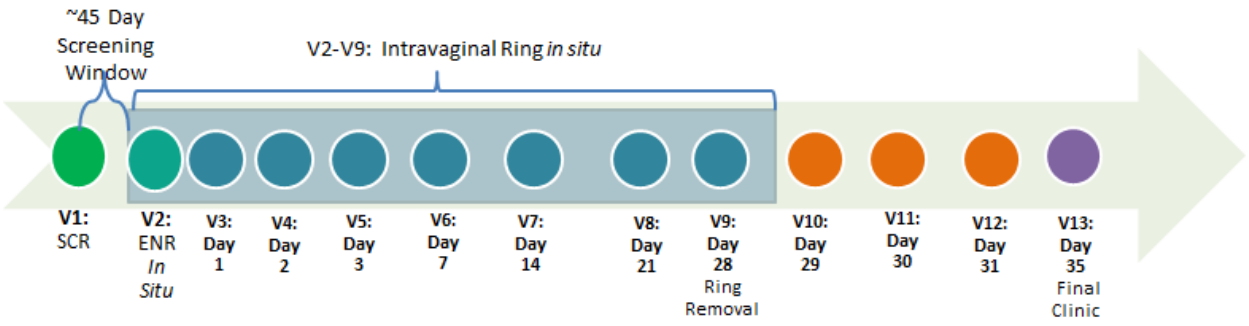


Table 1: PK Specimen Collection Schedule

STUDY VISIT	PK Specimen Collection
Screening	None
Enrollment	Blood (hr 1, 2, 4, 6), Vaginal fluid (hr 0,1, 2, 4, 6)
Day 1	Blood, Vaginal fluid
Day 2	Blood, Vaginal fluid
Day 3	Blood, Vaginal fluid
Day 7	Blood, Vaginal fluid
Day 14	Blood, Vaginal fluid
Day 21	Blood, Vaginal fluid
Day 28	Blood (hr 0, 1, 2, 4, 6), Vaginal fluid (hr 0, 1, 2, 4, 6), Cervical tissue (hr 0), Rectal fluid ♠ (hr 0)
Day 29	Blood, Vaginal fluid
Day 30	Blood, Vaginal fluid
Day 31	Blood, Vaginal fluid
Day 35	Blood, Vaginal fluid

1 KEY ROLES

1.1 Protocol Identification

Protocol Title:	Phase 1 Safety and Pharmacokinetics Study of MK 2048/Vicriviroc (MK-4176)/MK-2048A Intravaginal Rings
Protocol Number:	MTN-027
Short Title:	Safety and PK of IVRs Containing VCV (MK-4176) and/or MK-2048
Date:	December 20, 2014

1.2 Funders, Sponsor and Monitor Identification

Funding Agencies:	US Division of AIDS (DAIDS)/National Institute of Allergy and Infectious Diseases (NIAID) National Institutes of Health (NIH) 5601 Fishers Lane Rockville, MD 20892-9831 USA US National Institute of Mental Health (NIMH) 6001 Executive Boulevard Rockville, MD 20852 USA US *Eunice Kennedy Shriver* National Institute of Child Health and Human Development (NICHD) 6100 Executive Boulevard Bethesda, MD 20892 USA
IND Sponsor:	DAIDS 5601 Fishers Lane Rockville, MD 20892-9831 USA
Monitor:	Pharmaceutical Product Development (PPD), Inc. 929 North Front Street Wilmington, NC 28401-3331 USA
Pharmaceutical Collaborator:	Merck & Co.

1.3 Medical Officer

Medical Officer:	Jeanna Piper, MD DAIDS/NIAID 5601 Fishers Lane Rockville, MD 20892-9831 USA

1.4 Clinical Laboratories

Laboratory Center:	MTN Laboratory Center (LC) 204 Craft Avenue Pittsburgh, PA 15213 USA
Pharmacology:	MTN Pharmacology LC University of Colorado School of Pharmacy V20-4410 12850 E Montview Blvd Aurora, CO 80045 USA

1.5 Data Center

Data Center:	Statistical Center for HIV/AIDS Research & Prevention (SCHARP)/Fred Hutchinson Cancer Research Center (FHCRC) 1100 Fairview Avenue N., LE-400 PO Box 19024 Seattle, WA 98109-1024 USA
Qualitative Data Center:	RTI International 351 California Street, Suite 500 San Francisco, CA 94104 USA

1.6 Study Implementation

Study Implementation:	FHI 360 359 Blackwell Street, Suite 200 PO Box 21059 Durham, NC 27701 USA

2 INTRODUCTION

2.1 Human Immunodeficiency Virus (HIV) Prevention and Intravaginal Rings

The HIV/AIDS epidemic remains one of the world's most significant public health challenges. With approximately 2.3 million individuals newly infected each year, the development of safe and efficacious coitally-dependent and coitally-independent HIV prevention strategies remains a global health priority.[1] Extensive research efforts to develop novel vaginal and rectal antiretroviral (ARV) formulations and delivery systems are ongoing.[2] Undeniably linked to establishing the safety and efficacy of products in development is participant adherence to drug regimen. As products advance to late stage clinical trial development, the critical importance of participant adherence is evident. Across several large Phase 3 pre-exposure prophylaxis (PrEP) clinical trials, moderate to high participant adherence coupled with a highly effective ARV products led to the extension of Truvada®'s licensure to include PrEP,[3, 4] However, in trials testing the same drug, due to low participant adherence no efficacy was demonstrated.[5, 6]

Sustained drug delivery devices, such as IVRs, may offer an ideal method of drug delivery. IVRs are discreet, coitally independent, offer continuous delivery of drug and are acceptable.[7] IVRs have previously been developed and approved as delivery methods for various medications. For example, NuvaRing®, a contraceptive IVR made of ethylene vinyl acetate (EVA) copolymers (with a 28% w/w core and a 9% w/w skin layer) and magnesium stearate, in which 2.7 mg of ethinyl estradiol and 11.7 mg of etonogestrel are dispersed, has been found to be both effective and acceptable to women. In an acceptability study involving 1,950 NuvaRing® users, 45.5% of women cited that their reason for liking the IVR was "not having to remember anything".[8] In a randomized controlled trial testing two alternative delivery systems for combined hormonal contraceptive, women overwhelmingly preferred the IVR to oral contraceptives ($P<.001$).[9] It is likely that products that can be applied less frequently, such as a vaginal ring that is replaced every 28 days, may be more acceptable to users, resulting in higher user-adherence and may lead to increased effectiveness. IVRs that need to be replaced every 28 days may have benefits over dosage forms that need to be used more frequently, as well as offer a wider choice of microbicide formulations for women if proven effective.

2.2 Rationale

MTN-027 is a Phase 1 clinical trial designed to assess the safety and pharmacokinetics of a combination IVR called MK-2048A that contains VCV (MK-4176) and MK-2048, compared to IVRs containing VCV alone and MK-2048 alone. The safety of all three IVRs will be compared to the placebo IVR. The combination IVR is novel as it combines two different classes of ARV agents -- the CCR5-receptor antagonist, VCV (MK-4176), with an integrase inhibitor, MK-2048.

The design of a combination microbicide IVR is based on a strong clinical rationale for combining ARV drugs with different mechanisms of action to increase the breadth of protection and limit the emergence of resistant HIV viral strains. Highly active antiretroviral therapy (ART) and the use of ARV combinations in the prevention of mother-to-child HIV transmission have demonstrated the benefit of ARV combinations in the treatment and prevention of HIV infection. Although the effectiveness of the drug combination planned for this safety trial has not been evaluated clinically, both VCV and MK-2048 are highly potent ARVs.

VCV is a CCR5-receptor antagonist that is potent against CCR5-tropic viruses, which are the predominantly transmitted HIV viral strains, and is active against viruses resistant to other drug classes. Development of resistance to VCV maleate (MK-7690) during Phase 3 trials was uncommon, thereby making it an attractive microbicide candidate. However, VCV is not active against viruses using co-receptors other than CCR5. Two pharmaceutical forms of VCV have been developed: a free-base form, MK-4176, currently used for the development VCV-based microbicide IVRs, and the maleate salt form, MK-7690, previously used for the development of the VCV oral formulation.

MK-2048 is an HIV-1 integrase inhibitor that is highly potent against both wild type HIV-1 and raltegravir-resistant isolates. The combination of two highly potent ARVs not currently being used for HIV-1 treatment may serve as an ideal HIV prevention combination.

It is important to note that data from MTN-027 will be paired with MTN-028. MTN-028 is a Phase 1 safety and pharmacokinetic of two intravaginal rings (IVRs) containing a combination of the active ingredients; VCV (MK-4176) and MK-2048, that have been formulated with different dose strengths: one containing 91 mg of VCV (MK-4176) and 10 mg of MK-2048, and a second, containing 182 mg VCV (MK-4176) 30 mg MK-2048 . The study will enroll approximately 18 healthy, 18-45 year old HIV-uninfected, non-pregnant, sexually abstinent women who are using adequate contraception. Women will be randomized to one of two study regimens in a 2:1 ratio (low dose formulation to standard dose formulation). The IVR will be worn for approximately 28 consecutive days. Based on in vitro, in vivo, and ex vivo studies, VCV (MK-4176) and MK-2048 show promise as topically- applied microbicides. The safety and acceptability of these agents alone and in combination will be evaluated in the MTN-027 trial; however the optimal dose of MK-4176 and MK-2048 to achieve sufficient vaginal fluid concentrations for antiviral activity is unknown. Two different formulations of the MK-2048A combination IVR have been developed and will be evaluated in MTN-028 in an effort to inform in vitro and in vivo modeling to further optimize the drug release profiles of an IVR containing VCV and MK-2048 for use in future studies, including the potential development of a combination antiretroviral/contraceptive ring.

2.3 Vicriviroc (MK-4176) IVR

2.3.1 Description

VCV (MK-4176) is the VCV free-base formulation used for development of VCV as a microbicide administered in an IVR. VCV (MK-4176) is chemically described as 5-({4-[(3*S*)-4-{2-methoxy-1-[4-(trifluoromethyl)phenyl]ethyl}-3-methylpiperazin-1-yl]-4-methylpiperidin-1-yl}carbonyl)-4,6-dimethylpyrimidine. Further information is provided in the MK-2048A Investigator Brochure (IB).[10] The maleate salt formulation of VCV, known as MK-7690, represents the oral formulation of VCV. Further information on VCV (MK-7690) is available in the Vicriviroc Maleate IB.[11]

The VCV (MK-4176) IVR is an EVA copolymer IVR containing 182 mg of VCV (MK-4176). It contains an EVA 28 core loaded with VCV (MK-4176) and surrounded by an EVA 28 skin layer loaded with no drug. The VCV (MK-4176) only IVR also contains 0.1 wt% magnesium stearate as a processing aid/lubricant. The magnesium stearate does not impact the appearance of the ring. The VCV (MK-4176) IVR is smooth, flexible and translucent with an outer diameter of 54 mm and a cross-sectional diameter of 4 mm. Further information is available in the MK-2048A IB.[10]

2.3.2 Mechanism of Action

VCV is a potent *in vitro* CCR5 entry inhibitor of HIV-1. In PBMC infection assays the overall geometric mean IC_{50} and IC_{90} values for VCV were 0.61 nM and 6.1 nM. Originally developed by Schering-Plough, VCV binds to a small hydrophobic pocket between the transmembrane helices near the extracellular surface of the CCR5 receptor, preventing the binding of gp120 to the target cell thereby preventing the virus from entering the target cell.[12,11]

2.3.3 Strength of Study Product

The VCV (MK-4176) IVR contains 182 mg of VCV (MK-4176).

2.4 MK-2048 IVR

2.4.1 Description

MK-2048 is a second-generation HIV-1 integrase inhibitor that belongs to a novel class of compounds with antiretroviral activity. MK-2048 is chemically described as (6S)-2-(3-chloro-4-fluorobenzyl)-8-ethyl-10-hydroxy-N,6-dimethyl-1,9-dioxo-1,2,6,7,8,9-hexahydropyrazino[1',2':1,5]pyrrolo[2,3-d]-pyridazine-4-carboxamide. MK-2048 is a white crystalline non-hygroscopic powder that is highly soluble across the physiological pH range (pH 2 to 8).[13]

The MK-2048 IVR is an EVA copolymer IVR containing 30 mg of the drug MK-2048. It contains an EVA 28 core loaded with no drug and surrounded by an EVA 28 skin layer

loaded with MK-2048.[10] The MK-2048 IVR is a smooth, white to off-white, opaque IVR, with an outer diameter of 54 mm and a cross-sectional diameter of 4 mm.

Further information is available in the MK-2048 and MK-2048A IBs. [10,13]

2.4.2 Mechanism of Action

MK-2048 is a second-generation HIV-1 integrase inhibitor. HIV-1 integrase is an enzyme required for the catalysis of the insertion of the HIV-1 DNA into the genome of the host cell. Integration is required for stable maintenance of the viral genome as well as efficient viral gene expression. MK-2048 inhibits the insertion of HIV-1 DNA into host cells, thus preventing viral replication. [10,13]

2.4.3 Strength of Study Product

The MK-2048 IVR contains 30 mg of MK-2048.

2.5 MK-2048A IVR (VCV [MK-4176] + MK-2048)

2.5.1 Description

The MK-2048A IVR is a novel combination product (drug/device) designed to offer sustained-release of the combination of two active ingredients,—the ARVs VCV (MK-4176) and MK-2048.[10] MK-2048A IVR is an EVA 28 core loaded with VCV (MK-4176) surrounded by an EVA 28 skin layer loaded with MK-2048.[10]

The MK-2048A IVR is a smooth, white to off-white, opaque IVR, with an outer diameter of 54 mm and a cross-sectional diameter of 4 mm. Further information is available in the MK-2048A IB.[10]

2.5.2 Mechanism of Action

Please see Sections 2.3.2 and 2.4.2 above.

2.5.3 Strength of Study Product

The strength of the MK-2048A IVR will be 182 mg of VCV (MK-4176) and 30 mg of MK-2048.

2.6 Placebo IVR

2.6.1 Description

The placebo IVR is an EVA copolymer IVR with a vinyl acetate content of 28% w/w. The placebo IVR is a smooth, flexible, translucent IVR, with an outer diameter of 54 mm and

a cross-sectional diameter of 4 mm. Further information is available in the MK-2048A IBs.[10]

2.6.2 Mechanism of Action

The placebo IVR is designed to be inactive in the vagina.

2.6.3 Strength of Study Product

The placebo IVR contains no active drug.

2.7 *In vitro and Ex Vivo* Studies

2.7.1 *In Vitro and Ex Vivo* Studies of VCV (MK-4176)

VIROLOGY

Anti-HIV Activity

VCV maleate (MK-7690) inhibited the replication of genotypically diverse HIV primary isolates in human peripheral blood mononuclear cells (PBMCs), with geometric mean inhibitory concentration 50% percent of viral inoculum inhibited (IC_{50}) and inhibitory concentration 90% percent of viral inoculum inhibited (IC_{90}) concentrations of 0.61 nM and 6.1 nM, respectively.[10] The *in vitro* antiviral effect was additive to synergistic when used in combination with other classes of ARV agents, including a monoclonal antibody against the CCR5 receptor. *Note*: Preliminary data showed that the VCV free-base and maleate salt forms of VCV (MK-4176 and MK-7690, respectively) had similar activity in *in vitro* antiviral infection assays with CCR5-tropic virus.[10]

In vitro and *ex vivo* anti-HIV activity studies conducted to support the development of VCV for a microbicide indication are summarized in Table 2. ARVs that are delivered intravaginally and are intended to be active against male-to-female transmission of HIV during sexual activity are exposed to a variety of factors that may influence their efficacy. *In vitro* assays were conducted to assess these factors and included antiviral activity in the presence of vaginal and seminal fluid simulants, a simulated pH shift and in the presence of whole semen (Table 2). *In vitro* anti-viral activity of VCV (MK-4176) was not affected by any of the factors. Approximately 30-fold more drug was needed to protect against cell-associated virus transmission as compared to cell-free. VCV (MK-4176) was not able to prevent CCR5-mediated cell fusion likely due to the creation of a viral synapse.[14] Because VCV (MK-4176) is specific for CCR5, it was confirmed not to be effective against CXCR4 virus.

Table 2: *In vitro/Ex vivo* studies of VCV (MK-4176)/VCV (MK-7690)

In Vitro Efficacy	
CCR5-tropic cell-associated virus transmission	EC_{50}, 360 nM TC_{50}, >1 μM TI, >2.8
CCR5-tropic cell-free entry inhibition in TZM-bl	EC_{50}, 0.09-11.3 μM TC_{50}, >100 μM TI, >8,85-11.11
CXCR4-tropic cell-free entry inhibition	EC_{50}, >100 μM TC_{50}, >100 μM TI, I
CCR5-tropic fusion inhibition	EC_{50}, >100 μM TC_{50}, >100 μM TI, I
CCR5-tropic cell-free entry inhibition in the presence of semen in TZM-bl	EC_{50}, 2.58 nM TC_{50}, >200 nM TI, >77.52
CCR5-tropic cell-free entry inhibition in human PBMC	EC_{50}, 1.56 nM TC_{50}, >500 nM TI, >2293
CCR5-tropic cell-free entry inhibition in the presence of simulated seminal fluid in human PBMC	EC_{50}, 1.12 nM TC_{50}, >500 nM TI, >446
CCR5-tropic cell-free entry inhibition in the presence of simulated vaginal fluid in human PBMC	EC_{50}, 0.548 nM TC_{50}, >500 nM TI, >912
CCR5-tropic cell-free entry inhibition at pH 4.5 in human PBMC	EC_{50}, 9.17 nM TC_{50}, >200 nM TI, >21.81
Ex Vivo Efficacy	
API (VCV) antiviral activity in human ectocervical explant	Partial protection at 100 μM, 1/6 explants
API (VCV) antiviral activity in human colorectal explant	Full protection at 10 μM, 6/6 explants
IVR segments (VCV IVR) antiviral activity in human ectocervical explant	Full protection, 6/6 explants
Other Assays	
Lactobacillus toxicity	CC_{50}, > 1 mM

API= Active pharmaceutical agent; CC_{50}= 50% cytotoxic concentration 50%; CCR5= C-C Chemokine receptor type 5; EC_{50}= 50% effective concentration; I= Indeterminate; IVR= Intravaginal ring; PMBC= peripheral blood mononuclear cells; TC_{50}= Toxic concentration 50%; TI= therapeutic Index; . The TI could not be established based on the calculated EC50 and TC50 values.

Ex Vivo studies were conducted using both polarized and non-polarized tissue explant models, including a human cervical tissue explant model which allows for exposure of only the surface of the epithelium to IVR segments and virus in a multi-day challenge assay. VCV (MK-4176) was tested as the free drug substance [i.e., active pharmaceutical ingredient (API)] and as drug formulated into IVR segments at a dose relevant to IVR product (i.e., VCV 182 mg) (Table 2). More VCV (MK-4176) was needed to block HIV-1 infection of polarized ectocervical and colonic tissue compared to PBMCs or other cell lines (Dezzutti, unpublished data). However, protection at the 100 and 10 μM concentrations was below the amount of VCV (MK-4176) anticipated to be released daily from an IVR (2.8 mM). An ectocervical tissue explant model, as described above, was used to determine the activity of the IVR segments containing VCV (MK-4176). For the IVR segment testing, a multi-day challenge assay was used. [15] Full protection (6/6 explants) was observed (Table 2 and Table 6). These data suggest

that continuous delivery of drug protects mucosal tissue better than a single dose of drug. Other studies were conducted, such as the effect of VCV (MK-4176) on vaginal *Lactobacilli,* bacteria important to a healthy vaginal environment which serves as a natural defense against HIV infection. VCV (MK-4176) was not toxic to any of vaginal *Lactobacilli* species tested (Table 2).

Resistance

Several *in vitro* studies have shown that VCV (MK-7690) resistance is predominantly caused by mutations in the variable domain 3 of gp120 (V3). However, mutations in other variable loops of *env* can contribute to resistance. Mutations in V2 (V169M) and V3 (L317W) emerged after 4 passages of an R5-tropic subtype B primary isolate of USA origin[16] in the presence of VCV (MK-7690), and I840Y in gp120 was selected after an additional 12 passages. I408T conferred a 2-fold increase in IC_{50} and the triple mutant had an MPI of 94%. The passaged virus remained R5-tropic with decreased infectivity.[17] Site directed mutational analysis showed that the G516V change in gp41 accompanied by M518V or F519I is critical for VCV (MK-7690) resistance in this isolate.[18] Virus passaged in CD4+ T cells or PBMCs developed mutations that in combination caused >20,000-fold VCV (MK-7690) resistance after 6-20 passages. These combinations included K305R/A316V/G321E, V535M/G514V and/or F519L, and G516V/M518V/ F519I.[19, 20]

Cross-resistance

Several primary HIV-1 isolates were independently passaged in PBMC cultures in the presence of increasing concentrations of VCV (MK-7690) until phenotypic resistance emerged. Resistant isolates were evaluated for susceptibility to different CCR5 antagonists in PBMC infection assays. As indicated in Table 3, VCV-resistant isolates were also cross-resistant to other small molecule CCR5 antagonists.

Table 3: Summary of Cross-Resistance Profiles for VCV-Resistant Mutants

Inhibitor Class	Inhibitor	CC 1/85	CC101.6	RU570	JV1083
CCR5 Inhibitor	Vicriviroc	R	R	R	R
	Maraviroc	R	R	R	R
	Aplaviroc	R	R	R	R

R = Resistant; S = Sensitive

Phenotypically resistant variants, with the exception of one CC 1/85 culture, remained CCR5-tropic, as demonstrated by the continued ability to replicate in CCR5- but not CXCR4-expressing U-87-CD4 cell lines.

Condom Compatibility

Condom compatibility studies have not been conducted with the IVR. However, studies with high and low dose preparations of VCV (MK-4176) (2000 µM and 200 µM) dissolved in modified vaginal fluid simulant were conducted according to ASTM D7661 on the following types of condoms:

- Non-lubricated, latex condoms (Durex, LifeStyles, Trojan ENZ)
- Polyiosprene condom (LifeStyles SKYN)

- Polyurethane condom (Trojan Supra)

All types of condoms met the acceptance criteria established in the protocol for the mean values of the treated samples after treatment with compounds. The results of the condom compatibility testing indicated that VCV (MK-4176) has no deleterious effects on the integrity of condoms at any of the concentrations tested.

2.7.2 *In Vitro* and *Ex Vivo* Studies of MK-2048

VIROLOGY

Anti-HIV Activity

MK-2048 is a potent and selective inhibitor of HIV-1 integrase catalyzed strand transfer with an IC_{50} of 7 nM. MK-2048 exhibits >5000-fold selectivity to the human DNA polymerases α, β, and γ (IC_{50} ≥ 50μM) and ≥ 2000-fold selectivity against other HIV-1 phosphoryl transferases (RNase H and reverse transcriptase IC_{50} > 15μM). [13] MK-2048 exhibits potent inhibition in multiple-cycle HIV-1 replication assays performed in the presence of 10% fetal bovine serum (IC_{95} = 11 ± 6 nM) or 50% human serum (IC_{95} = 41 ± 26 nM). These antiviral potencies are comparable to many effective agents currently licensed for the treatment of HIV-1 infection (e.g., indinavir—IC_{95} of 50 nM). When tested in a single-cycle HIV infectivity assay, MK-2048 showed potency (IC_{50} range = 1.4 to 3.6 nM) against 23 viruses with integrase genes obtained from a broad range of clinical HIV isolates. Many of these isolates were highly resistant to licensed ARV drugs. These findings support the expectation that MK-2048 should be broadly active against primary HIV isolates. [13]

In vitro and *ex vivo* anti-HIV activity studies conducted to support the development of MK-2048 for a microbicide indication are summarized in Table 4. *In vitro* anti-viral activity of MK-2048 was not affected by the presence of vaginal and seminal fluid simulants, a simulated pH shift or the presence of whole semen (Table 4). *Ex Vivo* studies were conducted using both polarized and non-polarized tissue explant models, including a human cervical tissue explant model which allows for exposure of only the surface of the epithelium to drug/IVR segments and virus in a multi-challenge assay.

