A Randomized, Double-Blind, Placebo-Controlled Trial of TOL-463 Insert for Suppression of Bacterial Vaginosis (BV) [SUBVert]

DMID Protocol Number: 18-0017

DMID Funding Mechanism:

Contract – HHSN2722013000121

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Pharmaceutical Support: Toltec Pharmaceuticals LLC

IND Sponsor: NIAID

Lead Principal Investigator: Jeanne Marrazzo, MD, MPH

DMID Clinical Project Manager: Jonathan Glock, MPH

Version Number: 5.0

09 December 2020

STATEMENT OF ASSURANCE

Each Institution will hold a current Federal Wide Assurance (FWA) issued by the Office for Human Research Protections (OHRP) for federally-funded human subjects research. Each FWA will designate at least one Institutional Review Board (IRB)/Independent Ethics Committee (IEC) registered with OHRP, for which the research will be reviewed and approved by the IRB/IEC and will be subject to continuing review [45 CFR 46.103(b)]. The IRB/IEC designated under an FWA may include an institution's IRB/IEC, an independent IRB/IEC, or an IRB/IEC of another institution after establishing a written agreement with that other institution.

STATEMENT OF COMPLIANCE

The study trial will be carried out in accordance with Good Clinical Practice (GCP) and as required by the following:

- United States Code of Federal Regulations (CFR) 45 CFR Part 46: Protection of Human Subjects
- Food and Drug Administration (FDA) Regulations, as applicable: 21 CFR Part 50 (Protection of Human Subjects), 21 CFR Part 54 (Financial Disclosure by Clinical Investigators), 21 CFR Part 56 (Institutional Review Boards), 21 CFR Part 11, and 21 CFR Part 312 (Investigational New Drug Application)
- International Conference on Harmonisation, (ICH): Good Clinical Practice (ICH E6); 62 Federal Register 25691 (1997); and future revisions
- Belmont Report: Ethical Principles and Guidelines for the Protection of Human Subjects of Research, Report of the National Commission for the Protection of Human Subjects of Biomedical and Behavioral Research
- National Institutes of Health (NIH) Office of Extramural Research (OER), Research Involving Human Subjects, as applicable
- National Institute of Allergy and Infectious Diseases (NIAID) Clinical Terms of Award, as applicable
- Applicable Federal, State, and Local Regulations and Guidance

SIGNATURE PAGE

The signature below provides the necessary assurance that this trial will be conducted according to all stipulations of the protocol, including all statements regarding confidentiality, and according to local legal and regulatory requirements and applicable US federal regulations and ICH E6 Good Clinical Practice (GCP) guidelines.

I agree to conduct the study in compliance with GCP and applicable regulatory requirements.

I agree to conduct the study in accordance with the current protocol and will not make changes to the protocol without obtaining the sponsor's approval and IRB/IEC approval, except when necessary to protect the safety, rights, or welfare of participants.

Principal Investigator and Site Investigator:

Signed: _____ Date: _____

Jeanne Marrazzo, MD, MPH
University of Alabama at Birmingham

Site Investigator:

Signed: _____ Date: _____

Harold C. Wiesenfeld, MD, CM
University of Pittsburgh

Site Investigator:

Signed: _____ Date: _____

Audrey French, MD
Cook County Health

Site Investigator:

Signed: _____ Date: _____

Sheldon Morris, MD, MPH
UC San Diego AntiViral Research Center (AVRC)

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LIST OF ABBREVIATIONS

AE	Adverse Event/Adverse Experience
ASCUS	Atypical Squamous Cells of Undetermined Significance
BV	Bacterial Vaginosis
CDC	Centers for Disease Control and Prevention
CFR	Code of Federal Regulations
CI	Confidence Interval
CIN	Cervical Intraepithelial Neoplasia
CMS	Clinical Materials Services
CoC	Certificate of Confidentiality
CRF	Case Report Form
CROMS	Clinical Research Operations and Management Support
CTM	Clinical Trial Material
C. trachomatis	*Chlamydia trachomatis*
DCF	Data Collection Form
DHHS	Department of Health and Human Services
DMID	Division of Microbiology and Infectious Diseases, NIAID, NIH, DHHS
DSMB	Data and Safety Monitoring Board
eCRF	Electronic Case Report Form
EIC	Enrollment Informed Consent
FDA	Food and Drug Administration
FWA	Federal Wide Assurance

GCP	Good Clinical Practice
GLP	Good Laboratory Practice
HIV	Human Immunodeficiency Virus
HPF	High-power field
HPV	Human Papillomavirus
IATA	International Air Transport Association
IB	Investigator's Brochure
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
IEC	Independent or Institutional Ethics Committee
IND	Investigational New Drug Application
IP	Investigational Product
IRB	Institutional Review Board
ISM	Independent Safety Monitor
ITT	Intent-to-Treat
KOH	Potassium Hydroxide
LDPE	Low-Density Polyethylene
MedDRA®	Medical Dictionary for Regulatory Activities
mITT	Modified Intent-to-Treat
MOP	Manual of Procedures
N	Number (typically refers to participants)
NAAT	Nucleic Acid Amplification Test
N. gonorrhoeae	*Neisseria gonorrhoeae*