MK-2048 was tested as the free drug substance (i.e., API) and as drug formulated into IVR segments at a dose relevant to IVR product (i.e., 30 mg MK-2048). *Ex vivo* testing of the MK-2048 API in polarized mucosal tissues showed more than 50% of the ectocervical tissues were protected from infection at 100 μM and 100% of colonic tissues were protected at 1 μM (Table 4). These MK-2048 concentrations are lower than the expected 649 μM/day anticipated to be released from the IVR. Activity of IVR segments containing MK-2048 was determined using an ectocervical tissue explant model (as described above). For the IVR segment testing, a multi-challenge assay was used. [15] Full protection (6/6 explants) was observed (Table 4 and Table 6). MK-2048 was mildly toxic to vaginal *Lactobacilli* species tested (Table 4).

Table 4: *In vitro/Ex vivo* studies of MK-2048 supporting a microbicide indication

In Vitro Efficacy	
CCR5-tropic cell-associated virus transmission	EC_{50}, >1 μM TC_{50}, >1 μM TI, I*
CCR5-tropic cell-free entry inhibition in TZM-bl	EC_{50}, 180 nM TC_{50}, >100 μM TI, 555.56
R4-tropic cell-free entry inhibition	EC_{50}, 150 nM TC_{50}, > 100 μM TI, >666.67
CCR5-tropic fusion inhibition	EC_{50}, 20.57 μM TC_{50}, > 100 μM TI, >4.86
CCR5-tropic cell-free entry inhibition in the presence of semen in TZM-bl	EC_{50}, 0.628 nM TC_{50}, >200 nM TI, >318.47
CCR5-tropic cell-free entry inhibition in human PBMC	EC_{50}, 0.083 nM TC_{50}, >500 nM TI, >6053
CCR5-tropic cell-free entry inhibition in the presence of simulated seminal fluid in human PBMC	EC_{50}, 0.269 nM TC_{50}, >500 nM TI, >1859
CCR5-tropic cell-free entry inhibition in the presence of simulated vaginal fluid in human PBMC	EC_{50}, 0.753 nM TC_{50}, >500 nM TI, >664
CCR5-tropic cell-free entry inhibition at pH 4.5 in human PBMC	EC_{50}, 1.24 nM TC_{50}, >200 nM TI, > 161.29
Ex Vivo Efficacy	
API (MK-2048) antiviral activity in human ectocervical explant	Partial protection at 100 μM, 4/6 explants
API (MK-2048) antiviral activity in human colorectal explant	Full protection at 1 μM, 6/6 explants
IVR segment (MK-2048 IVR) antiviral activity in human ectocervical explant	Full protection, 6/6 explants
Other Assays	
Lactobacillus toxicity	CC_{50}, 0.870 – 1.61 mM

API= Active pharmaceutical agent; CC_{50}= 50% cytotoxic concentration 50%; CCR5= C-C Chemokine receptor type 5; EC_{50}= 50% effective concentration; I= Indeterminate; IVR= Intravaginal ring; PMBC= peripheral blood mononuclear cells; TC_{50}= Toxic concentration 50%; TI= therapeutic Index; The TI could not be established based on the calculated EC50 and TC50 values.

Resistance

In vitro selection experiments with MK-2048 initiated with virus from clinical isolates selected the N155H mutation in integrase. Q148H/K/R with other mutations isolated from patients failing raltegravir-containing ART were found to have reduced susceptibility to MK-2048.[21-23] In another *in vitro* resistance study, the G118R and E138K mutations have also been found to be selected by MK-2048 and to cause resistance to MK-2048 but not to raltegravir or elvitegravir, two related drugs.[24]

Cross-resistance

HIV-1 variants selected for resistance to integrase strand transfer inhibitors harbor mutations in the integrase active site, such as T66I, L74M, F121Y, V151I, S153Y, M154I, and N155S.[24, 25] Various integrase inhibitors select for similar, but not identical, mutations in integrase. *In vitro*, mutation in the integrase active site (N155S) was found

to reduce the potency of MK-2048 by 3-fold. However, the activity of MK-2048 was not effected by single or combinations mutations at other integrase sites including T66I/L74M/V151I, F121Y, T66I/S155Y or T66I/M154I.[25]

2.7.3 *In vitro* and *Ex vivo* Studies of VCV (MK-4176) and MK-2048 in Combination

VIROLOGY

Anti-HIV Activity

Limited data is currently available on the antiviral activity of MK-2048 and VCV (MK-4176) in combination. Preliminary studies indicate at least additive antiviral activity of the two compounds when tested at equimolar concentrations relative to single drug alone in a standard antiviral assay (Table 5).[10]

Table 5: Antiviral Activity of free drug VCV (MK-4176) and MK-2048 Alone or in Combination

Viral Isolate	VCV alone (IC_{50} nM)	MK-2048 only (IC_{50} nM)	VCV + MK-2048 (IC_{50} nM)
JR-FL	11.7	7.7	8.9
ASM 57	7.1	4.9	6.1

Ex vivo anti-HIV activity studies conducted to support the development of the combination of MK-2048/VCV (MK-4194) for a microbicide indication are summarized in Table 6. A polarized ectocervical tissue explant model which allows for exposure of only the surface of the epithelium to drug and virus (as described above) was used to determine the activity of the MK-2048A combination IVR as well as IVRs containing VCV (MK-4176) or MK-2048 alone (see Tables 1 and 3). IVR segments contained drug doses relevant/proportional to planned IVR products (30 mg MK-2048 and 182 mg VCV). For the IVR segment testing, a multi-challenge assay was used.[15] Full protection (6/6 explants) was observed against 4 independent HIV challenges over 11 days in explants cultured with the IVR segments of the combination MK-2048A IVR as well as the IVR segments of the single agent IVRs (containing either MK-2048 or VCV (MK-4176) only).

Table 6: Ex vivo studies of IVRs containing MK-2048 and/or VCV or placebo

***Ex Vivo* Efficacy: Antiviral activity in human ectocervical explant model**	
IVR segments (MK-2048A combination IVR)	Full protection, 6/6 explants
IVR segments (MK-2048 IVR)	Full protection, 6/6 explants
IVR segments (VCV IVR)	Full protection, 6/6 explants
IVR segments (placebo)	No Protection, 0/6 explants

2.7.4 In Vitro and Ex Vivo Studies of Placebo

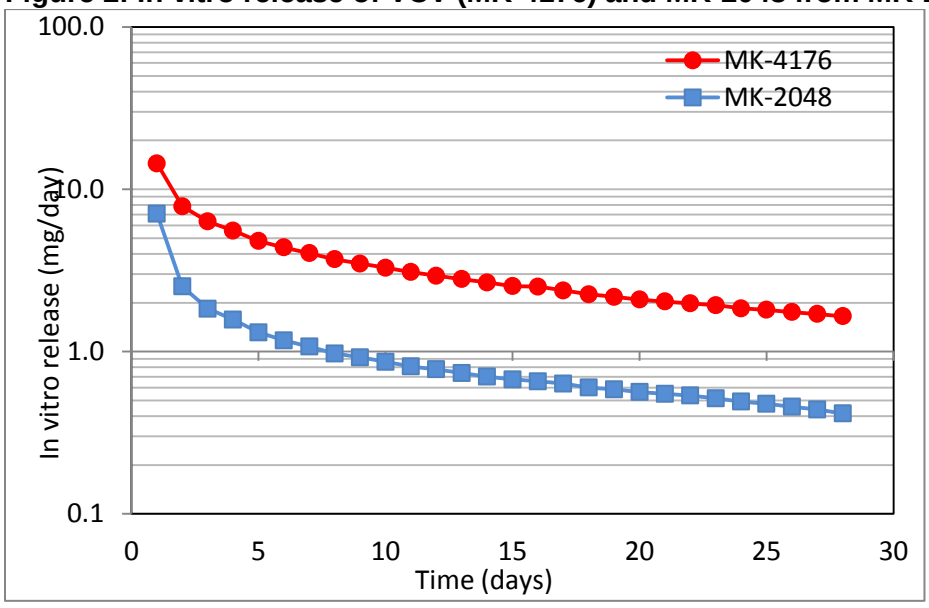
Anti-HIV-1 Potency of Placebo in microbicide models: *Ex vivo*
The placebo IVR segments were tested in a cervical tissue explant model (see details above). Placebo IVR segments provided no protection (0/6 explants) against multiple HIV challenges (Table 6).

2.7.5 *In Vitro* IVR Drug Release Studies: VCV (MK-4176) IVR, MK-2048 IVR and MK-2048A combination IVR

To evaluate the extent and rate of drug release from the EVA polymer IVRs, VCV only (MK-4176), MK-2048 only and MK-2048/VCV combination IVRs were incubated at 37°C in 100 mL *in vitro* release medium (0.05% (V/V) acetic acid in sodium dodecyl sulfate solution 1.0% (m/V). Samples for drug determination were collected every 24 hours and release medium was refreshed directly after each sampling.

A typical *in vitro* drug release profile of VCV (MK-4176) and MK-2048 over 28 days from the MK-2048A combination IVR (182 mg VCV (MK-4176) + 30 mg MK-2048) is shown in Figure 2. An initial burst release of both compounds was observed during the first 1 to 2 days of incubation with peak release approximately 14 and 7 mg/day for VCV (MK-4176) and MK-2048, respectively. Following this initial burst, drug release stabilized between days 5 to 28, with release rates ranging between 4.8 to 1.65 mg/day for VCV (MK-4176) and 1.3 to 0.415 mg/day for MK-2048. Assuming a total vaginal fluid volume of 10 ml, the predicted fluid levels of VCV (MK-4176) and MK-2048 on day 28 would be approximately 300 µM and 90 µM respectively; concentrations many fold above the *in vitro* antiviral IC_{95} for each compound.[10]

Figure 2: In vitro release of VCV (MK-4176) and MK-2048 from MK-2048A combination IVR



2.8 Nonclinical Studies

2.8.1 Non Clinical Studies of VCV (MK-7690)

Animal studies of the safety of VCV maleate form [VCV (MK-7690)] when administered via the oral route are available in the VCV (MK-7690) IB and MK-2048A IB.[10,11] Animal study data were conducted in multiple species and supported the safety of MK-7690 for further investigation in human trials. No treatment related effects on renal, respiratory or cardiovascular systems were observed. Central nerve system (CNS) studies showed that seizures were observed in some animals. Seizures in animals were self-limiting and preventable with standard anti-convulsant therapy. The plasma concentration in the most sensitive species (dog), below which no seizure occurred, was 4670 ng/mL. This concentration is substantially higher (>170X) than plasma levels measured in sheep dosed intravaginally with MK-2048A IVR or VCV (MK-4176) only IVR for 28 days.

A penile irritation study in male rabbits was conducted to evaluate the potential local toxicity to male partners of women using IVRs containing VCV (MK-4176). Hourly application of 0.2 mL of gels containing 0.231%, 0.461%, 0.922%, and 2.5% VCV (MK-4176) for 4 hours over 3 days resulted in no local or systemic toxicity.

A battery of biocompatibility tests was performed using extracts from the MK-2048A combination IVR containing both VCV (MK-4176) and MK-2048, as well as IVRs containing VCV (MK-4176) and MK-2048 alone. The following biocompatibility studies were conducted and did not identify any significant safety concerns in any of the IVRs.[10]

- **Cytotoxicity**
  - Cytotoxicity study using the elution method; extract in minimal essential medium (37°C for 24 hours)
- **Sensitization**
  - Guinea pig maximization study; extracts in 0.9% sodium chloride solution (50°C for 72 hours) and cotton seed oil (50°C for 72 hours)
- **Genotoxicity**
  - Bacterial reverse mutation study; extracts in 0.9% sodium chloride solution (50°C for 72 hours) and dimethyl sulfoxide (50°C for 72 hours)
  - Mouse lymphoma assay; extracts in 0.9% sodium chloride solution (50°C for 72 hours) and dimethyl sulfoxide (50°C for 72 hours)
- **Irritation and subacute/subchronic toxicity**
  - Rabbits dosed intravaginally, daily, for 35 days extracts in 0.9% sodium chloride solution (50°C for 72 hours) and cotton seed oil (50°C for 72 hours)

2.8.2 Non Clinical Studies of MK-2048

Animal studies of the safety of MK-2048 agents when administered via the oral route are available in the MK-2048A IB and MK-2048 IB.[10, 13] Animal study data were conducted in multiple species and supported the safety of MK-2048 for further investigation in human trials. No treatment related effects on renal, respiratory or

cardiovascular systems were observed in various animal models. In CNS studies, no treatment-related effects were observed.

A penile irritation study in male rabbits was conducted to evaluate the potential local toxicity to male partners of women using the various IVRs containing ARVs used in the study. The hourly application of 0.2 mL of gels containing 0.059%, 0.117%, and 0.234% MK-2048 for 4 hours over 3 days resulted in no local or systemic toxicity.

2.8.3 Non Clinical Studies of VCV (MK-4176) and MK-2048 Combination

Animal studies of the safety of MK-2048 and VCV (MK-7690) when tested as single agents via the oral or intravenous route are available in the MK-2048 IB, VCV (MK-7690) and MK-2048A IBs. [10, 11, 13]

Systemic and local toxicities during 28-day continuous intravaginal use of VCV (MK-4176) and MK-2048 as single agents or combination were conducted using various animal species, administered as an IVR or via gel formulations (Table 7).[10]

IVR Study: Sheep Model

The potential local effects, systemic toxicity and toxicokinetics of placebo and antiretroviral containing IVRs products (including VCV (MK-4176) IVR, MK-2048 IVR and the MK-2048A combination IVR) were evaluated in virginal Dorset sheep model for 28 days. These were the same IVR formulations to be used in MTN-027 study. Two feasibility studies were conducted: a non-GLP study (study no. S11727) and a GLP study (study no. S12284).

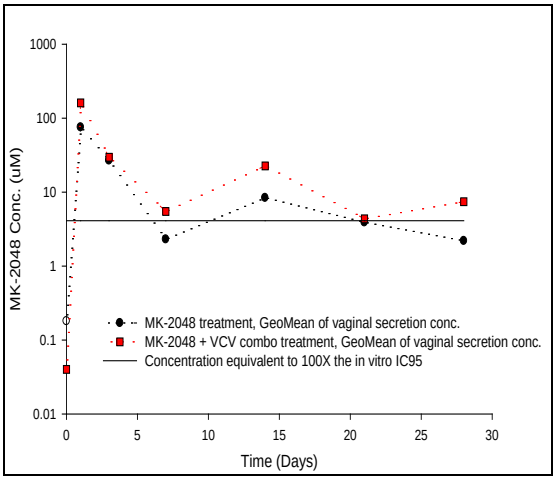
In the non-GLP feasibility study (Study S11727), the IVRs were generally well tolerated. Pharmacokinetic analysis indicated that mean plasma concentration of VCV (MK-4176) were low (≤0.013 µM) and plasma levels of MK-2048 in plasma were below the lower limit of quantitation (LLOQ) (0.0005 to 0.001 µM) at all-time points. The presence of the IVR was associated with mixed cell inflammation of the vaginal epithelial and sub-epithelial tissues and hypertrophy of the vaginal epithelium. However these changes were observed in all treatment groups, including the placebo IVR group, and therefore cannot be attributed to the presence of compounds in the IVRs. Of note, no sham treatment group was included in this non-GLP study. Therefore, the effect of manipulation of the animals during dosing/examination procedures could not be evaluated and compared to the findings in the IVR-treated animals. To better differentiate handling and manipulation effects from IVR related effects, a sham treated group was included in the GLP sheep study described below.

In a non-GLP study conducted in virginal Dorset sheep (Study S11727), IVR containing VCV (MK-4176), MK-2048, or a combination of both (182 mg of VCV (MK-4176) and/or 30 mg MK-2048) were inserted intravaginally (Figure 3). Pharmacokinetic analysis of plasma indicated low systemic exposures of VCV (Cmax≤0.013 µM) and no detectable levels of MK-2048 at all-time points during the 28-day study (LLOQ 0.0005 to 0.001 µM). The maximal concentrations of MK-2048 and VCV in local vaginal secretions were

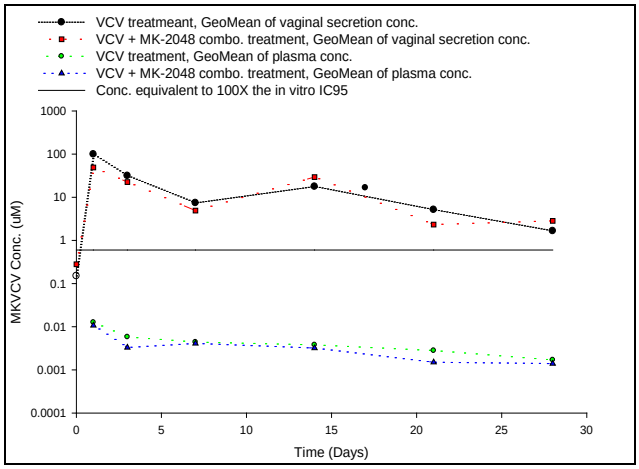
achieved a day after ring insertion. These concentrations were highly variable across animals and corresponded to a geometric mean of 75.3 µM and 160.7 µM for MK-2048 in the single-component and combination ring respectively and 99.3 µM and 49.1 µM for VCV (MK-4176) in the single-component and combination ring, respectively.

Twenty eight days after insertion of the IVRs, the geometric mean concentrations for MK-2048 were 2.19 µM and 7.41 µM in the single-component and combination ring respectively. For VCV (MK-4176), the concentrations 28 days after insertion were 1.66 µM and 2.84 µM in the single-component and combination IVR respectively. Remnant analysis of the IVRs recovered at the conclusion of the 28-day study indicated that 12.3 and 10.2 mg of MK-2048 were released from the single-component and combination IVRs respectively, at the conclusion of the study. For VCV (MK-4176), the amounts released were 111 and 105.7 mg, respectively.

Figure 3: Plasma and vaginal fluid concentrations for MK-2048 (A) and VCV (MK-4176) (B) following insertion of IVRs containing MK-2048 (30 mg) and/ VCV (MK-4176) (182 mg) to virginal Dorset sheep



(A)



(B)

In the GLP sheep study (SN12284) placebo, single ARV agent and combination agents IVRs, were used to evaluate the potential local effects, systemic toxicity and toxicokinetics in virginal Dorset sheep for 28 days in comparison to non-treated (sham) animals. Histopathological results showed that the vaginal mucosa among non-treated (sham) animals and animals exposed to the placebo IVR was comprised of normal squamous epithelium, sometimes exhibiting parakeratosis of the most superficial layer lining the vaginal lumen. The immediately subjacent propria-submucosa contained variable amounts of mixed cellular infiltrates composed of mononuclear cells (lymphocytes, monocytes) mixed with neutrophils. These cellular infiltrates also extended into the overlying stratified squamous epithelium. The vaginal and cervical tissues among animals exposed to IVRs containing VCV, MK-2048 or both had similar morphology as described for animals of non-treated and placebo controls. Because of the presence of similar findings in the sham, placebo and VCV/MK-2048 treated groups, it can be concluded that the microscopic observations in the vagina, cervix, uterus, or

ovaries were not related to the intravaginal placement of the IVRs. There were also no findings attributable to either VCV (MK-4176), MK-2048, or related to the combination of the two compounds among sheep evaluated at end of treatment on Day 28 or at end of recovery on Day 38. In this study, no systemic toxicity was observed in the sheep after 28 days of exposure to the IVRs.

MK-2048 plasma levels were below the LLOQ (0.0108 µM) at all-time points over 28 days of exposure to the IVR containing MK-2048 alone or in combination with VCV. VCV (MK-4176) was detected in the plasma at low levels with a C_{max} of 0.050 µM (3 times LLOQ of the analytical method: 0.0187 µM) achieved by 4 hours after IVR insertion. The plasma levels rapidly declined to below LLOQ thereafter.

The combination IVR contained 182 mg VCV (MK-4176) and 30 mg MK-2048. Remnant content analysis of the rings used in the 28-day study in sheep demonstrated an in vivo total exposure of 106 mg MK-4176 and 16 mg MK-2048. This *in vivo* release in sheep results in a 5.2 mg/kg and 0.5 mg/kg total cumulative 28 day dose in sheep for MK-4176 and MK-2048, respectively.

Repeat Dose Toxicity Studies: Gel Administration

Although studies using the IVRs in the sheep model allowed evaluation of toxicity of the drug product *in vivo*, no MK-2048 and very low levels of VCV (MK-4176) were detected in the plasma of treated animals. Therefore, to achieve higher systemic exposures than those observed with the drug product in sheep the compounds were formulated as gels to evaluate the potential local effects, systemic toxicity and toxicokinetics in Sprague-Dawley rats and New Zealand white rabbits when administered intravaginally for 28 days. Details of each study are provided below and summarized in Table 7.

In summary, there were no significant local or systemic findings following repeat administration of up to 2.5% VCV (MK-4176) and 0.234% MK-2048 up to 28 days In intravaginal toxicity studies in rats using gels. In intravaginal toxicity studies in rabbits there were no significant systemic findings or histomorphological findings in the vaginal proximal mucosa determined the NOEL at 0.231% of VCV (MK-4176) following repeat daily administration up to 28 days. There were no significant local or systemic findings following repeat administration up to 0.234% MK-2048 for 28 days. Although findings were reported at the higher dose levels for VCV (MK-4176) in the rabbit they were not reproduced in the equivalent studies in the rat and sheep model, therefore we consider these to be idiosyncratic for this model. The absence of relevant changes in distal mucosa and stratified squamous epithelium of the rabbit and the absence of changes in the rat and sheep models, which are more representative of vaginal tissues in women, are more relevant findings.

Table 7: Non-clinical safety/toxicity studies of VCV (MK-4176), MK-2048 and the combination of VCV (MK-4176) + MK-2048 via topical administration

Species	Dosage of ARV	Route/Mode of Administration	Duration	Study ID
Sheep[a]	Placebo VCV (MK- 4176) only, MK-2048 only VCV (MK- 4176) + MK-2048 combination VCV[b]: up to 18 mg/day[c] MK-2048[d]: up to 7mg/day	IVR	28 days	S11727[e]
Sheep	Placebo VCV (MK- 4176) only MK-2048 only VCV (MK- 4176) + MK-2048 combination VCV[f]: up to 18 mg/day[c] MK-2048[g]: up to 7 mg/day	IVR	28 days 10 day recovery	S12284
Rabbit	VCV (MK- 4176): 2.305 – 25 mg/rabbit/day[h] MK-2048: 0.59 – 2.34 mg/rabbit/day	Gel formulation applied vaginally	10 days	1726-020
Rabbit	VCV: 2.305 – 25 mg/rabbit/day MK-2048: 0.59 – 2.34 mg/rabbit/day	Gel formulation applied vaginally	28 days 10 day recovery	1726-027
Rabbit	VCV: 1.844 – 20 mg/rabbit/day MK-2048: 0.472 – 1.872 mg/rabbit/day	Gel formulation applied to penis	Gel applied hourly for 4 hours for 3 days, 2 day recovery	S12392
Rat	VCV: 0.2305 – 2.5 mg/rat/day MK-2048: 0.059 – 0.234 mg/rat/day	Gel formulation applied vaginally	28 days 10 day recovery	1726-026

[a]Study was performed under non-GLP quality standards.
[b]Total VCV present in the VCV only and VCV + MK-2048 combination IVR was 182 mg. The average daily drug present in local vaginal secretions over 28 days ranged between 1.66 and 99.31 µM.
[c]Daily release rate determined *in vitro*.
[d]Total MK-2048 present in the MK-2048 only and VCV + MK-2048 combination IVR was 30 mg. The average daily drug present local vaginal secretions over 28 days ranged between 2.19 and 160.67 µM.
[e]Final report assembled by Merck under the report designation: MK-2048 non-clinical report PK003.
[f]Total VCV present in the VCV only and VCV + MK-2048 combination IVR was 182 mg. The average daily drug present in local vaginal secretions over 28 days ranged between 0.28 and 79.24 µM with IVR present.
[g]Total MK-2048 present in the MK-2048 only and VCV + MK-2048 combination IVR was 30 mg. The average daily drug present in local vaginal secretions over 28 days ranged between 13.86 and 1036.3 µM with IVR present.
[h]Expressed as daily dose mg/animal for daily vaginal/penile gel administration.

A battery of biocompatibility tests was performed using extracts from the MK-2048A combination IVR containing VCV (MK-4176) and MK-2048, as well as IVR containing each of the drugs alone. The biocompatibility studies did not identify any significant safety concerns.

2.8.4 Non Clinical Studies of IVR Components

Since the ring is applied intravaginally, a program of specific toxicological studies was performed that paid special attention to local and systemic toxicity of its chemical components (i.e., EVA copolymer and potential leachables) *in vivo*, and to their toxic potential *in vitro*. Part of the evidence for the safe use of the IVR with respect to the EVA copolymer and potential leachables was obtained from studies performed with the EVA-containing radiopaque Multiload® (intrauterine device) and extracts prepared thereof. Other relevant data on the potential toxicity of the IVR constituents were derived from studies for the development of the contraceptive implant Implanon® and the vaginal ring Nuvaring®, both manufactured by N.V. Organon, now part of Merck. Detailed information about the non-clinical toxicology studies are provided in the MK-2048A IB.[10]

The placebo IVRs were used to evaluate the potential local effects, systemic toxicity and toxicokinetics in virginal Dorset sheep for 28 days in a feasibility non-GLP study (study no. S11727) and GLP study (study no. S12284) (see Table 7). In the non-GLP feasibility study, the IVRs were generally well tolerated. The presence of the IVR was associated with mixed cell inflammation of the vaginal epithelial and sub-epithelial tissues and hypertrophy of the vaginal epithelium. Similar changes in animals that received the placebo IVR compared to those that received IVRs containing drug eliminates the drugs alone or in combination as the sole cause of the microscopic observations. In the GLP study, no systemic toxicity was observed in the sheep model after 28 days of exposure to the placebo IVR.

These data support the use of the EVA copolymer as a delivery vehicle for VCV (MK-4176) and MK-2048.

2.9 Clinical Studies

2.9.1 Clinical Studies of VCV (MK-7690)

The pharmacokinetics, safety and tolerability of oral VCV maleate (MK-7690) have been evaluated in clinical studies (Phase 1 through Phase 3). Oral administration of VCV in healthy subjects, subjects with end stage renal disease, subjects with mild/moderate hepatic failure and patients with HIV or HIV/HCV coinfection was generally well tolerated, without identification of an exposure-related toxicity.

Pharmacokinetics

Following oral administration of VCV (MK-7690), maximum plasma concentrations were reached approximately 1 to 2 hours after VCV (dosed alone or in the presence of ritonavir). C_{max} and AUC increased in a linear fashion with an increase in VCV dose. C_{max} concentrations at VCV 150 mg, in the presence of ritonavir 100 mg, are the highest attained to date (2470 ng/mL); this dose was well tolerated with no safety concerns detected.

VCV (MK-7690) is extensively metabolized by CYP3A4. Unchanged parent drug was the major component of drug-derived radioactivity in plasma. The carboxylic acid metabolite of VCV was the predominant circulating metabolite in human subjects after multiple oral doses of VCV monotherapy (50 mg twice daily for 14 days).

Safety

The highest doses of VCV (MK-7690) administered to HIV-seronegative healthy subjects have been single doses of up to 300 mg alone and multiple doses up to 250 mg QD alone for 7 days and 100 mg with ritonavir for 10 days. Pooled safety data from seronegative subject studies examined treatment-emergent adverse events (TEAEs) from studies where VCV (MK-7690) was administered from 10 mg as a single-dose to 250 mg QD alone or 150 mg with ritonavir QD. Of the 729 HIV-seronegative healthy subjects treated with VCV (MK-7690), 19 (3%) discontinued due to an AE compared to none of the subjects who received placebo. The most frequently reported TEAEs were headache (14%) and diarrhea (10%) after any VCV (MK-7690) administration (alone or in combination). A relationship to VCV (MK-7690) dose was not apparent. The majority of TEAEs, regardless of treatment, were mild to moderate in severity. In general, there were no remarkable or unexpected safety concerns, or trends in clinical laboratory tests associated with VCV (MK-7690) across the studied drug combinations. Neurological assessments of safety using continuous EEG monitoring to evaluate the potential for VCV to induce aberrant EEG waveform activity were conducted in two placebo-controlled dose-escalating studies in HIV-negative healthy subjects (P03161 and P04873). Results of the healthy subject EEG studies and the absence of drug-related seizures in Phase 2/3 patient studies confirmed the assertion that concentrations of VCV that might lead to seizures in humans are significantly higher than those administered for clinical treatment. [11]

Phase 1b/2a HIV Infected Subjects: Safety and Activity

No dose-related increase in the number of AEs was reported among HIV-infected or HIV/HCV co-infected subjects in the Phase 1b/2a studies P02726 (a 14-day, rising-multiple-dose study in HIV-infected subjects) and P04416 (a 28-day safety study in HIV/HCV co-infected subjects). Most adverse events were mild and unrelated to study drug.[10]

Two subjects in P02726 exhibited detectable X4 viral isolates in the 50 mg cohort. The mixed/dual R5/X4 viral phenotype was identified in one subject prior to dosing and after 14 days of dosing in the second. The second subject experienced a >1.5 log10 reduction in viral ribonucleic acid (RNA) and did not have a decline in CD4+ cell (T helper cell) count. By Day 28, 14 days after the last VCV (MK-7690) dose, no CXCR4 virus was detected.[10]

The mean change from baseline in HIV RNA for all three dose cohorts in the P02726, rising, multiple-dose study in HIV-infected subjects demonstrated potent viral suppression of 0.9 to 1.6 log10 on treatment with VCV (MK-7690). After 10, 25, and 50

mg VCV (MK-7690) BID, 46%, 77%, and 82% of subjects, respectively, reached or surpassed a 1.0 $\log_{10}$ reduction in viral load by Day 14.[10]

Phase 2 and 3 Safety/Tolerability and Activity

As of May 2010, more than 1300 HIV-infected subjects took part in Schering-sponsored Phase 2 or 3 trials, with the majority (80%) of the HIV-infected participants being treatment-experienced (n=1207) at the time of enrollment. Most TEAEs were mild and not related to VCV (MK-7690). The most commonly reported AEs were diarrhea, nausea, headache, upper respiratory infection, nasopharyngitis, and fatigue, with no clear difference in incidence between VCV (MK-7690) recipients and control groups. Of the treatment-naive subjects (n=309), a total of 30 subjects experienced serious adverse events (SAEs); 17 of the 177 (9.6%) treated with VCV (MK-7690), and 13 of the 132 (9.8%) in the control group. Among the treatment-experienced subjects (n=1488) a total of 210 experienced SAEs (including 171/1092 subjects (16%) receiving VCV (MK-7690) and 39/396 subjects (10%) receiving placebo).

Resistance

In two phase 3 studies (Study P04405 and P04889), viral isolates from study subjects at selected time points (Baseline, Weeks 2, 24, and 48, or at the time of virologic breakthrough) were tested for susceptibility to VCV (MK-7690). For both studies, an assessment of the Overall Susceptibility Score (OSS) was made using the PhenoSense GT assay at Screening, at Week 48 or at the time of virologic failure. Only four subjects exhibited a significant change in susceptibility to VCV (MK-7690) (manifested as a decrease in maximum calculated percent inhibition [MPI]) over the course of therapy and these changes were not detected until Week 12 (Subject 1803) or later. Two of the four subjects had an OSS of 3, the third had an OSS of 2, and final subject (Subject 1803) had an OSS of 1. X4-using virus was not detected at the time of study discontinuation in any of the four subjects with phenotypic resistance to VCV (MK-7690).

As above, viral isolates from the four subjects who exhibited reduced susceptibility to VCV (MK-7690) were subjected to a clonal analysis. Although a comparison of Baseline and end of treatment samples (12 clones/time point) revealed sequence evolution throughout the gp160 glycoprotein during the time of treatment, there was no consistent pattern of genotypic changes across subjects.[11]

Several clinical studies have also evaluated VCV (MK-7690) resistance. Two studies examined VCV (MK-7690) resistance in R5-tropic isolates from treatment failures but found no consistent pattern of resistance to VCV (MK-7690) from treated patients with virologic failure.[26, 27] One subject from a clinical trial infected with subtype D HIV-1 developed resistance to VCV (MK-7690) via multiple changes in gp120, including Q315E and R321G in the V3 loop, and E328K and G429R in C4.[28] One of 29 subjects in a Phase 2b study of VCV (MK-7690) infected with subtype C HIV-1 also harbored V3 loop mutations.[29]

Effectiveness

Based upon the analysis of a Phase 2 study (Study P03672) which evaluated VCV (MK-7690) dosages of 20 mg and 30 mg QD, the 30-mg dose was selected for the Phase 3 studies (Studies P04405 and P04889). These Phase 3 trials were designed to confirm the efficacy of the 30 mg VCV (MK-7690) QD dosage in combination with Optimized Background Therapy (OBT) including a ritonavir-boosted protease inhibitor (PI/r) in a larger cohort of HIV-infected, treatment-experienced subjects. Following completion and full analysis of the Phase 3 VCV (MK-7690) studies, it became clear that VCV (MK-7690) 30 mg QD did not provide incremental benefit to subjects with three or more active drugs in their background ART regimen. However, a greater proportion of subjects achieved virologic suppression (HIV RNA <50 copies/mL) in the VCV (MK-7690) group than in the control group, when VCV (MK-7690) was administered with a background ART regimen containing two or fewer active agents, similar to the Phase 2 study (Study P03672). Clinical development of VCV (MK-7690) was subsequently terminated.[11]

2.9.2 Clinical Study of MK-2048

Safety

One Phase 1 clinical trial of MK-2048 in an oral formulation has been conducted.[10, 13] Merck enrolled 16 men in a double-blind, randomized, placebo-controlled, alternating panel, rising single-dose study to evaluate the safety, tolerability and PK of MK-2048 in healthy male participants between the ages of 18 to 45 years. Each participant received either a single oral dose of MK-2048 or placebo per treatment period over a course of 4 periods, with a 7 day washout interval between periods. Total duration of follow-up for each was 8 weeks. Dose escalation (50, 100, 200, 400, 800, and 1200 mg) continued sequentially proceeding to the highest dose. Blood samples were collected for determination of MK-2048 plasma concentrations for 120 hours post-dose in each treatment period.

Safety was monitored throughout the study by repeated clinical and laboratory evaluation. MK-2048 was generally well-tolerated after single dose administration. There were no serious clinical adverse experiences or discontinuations due to an adverse event. Adverse events were generally transient and mild in intensity. Only one adverse event was reported by more than one subject: mild headache.[10, 13]

Pharmacokinetics

In the aforementioned trial, plasma was analyzed at specified time points for determination of MK-2048 concentrations. At fasted doses up to 1200 mg, peak plasma concentrations of MK-2048 occurred within 1.5 hour post dose, and concentrations declined rapidly from C_{max} in a single log-linear phase with a half-life of approximately 1 to 1.5 hr. Plasma concentrations were largely undetectable after 12 and 24 hours post dose at all doses. AUC and C_{max} increased approximately dose proportionally over this dose range (50 to 1200 mg), although there was some variability between panels. MK-2048 failed to achieve target plasma concentrations, as the samples were largely undetectable (assay limit of quantitation = 2 ng/mL = 4.3 nM) after 12 and 24 hours

post-dose at all doses. Due to these results further development of the oral product was discontinued.[10, 13]

2.9.3 Clinical Studies of Similar IVR Delivery Systems and Placebo IVR

The vaginal delivery system used for the MK-2048A ring has a comparable copolymer composition and the same dimensions as the approved vaginal contraceptive ring, NuvaRing®.[10] NuvaRing® is an estrogen/progestin combination hormonal contraceptive (CHC) indicated for use by women to prevent pregnancy. The first marketing authorization for NuvaRing® was obtained in 2001 and since market introduction in 2002, estimated worldwide exposure exceeds 8.7 million woman-years. Clinical and marketing experience with NuvaRing® indicates that the vaginal delivery system is well-accepted, as the majority of women have not had any problems with the use of NuvaRing®.

In clinical studies with NuvaRing®, an increase in local vaginal side-effects has been reported, including vaginitis (13.2%), leukorrhoea (5.6%), device-related problems (5.0%), and vaginal discomfort (2.9%). Device-related problems mainly concerned expulsion (3.1%), coital problems (1.1%), and foreign body feeling (0.7%). Vaginal discomfort predominantly concerned vaginal dryness (0.6%). Comparative studies indicated that these events were all more common for NuvaRing® users than for combined oral contraception users, which is obvious for device-related problems, but maybe less so for the other types of events. When relationship to the contraceptive method, as judged by the investigator, was taken into account the incidence of vaginitis was 4.3% for NuvaRing® as compared to 1.0-2.1% for combined oral contraception. A similar trend, but at lower absolute incidences, was observed for leukorrhea, while vaginal dryness was uncommon altogether.

There was no placebo IVR used in the clinical development program for NuvaRing®. Merck has continued development of next generation rings (NGR) for contraception and has advanced candidates into Phase 2 clinical trials.

The design and manufacture of the NGR is based on the NuvaRing technology, with both rings utilizing an ethylene vinylacetate (EVA) copolymer. A placebo IVR has been administered to 90 subjects in a single clinical study in the NGR program. MK-8342B Protocol 057 was a multi-center, randomized, placebo-controlled, dose finding study to evaluate the treatment effect of MK-8342B compared to placebo in female subjects aged 18-50 years who had been diagnosed with primary dysmenorrhea. Each subject in the placebo arm (N=90) used the placebo IVR for 21 consecutive days followed by 7 days without the ring, over two consecutive treatment cycles. The adverse events reported for the placebo IVR included headache (7.8%), acne (5.6%), abdominal pain/discomfort (4.4%), breast pain/discomfort (3.3%), diarrhea (1.1%), nausea (1.1%), vaginal discharge (1.1%) and anxiety (1.1%). There were no severe adverse events in subjects that received placebo IVR. All events were reported as resolved by the completion of the trial.

2.10 Study Hypothesis and Rationale for Study Design

2.10.1 Study Hypothesis

MTN-027 hypothesizes that all four study IVRs will be safe and well-tolerated among healthy, sexually abstinent women. The null hypothesis is that there will be no difference in the safety profile between the active products and the placebo.

2.10.2 Rationale for Study Design

Based on *in vitro*, *in vivo*, and *ex vivo* studies, VCV (MK-4176) and MK-2048 show great promise as topical microbicide agents to prevent HIV-1 infection.

IVRs have already been developed and approved as delivery methods for medications. For example, NuvaRing®, a contraceptive IVR made of EVA copolymers (with a 28% w/w core and a 9% w/w skin layer) and magnesium stearate, in which 2.7 mg of ethinyl estradiol and 11.7 mg of etonogestrel are dispersed, has been found to be both effective and acceptable to women. In an acceptability study involving 1,950 NuvaRing® users, 45.5% of women cited that their reason for liking the IVR was “not having to remember anything”. In a randomized controlled trial testing two alternative delivery systems for combined hormonal contraceptive, women overwhelmingly preferred the ring to oral contraceptives ($P<.001$).[9] These contraceptive ring data are encouraging as microbicide ring effectiveness will likely correlate with consistent and correct use. It is likely that products that can be applied less frequently will be more acceptable to users, achieve better user-adherence, and may lead to increased effectiveness. Intravaginal rings that need only be replaced every 28 days may have benefits over dosage forms that need to be used more frequently, as well as offer a wider choice of microbicide formulations for women if proven effective.

MTN-027 participants will be randomized in a 1:1:1:1 ratio to receive either an EVA copolymer IVR containing 182 mg VCV (MK-4176), or 30 mg MK-2048, or 182 mg VCV (MK-4176) + 30 mg MK-2048, or placebo. IVRs are to be used continuously for a period of 28 days, followed by 7 days with no study product.

Safety

Careful assessments of local and systemic safety will be undertaken in MTN-027. The study products evaluated in this trial have potential adverse effects, and tolerance may vary depending on formulation. The design of MTN-027 will allow safety comparisons of each product to a placebo, and may provide some data suggesting relative safety among active products. Most importantly researchers will undertake these questions while protecting the safety of study participants.

3 OBJECTIVES

3.1 Primary Objectives

- Assess and compare the safety of ethylene-vinyl acetate IVRs containing 182 mg vicriviroc (MK-4176), or 30 mg MK-2048, or 182 mg vicriviroc (MK-4176) + 30 mg MK-2048, when used continuously for 28 days by healthy, HIV-uninfected, sexually abstinent women, as compared with the placebo IVR

- Examine local and systemic pharmacokinetics of vicriviroc (MK-4176) and MK-2048 in vaginal fluid, plasma and cervical tissue during and after 28 days continuous use of an IVR containing 182 mg vicriviroc (MK-4176), or 30 mg MK-2048, or 182 mg vicriviroc (MK-4176) + 30 mg MK-2048

3.2 Secondary Objectives

- Evaluate the acceptability of the study IVR in HIV-uninfected sexually abstinent women over 28 days of use

- Evaluate the adherence to the study IVR in HIV-uninfected sexually abstinent women over 28 days of use

3.3 Exploratory Objectives

- Evaluate the HIV inhibitory activity of cervical tissue after ring use

- Assess drug levels in rectal fluid

- Describe the genital microenvironment during 28 days of study product use and follow-up

- Evaluate the potential relationship between participant self-report of adherence, vicriviroc (MK-4176) and MK-2048 remnant content in returned IVRs, and PK levels

4 STUDY DESIGN

4.1 Identification of Study Design

MTN-027 is a multi-site, single blind, four-arm, randomized placebo-controlled trial.

4.2 Summary of Major Endpoints

Primary Endpoints:

- The primary safety endpoints are the proportion of women in each of the four IVR regimens with:
  - Genitourinary events Grade 1 or higher as defined by the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Version 1.0, Dec 2004 (Clarification dated August 2009), Addendum 1, (Female Genital Grading Table for Use in Microbicide Studies) judged to be related to study product
  - Adverse events Grade 2 or higher as defined by the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Version 1.0, Dec 2004 (Clarification dated August 2009)

- The pharmacokinetic endpoints are:
  - Local and systemic concentrations of vicriviroc (MK-4176) and MK-2048 in plasma, vaginal fluids and cervical tissue during and after 28 days of continuous use of an IVR containing 182 mg vicriviroc (MK-4176), or 30 mg MK-2048, or 182 mg vicriviroc (MK-4176) + 30 mg MK-2048 (MK-2048A)

Secondary Endpoints:

- Acceptability:
  - Participant report of acceptability including genitourinary and emotional (dis)comfort, awareness/feeling during daily activities, ring insertion/removal issues, and willingness to use in the future

- Adherence
  - Participant report of frequency of study IVR removal/expulsion and duration without IVR inserted in vagina

4.3 Description of Study Population

The study population will include 48 healthy 18-45 year old women (inclusive) who are HIV-uninfected, non-pregnant, sexually abstinent and using adequate contraception, as described in Sections 5.2 and 5.3.

4.4 Time to Complete Accrual

The approximate time to complete study enrollment is expected to be 6-9 months at each site.

4.5 Study Groups

Four study groups are planned. All study groups will be assigned to complete a total of 13 visits.

The four study groups are as follows:

1) VCV (MK-4176) IVR
2) MK-2048 IVR
3) MK-2048A IVR (VCV [MK-4176] + MK-2048)
4) Placebo IVR

4.6 Expected Duration of Participation

The expected duration for participants is approximately 5 weeks, not including the 45 day screening window. No study data will be collected after the 35 Day Final Clinic/Early Termination Visit unless the participant is pregnant at the 35 Day Final Clinic/Early Termination Visit. Participants who are pregnant at the 35 Day Final Clinic/Early Termination Visit may be followed as per Section 9.7, Pregnancy. Participants who have AEs at the 35 Day Final Clinic/Early Termination Visit that have not resolved or stabilized will be followed beyond the 35 Day Final Clinic/Early Termination Visit until a clinically acceptable resolution of the AE(s) is confirmed and documented. Clinical acceptability of resolution will be determined by the site Investigator of Record (IoR) in consultation with the Protocol Safety Review Team (PSRT).

4.7 Sites

Sites selected by the MTN Executive Committee will participate in MTN-027.

5 STUDY POPULATION

5.1 Selection of the Study Population

The inclusion and exclusion criteria in Sections 5.2 and 5.3 will be utilized to ensure the appropriate selection of study participants.

5.1.1 Recruitment

Participants will be recruited from a variety of sources including family planning and gynecology clinics, colleges and universities, online sites, faith communities, as well as community-based locations such as community-based organizations and street-based outreach. Participants also will be referred to the study from other local research projects and other health and social service providers serving the target study population. Recruitment strategies will be guided by input from the site's Community Advisory Group. Recruitment materials will be approved by site Institutional Review Boards/Ethics Committees (IRBs/ECs) prior to use. Site community representatives should advise on these materials before they are submitted to the IRB/EC for review.

5.1.2 Retention

Once a participant is enrolled/randomized in MTN-027, the study site will make every effort to retain the participants in follow-up to minimize possible bias associated with loss-to-follow-up. Each study site will establish and follow standard operating procedures (SOPs) for participant retention. An average retention rate of 95% will be targeted across sites.

5.2 Inclusion Criteria

Participants must meet all of the following criteria to be eligible for inclusion in the study.

1) Born female

   Note: Participants who were female at birth, who now identify as male, will not be excluded so long as they are not on female-to-male transition therapy.

2) Age 18 through 45 years (inclusive) at Screening, verified per site SOPs

3) Able and willing to provide written informed consent to be screened for and take part in MTN-027

4) Able and willing to provide adequate locator information, as defined by the site SOPs

5) HIV-uninfected, based on testing performed by study staff at Screening and Enrollment (per applicable algorithm in Appendix II) and willing to receive results

6) In general good health at Screening and Enrollment, as determined by the site IoR or designee

7) At Screening, participant states willingness to abstain from receptive sexual activity (including penile-vaginal intercourse, anal intercourse, receptive oral intercourse, finger stimulation, and the use of sex toys) for the 5 days prior to the Enrollment Visit and for the duration of study participation

8) Per participant report, using an effective method of contraception at Enrollment, and intending to continue the use of an effective method for the duration of study participation. Effective methods for MTN-027 include: hormonal methods (except contraceptive IVRs), intrauterine device (IUD) inserted at least 28 days prior to enrollment, engages in sex exclusively with women, sterilized (self or partner), and/or sexually abstinent for the past 90 days

9) Women over the age of 21 (inclusive) must have documentation of a satisfactory Pap within the past 3 years prior to Enrollment consistent with Grade 0 according to the Female Genital Grading Table for Use in Microbicide Studies Addendum 1 to the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009), or satisfactory evaluation with no treatment required of Grade 1 or higher Pap result

10) Per participant report at Screening and Enrollment, agrees not to participate in other research studies involving drugs, medical devices, or vaginal products for the duration of study participation

11) Per participant report at Screening, regular menstrual cycles with at least 21 days between menses

Note: This criterion is not applicable to participants who report using a progestin-only method of contraception at Screening, e.g., Depo-Provera or levonorgestrel-releasing IUD nor to participants using continuous combination oral contraceptive pills, as the absence of regular menstrual cycles is an expected, normal consequence in this context.