NIAID	National Institute of Allergy and Infectious Diseases, NIH, DHHS
NIH	National Institutes of Health
OER	Office of Extramural Research
OHRP	Office for Human Research Protections
PCR	Polymerase Chain Reaction
PD	Protocol Deviation
PI	Principal Investigator
PID	Pelvic Inflammatory Disease
PP	Per-Protocol
PPROM	Preterm Premature Rupture of Membranes
PVC	Polyvinyl Chloride
QA	Quality Assurance
QC	Quality Control
SAE	Serious Adverse Event/Serious Adverse Experience
SAP	Statistical Analysis Plan
SDCC	Statistical and Data Coordinating Center
SIC	Screening Informed Consent
SOP	Standard Operating Procedure
STI	Sexually Transmitted Infection
SubI	Sub Investigator
TEAE	Treatment-Emergent Adverse Event
TOC	Test of Cure
T. vaginalis	*Trichomonas vaginalis*

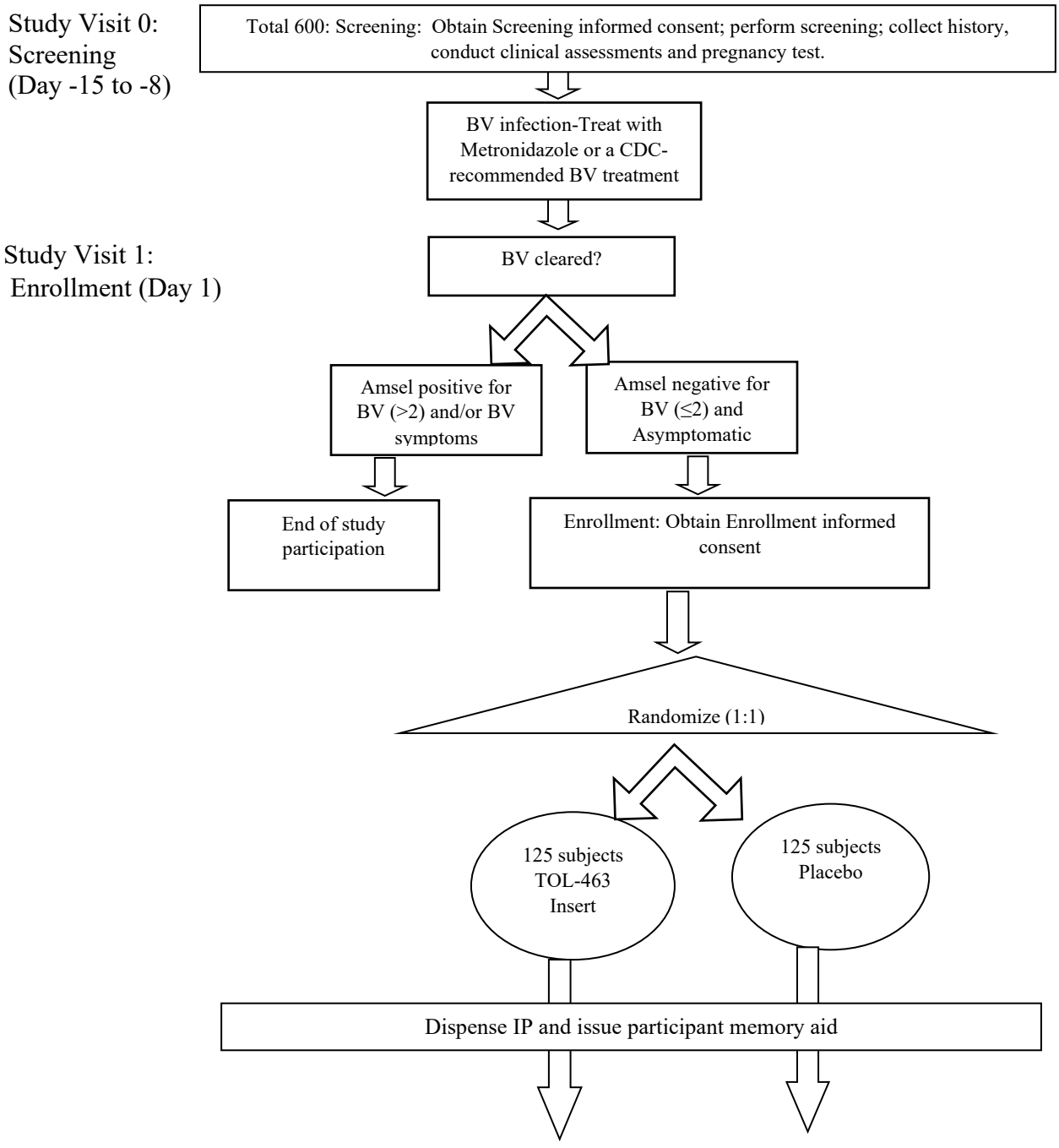
US	United States
USP-NF	United States Pharmacopeia and The National Formulary
WBC	White blood cell
VVC	Vulvovaginal Candidiasis
V0	Visit 0, Screening
V1	Visit 1, Enrollment
V2	Visit 2, First Follow-up, weeks 1-4
V3	Visit 3, Second Follow-up, weeks 5-8
V4	Visit 4, Third/Final Follow-up, weeks 9-12