12) At Screening, participant states a willingness to refrain from inserting any non-study vaginal products or objects into the vagina, including, but not limited to, spermicides, female condoms, diaphragms, contraceptive IVRs, vaginal medications, menstrual cups, cervical caps (or any other vaginal barrier method), douches, lubricants, sex toys (vibrators, dildos, etc.) for the 5 days prior to Enrollment and for the duration of their study participation

5.3 Exclusion Criteria

Women who meet any of the following criteria will be excluded from the study:

1) Participant report of any of the following at Screening and/or Enrollment:
   a. History of adverse reactions to any of the components of the study products
   b. Non-therapeutic injection drug use in the 12 months prior to Screening and Enrollment
   c. Post-exposure prophylaxis (PEP) for HIV exposure within 6 months prior to Enrollment
   d. Pre-exposure prophylaxis (PrEP) for HIV prevention within the 6 months prior to Enrollment
   e. Regular use and/or anticipated regular use during the period of study participation of CYP3A inducer(s) and/or inhibitor(s)
   f. Use and/or anticipated use during the period of study participation of female-to-male transition therapy
   g. Chronic and/or recurrent candidiasis
   h. Gonorrhea, chlamydia and/or syphilis diagnosis in the 6 months prior to Enrollment
   i. Last pregnancy outcome 90 days or less prior to Screening
   j. Currently breastfeeding
   k. Has had a hysterectomy
   l. Intends to become pregnant within the next 3 months
   m. Has plans to relocate away from the study site area in the next 3 months
   n. Current sexual partner is known to be HIV-positive

2) Reports participating in any other research study involving drugs, medical devices, or vaginal products within 60 days or less prior to enrollment

3) At Screening or Enrollment, as determined by the IoR/designee, any significant uncontrolled active or chronic cardiovascular, renal, liver, hematologic, neurologic, gastrointestinal, psychiatric, endocrine, respiratory, immunologic disorder or infectious disease

4) Has any of the following laboratory abnormalities at Screening:
   a. Aspartate aminotransferase (AST) or alanine transaminase (ALT) Grade 1 or higher*
   b. Calculated creatinine clearance less than 60 mL/min by the Cockcroft-Gault formula, where creatinine clearance (female) in mL/min = (140 – age in years) x (weight in kg) x (0.85)/72 x (creatinine in mg/dL)
   c. Hemoglobin Grade 1 or higher*
   d. Platelet count Grade 1 or higher*
   e. White blood count Grade 2 or higher*
   f. Positive HBsAg test result
   g. Positive Anti-HCV test result

h. International normalized ratio (INR) > 1.5 × the site laboratory upper limit of normal (ULN)

Note: Otherwise eligible participants with an exclusionary test result (other than HIV, HBV or HCV) can be re-tested during the screening process. If a participant is re-tested and a non-exclusionary result is documented within 45 days of providing informed consent for screening, the participant may be enrolled.

*As per the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events Version 1.0, December, 2004 (Clarification dated August 2009)

5) Pregnant at either Screening or Enrollment
Note: A documented negative pregnancy test performed by study staff is required for inclusion; however a self-reported pregnancy is adequate for exclusion from screening/enrollment into the study.

6) Diagnosed with urinary tract infection (UTI) at Screening or Enrollment

Note: Otherwise eligible participants diagnosed with UTI during screening will be offered treatment. If within the 45 day screening window treatment is complete and symptoms have resolved the participant may be enrolled.

7) Diagnosed with pelvic inflammatory disease, reproductive tract infection (RTI) or a sexually transmitted infection (STI) requiring treatment per current Centers for Disease Control and Prevention (CDC) guidelines (http://www.cdc.gov/std/treatment/) at Screening or Enrollment

Note: With the exception of gonorrhea, chlamydia and/or syphilis, otherwise eligible participants diagnosed with a RTI during screening will be offered treatment. If within the 45 day screening window treatment is complete and symptoms have resolved the participant may be enrolled.

8) At Enrollment, has a clinically apparent Grade 1 or higher pelvic exam finding (observed by study clinician or designee) per the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009), Addendum 1, Female Genital Grading Table for Use in Microbicide Studies

Note: Cervical friability bleeding associated with speculum insertion and/or specimen collection judged to be within the range of normal according to the clinical judgment of the IoR/designee is considered expected non-menstrual bleeding and is not exclusionary.

9) At Screening, severe pelvic relaxation such that either the vaginal walls or the uterine cervix descend beyond the vaginal introitus with valsalva maneuver or has pelvic anatomy that compromises the ability to adequately assess vaginal safety

10) Has any other condition that, in the opinion of the IoR/designee, would preclude informed consent, make study participation unsafe, complicate interpretation of study outcome data, or otherwise interfere with achieving study objectives.

5.4 Co-enrollment Guidelines

As indicated in Section 5.2, participants should not take part in other research studies involving drugs, medical devices, or vaginal products after the Screening Visit and while taking part in MTN-027. Participation in the following types of studies may be allowed at the discretion of the IoR/designee:

- Participants may take part in ancillary studies approved by MTN-027 Protocol Chair
- Participants who become infected with HIV may take part in observational and/or interventional studies for HIV-positive persons
- Participants who become pregnant may take part in observational studies, including registries

Should any participant report concurrent participation in contraindicated studies after enrolling in MTN-027, the IoR/designee must immediately notify the MTN-028 Management Team and consult the PSRT regarding ongoing product use and other potential safety considerations associated with co-enrollment.

6 STUDY PRODUCT

6.1 Regimen

Each participant will be randomized to one of four study regimens:

Table 8: Study Regimen

Group	N	Group Description
A	12	VCV (MK-4176) IVR, containing 182 mg VCV (MK-4176)
B	12	MK-2048 IVR, containing 30 mg MK-2048
C	12	MK-2048A IVR, containing 182 mg VCV (MK-4176) and 30 mg MK-2048
D	12	Placebo IVR

Each participant will receive an IVR containing 182 mg VCV (MK-4176), 30 mg MK-2048, 182 mg VCV (MK-4176) + 30 mg MK-2048 or placebo. The IVR will be worn for approximately 28 consecutive days. It will be inserted into the vagina, by the participant (or clinician, if necessary) at the Enrollment Visit and removed by participant (or clinician, if necessary) on Study Visit Day 28. The participant will be followed for approximately 7 days following IVR removal.

6.2 Administration

Study participants will be given detailed instructions in the clinic on proper IVR insertion and removal procedures. Hands should be thoroughly washed before and after study IVR insertion and/or removal. Additional details on administration (IVR insertion, removal, procedures in the event of expulsion or loss) will be provided.

6.3 Study Product Formulation

6.3.1 Study IVR

The IVRs containing MK-2048 only and MK-2048/VCV (MK-4176) are white to off-white, opaque; the placebo and VCV (MK-4176)-alone IVRs are clear or translucent in appearance. The IVRs are smooth and flexible. The IVR dimensions are as follows: 54 mm and 4.0 mm, outer diameter and cross-sectional diameter, respectively. The IVRs consist of a closed-ring fiber having two coaxial layers: a core layer and a skin layer. In the combination and VCV only IVRs, the core layer is loaded with VCV (MK-4176). In the MK-2048 and combination IVRs, the skin layer is loaded with MK-2048. The core layer contains 10% MK-4176 and the skin layer contains 30% MK-2048. The VCV (MK-4176) only IVR also contains 0.1 wt% magnesium stearate as a processing aid/lubricant. This is not included in the other IVRs.

The drug substance will include either 182 mg VCV (MK-4176), 30 mg MK-2048, both of these combined or no drug (placebo). VCV (MK-4176) w/w and MK-2048 w/w, alone or in combination are dispersed in an ethylene vinyl acetate (EVA) copolymer delivery device. The IVR is designed to provide sustained release of drug over a 28-day period.

Placebo IVRs contain only EVA polymer and no compound.

6.4 Supply and Accountability

6.4.1 Supply

MSD in Oss, The Netherlands, will manufacture the study IVRs and analyze/release the IVRs under Good Manufacturing Practices (GMP). The MTN-027 pharmaceutical collaborator will package, label and ship all of the study IVRs directly to the Pharmacist of Record (PoR) at each study site.

6.4.2 Storage and Dispensing

IVRs should be stored at 2-8°C. IVRs are dispensed from the pharmacy only to enrolled study participants or to clinic staff on behalf of the participant, upon receipt of a written prescription from an authorized prescriber. If an IVR is dispensed from the pharmacy to clinic staff, then a clinic staff member will subsequently provide the IVR to the study participant in the clinic. A small zip bag will be dispensed from the pharmacy with each

IVR. In the event that the study participant must remove the IVR outside the clinic setting, this bag is available for storage of the used IVR.

Participants will be randomized to receive an IVR containing 182 mg VCV (MK-4176), 30 mg MK-2048, 182 mg VCV (MK-4176) + 30 mg MK-2048 or placebo, in a blinded fashion. The site staff and the participant will not know which product the participant is assigned to receive. Although the appearance may suggest the participant has received either placebo or VCV-only, or the MK-2048 or the combination ring, confirmation of which ring the participant ring assignment will not be known during the trial.

6.4.3 Accountability

Each site PoR is required to maintain complete records of all study IVRs received and subsequently dispensed. All unused study products must be returned to MTN LOC Pharmacist after the study is complete unless otherwise instructed by the MTN LOC Pharmacist. Accountability procedures to be followed will be provided in the MTN-027 Pharmacy Study Product Management Procedures Manual.

6.4.4 Retrieval of Study Product

Table 9: Retrieval of Study Product

	Retrieve Study Product Within:
Permanent discontinuation or temporary hold due to potential HIV seroconversion	24 hours
Permanent discontinuation for any other reason or IoR discretion	5 working days
Temporary hold for reasons with expected duration of greater than 7 days	7 working days
End of Study	2 working days

If a study product hold extends for 7 days or more, and product has not been retrieved as of the seventh day, study staff members must make every effort to retrieve study product as soon as possible.

It is not necessary to retrieve study products from participants for whom study product use is being temporarily held for less than 7 days. However, study products may be retrieved from such participants, to protect their safety, if there is concern that the participant may not comply with clinic staff instructions to refrain from study product use for the duration of the temporary hold.

For all study product holds requiring retrieval of study product(s), if the study product(s) are not retrieved within the timeframe stated in the table above, the MTN-027 PSRT must be informed.

The IVR must be worn for approximately 28 consecutive days at a time. If prolonged use of the study IVR has occurred, attempts must be made to contact the participant and retrieve the study product and the PSRT must be informed.

For each participant, all IVRs remaining in the participant's possession must be retrieved at/by Visit Day 28. If the participant does not bring her study product to this visit, study staff must arrange to retrieve the IVR within 2 business days. If the IVR is not retrieved within that timeframe, the MTN-027 PSRT must be informed.

6.5 Concomitant Medications

Enrolled study participants may use concomitant medications during study participation. All concomitant medications reported throughout the course of the study will be recorded on case report forms designated for that purpose. All prescription medications, over-the-counter preparations, vitamins, nutritional supplements, and herbal preparations will be recorded on forms for concomitant medications.

6.6 Prohibited Medications and Practices

Several concomitant medications/practices will not be permitted. Participants are asked to avoid using certain CYP3A inhibitors and CYP3A inducers. These medications are not recommended because VCV (MK-4176) is a CYP3A substrate. A listing of CYP3A inhibitors and inducers to be avoided are provided in the MTN-027 SSP Manual available at www.mtnstopshiv.org. Please note single dose oral fluconazole for the treatment of vaginal fungal infections is permitted. Participant use of female-to-male transition medications is prohibited. Effects of female-to-male transition medications can include vaginal dryness and thinning of the epithelium which may impact product safety, confound adverse event determination and may impact pharmacokinetic parameters.

Participants will be asked to refrain from using tampons during the first week of study participation (starting at the enrollment visit) and for 24 hours prior to each clinic visit following enrollment. Prohibited non-study intravaginal products and other devices include, but are not limited to, spermicides, female condoms, diaphragms, contraceptive intravaginal rings, vaginal medications, menstrual cups, cervical caps, douches, lubricants, and sex toys (e.g., vibrators, dildos, etc.). Participants are expected to be sexually abstinent for the duration of their trial participation and for 5 days preceding Enrollment, i.e., no receptive intercourse (vaginal, anal, oral and finger simulation). These medications and practices are restricted to protect the integrity of the lower genital tract and reduce the possibility of adverse events due to agents other than the study product.

Participants will be counseled to avoid such use and practices. Participant use of prohibited medications and engagement in prohibited practices will be documented.

Additional information for participants who report the use of prohibited medications can be found in Section 9.3.

7 STUDY PROCEDURES

An overview of the study visit and evaluations schedule is presented in Appendix I. Presented in this section is additional information on visit-specific study procedures. Detailed instructions to guide and standardize procedures across sites are provided in the MTN-027 SSP Manual available at www.mtnstopshiv.org.

7.1 Pre-screening

As part of participant outreach and recruitment strategies, study staff can pre-screen potential study participants at either on-site or off-site locations. During these interactions, study staff may explain the study to potential participants and ascertain elements of presumptive eligibility, (e.g., willingness to use a VR, willingness to be sexually abstinent for the duration of study participation, etc.), to be confirmed at an on-site screening visit. Process information (e.g., number of potential participants contacted, number presumptively eligible) may be recorded and stored at the study site in the absence of written informed consent from potential participants, provided the information is collected in such a manner that it cannot be linked to participant identifiers, unless a waiver is granted from the local IRB. Procedures and documentation will comply with local IRB requirements.

7.2 Visit 1- Screening

Screening can take place up to 45 days prior to Enrollment. Multiple visits may be conducted to complete all required procedures, if necessary. Written informed consent for screening will be obtained before any screening procedures are initiated. For participants who do not meet the eligibility criteria, screening will be discontinued once ineligibility is determined. Detailed information regarding visit windows will be described in the MTN-027 SSP Manual.

Table 10: Visit 1- Screening Visit

Visit 1- Screening Visit		
Component		**Procedures**
Administrative and Regulatory		• Obtain written informed consent • Assess consent form comprehension • Assign participant ID (PTID) • Collect locator information • Collect demographic information • Assess eligibility • Provide reimbursement for study visit • Schedule next visit*
Behavioral		• Provide counseling – HIV pre- and post-test – Protocol requirements (To include adherence, product use and contraceptive counseling, as needed)
Clinical		• Collect medical and menstrual history • Collect concomitant medications • Perform physical examination • Perform pelvic examination • Provide available test results • Treat or prescribe treatment for UTIs/RTIs/STIs or refer for other findings*
Laboratory	**Urine**	• Collect urine – Human Chorionic Gonadotropin (hCG) – Dipstick urinary analysis (UA) – Urine culture*
	Blood	• Collect blood – Complete blood count (CBC) with differential and platelets – HIV-1 serology – HBsAg – Coagulation (INR) – Anti-HCV – Chemistries (AST, ALT, Creatinine) – Syphilis serology
	Genital	• Collect pelvic specimens – Rapid test for Trichomonas – Nucleic Acid Amplification Test (NAAT) for *Neisseria gonorrhoeae* and *Chlamydia trachomatis* (GC/CT) – Vaginal fluid pH* – Potassium hydroxide (KOH) wet mount for candidiasis* – Saline wet mount for bacterial vaginosis (BV)* – Collect Pap test*

* If indicated

7.3 Visit 2- Enrollment (Day 0)

Menses must not coincide with Study Visits 2-6 (Days 0, 1, 2, 3, 7), therefore participant's menstrual cycle must be considered when scheduling Visit 2- Enrollment Visit (Day 0).

Table 11: Enrollment Visit- Visit 2 (Day 0)

Enrollment Visit- Visit 2 (Day 0)		
Component		**Procedures**
Administrative and Regulatory		• Confirm eligibility • Review/update locator information • Randomization • Provide reimbursement for study visit • Schedule next visit*
Behavioral		• Behavioral assessment (See Section 7.8) • Provide counseling – HIV pre- and post-test – HIV/STI risk reduction – Protocol requirements
Clinical		• Review/update medical and menstrual history • Review/update concomitant medications • Document pre-existing conditions • Perform physical examination • Perform pelvic examination • Provide available test results • Treat or prescribe treatment for UTIs/RTIs/STIs or refer for other findings*
Laboratory	**Urine**	• Collect urine – hCG – Dipstick UA* – Urine culture*
	Blood	• Collect blood – CBC with differential and platelets – HIV-1 serology – Chemistries – Plasma archive – PK (Post ring insertion at time points: Hours 1, 2, 4, 6) – Syphilis serology*
	Genital	• Collect pelvic specimens – Cytobrush for flow cytometry – Vaginal fluid for PK (Post ring insertion at time points: Hours 0, 1, 2, 4, 6) – Vaginal fluid pH – Gram stain – Vaginal swab for vaginal biomarkers – Quantitative vaginal culture – KOH wet mount for candidiasis* – Saline wet mount for BV* – Rapid test for Trichomonas* – NAAT for GC/CT*
	Rectal	• Collect rectal specimen – Rectal fluid via sponge for PK ♿
Study Product Supply		• Participants will receive study IVR, study IVR use instructions and will be instructed to self-insert the study IVR, followed by pelvic exam to check placement

* If indicated ♿ To be collected on a subset of participants who opt in

7.4 Follow-up Visits

The following procedures will occur on Days 1, 2, 3, 7, 14, 21, 28, 29, 30, 31, and Day 35 /Final Clinic or Early Termination Visit.

7.4.1 Visits 3-8 (Days 1, 2, 3, 7, 14, 21)

Table 12: Visits 3-8 (Days 1, 2, 3, 7, 14, 21)

Follow-up Visits 3-9 (Days 1, 2, 3, 7, 14, 21)		
Component		**Procedures**
Administrative and Regulatory		• Review/update locator information • Provide reimbursement for study visit • Record/update AEs • Schedule next visit
Behavioral		• Behavioral assessment (See Section 7.8) ϴ • Provide modified counseling – HIV pre- and post-test counseling* – Protocol requirements*
Clinical		• Review/update medical and menstrual history • Review/update concomitant medications • Perform modified physical examination • Perform pelvic examination • Provide available test results • Treat for UTIs/RTIs/STIs or refer for other findings*
Laboratory	**Urine**	• Collect urine – hCG₪* – Dipstick UA* – Urine culture*
	Blood	• Collect blood – PK – HIV-1 serology* – Syphilis serology*
	Genital	• Collect pelvic specimens – Collect vaginal fluid for PK analysis – Vaginal fluid pH♦* – Gram Stain♦ – Vaginal swab for vaginal biomarker assessment♦ – Quantitative vaginal culture♦ – Rapid test for Trichomonas* – KOH wet mount for candidiasis* – Saline wet mount for BV* – NAAT for GC/CT*

* If indicated ♦ Day 3 only, ϴ Day 7, 14, 21 only, ₪ Day 14 only

7.4.2 Visit 9 (Day 28) Ring Removal Visit

The following procedures will occur at Visit Day 28 or the Ring Removal Visit.

Table 13: Visit 9 (Day 28) Ring Removal Visit

Follow-up Visit 9 (Day 28) Ring Removal Visit		
Component		**Procedures**
Administrative and Regulatory		• Review/update locator information • Provide reimbursement • Record/update AEs • Schedule next visit
Behavioral		• Behavioral assessment (See Section 7.8) • Provide counseling – HIV pre- and post-test* – Protocol requirements*
Clinical		• Review/update medical and menstrual history • Review/update concomitant medications • Perform modified physical examination • Perform pelvic examination • Provide available test results • Treat or prescribe treatment for UTIs/RTIs/STIs or refer for other findings*
Laboratory	**Urine**	• Collect urine – hCG – Dipstick UA – Urine culture*
	Blood	• Collect blood – CBC with differential and platelets – Chemistries – PK (Post ring removal at time points: Hours 0, 1, 2, 4, 6) – HIV-1 serology* – Syphilis serology*
	Genital	• Collect pelvic specimens – Collect vaginal fluid for PK analysis (Post ring removal at time points: Hours 0, 1, 2, 4, 6) – Collect cervical tissue for PK analysis – Collect cervical tissue for PD analysis at site(s) with capacity – Cytobrush for flow cytometry – Vaginal fluid pH – Quantitative vaginal culture – Gram stain – Vaginal swab for vaginal biomarker assessment – KOH wet mount for candidiasis* – Saline wet mount for BV* – Rapid test for Trichomonas* – NAAT for GC/CT*
	Rectal	• Collect rectal specimen – Rectal fluid via sponge for PK ♿
Study Product		• Collect IVR

* If indicated ♿ To be collected on a subset of participants who opt in

7.4.3 Visits 10-12 (Days 29, 30, 31)

Table 14: Visits 10-12 (Days 29, 30, 31)

Follow-up Visits 10-12 (Days 29, 30, 31)		
Component		**Procedures**
Administrative and Regulatory		• Review/update locator information • Provide reimbursement for study visit • Record/update AEs • Schedule next visit
Behavioral		• Provide modified counseling – HIV pre- and post-test* – Protocol requirements*
Clinical		• Review/update medical and menstrual history • Review/update concomitant medications • Perform modified physical examination • Perform pelvic examination • Disclosure of available test results • Treat or prescribe treatment for UTIs/RTIs/STIs or refer for other findings*
Laboratory	**Urine**	• Collect urine* – hCG* – Dipstick UA* – Urine culture*
	Blood	• Collect blood – PK – HIV-1 serology* – Syphilis serology*
	Genital	• Collect pelvic specimens – Vaginal fluid pH – Collect vaginal fluid for PK analysis – Rapid test for Trichomonas* – KOH wet mount for candidiasis* – Saline wet mount for BV* – NAAT for GC/CT*

* If indicated

7.4.4 Visit 13 (Day 35) Final Clinic/Early Termination Visit

Table 15: Visit 13 (Day 35) Final Clinic/Early Termination Visit

Visit 13 (Day 35) Final Clinic/ Early Termination Visit		
Component		**Procedures**
Administrative and Regulatory		• Review/update locator information • Provide reimbursement for study visit • Record/update AEs • Schedule next visit*
Behavioral		• Behavioral assessment (See Section 7.8) • In-depth interview • Provide counseling – HIV pre- and post-test – Protocol requirements*
Clinical		• Review/update medical and menstrual history • Review/update concomitant medications • Perform physical examination • Perform pelvic examination • Disclosure of available test results • Treat or prescribe treatment for UTIs/RTIs/STIs or refer for other findings*
Laboratory	**Urine**	• Collect urine – hCG*
	Blood	• Collect blood – CBC with differential and platelets – HIV-1 serology – Chemistries – PK – Syphilis serology*
	Genital	• Collect pelvic specimens – Vaginal fluid pH – Vaginal fluid for PK analysis – Quantitative vaginal culture – Gram stain – Vaginal swab for vaginal biomarker assessment – Rapid test for Trichomonas* – KOH wet mount for candidiasis* – Saline wet mount for BV* – NAAT for GC/CT*
Study Product		• Collect IVR ▲

* If indicated, ▲ If Early Termination Visit and not already performed

7.5 Follow-up Procedures for Participants Who Permanently Discontinue Study Product

7.5.1 Participants Who Become Infected with HIV-1

If a participant becomes infected with HIV-1 after the Enrollment Visit, she will be referred to local care and treatment services and may return to the research clinic for additional counseling and other support services, as needed. Continued study participation would be of no added benefit, thus follow-up visits will be discontinued and the participant will be considered terminated from the study. Participants who seroconvert after randomization may be offered additional laboratory testing (such as HIV RNA and HIV drug resistance testing), as clinically indicated per discussions between IoR and LC. Please reference the MTN-027 SSP Manual for additional details (www.mtnstopshiv.org).

7.5.2 Participants Who Become Pregnant

If a participant becomes pregnant, she will be referred to local health care services and may return to the research clinic for additional counseling, as needed. Continued study participation would be of no added benefit to the participant, thus follow-up visits and procedures will be discontinued and the participant will be considered terminated from the study. A participant who is pregnant at study termination will continue to be followed until the pregnancy outcome is ascertained, see Section 9.7 for additional details. For additional details regarding obtaining pregnancy outcome, please reference the MTN-027 SSP (www.mtnstopshiv.org).

Participants who become pregnant while on study product may be offered enrollment in MTN-016 (www.mtnstopshiv.org), provided their study site is taking part in MTN-016.

7.5.3 Participants Who Permanently Discontinue Study Product for Other Reasons

For participants who permanently discontinue study product use for any other clinician initiated reason other than HIV seroconversion or pregnancy, site investigators may, after consultation with the PSRT and MTN-027 Management Team, decide to discontinue study follow-up visits and procedures. However, participants who permanently discontinue study product use due to an AE must continue to be followed until the resolution or stabilization of the AE is documented.

In the event study follow-up is continued, participants will have the protocol-specified weekly visits through Day 35, specifically those visits at Day 7, Day 14, Day 21, Day 28 and Day 35. Protocol-specified procedures will continue except the following:

- Pelvic exams*
- Collection of blood for safety assessments*
- Collection of samples for PK and PD
- Behavioral assessments
- Protocol requirements counseling will be modified

*Unless required for AE follow-up

The above procedures should be collected/conducted at the visit in which study product is discontinued and omitted thereafter, unless the participant was previously on a temporary hold.

The MTN-027 Management Team, in consultation with the MTN Pharmacology Core, may provide real-time guidance to the site regarding a modified study visit schedule, in an effort to ensure that PK samples are collected at the appropriate time points. Participants' duration of use and timing of study product permanent discontinuation will be factored into the modified schedule. See SSP for additional details.

7.6 Follow-up Procedures for Participants Who are on a Temporary Clinical Study Product Hold

All protocol-specified study visits and procedures will continue except the following:

- Collection of samples for PD
- Behavioral assessment
- Provision of product use/protocol adherence counseling
- Pelvic exams*

*Unless required for AE follow-up

The collection of samples for PK should be collected/conducted at the visit in which study product is temporarily held. The MTN-027 Management Team, in consultation with the MTN Pharmacology Core, will provide real-time guidance to the sites regarding a modified study visit schedule, in an effort to ensure that PK samples are collected at the appropriate time points. Participants' duration of use and timing of study product discontinuation will be factored into the modified schedule. See SSP for additional details.

The behavioral assessment should be collected/conducted at the visit in which study product is temporarily held and omitted thereafter. Completion of these procedures will resume if and when the participant resumes study product use.

7.6.1 Interim Visits

Interim visits may be performed at any time during the study and any procedures may be conducted. All interim contacts and visits will be documented in participants' study records and on applicable CRFs.

7.7 Pharmacokinetics

All enrolled participants randomized will undergo PK specimen collection procedures. These collections will occur at study visits as described in the table below. Blood, vaginal fluid, and cervical biopsies will be collected to assay for VCV (MK-4176) and MK-2048 concentrations.

Blood and pelvic PK specimens should be collected within approximately one hour of each other in the sequence listed below, if a single time point collection is planned. Participants will report ring adherence, including details regarding any ring expulsions including the date and approximate length of time that the ring was out of the vagina. Study staff will record this information. These data will be collected at study visits in which PK assessments are scheduled to occur, see Table 16 below. Staff will also record all PK specimen collection times. The SSP will provide information regarding the study visit windows.

Table 16: PK Specimen Collection Schedule

STUDY VISIT	PK Specimen Collection
Screening	None
Enrollment	Blood (hr 1, 2, 4, 6), Vaginal fluid (hr 0,1, 2, 4, 6) , Rectal fluid ♿ (hr 0)
Day 1	Blood, Vaginal fluid
Day 2	Blood, Vaginal fluid
Day 3	Blood, Vaginal fluid
Day 7	Blood, Vaginal fluid
Day 14	Blood, Vaginal fluid
Day 21	Blood, Vaginal fluid
Day 28	Blood (hr 0, 1, 2, 4, 6), Vaginal fluid (hr 0, 1, 2, 4, 6), Cervical tissue (hr 0), Rectal fluid ♿ (hr 0)
Day 29	Blood, Vaginal fluid
Day 30	Blood, Vaginal fluid
Day 31	Blood, Vaginal fluid
Day 35	Blood, Vaginal fluid

♿ To be collected on a subset of participants who opt-in

7.8 Behavioral Measures

The behavioral measures of this protocol will focus on acceptability of and adherence to the IVR. The primary behavioral objective is to assess the acceptability of the IVR as a new HIV prevention modality in its own right *and* in comparison to other vaginal products used by trial participants in the past. The secondary objective is to understand

how women's experiences with the IVR promote or hinder adherence over 28 days of use.

Acceptability of Vaginal Ring

Microbicide acceptability has been defined as the voluntary sustained use of a microbicide product in the context of alternatives.[30] As described by Morrow and Ruiz [31] (with the addition of insertion and expulsion-associated items appropriate to the IVR modality), microbicide acceptability in MTN-027 will be assessed considering the following factors:

- **Vehicle-associated:** Texture, product scent and color, and desirable/appealing elements of IVR
- **Insertion-associated:** Clarity of instructions for insertion, and ease of insertion
- **Expulsion-associated**: Clarity of instructions for what to do upon expulsion
- **Use-associated:** Odor post-application, lubrication and drying effects post-insertion, desirable/appealing elements of use, changes in hygiene practices secondary to use, genitourinary comfort/discomfort, emotional comfort/discomfort, awareness/feeling during daily activities
- **Related Covariates:** History of vaginal product use, likelihood of future use of IVR, desired changes to the IVR, feelings of control, peace of mind, communication with others (e.g., friends, family or partners) regarding IVR use

Adherence to Study Protocol

We will assess women's adherence to the IVR, as well as to the study protocol guidelines considering the following factors:

- **Expulsion-associated:** adherence to protocol regarding response to expulsion events, reasons for expulsion, type of expulsion (natural vs. voluntary), frequency of expulsion, duration of IVR outside of the vagina during each expulsion event
- **Sexual activity-associated:** Self-efficacy towards abstinence, engagement in sex, number of sexual partners, types of sex (i.e., oral, vaginal, anal), gender of partner(s), frequency of sex
- **Vaginal product-associated:** Self-efficacy towards not using any other vaginal products, use of other vaginal products, frequency, duration, reason
- **Related Covariates:** Concerns regarding medication use, perception of partner(s)' attitudes towards trial participation, stressful life events (e.g., concomitant injury or illness), social network communication

We will use the following assessments to evaluate acceptability and adherence:

1. Behavioral Assessment
2. In-depth interviews

Behavioral Assessment

All participants will complete a CASI questionnaire at Enrollment, Day 7, Day 14, Day 21, Day 28, and finally at Final Clinic Visit. The questionnaire administered at Day 7, Day 14, Day 21 will be brief so as to reduce participant burden yet still capture data over time. CASI questionnaires will be offered at a private computer terminal located at the study site. A few of the survey items in the questionnaire will be asked in a cross-sectional fashion (in a single questionnaire), whereas the primary acceptability and adherence items at the core of the survey will be asked each time. In addition to demographics, these questionnaires will explore, among other topics, participants' experiences using the product, likes and dislikes, experiences of (dis)comfort, and likelihood of future use. (*See acceptability and adherence domains above*).

In-depth Interviews

Each participant will take part in an in-depth interview (IDI) conducted via video conferencing from a central location at the Final Clinic Visit. Using an open-ended interview guide, the in-depth interview will cover questions regarding experiences with product use, participant concerns, and exploration of various factors that may have encouraged or hindered adherence. Immediately following the interview, the interviewer will complete a summary sheet detailing the major points of interest regarding acceptability and adherence elicited in the course of the interview. The interviewer will be blinded to the participant's study product assignment and the interview will be audio recorded and transcribed.

Summary Database

The summary database will be based on the results of each of the assessments noted above and will detail those factors that most influence acceptability and adherence.

Figure 4: Methods of Assessing Acceptability and Adherence

7.9 Adherence Counseling and Assessment

Participants will receive study IVR adherence counseling at the Enrollment Visit and modified counseling at additional follow-up visits. Site staff will counsel participants to refrain from removing the ring (except as directed) and from using prohibited medications and practices as described in Section 6.6. Site staff will also provide counseling for re-insertion in case of ring removal/expulsion.

7.10 Clinical Evaluations and Procedures

Physical exams will include the following assessments:

- General appearance
- Weight
- Vital signs
  - Temperature
  - Pulse
  - Blood pressure
  - Respirations
- Height *
- Abdomen*
- Head, Eye, Ear, Nose and Throat (HEENT) Examination*
- Lymph nodes*
- Neck*
- Heart*
- Lungs*
- Extremities*
- Skin*
- Neurological*

*may be omitted after the Enrollment Visit

Additional clinical assessments may be performed at the discretion of the examining clinician in response to symptoms or illnesses present at the time of the exam.

Pelvic exams will be conducted per guidelines for naked eye inspection described in the WHO/CONRAD Manual for Standardization of Colposcopy for the Evaluation of Vaginal Products, Update 2004, available at http://www.conrad.org/publications-13.html. The required sequence of procedures and specimen collection performed during pelvic exams will be specified in the MTN-027 SSP Manual.

7.11 Laboratory Evaluations

Local Laboratory

- Urine
  - hCG
  - Dipstick UA
  - Urine culture

- Blood
    - Syphilis
    - HIV serology
    - HBsAg
    - INR
    - Anti-HCV
    - CBC with differential and platelets
    - Chemistries
        - Creatinine clearance
        - AST
        - ALT
- Vaginal
    - pH
    - Rapid test for Trichomonas
    - Saline wet mount for BV
    - KOH wet mount for candidiasis
    - Chlamydia and gonorrhea

- Cervical
    - Pap test

<u>Laboratory Center</u>

- Blood
    - Plasma archive
    - Standardized and specialized HIV-1 resistance tests
    - Blood PK

- Genital
    - Gram stain assessment
    - Vaginal fluid for PK
    - Cervical tissue for PK and PD*
    - Quantitative culture
    - Biomarker assessment
    - Cytobrush for flow cytometry

- Rectal
    - Rectal fluid via sponge (in a subset)

<u>Merck & Co. Designated Laboratory</u>

- Study Product
    - Remnant content assessment

*Cervical tissue for PD is only to be collected at site(s) with capacity

7.12 Specimen Collection and Processing

Each study site will adhere to the standards of good clinical laboratory practice in accordance with current DAIDS Laboratory Requirements and the MTN-027 Study Specific Procedures Manual (www.mtnstopshiv.org) for proper collection, processing, labeling, transport, and storage of specimens at the local laboratory. Specimen collection, testing, and storage at the site laboratories will be documented when applicable using the Laboratory Data Management System (LDMS).

In cases where laboratory results are not available due to administrative or laboratory error, sites are permitted to re-draw specimens. Further, as part of quality control, researchers may need to look at short pieces of non-coding repetitive DNA sequence (3-7 base pairs) from blood in the event of sample mix-up. This test will only let researchers know the number of times this short segment is repeated and not specific genes or specific sequences of base pairs. This sequence element does not contain any information about genes, therefore researchers will not be able to identify if participants are predisposed to specific diseases or any other genetic information based on this information. This test will be an important tool for distinguishing whether two samples collected at the same or different time points are likely from the same person. The test will only be used as part of a sample investigation with the knowledge of the site in situations where a known or suspected sample mix-up has occurred. No genetic testing (limited or genome-wide) is planned on leftover samples that are stored for the purposes of future research.

7.13 Specimen Handling

Specimens will be handled in accordance with current requirements for DAIDS Sponsored and/or Funded Laboratories in Clinical Trials. (http://www.niaid.nih.gov/labsandresources/resources/daidsclinrsrch/documents/labpolicy.pdf)

7.14 Biohazard Containment

As the acquisition of HIV and other blood-borne pathogens can occur through contact with contaminated needles, blood, and blood products, appropriate blood and secretion precautions will be employed by all personnel in the drawing of blood and shipping and handling of all specimens for this study as recommended by the CDC and National Institutes of Health (NIH). All biological specimens will be transported using packaging mandated by Code of Federal Regulations (CFR) 42 Part 72. All dangerous goods materials, including diagnostic specimens and infectious substances, must be transported according to instructions detailed in the International Air Transport Association (IATA) Dangerous Goods Regulations. Biohazardous waste will be contained according to institutional, transportation/carrier, and all other applicable regulations.

8 ASSESSMENT OF SAFETY

8.1 Safety Monitoring

Site IoRs are responsible for continuous close safety monitoring of all study participants and for alerting the Protocol Team if unexpected concerns arise. A sub-group of the Protocol Team, including the Protocol Chair, DAIDS Medical Officer, Protocol Safety Physicians, and SCHARP Clinical Affairs Safety Associates and other team designees will serve as the PSRT. The MTN Statistical Data Management Center (SDMC) prepares routine AE and clinical data reports (blinded to treatment assignment) for review by the PSRT, which meets via conference call approximately once per month or as needed throughout the period of study implementation to review safety data, discuss product use management, and address any potential safety concerns.

8.2 Clinical Data and Safety Review

A multi-tiered safety review process will be followed for the duration of this study. The study site investigators are responsible for the initial evaluation and reporting of safety information at the participant level and for alerting the PSRT if unexpected concerns arise. Participant safety is also monitored at the Network level through a series of routine reviews conducted by the SDMC Clinical Affairs staff, the PSRT and study sponsors. Additional reviews may be conducted at each of these levels as dictated by the occurrence of certain events.

MTN SDMC Clinical Affairs staff will review incoming safety data on an ongoing basis. Events identified as questionable, inconsistent, or unexplained will be queried for verification. Adverse event reports submitted in an expedited manner to the DAIDS Safety Office will be forwarded to the DAIDS Medical Officer and SDMC Clinical Affairs staff for review.

The PSRT will meet approximately every month via conference call to review clinical data reports generated by the MTN SDMC. The content, format and frequency of the clinical data reports will be agreed upon by the PSRT and the SDMC in advance of study implementation. In addition to the routine safety data reviews, the PSRT will convene on an ad hoc basis to make decisions regarding the handling of any significant safety concerns. If necessary, experts external to the MTN representing expertise in the fields of microbicides, biostatistics, HIV acquisition and medical ethics may be invited to join the PSRT safety review. A recommendation to pause or stop the trial may be made by the PSRT at this time or at any such time that the team agrees that an unacceptable type and/or frequency of AEs has been observed.

The Study Monitoring Committee (SMC) will review participant safety data as part of their regular reviews (see Section 10.8.1), since no Data and Safety Monitoring Board oversight is planned for MTN-027. The SMC may recommend that the study proceed as

designed, proceed with design modifications, or be discontinued. Members of the SMC will be independent investigators with no interest (financial or otherwise) in the outcomes of this study. If at any time a decision is made to discontinue enrollment and/or study product use in all participants, DAIDS will notify the FDA and the Site IoR will notify the responsible IRB expeditiously.

8.3 Adverse Events Definitions and Reporting Requirements

8.3.1 Adverse Events

An AE is defined as any untoward medical occurrence in a clinical research participant administered an investigational product and which does not necessarily have a causal relationship with the investigational product. As such, an AE can be an unfavorable or unintended sign (including an abnormal laboratory finding, for example), symptom or disease temporally associated with the use of an investigational product, whether or not considered related to the product. This definition is applied to all study groups, and is applied to all groups beginning at the time of enrollment (i.e., once a participant has been randomized). The term “investigational product” for this study refers to all study products.

Study participants will be provided instructions for contacting the study site to report any untoward medical occurrences they may experience. In cases of potentially life-threatening events, participants will be instructed to seek immediate emergency care. Where feasible and medically appropriate, participants will be encouraged to seek evaluation where a study clinician is based, and to request that the clinician be contacted upon their arrival. With appropriate permission of the participant, whenever possible, records from all non-study medical providers related to untoward medical occurrences will be obtained and required data elements will be recorded on study CRFs. All participants reporting an untoward medical occurrence will be followed clinically until the occurrence resolves (returns to baseline) or stabilizes.

Study site staff will document in source documents and the appropriate AE Log CRF all AEs reported by or observed in enrolled study participants regardless of severity and presumed relationship to study product. AE severity will be graded per the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009) and the Female Genital Grading Table for Use in Microbicide Studies (Addendum 1 to the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009)), except that asymptomatic BV and asymptomatic candidiasis will not be reportable AEs. In addition, changes in genital bleeding judged to be related to a woman's contraceptive use will not be considered an AE. In cases where a genital AE is covered in both tables, the Female Genital Grading Table for Use in Microbicide Studies will be the grading scale utilized.

8.3.2 Serious Adverse Events

SAEs will be defined by the Manual for Expedited Reporting of Adverse Events to DAIDS (Version 2.0, January 2010), as AEs occurring at any dose that:

- Results in death
- Is life-threatening
- Requires inpatient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect
- Is an important medical event that may not result in death, be immediately life-threatening, or require hospitalization but may jeopardize the participant or require intervention to prevent one of the outcomes listed in the definition above.

8.3.3 Adverse Event Relationship to Study Product

Relatedness is an assessment made by a study clinician of whether or not the event is related to the study agent. Degrees of relatedness will be categorized according to current DAIDS-approved guidelines. Per the Manual for Expedited Reporting of Adverse Events to DAIDS (Version 2.0, January 2010), the relationship categories that will be used for this study are:

- *Related:* There is a reasonable possibility that the AE may be related to the study agent(s)
- *Not Related:* There is not a reasonable possibility that the AE is related to the study agent(s)

8.4 Expedited Adverse Event Reporting Requirements

8.4.1 Adverse Event Reporting to DAIDS

Requirements, definitions and methods for expedited reporting of Adverse Events (AEs) are outlined in Version 2.0 of the DAIDS Expedited Adverse Event (EAE) Manual, which is available on the Regulatory Support Center (RSC) website at http://rsc.tech-res.com/safetyandpharmacovigilance/.

The DAIDS Adverse Experience Reporting System (DAERS), an internet-based reporting system, must be used for expedited AE reporting to DAIDS. In the event of system outages or technical difficulties, expedited AEs may be submitted via the DAIDS EAE Form. For questions about DAERS, please contact DAIDS-ES at DAIDS-ESSupport@niaid.nih.gov. Site queries may also be sent from within the DAERS application itself.

Where DAERS has not been implemented, sites will submit expedited AEs by documenting the information on the current DAIDS EAE Form. This form is available on the RSC website, http://rsc.tech-res.com/safetyandpharmacovigilance/. For questions about EAE reporting, please contact the RSC (DAIDSRSCSafetyOffice@tech-res.com).

8.4.2 Reporting Requirements for this Study

The SAE Reporting Category, as defined in Version 2.0 of the DAIDS EAE Manual, will be used for this study. The study agents for which expedited reporting are required are VCV (MK-4176) IVR, MK-2048 IVR, MK-2048A IVR, placebo IVR.

8.4.3 Grading Severity of Events

The most current Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events (DAIDS AE Grading Table), Version 1.0, Dec 2004 (clarification dated August 2009), and the Female Genital Grading Table for Use in Microbicide Studies (Addendum 1 to the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004, clarification dated August 2009) will be used and are available on the RSC website at http://rsc.tech-res.com/safetyandpharmacovigilance/.

8.4.4 Expedited AE Reporting Period

The expedited AE reporting period for this study begins at enrollment (i.e., randomization) and continues through the participant's termination from the study.

After the protocol-defined AE reporting period, unless otherwise noted, only Suspected, Unexpected Serious Adverse Reactions (SUSARs) as defined in Version 2.0 of the EAE Manual will be reported to DAIDS if the study staff become aware of the events on a passive basis (from publicly available information).

8.5 Pregnancy and Pregnancy Outcomes

Pregnant women are excluded from this study.

Pregnancy-related data will be collected using pregnancy CRFs for all pregnancies detected during the study. Pregnancy outcomes will not be expeditiously reported to Merck or the DAIDS Medical Officer (MO) unless there is an associated AE in the pregnant participant that meets expedited reporting criteria or the pregnancy results in a congenital anomaly meeting the Manual for Expedited Reporting of EAEs to DAIDS (Version 2.0, January 2010) guidelines for expedited reporting.

8.6 Regulatory Requirements

Information on all reported AEs will be included in reports to the FDA and other applicable government and regulatory authorities. Site IoRs/designees will submit AE and any relevant safety information in accordance with local regulatory requirements.

8.7 Social Harms Reporting

Although study sites make every effort to protect participant privacy and confidentiality, it is possible that participants' involvement in the study could become known to others and that social harms may result. Social harms that are judged by the IoR/designee to be serious or unexpected will be reported to the PSRT and responsible site IRBs according to their individual requirements.

9 CLINICAL MANAGEMENT

Guidelines for clinical management and temporary product hold/permanent discontinuation of study product are outlined in this section. In general, the IoR/designee has the discretion to hold study product temporarily at any time if s/he feels that continued product use would be harmful to the participant or interfere with treatment deemed clinically necessary. Unless otherwise specified below, the IoR/designee should immediately consult the PSRT for further guidance on resuming study product, continuing the hold temporarily, or progressing to permanent discontinuation of study product. The IoR/designee will document all temporary product holds and permanent discontinuations on applicable CRFs.

9.1 Grading System

AE severity grading is described in Section 8.3.1.

9.2 Dose Modification Instructions

No dose modifications will be undertaken in this study.

9.3 General Criteria for Temporary Hold and Permanent Discontinuation of Study Product

Temporary Hold

The IoR/designee must consult the PSRT on all temporary product holds for further guidance on resuming product use, continuing the temporary hold, or progressing to permanent discontinuation. If the underlying reason for the temporary hold later resolves, the IoR/designee should consult the PSRT to resume product use at that time. A participant will be temporarily held from product use by the IoR/designee for any of the following reasons:

- Report of use of prohibited medications and medications to be avoided as described in Section 6.6; product use may resume when the participant reports no longer taking the prohibited medication, provided other reasons for temporary product hold/permanent discontinuation do not apply.
- Participant is unable or unwilling to comply with required study procedures, or otherwise might be put at undue risk to their safety and well-being by continuing product use, according to the judgment of the IoR/designee.

Permanent Discontinuation

A participant will be permanently discontinued from product use by the IoR/designee for any of the following reasons:

- Exposure to or acquisition of HIV infection; for those who acquire HIV study product should be held beginning immediately upon recognition of the first reactive rapid HIV test
- Pregnancy
- Breastfeeding
- Participant is unable or unwilling to comply with required study procedures, or otherwise might be put at undue risk to their safety and well-being by continuing product use, according to the judgment of the IoR/designee.

9.4 Temporary Product Hold/Permanent Discontinuation in Response to Adverse Events

Grade 1 or 2

In general, a participant who develops a Grade 1 or 2 AE as defined by the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009) or the Female Genital Grading Table for Use in Microbicide Studies, Addendum 1 to the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009) regardless of relationship to study product that is not specifically addressed in Section 9.5 below may continue product use. If the IoR/designee opts to temporarily hold study product the PSRT must be notified.