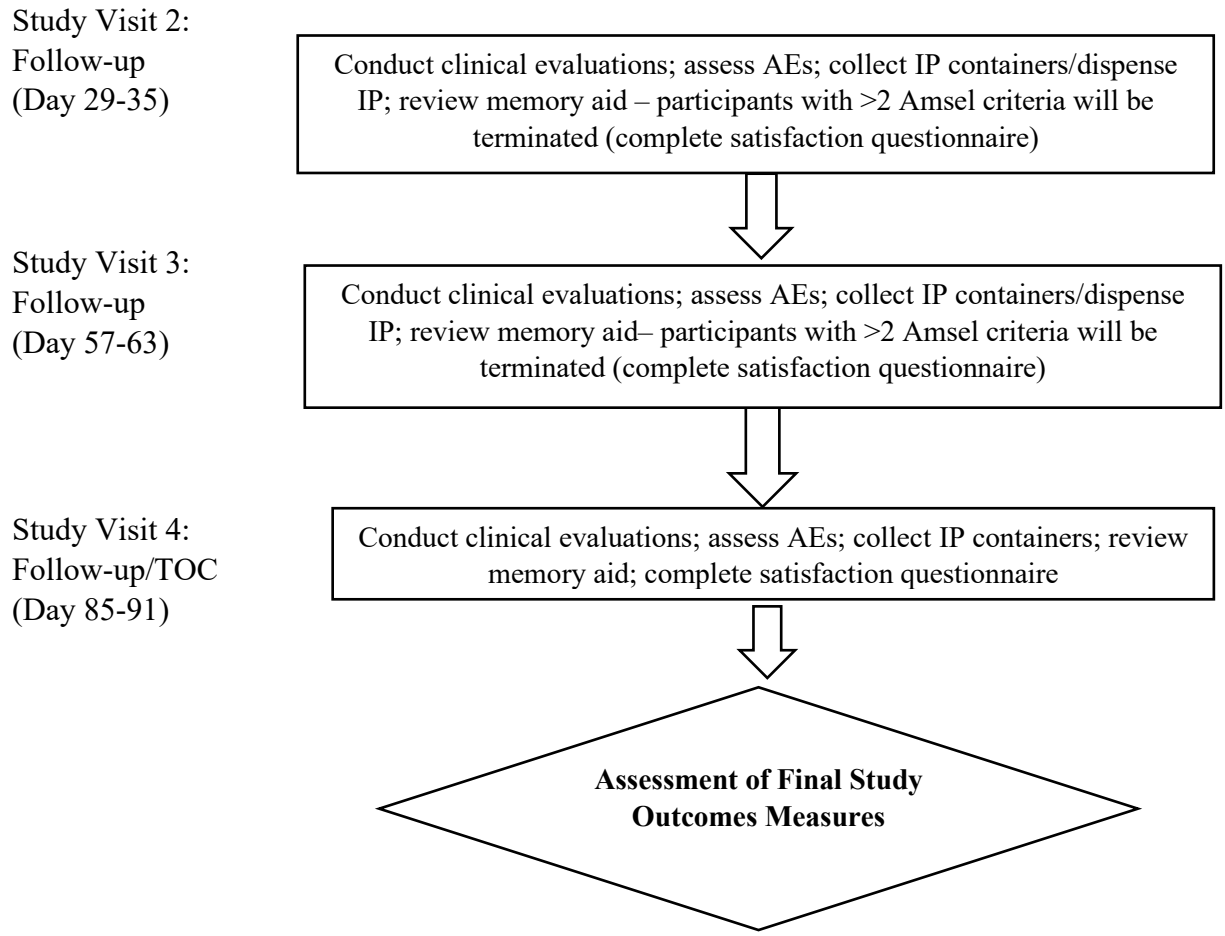
PROTOCOL SUMMARY

Title:	A Randomized, Double-Blind, Placebo-Controlled Trial of TOL-463 Insert for Suppression of Bacterial Vaginosis (BV)
Design of the Study:	TOL-463 Insert or placebo as determined by randomization, administered twice-weekly for 12 weeks, with 3 in-clinic follow-up visits post enrollment (V1) at Visit 2 (Day 29), Visit 3 (Day 57) and Visit 4 /TOC (Day 85) for safety, clinical, and laboratory assessment.
Study Phase:	II
Study Population:	Approximately 600 adult females, 18-55 years of age, with at least three diagnoses of BV in the prior 12 months, including the diagnosis at the initial screening visit to enroll approximately 250 participants (to achieve 200 evaluable participants).
Number of Sites:	Four (Birmingham, Alabama; Pittsburgh, Pennsylvania; Chicago, Illinois; San Diego, California)
Description of Study Product:	TOL-463 Insert or placebo administered vaginally twice-weekly for 12 weeks
Study Objectives:	Primary: • To evaluate the clinical efficacy of a twice-weekly application of TOL-463 vaginal insert in suppression of BV in women with a history of recurrent BV (RBV) following successful induction with oral metronidazole or a CDC-recommended BV treatment