Grade 3

For participants who develop a Grade 3 AE as defined by the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009) or the Female Genital Grading Table for Use in Microbicide Studies, Addendum 1 to the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009) that is judged by the IoR/designee to be unrelated to study product, a temporary product hold must be initiated and the PSRT must be notified.

The study product must be permanently discontinued for participants who develop a Grade 3 AE judged by the IoR/designee to be related.

Grade 4

For participants who develop a Grade 4 as defined by the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009) or the Female Genital Grading Table for Use in Microbicide Studies, Addendum 1 to the DAIDS Table for Grading Adult and Pediatric Adverse Events, Version 1.0, December 2004 (Clarification dated August 2009) AE regardless of relationship to study product, study product must be permanently discontinued.

9.5 Other Clinical Events

Genital Tract Infections

Management of sexually transmitted infections (STIs) and other forms of vaginitis and cervicitis will be in accordance with current CDC guidelines (http://www.cdc.gov/std/treatment/). When clinically appropriate, investigators should use oral or parenteral medications when at all possible to avoid intravaginal medication use. Observed single dose treatment should be provided whenever possible, per clinician discretion.

- Study IVR need not be held in the event of an STI/RTI requiring treatment, unless other temporary product hold/permanent discontinuation guidelines apply.

- Should the IoR/designee determine that a temporary hold is warranted, for reasons not noted below, consultation with the PSRT is required.
  Note: One exception, per management guidelines below, consultation with the PSRT is not required for the initial management of deep epithelial disruption (ulceration)

If a suspected finding is reported by a participant between scheduled visits, an interim visit may be scheduled at the discretion of the site investigator.

Management of genital events observed at scheduled or interim visits will be in accordance with the following:

Superficial epithelial disruption (abrasion/peeling)

- Continue study IVR use
- Perform naked eye evaluation
- Re-evaluate in 3-5 days
- If condition worsens, temporarily hold study IVR use and consult the PSRT; otherwise continue study IVR use

Deep epithelial disruption (ulceration)

- Temporarily hold study IVR for deep epithelial disruption confirmed by site investigator
- Re-evaluate in 3-5 days and resume study IVR use if resolved

- If resolved at that time, may resume study IVR use. If unresolved at this second reevaluation, continue temporary product hold, consult with PSRT regarding permanent discontinuation, and provide care per local standard. If unresolved at 3-5 days, re-evaluate within 2-3 days.
- If there is reoccurrence with no identified etiology, continue temporary product hold and consult the PSRT regarding permanent discontinuation

Localized erythema or edema: area of less than 50% of vulvar surface or combined vaginal and cervical surface

- Continue study IVR use
- Perform naked eye evaluation
- If asymptomatic, re-evaluate at next regularly scheduled visit
- If symptomatic, re-evaluate in 3-5 days
- If worsened significantly, temporarily hold study IVR use and consult the PSRT; otherwise continue study IVR use

Generalized erythema or severe edema: area of more than 50% of vulvar surface or combined vaginal and cervical surface affected by erythema

- Temporarily hold study IVR
- Perform naked eye evaluation
- Re-evaluate in 3-5 days and resume study IVR use if resolved
- If unresolved at 3-5 days, re-evaluate within 2-3 days. If resolved at that time may resume use. Safety follow up by phone after IVR use has resumed may be performed if indicated. If unresolved at this second reevaluation, continue temporary product hold, consult with PSRT regarding permanent discontinuation, and provide care per local standard

Unexpected genital bleeding

- Continue study IVR use (at study clinician's discretion)
- Perform naked eye evaluation
- If determined to be due to deep epithelial disruption, refer to guidelines above; otherwise continue study IVR use

Genital petechia(e)

- Continue study IVR use
- Perform naked eye evaluation
- No further evaluation or treatment is required

Genital ecchymosis

- Continue study IVR use
- Perform naked eye evaluation
- No further evaluation or treatment is required

9.6 HIV-1 Infection

A participant who has a positive test for HIV must have study product permanently discontinued and will be terminated, as per Section 7.5.1.

9.7 Pregnancy

All study participants are required to be sexually abstinent during MTN-027 participation.

Pregnancy testing will be performed at scheduled study visits and participants will be encouraged to report all signs or symptoms of pregnancy to study staff. The IoR/designee will counsel any participant who becomes pregnant regarding possible risks to the fetus according to site SOPs. The IoR/designee also will refer the participant to all applicable services; however, sites will not be responsible for paying for pregnancy-related care.

A participant who becomes pregnant during the course of the study will have study product discontinued and will be terminated from the study, as per Section 7.5.2. A participant who is pregnant at study termination will continue to be followed until the pregnancy outcome is ascertained (or, in consultation with the PSRT, it is determined that the pregnancy outcome cannot be ascertained). Pregnancy outcomes will be reported on relevant CRFs; outcomes meeting criteria for EAE reporting also will be reported on EAE forms.

A participant who becomes pregnant during the course of study participation may be offered participation in MTN-016, HIV Prevention Agent Pregnancy Exposure Registry: EMBRACE Study, at sites participating in MTN-016. This registry study captures pregnancy outcomes as well as infant health information, (including growth), to evaluate the safety and teratogenic risks of microbicide and oral PrEP exposure in pregnancy. In the event that a study site is not taking part in MTN-016, participants may be contacted to collect the outcome of pregnancies up to one year after the birth of the infant.

9.8 Criteria for Early Termination of Study Participation

Participants may voluntarily withdraw from the study for any reason at any time. The IoR/designee also may withdraw participants from the study to protect their safety and/or if they are unwilling or unable to comply with required study procedures, after consultation with the PSRT. Participants also may be withdrawn if Merck & Co., NIAID, MTN, government or regulatory authorities, including the FDA and Office for Human Research Protections (OHRP), or site IRBs/ECs terminate the study prior to its planned end date. Every reasonable effort is made to complete a final evaluation of participants who withdraw or are withdrawn from the study prior to completing follow-up. Study staff members will record the reason(s) for all withdrawals in participants' study records. In the event that participants who voluntarily withdraw from the study wish to re-join the

study, they may resume product use (if applicable) and follow-up through their originally scheduled study exit date, pending consultation with the PSRT.

10 STATISTICAL CONSIDERATIONS

10.1 Overview and Summary of Design

This is a multi-site, single-blind, four arm, 1:1:1:1 randomized, placebo-controlled trial to assess and compare the safety and PK of IVRs containing 182 mg VCV (MK-4176), or 30 mg MK-2048, or 182 mg VCV (MK-4176) + 30 mg MK-2048, when used continuously for 28 days by healthy, HIV-uninfected, sexually abstinent women, as compared with a placebo IVR. A total of approximately 48 women (12 in each arm) will be randomized.

10.2 Study Endpoints

Primary endpoints

Consistent with the primary study objective to assess and compare safety of ethylene-vinyl acetate IVRs containing 182 mg VCV (MK-4176), or 30 mg MK-2048, or 182 mg VCV (MK-4176) + 30 mg MK-2048, when used continuously for 28 days by healthy, HIV-uninfected, sexually abstinent women, as compared with the placebo IVR, the primary safety endpoints are the proportion of women in each of the four IVR regimens with:

- Evidence of a Grade 1 or higher genitourinary events as defined by the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Version 1.0, Dec 2004 (Clarification dated August 2009), Addendum 1, (Female Genital Grading Table for Use in Microbicide Studies) judged to be related to study product
- Evidence of a Grade 2 or higher adverse events as defined by the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, Version 1.0, Dec 2004 (Clarification dated August 2009)

Consistent with the primary study objective to examine local and systemic pharmacokinetics of VCV (MK-4176) and MK-2048 in vaginal fluid, plasma and cervical tissue during and after 28 days continuous use of an ethylene-vinyl acetate (EVA) IVR containing 182 mg VCV (MK-4176), or 30 mg MK-2048, or 182 mg VCV (MK-4176) + 30 mg MK-2048 the pharmacokinetic endpoints will be:

- Local and systemic concentrations of VCV (MK-4176) and MK-2048 in vaginal fluid, cervical tissue, plasma and rectal fluid respectively, during and after 28 days of continuous use of an IVR containing 182 mg VCV (MK-4176), or 30 mg MK-2048, or 182 mg VCV (MK-4176) + 30 mg MK-2048

Secondary endpoints

Consistent with the secondary objective to evaluate the acceptability of the study IVR in HIV-uninfected sexually abstinent women over 28 days of use, the following endpoint will be assessed:

- Participant report of acceptability including genitourinary and emotional (dis)comfort, awareness/feeling during daily activities, ring insertion/removal issues, and willingness to use in the future

Consistent with the secondary objective to adherence to the study IVR in HIV-uninfected sexually abstinent women over 28 days of use, the following endpoint will be assessed:

- Participant report of frequency of study IVR removal/expulsions and duration without IVR inserted in vagina

10.3 Primary Study Hypothesis

MTN-027 hypothesizes that the IVRs containing VCV (MK-4176) and/or MK-2048 will be as safe and as well-tolerated as the IVR containing placebo.

10.4 Sample Size and Power Calculations

10.4.1 Safety Endpoints

The proposed total sample size is approximately N=48 women randomized into 4 arms in a 1:1:1:1 ratio giving 12 women per group. This sample size is based upon the size of similar Phase 1 studies of vaginal microbicide products.

As a means to characterize the statistical properties of this study the table below presents the probability of observing zero, at least one, and two or more safety endpoints among the 12 women in each group for various "true" event rates:

Table 17: Analysis of Safety Event Frequency

Event Rate	P (0 events | n=12)	P (≥1 event | n=12)	P (≥2 events | n=12)
1%	88.6	11.4	0.62
5%	54.0	46.0	11.8
10%	28.2	72.0	34.0
15%	14.2	85.8	55.7
25%	3.2	96.8	84.2
35%	0.7	99.4	95.8
45%	0.1	99.9	99.2

An alternative way of describing the statistical properties of the study design is in terms of the 95% confidence interval for the true rate based on the observed data. The table below shows the exact 2-sided 95% confidence intervals for the probability of an event based on a particular observed rate. If none of the 12 participants receiving a treatment regimen experience a safety event, the 95% exact 2-sided upper confidence bound for the true rate of such events in a particular arm of the study is 26.4%.

Table 18: Exact 2-sided 95% Confidence Intervals Based on Observing a Particular Rate of Safety Endpoints for Arms of Size 12

Observed event rate	Confidence interval (%)
0/12	0.0, 26.4
1/12	0.2, 38.4
2/12	2.1, 48.4

An additional aim of the study is to compare the safety between each of the drug containing IVR arms of the study and the placebo IVR. Assuming a one-sided test with α=0.05 and 90% power, the table below provides the difference in the rates of safety events (proportion of women experiencing the safety event of interest) between a drug containing IVR arm and the placebo IVR arm that is detectable with 90% power for a given rate in the placebo IVR arm. For example, if the true rate of a given toxicity endpoint in the placebo IVR arm is 16.7% (2 of 12 women experiencing a safety event), the proposed sample size provides 90% power to exclude safety endpoint rates greater than 79.0% (72.6% with 80% power). Hence, while comparisons will be made between the drug containing IVR arms of the study and the placebo IVR arm, the study will only have power to detect very large differences in safety event rates.

Table 19: Difference in the Rates of Safety Events

Rate in Placebo IVR Arm	Rate in a Drug Containing IVR Arm Detectable with 90% Power
8.3%	69.3%
16.7%	79.0%
25.0%	87.0%
33.3%	93.4%
41.7%	99.2%

10.4.2 Pharmacokinetic Endpoints

Pharmacokinetic endpoints will include mean and variance concentration-time course, maximum concentration (C_{max}), and time to maximum concentration (T_{max}), half-life (T-1/2), and steady-state concentrations, assessed in blood and vaginal swabs for each study drug. The mean and variance in cervical tissue concentration will be measured at day 28. The sample size of the study is driven by the safety endpoints as described above.

10.4.3 Acceptability Endpoints

Microbicide acceptability has been defined as the voluntary sustained use of a microbicide product in the context of alternatives.[30] As described by Morrow and Ruiz [31](with the addition of insertion and expulsion-associated items appropriate to the IVR

modality), microbicide acceptability in MTN-027 will be assessed considering the following factors:

- **Vehicle-associated:** Texture, product scent and color, and desirable/appealing elements of IVR
- **Insertion-associated:** Clarity of instructions for insertion, and ease of insertion
- **Expulsion-associated**: Clarity of instructions for what to do upon expulsion
- **Use-associated:** Odor post-application, lubrication and drying effects post-insertion, desirable/appealing elements of use, changes in hygiene practices secondary to use, genitourinary comfort/discomfort, emotional comfort/discomfort, awareness/feeling during daily activities
- **Related Covariates:** History of vaginal product use, likelihood of future use of IVR, desired changes to the IVR, feelings of control, peace of mind, communication with others (e.g., friends, family or partners) regarding IVR use

Several components of acceptability (e.g., ease of insertion and removal, genitourinary discomfort, awareness/feeling the study IVR during daily activities, emotional comfort wearing the ring continuously for 28 days, willingness to use an HIV protective ring in the future, if one were available) will be used to assess overall acceptability. Each component will be assessed by a combination of dichotomous measures and rating scales where women will be categorized into (1) those reporting no acceptability issues during the 28 days of study IVR use (e.g., those reporting no genitourinary discomfort) and (2) those reporting at least one issue during the 28 days of study IVR use (e.g., those reporting some discomfort), and if yes, the intensity or severity of the issue (on a scale from 1 to 10). An acceptability endpoint is defined as a negative report by a participant, on any of the above components for acceptability. A sample size of 48 women will provide a precision of 13.0% (i.e., half the width of the 95% confidence interval) assuming an observed acceptability of 75%.

10.4.4 Adherence Endpoints

Adherence will be measured by the percentage of women who keep the IVR inserted at all times in the vagina over the course of 28 days. A sample size of 48 women will provide a precision of 13.0% (i.e., half the width of the 95% confidence interval) assuming an observed adherence of 75%.

We will assess women's adherence to the IVR, as well as to the study protocol guidelines (i.e., remaining sexually abstinent for trial duration), considering the following factors:

- **Expulsion-associated:** adherence to protocol regarding response to expulsion events, reasons for expulsion, type of expulsion (natural vs. voluntary), frequency of expulsion, duration of IVR outside of the vagina during each expulsion event
- **Sexual activity-associated:** Self-efficacy towards abstinence, engagement in sex, number of sexual partners, types of sex (i.e., oral, vaginal, anal), gender of partner(s), frequency of sex

- **Vaginal product-associated:** Self-efficacy towards not using any other vaginal products, use of other vaginal products, frequency, duration, reason
- **Related Covariates:** Concerns regarding medication use, perception of partner(s)' attitudes towards trial participation, stressful life events (e.g., concomitant injury or illness), social network communication

10.5 Participant Accrual, Follow-up and Retention

Based on previous studies of vaginal products with similar eligibility requirements, the accrual of 48 eligible participants will take approximately 6-9 months. Women lost to follow-up and/or on temporary hold or permanent product discontinuation will not be replaced. However, every effort will be made to complete their regularly scheduled safety evaluations. Each site will target retention of 95% of enrolled participants over the follow-up period.

10.6 Randomization

Participants will be randomized in a 1:1:1:1 ratio to the four arms of the study. Study arm randomization will be stratified by site to ensure balanced assignment of products at each site. This randomization scheme will be generated and maintained by the MTN SDMC and will be specified in the SSP manual.

10.7 Blinding

Participants will be blinded to the treatment assignment. All IVRs will be individually packaged and labeled.

There are no circumstances under which it is expected that unblinding of the participant will be necessary for the provision of medical treatment or to otherwise protect the safety of study participants. If, however, an Investigator feels that specific product knowledge is necessary to protect participant safety, the Investigator will notify the PSRT to consider and rule upon the request.

10.8 Data and Safety Monitoring and Analysis

10.8.1 Study Monitoring Committee

No Data and Safety Monitoring Board oversight is planned for this study. The MTN SMC will conduct interim reviews of study progress, including rates of participant accrual, retention, completion of primary and main secondary endpoint assessments, study or lab issues, and, in a closed report, safety data by arm of the study. These reviews will take place approximately every 4-6 months, and as needed. At the time of these reviews, or at any other time, the SMC may recommend that the study proceed as designed, proceed with design modifications, or be discontinued.

Note: Safety data from MTN-027 will be shared with the MTN-028 SMC.

10.8.2 Primary Analysis

When the use of descriptive statistics to assess group characteristics or differences is required, the following methods will be used: for categorical variables, the number and percent in each category; for continuous variables, the mean, median, standard deviation, quartiles and range (minimum, maximum). Within-treatment group assessment of the change from the baseline measurement to a follow-up measurement will be analyzed using McNemar's test (for categorical response variables) or the paired t-test or Wilcoxon signed-ranks test (for continuous variables). When use of formal testing to assess differences between users of the placebo IVR and users of one of the drug containing IVRs is required, the following methods will be used: for binomial response variables, chi-square tests and logistic regression (or exact testing methods); for continuous variables, t-tests and linear regression or nonparametric methods if data are non-Normal.

To assess the adequacy of the randomization, participants in each of the four arms will be compared for baseline characteristics including demographics and laboratory measurements using descriptive statistics. Due to the small sample size, formal comparisons will not be done.

Safety Endpoints

All visits in which a woman has been exposed to the study product will be included in the primary analyses of safety. Secondary intent to treat analyses may also be performed. To assess genitourinary safety, the number and the percentages of participants experiencing each safety endpoint (see Section 10.2) will be tabulated by study arm. Each participant will contribute once in each category (i.e., only for highest severity AE for each participant) for the calculation of event rates. Exact binomial confidence intervals will be calculated for each safety endpoint for each arm and Fisher's Exact test used to test for differences in event rates between each drug containing IVR arm and the placebo IVR arm.

Pharmacokinetic Analysis

Blood and CVF will be analyzed for routine PK parameters - C_{max}, T_{max}, Css, half-life and AUC - and described using descriptive statistics. Css will be described for cervical biopsies following ring removal. Measures of drug exposure (e.g., AUC, C_{max}) will be compared between single drug and combination drug arms and concentration differences will be analyzed.

10.8.3 Secondary Analyses

Acceptability

To assess acceptability of the study IVR, the number and percentage of participants experiencing at least one negative report of acceptability, including genitourinary discomfort and ring insertion/removal issues will be presented. This binomial proportion

will be used to assess the acceptability of the study IVR along with its corresponding 95% confidence interval.

The acceptability analysis will be supplemented by presenting the above proportion by randomization arm along with its corresponding 95% confidence interval.

Adherence

To assess adherence of women to the IVR, the proportion of participants who kept the study IVR inserted at all times during the 28 days will be calculated along with a 95% confidence interval. For women who were not fully adherent, the number of removal/expulsion events and average duration of these events will be reported. Additionally, the average cumulative period of time during the study period when the study IVR was outside the vagina will be calculated. Number, type and circumstances of expulsions (voluntary and involuntary) will be described.

10.8.4 Missing Data

We are targeting a retention rate of 95% over the 35 day study period. Based on previous MTN trials, we expect to have minimal missing data. If missing data rates are higher than anticipated (over 10%), robust methods such as nonparametric tests and generalized estimating equations (GEE) using all available baseline predictors of the missing outcomes as covariates will be used to obtain less biased estimates of the treatment effect.

11 DATA HANDLING AND RECORDKEEPING

11.1 Data Management Responsibilities

Study CRFs will be developed by the MTN SDMC in conjunction with the protocol team. Electronic study questionnaires (CASI questionnaires) will be developed by the protocol Behavioral Scientist in conjunction with the protocol team. Quality control reports and queries routinely will be generated and distributed by the SDMC to the study sites for verification and resolution. As part of the study activation process, each study site must identify all CRFs to be used as source documents. CRF data are transferred to the MTN SDMC, entered, and cleaned using the DataFax data management system. CASI questionnaire data are entered directly into and stored on a secure MTN SDMC-hosted web server.

11.2 Source Documents and Access to Source Data/Documents

All study sites will maintain source data/documents in accordance with current DAIDS policies. (http://rsc.tech-res.com/policiesandregulations/)

Each IoR/designee will maintain, and store securely, complete, accurate and current study records throughout the study. In accordance with U.S. regulations regarding testing investigational products, the IoR/designee will maintain all study documentation for at least two years following the date of marketing approval for the study products being tested for the indication in which they were studied. If no marketing application is filed, or if the application is not approved, the records will be retained for two years after the investigation is discontinued and the US FDA is notified.

Study records must be maintained on site for the entire period of study implementation. Thereafter, instructions for record storage will be provided by DAIDS. No study records may be moved to an off-site location or destroyed prior to receiving approval from DAIDS.

11.3 Quality Control and Quality Assurance

All study sites will conduct quality control and quality assurance procedures in accordance with current DAIDS policies. (http://rsc.tech-res.com/policiesandregulations/)

12 CLINICAL SITE MONITORING

Study monitoring will be carried out by Pharmaceutical Product Development, Inc. (PPD) (Wilmington, NC) in accordance with current DAIDS policies. Study monitors will visit the site to do the following:

- Review informed consent forms, procedures, and documentation
- Assess compliance with the study protocol, Good Clinical Practices (GCP) guidelines, and applicable regulatory requirements (US and non-US), including CFR Title 45 Part 46 and Title 21 Parts 50, 56, and 312
- Perform source document verification to ensure the accuracy and completeness of study data
- Verify proper collection and storage of biological specimens
- Verify proper storage, dispensing, and accountability of investigational study products
- Assess implementation and documentation of internal site quality management procedures

The IoR/designee will allow study monitors to inspect study facilities and documentation (e.g., informed consent forms, clinic and laboratory records, other source documents, CRFs), as well as observe the performance of study procedures. The IoR/designee also will allow inspection of all study-related documentation by authorized representatives of the MTN LOC, Merck & Co., SDMC, LC, NIAID, FDA, OHRP, IRBs/ECs and other local

and US regulatory authorities. A site visit log will be maintained at the study site to document all visits.

13 HUMAN SUBJECTS PROTECTIONS

Site investigators will make efforts to minimize risks to participants. Participants and study staff members will take part in a thorough informed consent process. Before beginning the study, the IoR/designee will have obtained IRB approval and the protocol will have been submitted to the FDA. The IoR/designee will permit audits by the NIH, Merck & Co., the FDA, OHRP, MTN LOC, IRBs/ECs, SDMC, and other local and US regulatory authorities or any of their appointed agents.

13.1 Institutional Review Boards

Each participating institution is responsible for assuring that this protocol, the associated site-specific informed consent forms, and study-related documents (such as participant education and recruitment materials) are reviewed by an IRB responsible for oversight of research conducted at the study sites. Any amendments to the protocol must be approved by DAIDS and the responsible IRBs prior to implementation.

Subsequent to the initial review and approval, the responsible IRBs must review the study at least annually. Each IoR/designee will make safety and progress reports to the IRBs at least annually and within three months after study termination or completion. These reports will include the total number of participants enrolled in the study, the number of participants who completed the study, all changes in the research activity, and all unanticipated problems involving risks to human subjects or others. In addition, the results of all SMC reviews of the study will be provided to the IRBs. Study sites will submit documentation of continuing review to the DAIDS Protocol Registration Office in accordance with the DAIDS Protocol Registration Policy and Procedures Manual.

13.2 Protocol Registration

Prior to implementation of this protocol, and any subsequent full version amendments, each site must have the protocol and the protocol consent forms approved, as appropriate, by their local IRB and any other applicable regulatory entity (RE). Upon receiving final approval, sites will submit all required protocol registration documents to the DAIDS Protocol Registration Office (PRO) at the Regulatory Support Center (RSC). The DAIDS PRO will review the submitted protocol registration packet to ensure that all of the required documents have been received.