	Secondary: • To evaluate the time to BV recurrence as defined by clinical criteria • To evaluate the incidence of vaginal symptoms over study participation • To evaluate acceptability of a twice-weekly application of TOL-463 vaginal insert in suppression of BV • To evaluate the safety of TOL-463 vaginal insert compared to placebo, including the incidence of secondary VVC Exploratory: • To evaluate the performance of the BD Max Vaginal Panel in characterizing clinical and microbiologic outcomes of the intervention
Duration of Individual Participant Participation:	Participation will include a Screening Visit, an Enrollment Visit, and 3 clinical follow-up visits. Total participation for each participant will be approximately 100 days
Estimated Time to Last Participant/Last Study Day:	Approximately 24 months.

Figure 1: Schematic of Study Design




Study Visit 2:
Follow-up
(Day 29-35)
Conduct clinical evaluations; assess AEs; collect IP containers/dispense IP; review memory aid – participants with >2 Amsel criteria will be terminated (complete satisfaction questionnaire)
Study Visit 3:
Follow-up
(Day 57-63)
Conduct clinical evaluations; assess AEs; collect IP containers/dispense IP; review memory aid– participants with >2 Amsel criteria will be terminated (complete satisfaction questionnaire)
Study Visit 4:
Follow-up/TOC
(Day 85-91)
Conduct clinical evaluations; assess AEs; collect IP containers; review memory aid; complete satisfaction questionnaire
Assessment of Final Study Outcomes Measures

1 KEY ROLES

Lead Principal Investigator:	Jeanne Marrazzo, MD, MPH University of Alabama at Birmingham School of Medicine Tinsley Harrison Tower 215 1900 University Blvd. Birmingham, AL 35294-0006
DMID Clinical Project Manager:	Jonathan Glock, MPH DMID/NIAID/NIH 5601 Fishers Lane, 8E48 Rockville, MD 20852
Statistical and Data Coordinating Center (SDCC):	The Emmes Company, LLC 401 N. Washington St. Suite 700 Rockville, MD 20850 (301) 251-1161

2 BACKGROUND AND SCIENTIFIC RATIONALE

2.1 Background

Bacterial vaginosis (BV) and vulvovaginal candidiasis (VVC), collectively referred to as vaginitis, are the two most prevalent lower reproductive tract infections, affecting tens of millions of women annually in the United States (US) and far more globally. BV is characterized by a general depletion of specific *Lactobacillus* species that are necessary for maintaining vaginal health and a dramatic increase in commensal organisms, notably *Gardnerella vaginalis* and other anaerobes; a local inflammatory response to *Candida* species (primarily *C. albicans*) characterizes VVC. Although currently recommended regimens afford reliable symptom relief, failure rates for BV and VVC are as high as 83% and 60%, respectively, [1-3] and in which the survival of resistant biofilm communities have been increasingly implicated. These failure rates represent an important public health concern because these infections are associated with a host of serious complications including increased risk of acquiring and transmitting human immunodeficiency virus (HIV) and other sexually transmitted infections (STIs) and adverse pregnancy outcomes, including preterm premature rupture of membranes (PPROM), preterm birth and low birth weight infants.[4-6]

This study will assess the efficacy of TOL-463 insert in suppressing BV.

2.2 Scientific Rationale

2.2.1 Purpose of Study

BV is the most common reason that women in the US seek clinical care for vulvovaginal complaints and is the most commonly diagnosed cause of this syndrome across all care settings, including prenatal clinics, primary care clinics, and women's health settings.[4, 7] Well-executed studies indicate that up to one-third of adult women in the US may have BV at any given time,[8] and the population attributable risk in the US of BV for pre-term (early) deliveries, which are associated with substantial infant and maternal morbidity, has been estimated to be as high as 30%.[7] This is likely mediated by ascendant infection with the anaerobic bacteria associated with BV and their precipitation of intra-amniotic infection and subsequently PPROM and preterm delivery. Moreover, it is well established that BV increases women's risk of upper reproductive tract infection (pelvic inflammatory disease (PID)) and of acquiring and transmitting other genital infections, notably HIV, human papillomavirus (HPV), and genital herpes.[4] Unfortunately, current treatment options are limited to nitroimidazole and

clindamycin-based regimens, and despite the broad scientific consensus recognizing the serious adverse outcomes associated with BV, these treatment options are suboptimal. While some women derive immediate relief from the available antibiotic-based therapies, therapeutic cure rates remain low; moreover, recurrence after initially successful treatment is the rule. Over 70% of women in prospective studies experience recurrence in the year following treatment, often multiple times.[4, 9] New and innovative approaches for the immediate treatment and ongoing suppression for BV are urgently needed, especially ones that can address the exceedingly common problem of BV persistence and recurrence.