Site-specific informed consent forms (ICFs) will be reviewed and approved by the DAIDS PRO and sites will receive an Initial Registration Notification from the DAIDS PRO that indicates successful completion of the protocol registration process. A copy of the Initial Registration Notification should be retained in the site's regulatory files.

Upon receiving final IRB and any other applicable RE approval(s) for an amendment, sites should implement the amendment immediately. Sites are required to submit an amendment registration packet to the DAIDS PRO at the RSC. The DAIDS PRO will review the submitted protocol registration packet to ensure that all the required documents have been received. Site-specific ICF(s) *will not* be reviewed and approved by the DAIDS PRO and sites will receive an Amendment Registration Notification when the DAIDS PRO receives a complete registration packet. A copy of the Amendment Registration Notification should be retained in the site's regulatory files.

For additional information on the protocol registration process and specific documents required for initial and amendment registrations, refer to the current version of the DAIDS Protocol Registration Manual.

13.3 Study Coordination

DAIDS holds the Investigational New Drug (IND) applications for this study. Assignment of all sponsor responsibilities for this study will be specified in a Clinical Trials Agreement (CTA) executed by NIAID and Merck & Co.

Study implementation will be directed by this protocol, which may not be amended without prior written approval from the Protocol Chairs and DAIDS Medical Officer. Study implementation will also be guided by a common study-specific procedures manual that provides further instructions and operational guidance on conducting study visits; data and forms processing; specimen collection, processing, and shipping; AE assessment, management and reporting; dispensing study products and documenting product accountability; and other study operations. Standardized study-specific training will be provided to all sites by the MTN LOC, SDMC, LC and other designated members of the Protocol Team.

Close coordination between protocol team members is necessary to track study progress, respond to queries about proper study implementation, and address other issues in a timely manner. The PSRT will address issues related to study eligibility and AE management and reporting as needed to assure consistent case management, documentation, and information-sharing across sites. Rates of accrual, adherence, follow-up, and AE incidence will be monitored closely by the team as well as the SMC.

13.4 Risk Benefit Statement

13.4.1 Risks

<u>General</u>

Phlebotomy may lead to excessive bleeding, discomfort, feelings of dizziness or faintness, and/or bruising, swelling and/or infection.

Pelvic examination and vaginal fluid collection may cause mild discomfort and/or vaginal bleeding or spotting. Cervical biopsies carry the risk of discomfort or pain during the procedure and for a few hours afterwards. Participants may have mild vaginal spotting (bleeding) for one or two days, and will be instructed to avoid sexual intercourse and product use for the duration of the study. While abstinence is a requirement of this study, if participants are sexually active they may also be at increased risk for STIs and HIV acquisition, if exposed. There is a small risk of infection and heavier bleeding. Participants will be instructed to contact the clinic if symptoms are bothersome, if heavy bleeding is noted (soaking through a pad in an hour or less) or if the participant develops any abnormal odor or discharge from the vagina.

Disclosure of HIV and STI status may cause worry, sadness or depression.

Disclosure of HIV-positive status has been associated with depression, suicidal ideation, and denial as well as social isolation. Trained counselors will be available to help participants deal with these feelings. Participation in clinical research includes the risks of loss of confidentiality and discomfort with the personal nature of questions when discussing sexual behaviors.

Participants at sites requiring partner notification in response to diagnosed STI or HIV infection could have problems in their relationships with their sexual partners. Participants also could have problems in their partner relationships associated with maintenance of study-required abstinence.

Insertion of a lubricated anoscope will likely cause some discomfort and there is the risk of discomfort and a small risk of bleeding with the insertion of rectal sponges for the collection of rectal fluid.

Use of the study IVR may lead to vaginal symptoms, including irritation, increased discharge, and discomfort (including with vaginal intercourse). As with any vaginally retained product, the possibility of toxic shock syndrome, although rare, exists.

With any drug, there is the potential risk for an allergic reaction. The most commonly reported symptoms associated with allergic reactions are:

- Rash
- Dizziness
- Itching
- Muscle aches
- Nausea
- Fainting
- Facial flushing
- Chest tightness
- Cough
- Hives

- Fever
- Shortness of breath

The following side effects have been associated with the use of oral VCV in patients being treated for HIV. These side effects may or may not be associated with the use of VCV when the drug is placed into a vaginal ring and worn by HIV-negative women:

- Hepatocellular events (Liver problems)
- Ischemic cardiovascular events (Heart problems, such as a heart attack)
- Dyslipidemias (High blood lipid or cholesterol levels)
- Herpes simplex virus (HSV) infections
- Upper respiratory infections
- Seizures
- Malignancies

The most common AEs associated with VCV are: diarrhea, nausea, headache, upper respiratory infection, nasopharyngitis, and fatigue.

However, there was no clear difference in incidence of side effects or AEs between VCV recipients and control groups.

The following side effect has been associated with the use of oral MK-2048. This side effect may or may not be associated with MK-2048 when formulated in an intravaginal ring.

- Mild headache

MTN-027 will ask questions of participants during the in-depth interview that may cause individuals discomfort. This could result in undesired changes in thought and emotion.

13.4.2 Benefits

Participants in this study may experience no direct benefit. Participants and others may benefit in the future from information learned from this study. Specifically, information learned in this study may lead to the development of safe and effective interventions to prevent HIV acquisition. Participants also may appreciate the opportunity to contribute to the field of HIV prevention research.

Participants will receive HIV/STI risk reduction counseling, HIV and STI testing, physical examination, pelvic examination, and routine laboratory testing related to blood, liver, and kidney function. Participants may be provided or referred for STI treatment in accordance with CDC guidelines. For other medical conditions identified as part of the study screening and/or follow-up procedures, participants will be referred to other sources of care available in their community. Some volunteers may have the opportunity to access expedient treatment and decreased morbidity due to early

diagnosis and treatment of abnormalities identified during tests, examinations and referrals.

13.5 Informed Consent Process

Written informed consent will be obtained from each study participant prior to screening. Written informed consent also will be obtained for long-term specimen storage and possible future testing, although consent for long-term specimen storage is not required for study participation. In obtaining and documenting informed consent, the IoR and their designees will comply with applicable local and US regulatory requirements and will adhere to GCP and to the ethical principles that have their origin in the Declaration of Helsinki. Study staff must document the informed consent process in accordance with the Requirements for Source Documentation in DAIDS Funded and/or Sponsored Clinical Trials (http://rsc.tech-res.com/policiesandregulations/). Participants will be provided with copies of the informed consent forms if they are willing to receive them.

In addition to informed consent forms, the Protocol Team will work with study staff and community representatives to develop appropriate materials about the study and a standardized approach to the informed consent process to be implemented at all study sites, which will be detailed in the study-specific procedures manual.

The informed consent process will cover all elements of informed consent required by research regulations. In addition, the process specifically will address the following topics of importance to this study:

- The unknown safety and unproven efficacy of the study products
- The need to abstain from sexual intercourse, regardless of study treatment group
- The importance of participants in all four study groups to the success of the study
- The importance of adherence to the study visit and procedures schedule
- The potential medical risks of study participation (and what to do if such risks are experienced)
- The potential social harms associated with study participation (and what to do if such harms are experienced)
- The real yet limited benefits of study participation
- The distinction between research and clinical care
- The right to withdraw from the study at any time

13.6 Participant Confidentiality

All study procedures will be conducted in private, and every effort will be made to protect participant privacy and confidentiality to the extent possible. Each study site will implement confidentiality protections that reflect the local study implementation plan and the input of study staff and community representatives to identify potential confidentiality issues and strategies to address them. In addition to local considerations, the protections described below will be implemented at all sites.

All study-related information will be stored securely at the study site. All participant information will be stored in locked areas with access limited to study staff. All laboratory specimens, study data collection, and administrative forms will be identified by coded number only to maintain participant confidentiality. All records that contain names or other personal identifiers, such as locator forms and informed consent forms, will be stored separately from study records identified by code number. All local databases will be secured with password protected access systems. Forms, lists, logbooks, appointment books, and any other listings that link participants' ID numbers to identifying information will be stored in a locked file in an area with limited access. Participants' study information will not be released without their written permission, except as necessary for review, monitoring, and/or auditing by the following:

- Representatives of the US Federal Government, including the US FDA, the US OHRP, NIH, and/or contractors of the NIH, and other local and US regulatory authorities
- Representatives of Merck & Co.
- Study staff
- Site IRBs/ECs

All study documents/data will be properly disposed of, including the proper destruction and/or deletion of paper files, electronic study data, and electronic documents. Audio files of In-depth Interviews will be transcribed and destroyed following a transcription quality assurance check. A member of the MTN BRWG or designee is responsible for ensuring that these files have been destroyed.

The MTN has a Certificate of Confidentiality from the US Department of Health and Human Services that is applicable to this study. This Certificate protects study staff from being compelled to disclose study-related information by any US Federal, State or local civil, criminal, administrative, legislative or other proceedings. It thus serves to protect the identity and privacy of study participants.

13.7 Special Populations

13.7.1 Pregnant Women

Women who test positive for pregnancy at Screening or Enrollment Visits will not be eligible to participate in this study. Should a woman test positive for pregnancy after Enrollment, study product will be permanently discontinued and participants will be withdrawn from the study, per Section 7.5.2. During the informed consent process, women will be informed that the study IVR is not a method of contraception and the effects of the study IVR on a developing human fetus are unknown.

All potential participants are required by the eligibility criteria for Screening and Enrollment to be currently sexually abstinent for the duration of study participation and using effective contraception.

13.7.2 Children

The NIH has mandated that children be included in research trials when appropriate. This study meets “Justifications for Exclusion” criteria for younger children as set forth by the NIH. Specifically, “insufficient data are available in adults to judge potential risk in children” and “children should not be the initial group to be involved in research studies.” This study does not plan to enroll children under 18 years old.

13.8 Compensation

Pending IRB approval, participants will be compensated for time and effort in this study, and/or be reimbursed for travel to study visits and time away from work. Site specific reimbursement amounts will be specified in the site specific informed consent forms.

13.9 Communicable Disease Reporting

Study staff will comply with local requirements to report communicable diseases including HIV-1 identified among study participants to health authorities. Participants will be made aware of reporting requirements during the informed consent process.

13.10 Access to HIV-related Care

13.10.1 HIV Counseling and Testing

HIV test-related counseling will be provided to all potential study participants who consent to undergo HIV-1 screening to determine their eligibility for this study, and to all enrolled participants at each follow-up HIV-1 testing time point. Testing will be performed in accordance with the algorithm in Appendix II. Counseling will be provided in accordance with standard HIV counseling policies and methods at each site and additionally will emphasize the unknown efficacy of the study products in preventing HIV-1 infection. In accordance with the policies of the NIH, participants must receive their HIV-1 test results to take part in this study.

13.10.2 Care for Participants Identified as HIV-Positive

An individual who has been identified as infected with HIV-1 will be managed or referred for management according to the local standard of care. Should a participant test positive for HIV after Visit 2, follow-up procedures will be performed as per Section 7.5.1. Please refer to Section 9.6 for additional details.

13.11 Study Discontinuation

This study may be discontinued at any time by NIAID, the MTN, Merck & Co., the US FDA, the OHRP, other government or regulatory authorities, or site IRBs/ECs.

14 PUBLICATION POLICY

DAIDS/NIAID and MTN policies and a CTA between Merck & Co. and NIAID will govern publication of the results of this study. Any presentation, abstract, or manuscript will be submitted by the investigator to the MTN Manuscript Review Committee, DAIDS, NIAID, NIMH, and Merck & Co. for review prior to submission.

15 APPENDICES

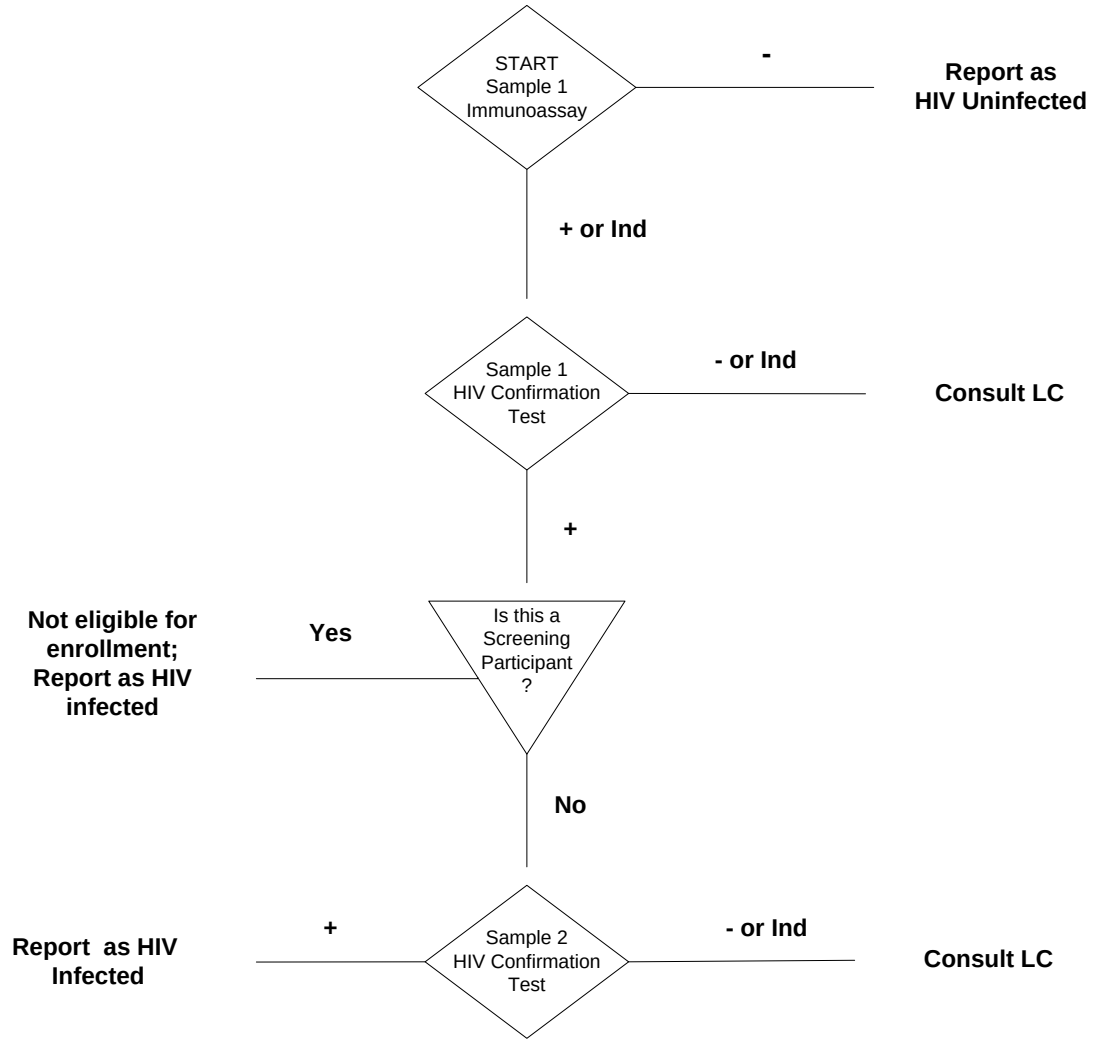
APPENDIX I: SCHEDULE OF STUDY VISITS AND EVALUATIONS

		SCR	ENR	VISIT Days 1, 2, 3, 7, 14, 21	VISIT Day 28	VISIT DAYS 29,30, and 31	DAY 35 Final Clinic/ Term.
ADMINISTRATIVE AND REGULATORY							
Informed consent(s)		X					
Assess informed consent comprehension		X					
Assignment of PTID		X					
Locator information		X	X	X	X	X	X
Demographic information		X					
Eligibility assessment		X					
Eligibility confirmation			X				
Randomization			X				
Reimbursement		X	X	X	X	X	X
Record/ update AEs				X	X	X	X
Schedule next visit		*	*	X	X	X	*
BEHAVIORAL							
Behavioral assessment			X	ϴ	X		X
In-depth interview							X
HIV pre- and post- test counseling		X	X	*	*	*	X
Protocol requirements counseling (To include adherence, product use and contraceptive counseling, as needed)		x	x	*	*	*	*
CLINICAL							
Medical and menstrual history		X	X	X	X	X	X
Concomitant medications		X	X	X	X	X	X
Document pre-existing conditions			X				
Physical examination (full or modified)		X	X	X	X	X	X
Pelvic examination		X	X	X	X	X	X
Provide available test results		X	X	X	X	X	X
Treat or prescribe treatment for UTI/RTI/STIs or refer		*	*	*	*	*	*
LABORATORY							
Urine	*Collect Urine*						
	hCG	X	X	₪ *	X	*	*
	Dipstick UA	X	*	*	X	*	
	Urine culture	*	*	*	*	*	
Blood	*Collect Blood*						
	CBC with differential and platelets	X	X		X		X
	HIV-1 serology	X	X	*	*	*	X
	HBsAg	X					
	INR	X					
	Anti-HCV	X					
	Chemistries (Creatinine, AST, ALT)	X	X		X		X
	PK- Blood		X	X	X	X	X
	Syphilis serology	X	*	*	*	*	*
	Plasma archive		X				
Genital	*Collect pelvic specimens*						
	Vaginal fluid pH	*	X	♦ *	X	X	X
	Rapid Trichomonas test	X	*	*	*	*	*
	KOH wet mount for candidiasis	*	*	*	*	*	*
	Saline wet mount for BV	*	*	*	*	*	*
	Vaginal NAAT for GC/CT	X	*	*	*	*	*
	PK- Vaginal fluid		X	X	X	X	X

		SCR	ENR	VISIT Days 1, 2, 3, 7, 14, 21	VISIT Day 28	VISIT DAYS 29,30, and 31	DAY 35 Final Clinic/ Term.
	Cytobrush		X		X		
	PK- Cervical tissue				X		
	PD- Cervical tissue				µ		
	Collect Pap test	*					
	Quantitative vaginal culture		X	♦	X		X
	Gram stain		X	♦	X		X
	Vaginal swab for vaginal biomarkers		X	♦	X		X
Rectal	*Collect rectal specimens*						
Rectal	Rectal fluid for PK		♿		♿		
STUDY PRODUCT							
Participants will receive study IVR, study IVR use instructions and will be instructed to self-insert the study IVR, followed by pelvic exam to check placement			X				
Collect IVR					X		▲

* If indicated, ϴ Required at Day 7, 14, 21, ▲ If Early Termination Visit and not already performed, ♦ Required at Day 3, ₪ Required at Day 14 only µ To be collected at site(s) with capacity ♿ To be collected on a subset of participants who opt in

APPENDIX II: ALGORITHM FOR HIV ANTIBODY TESTING



Ind: Indeterminate test results
LC: Laboratory Center

APPENDIX III: SAMPLE INFORMED CONSENT FORM (SCREENING, ENROLLMENT, LONG-TERM STORAGE AND FUTURE TESTING)

SAMPLE INFORMED CONSENT FORM
DIVISION OF AIDS, NIAID, NIH

MTN-027

Phase 1 Safety and Pharmacokinetics Study of MK-2048/Vicriviroc (MK-4176)/MK-2048A Intravaginal Rings

Version 1.0

December 20, 2014

PRINCIPAL INVESTIGATOR: ***[Sites to insert]***
PHONE: ***[Sites to insert]***
Short Title for the Study: Safety and PK of a Vicriviroc/MK-2048 VR

INFORMED CONSENT

You are being asked to take part in this research study because you are a healthy, HIV-negative woman between the ages of 18 and 45 years old. Approximately 48 women will participate in this study at two sites in the United States. This Microbicide Trials Network (MTN) study is sponsored by the US National Institutes of Health (NIH). The study products in this clinical trial include MK-2048, vicriviroc, MK-2048/vicriviroc, and the placebo vaginal rings. A vaginal ring is a flexible plastic ring that is inserted in the vagina. In this study, participants are asked not to remove the ring for approximately 28 days. The study products are supplied by Merck. At this site, the person in charge of this study is *[INSERT NAME OF PRINCIPAL INVESTIGATOR]*.

Before you decide if you want to join this study, we want you to learn more about it. This consent form gives you information about the study. Study staff will talk with you and answer any questions you may have. Once you read and understand the study and its requirements, you can decide if you want to join. If you do decide to take part in the trial, you will sign your name on this form. A copy of this document will be offered to you. Signing this consent form does not mean you will be able to join the study. You must first complete the screening tests and exams to see if you are eligible.

It is important to know that your participation in this research is your decision and taking part in this study is completely voluntary (see Your Rights as a Research Participant/Volunteer for more information).

WHAT IS THE PURPOSE OF THIS STUDY?

The main purpose of this research study is to find out if vaginal rings containing the study drugs vicriviroc, MK-2048, and combination of vicriviroc/MK-2048 are safe when inserted into the vagina for approximately 28 days. Another purpose of this study is to better understand how the study drugs enter and exit the body.

STUDY PRODUCTS
All of the study drugs have been previously tested for HIV treatment. HIV is the virus that causes AIDS. Now researchers would like to know if these drugs can work to prevent HIV. To do this they first need to better understand what effect these drugs have on the body, including the vagina.

The study drugs in the vaginal rings work in different ways to potentially prevent HIV:

- Vicriviroc works by preventing HIV from binding to cells. By preventing the virus from connecting to the cell, HIV is unable to enter the cell and begin reproducing. Vicriviroc has been studied for HIV treatment in several clinical trials and has been found to be safe.

- MK-2048 blocks the integration of HIV to the body's cells, specifically into the DNA, which is the body's genetic material. By preventing this step, the medication prevents HIV from taking hold in cells and begin reproducing. Oral tablets of MK-2048 were studied in one clinical trial involving 16 men and were generally well-tolerated.

This study is <u>not</u> testing to see if the study drugs prevent HIV infection. Researchers do not yet know if one or both of the two drugs will work in humans to protect against HIV.
There are only two known effective ways to prevent HIV: condoms and/or the use of pre-exposure prophylaxis (PrEP). Condoms and/or the use of pre-exposure prophylaxis (PrEP) are the only known ways to prevent HIV currently. PrEP is a new HIV prevention method in which people who do not have HIV take an oral tablet to reduce their risk of becoming infected. Study staff can provide you with additional information about PrEP if you are interested in learning more.

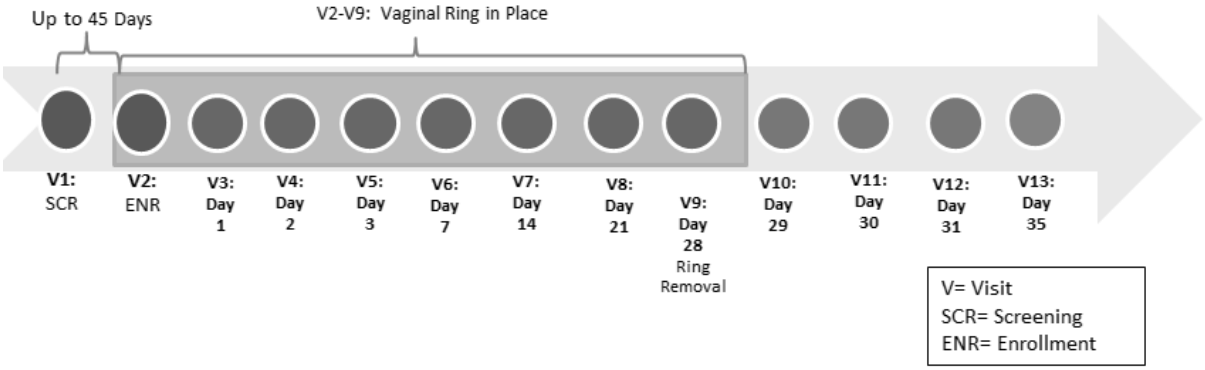
While the study drugs have been tested before in humans, this is the first time vicriviroc and MK-2048 have been tested in a vaginal ring formulation. This is also the first time vicriviroc and MK-2048 have been tested in combination in humans.