2.2.2 Purpose of Study

This study will evaluate the safety and efficacy of twice-weekly doses of TOL-463 Insert as an agent for the suppression of BV following successful induction therapy with a standard course oral metronidazole or a CDC-recommended BV treatment. Participants will be randomized to receive either TOL-463 Insert or placebo.

2.2.3 Study Population

Women between the ages of 18 and 55 presenting to US health clinics and responding to advertisements will be invited to participate in this study. Women who are pregnant will not be permitted to enroll as TOL-463 Insert has not been studied in pregnancy.

2.3 Potential Risks and Benefits

2.3.1 Potential Risks

There may be some physical discomfort associated with pelvic examination and with obtaining vaginal secretions for microscopy and diagnostic tests. If a Pap smear is needed it may cause minor cramping or minimal bleeding afterwards. Information concerning participants' sexual history is necessarily detailed given the study's objectives and may provoke some minor psychological or emotional stress when requested; women who receive a new diagnosis of a STI during the study may also experience minor psychological or emotional stress. The oral metronidazole that eligible screened participants will take is recommended by the CDC for the treatment of BV but does have side effects as described in the packaging insert. If a different regimen recommended for the treatment of BV by the CDC is used, refer to the packaging insert for the potential side effects. Participants who develop VVC during study participation and use oral fluconazole may experience possible side effects as described in the packaging insert. It is possible that a participant's cleared BV may not be suppressed by TOL-463 Insert or placebo. If this happens, study staff will provide alternative treatment at no cost to the participant.

2.3.1.1 Potential Risks Associated with TOL-463

The safety and tolerability of TOL-463 are supported by the results of completed Phase I and Phase II clinical studies sponsored by the NIH through the NIAID, Division of Microbiology and Infectious Diseases (DMID Protocol No. 11-0077 and 15-0039, respectively). The findings are consistent with the two-week Good Laboratory Practice (GLP) vaginal irritation and toxicity study of TOL-463 with toxicokinetic endpoints in rabbits, also sponsored by the NIH, demonstrating absence of safety signals and minimal systemic exposure to TOL-463. The most common treatment-related adverse events, (AEs), reported in TOL-463 clinical trials were localized in nature, e.g., vulvovaginal burning and pruritus, and none led to discontinuation of study product. All treatment-related AEs were mild to moderate in intensity and self-limiting. No serious or severe AEs related to TOL-463 were reported. Please refer to Investigator's Brochure (IB) for additional information.

2.3.2 Potential Benefits

Participants may benefit from this study by finding out more about the specific cause of any vaginal symptoms they may be experiencing. Participants may benefit from pre- and post-test counseling, treatment, and referrals, as necessary. It has previously been observed that participants are empowered by knowing, for example, more about what BV is and how it can be prevented.

The main therapeutic component of the study product is recognized as an effective component of multi-drug regimens to prevent recurrent BV.[10-12] Thus, women may benefit from the use of a novel agent that incorporates this chemical compound into therapeutic form for these infections.

3 STUDY DESIGN, OBJECTIVES AND ENDPOINTS OR OUTCOME MEASURES

3.1 Study Design Description

Schematic of Study Design - See Figure 1.

This is a Phase II randomized, double-blind, placebo-controlled study screening approximately 600 adult females 18-55 years of age to enroll approximately 250 participants to achieve 200 evaluable participants at the test of cure (TOC) visit. The study is designed to determine the clinical efficacy of an investigational product (IP), TOL-463 Insert, manufactured by Toltec Pharmaceuticals, in suppressing RBV when administered to women who have a history of RBV and have been successfully cleared of their current BV infection using oral metronidazole, 500 mg twice a day for 7 days or another CDC-recommended BV treatment.

Participants who consent to screening will have baseline clinical information collected at Visit 0, screening (Day -15 to -8), (V0). This information will consist of demographics, current vaginal symptoms, medical and sexual history, concomitant medications, and clinical and laboratory findings. Participants will undergo a pelvic examination and have vaginal specimens collected for confirmation of BV and for diagnostic testing for common STIs (*C. trachomatis, N. gonorrhoeae,* and *T. vaginalis*). Women with a clinical diagnosis of BV who meet all other screening Inclusion/Exclusion criteria will be treated with oral metronidazole, 500 mg twice a day for 7 days, or another CDC-recommended BV treatment, and scheduled for a Visit 1, enrollment, (V1) in 8 to 15 days. At the enrollment visit, women who test negative for BV, defined as ≤ 2 of 4 Amsel criteria and absence of symptoms of BV (absence of homogenous discharge and odor), will be offered to review and sign the Enrollment consent. Women who consent and qualify will be enrolled and randomized (1:1) to one of two groups (TOL-463 Insert or placebo). IP will be administered vaginally, twice-weekly for 12 weeks. Follow-up clinic visits will occur at Visit 2 (V2), Visit 3 (V3), and Visit 4 (V4). For women with cleared BV at enrollment, the primary endpoint will be clinical failure, RBV occurring by V4.