STUDY GROUPS
All of the eligible women will be randomized to one of four vaginal ring study groups:
- Vicriviroc
- MK-2048
- A combination ring containing vicriviroc and MK-2048, or
- A placebo ring. The placebo ring has no study drug or any other active ingredient(s) in it.

Twelve women will be in each study group and will be assigned to a group by random chance (the equivalent of throwing dice). It is important that you know that you will receive a ring that may or may not contain a drug. This is a single-blind study, which means you will not know which group you are in until the study is completed. The study clinician who provides the vaginal ring to participants may know whether the ring you receive contains MK-2048. Women will be asked to insert a ring in their vagina for a duration of approximately 28 days.

All of the study groups are important to this study. No matter which study group you are in, you must remember that we do not know if the drugs contained within the rings work to protect women from getting HIV.

WHAT WILL HAPPEN DURING THE MTN-027 STUDY VISITS?



Screening Procedures:

The MTN-027 study includes a total of 13 study visits including the Screening Visit which is taking place today after you sign this informed consent form. Visits will take place here, at this study clinic.

The procedures done at this visit (the screening visit) will take about [sites to insert time].

- Study staff will ask you where you live and other questions about you, your medical health (including what medications you are taking), menstrual history, your sexual practices and your understanding of the study requirements.
- Study staff will:
  - Perform a physical exam.
  - Talk with you about the requirements of the study including, but not limited to:
    - Keeping the VR in place and not removing it between visits;
    - Being sexually abstinent for the duration of this study. This means no receptive penile-vaginal intercourse, anal intercourse, receptive oral intercourse, finger stimulation, or the use of sex toys.
    - Refraining from inserting any non-study vaginal products or objects into the vagina for the 5 days prior to Enrollment and for the duration of your study participation. These include but are not limited to: spermicides, female condoms, diaphragms, contraceptive vaginal rings, vaginal medications, menstrual cups, cervical caps (or any other vaginal barrier method), douches, lubricants, or sex toys (vibrators, dildos, etc.) or fingers.
  - Test your urine for pregnancy and other conditions
    - If you are pregnant you cannot join this study.
    - Study staff will talk with you about ways to avoid becoming pregnant.
  - Take a blood sample [Sites to insert amount]:
    - To test the health of your blood, liver and kidneys.
    - To test for infections that typically are passed through sex, including HIV, hepatitis B, hepatitis C, syphilis, etc.

      - You will be told your test results as soon as they are available. You will talk with the study staff about the meaning of your results, how you feel about them, and learn about ways to prevent HIV and other sexually transmitted infections. Sometimes HIV tests are not clearly positive, but also not clearly negative. In that case, we will do more tests until we are sure of your status. To participate in the study you

must receive the results of your HIV test. If the test shows you have HIV, you cannot join the study. We will refer you to available sources of medical care and other services you may need. The study staff will tell you about other studies you may be eligible for, if any.

- Perform a pelvic examination:
  - The study clinician will use a speculum, a plastic or metal instrument used to separate the walls of the vagina. The study clinician will check your vagina and cervix (the tissue that attaches the vagina to the uterus) for signs of infection, and other problems. They will also take some fluids to test for sexually transmitted infections and diseases (commonly known as STIs or STDs) and other problems.
  - The study staff may also collect samples from your cervix for a "Pap test" or "Pap smear". Study staff will inform you of the results of your Pap test. It takes about ***[SITES TO INSERT AMOUNT OF TIME]*** before Pap test results are ready. If you are 21 years of age or older and have a written report confirming a normal Pap test in the past 3 years, or if you had an abnormal Pap test but had follow-up indicating no treatment was required, you will not need to have a Pap test taken at this screening visit. The results of your Pap test may affect whether or not you can join the study.
- Give you treatment or refer you for treatment for infections passed through sex, if needed.
- Inform you about other services, if needed.
- Provide you with the results of your tests, when available. It is expected that all of your results will be available by ***[SITES TO SPECIFY TIMEFRAME]***. .
- Schedule your next visit to enroll in the study, if you are willing and eligible.

If you decide not to join MTN-027, blood collected at this visit will not be kept or used for any tests other than those listed above.

Enrollment and Follow-up Procedures:

At your Enrollment visit (the visit where you enter the study) you will:

- Answer questions to confirm you are able to join the study
- Be randomly assigned to one of four study groups. Neither you nor the study staff will be able to choose your group or change the group you have been placed into. Women in all study groups will have the same study visit schedule.
- Discuss any health or medical problems you may have had in the past or are currently experiencing.
- Have a blood sample collected ***[SITE TO INSERT AMOUNT]*** to test the health of your blood, liver and kidneys and for HIV infection. We also will collect blood in the event there is a question about your lab results in the future. Finally blood will be collected at multiple time points to measure the amount of study drug in your body over time, specifically at hours 1, 2, 4, and 6 hours after the ring has been in place. To collect these blood samples, an intravenous cannula (IV tube) may be placed, flushed and kept in place for the blood draws during the 6 hours after ring placement.
- Provide vaginal fluid at multiple time points to measure the amount of study drug in your vagina. This will be collected when the ring is inserted and approximately 1, 2, 4, and 6 hours after the ring has been in place. You will be asked to collect these samples. Study staff will provide you with more information as to how and where to place the swab and how to collect the fluid.

- Receive and insert the study ring. Study staff may help you insert the study ring if you cannot do it on your own. All participants will have an exam to ensure the ring is inserted correctly.

  This visit will take longer than most because we will check your blood and vaginal fluids over a period of 6 hours.

At most study visits, including the Enrollment visit, you will:

- Provide and/or update study staff with your contact information (i.e. about where you live and how we can contact you).
- Tell study staff about your health, any changes in your health and/or any other problems.
- Talk with study staff about the following:
  - The rules of the study and how to follow the rules, including how to properly use the vaginal ring for example, how to clean and reinsert the ring if it falls out anytime between your visits to the clinic. Another rule of the study is sexual abstinence. We ask that you abstain from receptive sexual activity (including receptive penile-vaginal intercourse, anal intercourse, receptive oral intercourse, finger stimulation, and the use of sex toys) for the 5 days prior to Enrollment and for the duration of your study participation. **If you do not think you can be sexually abstinent for 40 days then you should not join this study.**
  - Sexually transmitted infections (STIs), HIV, HIV/STI testing, and ways to avoid HIV and other infections passed through sex.
- After you begin using the vaginal ring, you will be asked to have blood and vaginal fluid samples collected at all clinic visits to measure the amount of study drug in your body. When the ring is removed at Day 28 you will provide blood and vaginal fluid at multiple time points over a 6 hour period. An intravenous cannula (IV tube) may be placed for approximately 6 hours after ring removal for the blood draws. This visit, like your Enrollment visit, will be longer.
- Have a physical exam.
- Have a pelvic exam.
  - The study clinician will use a speculum. Study staff will ask if you are experiencing symptoms of an infection. They will check your vagina and cervix for signs of problems due to the ring or infection. They will also take samples to test for bacteria and organisms in the vagina and, if necessary, look for any other problems.
  - A small amount of vaginal fluid will be collected via swab(s), like a Q-tip. This vaginal fluid collected will be used for research purposes only, i.e. to measure the amount of study drug in your body.
  - When the vaginal ring is removed on Day 28, 2 small tissue samples will be taken from your cervix, each about the size of a grain of rice. These samples will be used to measure the amount of the study drug in your tissue and understand the drug's effects on the body.
  - Study researchers will keep the ring at the end of the study and will run additional tests on it for research purposes only.
- Receive treatment or be referred for treatment issues that the study staff may find.
- Receive test results, if available.
- Schedule your next visit.
- Answer questions about your experience using the ring, including whether or not the ring was removed or fell out of your vagina. You may use a computer to answer these questions or a staff member may ask you these questions.

- At your Final Clinic Visit (Day 35) you will be interviewed about your experience using the ring. This interview may take approximately 45-60 minutes and will occur over video chat, e.g., Skype, FaceTime, etc. This conversation will be recorded, but your responses will be kept private and confidential, and the audio-recording will be destroyed after they have been transcribed and checked.

At some visits you will be asked to provide a sample of blood to:
- Check the health of your blood, liver and kidneys.
- Test for HIV infection

Study staff can answer any questions you may have about the procedures mentioned above.

Additional Visits and Procedures

It may be necessary for you to have additional visit(s) and/or provide additional samples if any of the above procedures need to be repeated due to issues sample processing, testing or shipping, and/or if you are experiencing any symptoms or changes in your physical condition.

Additional testing may be performed as part of quality control.

Extra Sample Group: Optional Study Procedure

If you agree to take part in the Extra Sample Group, you will be asked to provide rectal fluid. At your Enrollment Visit and on Day 28 a short hollow plastic tube will be placed inside your rectum so that the study doctor can take samples. . You will consent to this procedure separately.

If you become infected with HIV

As a requirement of this study you are asked to **NOT** engage in any sexual activity. Your participation in this study will not cause HIV infection. However, there is always a chance that through sexual activity or other activities you may become HIV-positive. In the unlikely event that you become HIV-positive, study staff will give you counseling and refer you for medical care and other available services. You will continue to be counseled while you are in this study. Tests will be performed to see if you have HIV drug resistance. This will allow doctors to know what HIV drugs would be best for the treatment of your type of HIV. If the HIV tests confirm that you have been infected with HIV, you will stop using the VR. You may be referred to other research studies.

RISKS AND/OR DISCOMFORTS

Whenever your blood is drawn, you may have:
- Excessive bleeding
- Discomfort
- Feelings of dizziness or faintness
- Bruising, swelling and/or infection

During pelvic exams and vaginal fluid collection you may feel discomfort or pressure in your vagina and/or pelvis. Due to the pelvic exam you may also have vaginal bleeding or spotting, which will stop shortly after the examination. Cervical biopsies carry the risk of discomfort or pain during the procedure and for a few hours afterwards. Participants may

have spotting (bleeding) for one or two days. With cervical biopsies there is also a small risk of infection and heavier bleeding. You may also be at increased risk for STIs and HIV acquisition, if exposed. You will be encouraged to call the clinic to report any problems after the collection, especially if heavy bleeding is noted (soaking through a pad or tampon in an hour or less) or if you develop any abnormal vaginal odor or discharge.

Study Rings
The study rings can cause some side effects. We do not yet know all the side effects of the rings. Some, but not all, women who used rings in other studies have had:

- Discharge from the vagina
- Irritation and discomfort

With any product inserted vaginally, it is possible you could experience toxic shock syndrome. Toxic shock syndrome is a rare but serious illness caused by poisons (toxins) released by some types of Staphylococcus aureus, a common bacteria. The likelihood of this occurring is rare.

Study Drugs
The following side effects have been associated with the use of vicriviroc in patients being treated for HIV. These side effects may or may not be associated with the use of vicriviroc when the drug is placed into a vaginal ring and worn by HIV-negative women:

- Hepatocellular events (Liver problems)
- Ischemic cardiovascular events (Heart problems, such as a heart attack)
- Dyslipidemias (High blood lipid or cholesterol levels)
- Herpes simplex virus (HSV) infections
- Upper respiratory infections
- Seizures
- Malignancies (Cancerous tumor)

The most common side effects associated with vicriviroc are: diarrhea, nausea, headache, upper respiratory infection, nasopharyngitis (infectious swelling/redness of the nose and throat), and fatigue.

However, there was no clear difference in the rate of side effects among people who received vicriviroc compared with those who did not receive this medication.

The following side effect has been associated with the use of oral MK-2048. This side effect may or may not be associated with MK-2048 when it is placed into the vaginal ring:

- Mild headache

Currently, no risk information is available for these two drugs combined.

It is also possible that you may have an allergic reaction to the study product. Signs of allergic reaction may include: rash, dizziness, itching, muscle aches, nausea, fainting, facial flushing, chest tightness, cough, hives, fever, shortness of breath.

Other Possible Risks
You may become embarrassed and/or worried when discussing your sexual practices, ways to protect against HIV and other infections passed through sex, and your test results. You

may be worried while waiting for your test results. If you have HIV or other infections, learning this could make you worried. Trained study counselors will help you deal with any feelings or questions you have.

We will make every effort to protect your privacy and confidentiality during the study visits. Your visits will take place in private. However, it is possible that others may learn of your participation here and, because of this, may treat you unfairly or discriminate against you. For example, you could have problems getting or keeping a job, or being accepted by your family or community. Finding out your HIV-positive could cause depression and/or suicidal thoughts. Finding out your HIV-status could also cause problems between you and your partner. If you have any problems, study counselors will talk with you and/or your partner to try to help resolve them.

It is possible that if you and/or your partner(s) may experience problems in your relationships associated with maintenance of the study-required abstinence.

The interview that takes place at your last clinic visit will be audio recorded and questions of a personal nature may be asked. Responding to these questions may make you uncomfortable. The audio files will be put into writing by the person interviewing you or by another person who does not know you and does not have your personal information. The audio recordings will be destroyed as soon as they have been put into writing and ensured that an accurate copy has been made. The person in charge at this site will make sure that these records have been destroyed. You should NOT identify anyone in the interviews and any names that might be mentioned on the recording will NOT be noted. Instead a generic description will be used in the transcript (i.e., if you refer to a friend's name, "FRIEND1" will be noted).

Sexual Practices, Pregnancy, and Breastfeeding

The rings with study drug and the placebo ring are not birth control methods. You must agree to use an effective method of birth control such as birth control pills or another hormonal-based method (except for vaginal rings), or an intrauterine device (IUD), unless you or your partner have been sterilized (i.e., no longer able to have babies), you have sex with women exclusively; and/or you have been sexually abstinent for more than 90 days. You must also agree to not insert anything into your vagina for the duration of this study; this means that you may not have sex for the duration of this study. Sex for this study is defined as receptive penile intercourse, anal intercourse, receptive oral intercourse and the use of sex toys. You must also agree to not use tampons during the first week of study participation (starting at the enrollment visit) and for 24 hours prior to each clinic visit following enrollment.

We do not know what effect the study drugs have on pregnancy, including the effect of the study drug on the fetuses of women who use the vaginal ring when pregnant, or the babies of women who use the vaginal ring when breastfeeding. Because of this, pregnant women and women who are breastfeeding may not join this study. Women who join the study must agree to be sexually abstinent and use an effective method of contraception. Women who join this study will have pregnancy tests while in the study.

If you become pregnant during the study, study staff will refer you to available medical care and other services you or your baby may need. The study does not pay for this care. You will stop using the ring and you will exit the study. We will contact you to find out about your

pregnancy and the outcome of your pregnancy. The outcome of your pregnancy is important to study staff; therefore your pregnancy will be followed until the results of your pregnancy are known.

BENEFITS

No one knows if the study ring will prevent HIV infection. Information learned from this study may help in the development of ways to prevent the spread of HIV in the future. You may receive no direct benefit from your participation in this study. You will receive pelvic exams and counseling and testing for HIV and STIs. You will also have tests to check the overall health of your liver, kidneys, and blood cells.

This study cannot provide you with general medical care, but study staff will refer you to other available sources of care.

You will be counseled and tested for HIV and STIs. You will receive free male condoms at your Final Clinic Visit, if you need them. If you are infected with HIV, you will be referred for medical care, counseling, and other services available to you. Medical care for HIV infection will not be part of this study. You will need to receive care for HIV infection from your own health care provider or we will provide you with a referral. If you have an STI diagnosed, you will receive medicine or a referral, if needed, and study staff will discuss options available for counseling and treatment of your partner.

NEW INFORMATION

You will be told of any new information learned during this study that might affect your willingness to stay in the study. For example, if information becomes available that shows that the ring may be causing bad effects, you will be told about this. You will also be told when study results may be available, and how to learn about them.

WHY YOU MAY STOP TAKING THE STUDY DRUG EARLY OR BE WITHDRAWN FROM THE STUDY WITHOUT YOUR CONSENT

A study doctor may need to remove you from the study early without your permission if:

- The study is cancelled by the US FDA, US NIH, Merck (the company that supplies the vaginal rings), the US Office for Human Research Protections (OHRP), MTN, the local government or regulatory agency, or the Institutional Review Board (IRB)/ the Ethics Committee (EC). An IRB is a committee that watches over the safety and rights of research participants
- The Study Monitoring Committee (SMC) recommends that the study be stopped early (A SMC reviews the progress of the study and the kinds of effects that people report while they are participating in the study)
- You are found to be infected with HIV
- You become pregnant
- You are not able to keep appointments
- Other reasons that may prevent you from completing the study successfully

The study doctor will ask you to stop using the study vaginal ring but continue to come in for your follow-up visits and procedures if:

- A study doctor decides that using the vaginal ring would be harmful to you
- You require a treatment that you may not take while using the study ring
- You have a bad reaction to the study ring

If a study doctor asks you to stop using the ring, you will be asked to continue to come in for all scheduled visits and undergo some of the procedures described above, including the physical examination, vital signs, and blood tests. You will stop using the ring until the study doctor decides it is safe for you to start using it again, if possible.

In the event that you are removed from or choose to leave this study, you will be asked to complete a final evaluation and return your vaginal ring. If you do not have the vaginal ring with you at the time of your contact with staff, staff members will make every effort to assist you in returning the ring as soon as possible. *[Sites to specify allowances for special circumstances.]*

ALTERNATIVE OPTIONS
We do not know if the drugs contained within the rings work to protect women from getting HIV. Currently there are two known methods to reduce your risk of contracting HIV, the use of condoms and/or the use of oral pre-exposure prophylaxis (PrEP) medication, Truvada®. If you are interested in these alternative options you may want to discuss them with your doctor.

COSTS TO YOU
[Site to complete according to site capacity] There is no cost to you for study related visits, the vaginal ring, physical/pelvic examinations, laboratory tests or other procedures. Treatments available to you from the study site for infections passed through sex will be given to you free of charge or you will be referred for available treatment for the duration of the study.

REIMBURSEMENT
[*Sites to insert information about local reimbursement:*] You will receive [*Sites to insert amount* $xx] for your time, effort, and travel to and from the clinic at each scheduled visit. You may receive [Sites to insert amount $xx] for any visits which occur in between your normally scheduled visits.

CONFIDENTIALITY
Efforts will be made to keep your information confidential. However, it is not possible to guarantee confidentiality. Your personal information may be disclosed if required by law. The study staff may use your personal information to verify that you are not in any other research studies. This includes studies conducted by other researchers that study staff know about. Any publication of this study will not use your name or identify you personally.

Your records may be reviewed by:
- Representatives of the US Federal Government, including the US Food and Drug Administration (FDA), US Office for Human Research Protections (OHRP), NIH, and/or contractors of NIH
- Other local and US regulatory authorities
- Merck, the company that supplies the vaginal rings
- Study monitors
- Site IRB/EC
- Study staff

[Sites to include/amend the following:] [LOCAL/STATE/NATIONAL] regulations require study staff to report the names of people who test positive for HIV and other infections passed

during sex to the *[LOCAL HEALTH AUTHORITY]*. Outreach workers from the *[LOCAL HEALTH AUTHORITY]* may then contact you about informing your partners, since they also should be tested. If you do not want to inform your partners yourself, the outreach workers will contact them, according to the confidentiality guidelines of the *[HEALTH AUTHORITY].*

The researchers will do everything they can to protect your privacy. In addition to the efforts of the study staff to help keep your personal information private, we have obtained a Certificate of Confidentiality from the US Federal Government. This Certificate protects study staff from being forced to tell people who are not connected with this study, such as the court system, about your participation or information you give for study purposes. However, if the study staff learns of possible child abuse and/or neglect or a risk of harm to you or others, they will be required to tell the proper authorities. This Certificate does not prevent you from releasing information about yourself and your participation in the study.

RESEARCH-RELATED INJURY

[Sites to specify institutional policy:] It is unlikely that you will be injured as a result of study participation. If you are injured, the *[institution]* will give you immediate necessary treatment for your injuries. You *[will/will not]* have to pay for this treatment. You will be told where you can receive additional treatment for your injuries. The U.S. National Institutes of Health (NIH) does not have a mechanism to pay money or give other forms of compensation for research related injuries. You do not give up any legal rights by signing this consent form.

YOUR RIGHTS AS A RESEARCH PARTICIPANT/VOLUNTEER

[Sites to specify institutional policy:] Taking part in this study is completely voluntary. You may choose not to take part in this study or leave this study at any time. If you choose not to participate or to leave the study, you will not lose the benefit of services to which you would otherwise be entitled at this clinic. If you want the results of the study after the study is over, let the study staff members know.

PROBLEMS OR QUESTIONS

If you ever have any questions about the study, or if you have a research-related injury, you should contact *[insert name of the investigator or other study staff]* at *[insert telephone number and/or physical address]*.

If you have questions about your rights as a research participant, you should contact *[insert name or title of person on the IRB/EC or other organization appropriate for the site]* at *[insert physical address and telephone number]*.

Extra Sample Group

You may choose to provide rectal fluid to help researchers better understand where the study drugs go and how they work against HIV in the laboratory, but you do not have to agree to these extra procedures to participate in MTN-027. Participants who agree to provide rectal fluid samples at this site will be asked to provide these samples. Rectal fluid will be collected using a sponge.

If you agree to participate in the Extra Sample Group, the following risks apply:

- With the insertion of the lubricated short hollow plastic tube you may experience discomfort.
- There is the risk of discomfort and a small risk of bleeding with the insertion of rectal sponges.

_____ Yes, if chosen, I want to take part in the Extra Samples Group
Initials & Date

_____ No, I do not want to take part in the Extra Samples Group
Initials & Date

CONSENT FOR LONG-TERM STORAGE AND FUTURE TESTING OF SPECIMENS

There might be a small amount of blood, vaginal fluid, cervical tissue and cervical fluid left over after we have done all of the study related testing. We would like to ask your permission to store these leftover samples and related health information for use in future studies. This health information may include personal facts about you such as your race, ethnicity, sex, medical conditions and your age range. If you agree, your samples and related health data will be stored safely and securely at facilities that are designed so that only approved researchers will have access to the samples. Some employees of the facilities will need to have access to your samples to store them and keep track of where they are, but these people will not have information that directly identifies you. You can still enroll in this study if you decide not to have leftover samples stored for future studies. If you do not want the leftover samples stored, we will destroy these specimens. The type of testing planned for your leftover specimens is not yet known, however no genetic testing (limited or genome-wide) is planned on leftover samples that are stored for the purposes of future research. It is important that you know that any future testing/studies planned for these specimens must be approved by an Ethics Committee/ Institutional Review Board before they can be done. You can withdraw your consent for the storage and future testing of specimens at any time by providing your request in writing to the person in charge of this study.

_____ Initials and Date	I DO agree to allow my biological specimens and health data to be stored and used in future research studies.
_____ Initials and Date	I DO NOT agree to allow my biological specimens and health data to be stored and used in future research studies.

SIGNATURES- VOLUNTARY CONSENT

[INSERT SIGNATURE BLOCKS AS REQUIRED BY THE LOCAL IRB/EC] If you have read this consent form, or had it read and explained to you, and you understand the information, and you voluntarily agree to take part in the study, please sign your name or make your mark below.

_____	_____	_____
Participant Name (print)	Participant Signature/Mark	Date
_____	_____	_____
Study Staff Conducting Consent Discussion (print)	Study Staff Signature	Date
_____	_____	_____
Witness Name (print)	Witness Signature	Date

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