Safety will be measured by participant report, targeted physical examination, and pelvic examination. All AEs will be collected through V4. Safety oversight will be provided by a Data and Safety Monitoring Board (DSMB).

Laboratory testing will include urine pregnancy, nucleic acid amplification tests (NAATs) for chlamydia, gonorrhea, and trichomoniasis, and Gram stain for Nugent scoring, as well as assessment of vaginal pH, saline microscopy (clue cells, motile trichomonads, yeast forms), and

KOH preparation at each site's local lab. *Candida* culture will be performed as clinically indicated. Polymerase Chain Reaction, PCR-based molecular diagnosis of BV through a multi-organism PCR algorithm on the BD Max Vaginal Panel will be performed at a certified laboratory for all scheduled study visits.

The duration of the study for participants who are enrolled and randomized will be approximately 100 days. For those not enrolled, participation will end at their V1, Day 1. Enrollment is expected to be completed in approximately twenty four months.

All participants will be asked to abstain from anal, receptive oral, and vaginal sexual intercourse within one hour of IP administration and for 24 hours after. Participants who are sexually active will be required to use an effective form of birth control during study participation through the final visit/V4. A simplified participant memory aid will be provided to study participants to document symptoms, AEs, and protocol compliance.

For additional details on study procedures and evaluations and study schedule by study visits, see Sections 6 and 7 and Appendix A

3.2 Study Objectives

3.2.1 Primary

- To evaluate the clinical efficacy of a twice-weekly application of TOL-463 vaginal insert in suppression of BV in women with a history of recurrent BV (RBV) following successful induction with oral metronidazole or another CDC-recommended BV treatment.

3.2.2 Secondary

- To evaluate the time to BV recurrence as defined by clinical criteria.
- To evaluate the incidence of vaginal symptoms over study participation.
- To evaluate acceptability of a twice-weekly application of TOL-463 vaginal insert in suppression of BV.
- To evaluate the safety of TOL-463 vaginal insert compared to placebo, including the incidence of secondary VVC.

3.2.3 Exploratory

- To evaluate the performance of the BD Max Vaginal Panel in characterizing clinical and microbiologic outcomes of the intervention.

3.3 Study Endpoints

3.3.1 Primary Endpoint

- Among participants with RBV at baseline, the proportion of participants with recurrent BV by V4 as defined by clinical cure [Study day 85-91].

  Recurrence of BV at each follow-up visit is defined as > 2 of the following Amsel criteria:

  a. Positive KOH whiff test
  b. Homogeneous discharge characteristic of BV
  c. Clue cells ≥ 20% of vaginal squamous epithelial cells
  d. Vaginal pH > 4.5

3.3.2 Secondary Endpoints

1. Time to BV recurrence.
2. Proportion of participants reporting BV symptoms through the follow-up period.
3. Number of participants satisfied with the study treatment through the follow-up period.
4. The occurrence of AEs considered product-related following initiation of study treatment and through the final study visit.
5. The occurrence of culture confirmed secondary VVC following initiation of study treatment and through the final study visit.

3.3.3 Exploratory Endpoint

- To evaluate the performance of the BD Max Vaginal Panel in characterizing clinical and microbiologic outcomes of the intervention.

4 STUDY INTERVENTION/INVESTIGATIONAL PRODUCT

Oral metronidazole, 500mg, is acquired locally at each site and provided to participants as standard of care for women with BV who are eligible during the Screening Visit, V0. Store per manufacturer's instructions. If another CDC-recommended BV treatment is provided to participants, it would be acquired locally at each site and stored per manufacturer's instructions.

Oral fluconazole is acquired locally at each site and provided to participants as standard of care for women with VVC infections. Store per manufacturer's instructions.

4.1 Study Product Description

TOL-463 is a non-azole vaginal anti-infective drug candidate designed as a dual-indication therapy for BV and VVC. The product is formulated as a hydrophilic melt insert. All excipients are United States Pharmacopeia and The National Formulary (USP-NF) materials and are formulated at levels consistent with the acceptable use ranges specified within the FDA Inactive Ingredient Guide.

4.1.1 Formulation, Packaging, and Labeling

TOL-463 Vaginal Inserts and Placebo

TOL-463 vaginal inserts and matching Placebo vaginal inserts will be supplied in 2.0 cc/g low-density polyethylene (LDPE) + polyvinyl chloride (PVC) plastic unit-dose insert molds with ten vaginal applicators.

All study product will be labeled according to manufacturer or regulatory specifications and include the statement "Caution: New Drug – Limited by Federal law to Investigational Use." Clinical trial material (CTM) will be double blinded and ongoing stability studies conducted on those CTM lots to support the conduct of the clinical trial.

Please see IB for additional information.

4.1.2 Product Storage and Stability

TOL-463 Vaginal Inserts

The TOL-463 inserts should be stored at room temperature (20-25°C/68-77°F) and are expected to be stable for at least 24 months under these conditions based on stability data for TOL-463

research lots and Phase I and Phase II CTM. Stability testing of the TOL-463 insert lot manufactured for this study will remain ongoing throughout the course of the trial to ensure product integrity under the specified storage conditions.

Placebo for Vaginal Inserts

Placebo inserts should also be stored at room temperature (20-25°C/68-77°F). Stability testing of the placebo insert lot manufactured for this study will remain ongoing throughout the course of the trial to ensure product integrity under the specified storage conditions.

4.2 Acquisition/Distribution

TOL-463 Vaginal Inserts

TOL-463 Vaginal Inserts are manufactured and provided by Toltec Pharmaceuticals.

Placebo for Vaginal Inserts

Placebo Vaginal Inserts are manufactured and provided by Toltec Pharmaceuticals.

Upon request by DMID, the investigational products will be transferred to the following address:

DMID Clinical Materials Services (CMS)

Fisher BioServices

20439 Seneca Meadows Parkway

Germantown, MD 20876

Phone: (240) 477-1350

Fax: (240) 477-1360

Email: DMID.CMS@ThermoFisher.com

Study products will be shipped from the DMID Clinical Materials Services (CMS) to the investigational site upon request and approval by NIAID DMID.

4.3 Dosage, Preparation, Dispensing and Administration of Study Intervention/Investigational Product

The Research Pharmacist or delegated study staff is responsible for ensuring that the policies and regulations within their jurisdiction are followed when preparing and dispensing study products. This includes following requirements as they pertain to prescriptions, medication prescribing orders and labeling. In addition, instructions specific to a trial or about a study product must also be followed. A Site Research Pharmacist may be delegated the responsibility of IP dispensation. The Research Pharmacist must be a licensed, registered pharmacist and is the preferred healthcare practitioner to be delegated to perform this activity. If a Research Pharmacist is not available, a physician, nurse practitioner, physician assistant, registered nurse, or other authorized healthcare practitioner who is a member of the clinical study staff may be delegated to dispense the IP. These personnel must be licensed, trained, and qualified to prepare investigational study product and must be authorized to dispense the IP under state and local rules and regulations.

Participants will be randomized to one of two IPs, as follows:

- TOL-463 Vaginal Insert: Administered vaginally as a single 2 g unit dose twice a week with a minimum of a two-day duration in between doses for twelve weeks with the aid of a disposable applicator.
- Placebo: Administered vaginally as a single 2 g unit dose twice a week with a minimum of a two-day duration in between doses for twelve weeks with the aid of a disposable applicator.

4.4 Accountability Procedures for the Study Intervention/Investigational Product(s)

Fisher will ship packaged IP to the investigational sites. At each site, the IP will be stored appropriately, including segregation in a secure location and under controlled storage condition and in accordance with applicable regulatory requirements and administered by the delegated study personnel. A drug accountability log will be filled out for all IP at the sites.

After receipt of the IP, the site PI is responsible for its distribution and disposition and has ultimate responsibility for IP accountability. The site PI may delegate to a site research pharmacist or an appropriately qualified staff member the responsibility for IP accountability. The designee will be responsible for maintaining complete records and documentation of product receipt, accountability, dispensation, temperature and storage conditions, and final disposition of IP. All IP, whether administered or not, must be documented on the appropriate IP accountability

record or dispensing log. The sponsor's monitoring staff will verify the participating clinical sites' IP accountability records and dispensing logs per the study monitoring plan.

Used and unused IP will be retained until monitored and released for disposition, as applicable. This can occur on an ongoing basis for used IP.

Used IP and its packaging may be destroyed in accordance with site-specific SOPs following each monitoring visit where IP accountability is monitored, and discrepancies resolved. However, retaining used IP and/or its packaging until a monitoring visit occurs is only required when the local institution's SOP mandates. If local SOPs allow destruction of IP and its packaging, both can be destroyed per the site's SOPs prior to monitoring visits and after written permission is given by DMID. The site will ensure a double count of the IP and the packaging is performed and documented before destruction for use in monitor reconciliation. Further instructions are described in the protocol-specific MOP.

Unused dispensed IP and its packaging returned (from study participants) to the site will be accounted for at the site and participant compliance will be recorded. Any unused returned IP and its packaging will be disposed in accordance with site-specific SOPs.

Upon completion or termination of the study and after the final monitoring visit where IP accountability is monitored, and resolution of any discrepancies, final disposition of the unused (never dispensed IP) will be disposed in accordance with the Manual of Procedures (MOP) and site-specific SOPs.

5 SELECTION OF PARTICIPANTS AND STUDY ENROLLMENT AND WITHDRAWAL

This study will screen approximately 600 women 18-55 years of age who have recurrent RBV, defined as a current BV infection and at least two previous episodes of BV by documentation or self-report in the past year. Women who meet Screening eligibility will be treated with a standard course of oral metronidazole or another CDC-recommended BV treatment. Participants will return within approximately one week of completing the metronidazole treatment or other CDC-recommended BV treatment, for assessment of clinical cure. Clinical cure of BV is defined as ≤ 2 of 4 Amsel criteria and absence of reported symptoms of BV (i.e. no malodorous discharge consistent with BV).

Participants who return for the Enrollment Visit, V1 and are determined to be clinically cured will be offered to review and sign the Enrollment consent. Participants who consent and meet Enrollment eligibility will be randomized in a 1:1 ratio to twice weekly blinded suppressive therapy with TOL-463 or placebo. It is estimated that approximately 250 participants will be eligible for Enrollment with approximately 125 participants in each suppressive therapy group.

No exemptions will be granted on Screening or Enrollment eligibility criteria for this DMID-sponsored trial.

Participants will be recruited from health clinics in the U.S.

Those participants who enroll in the study will undergo up to 3 scheduled Follow-up visits to evaluate clinical diagnosis of BV at each visit. Participants who develop vaginal symptoms between scheduled visits will be asked to return for an unscheduled visit. If at any time during a follow-up visit a participant is diagnosed with clinical BV (> 2 Amsel criteria), the study endpoint of BV reoccurrence is met, and participation in the study will end.

The study will target a total of 200 evaluable participants.

5.1 Eligibility Criteria

5.1.1 Participant Inclusion Criteria

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

5.1.1.1 Screening Visit(s) Inclusion Criteria:

1. Women with a current BV infection and a history of at least two previous episodes of BV by documentation or self-report in the past year. V0 diagnosis is based on the presence of ≥ 3Amsel criteria*;
   *Homogeneous vaginal discharge; positive KOH whiff test; vaginal pH of > 4.5; and ≥ 20% clue cells.
2. Willing and able to provide written informed consent;
3. Age 18-55 years of age at the time of V0;
4. General good health based on medical history, targeted physical examination, and pelvic examination;
5. For participants 21 years of age or older, Pap test performed within the past 3 years, with the most recent result being normal or ASCUS HPV negative OR Pap smear collected at V0*;
   *Consistent with current Pap screening guidelines, a Pap smear must be performed at V0, for women who meet the following criteria and cannot provide documentation or self-report of a normal or ASCUS HPV negative Pap smear, conducted within the prior 3 years: (a) has not had a hysterectomy or (b) has had a hysterectomy and has a history of cervical intraepithelial neoplasia grade 2+ (CIN2+) in the past 20 years.
   Note: If a Pap smear is conducted at V0, the results are not required prior to enrollment.
6. Have a negative urine pregnancy test at V0, if of childbearing potential;
7. Must be of non-childbearing potential* OR must be using an effective method of birth control** and must be willing to continue the method through the end of IP administration.
   *Defined as post-menopausal, or status post bilateral tubal ligation, or status post bilateral oophorectomy or status post hysterectomy.
   **Acceptable methods are defined as:
   a. IUDs or hormonal contraceptives for at least 22 days prior to screening. Note: Intravaginal contraceptive rings (e.g., NuvaRing) are not acceptable forms of birth control for this study.
   b. Consistent use of a barrier method, including diaphragms or condoms, for at least 13 days prior to screening.
   c. Abstinence from vaginal sexual intercourse for at least 13 days prior to screening.
   d. Exclusively same-sex relationship.
   e. Monogamous relationship with vasectomized partner.
8. Willing and able to cooperate to the extent and degree required by this protocol at the discretion of the investigator.

5.1.1.2 Enrollment Visit Inclusion Criteria:

In addition to confirming all relevant Screening Visit Inclusion Criteria, women must meet all of the following criteria to be eligible for enrollment in the study.

1. Willing and able to provide Enrollment written informed consent;
2. After completion of metronidazole induction therapy or another CDC-recommended BV treatment, no clinical evidence of BV* and absence of symptoms of BV** at V1;
   *As defined by ≤ 2 of 4 Amsel criteria.
   **Defined as absence of vaginal discharge and odor consistent with BV.
3. Must have a negative urine pregnancy test at V1, if of childbearing potential;
4. Willing to refrain from any intravaginal products/medications* other than the IP throughout the course of the trial;
   * For example: douches, antifungal or antibacterial preparations, lubricants, contraceptive creams, gels, foams, sponges, spermicides.
5. Must agree to abstain from receptive oral, anal, and vaginal sexual intercourse one hour prior to IP administration and for 24 hours after;
6. Willing to refrain from using tampons or menstrual cups for 24 hours after IP administration.
7. Must be of non-childbearing potential* OR must be using an effective method of birth control** and must be willing to continue the method through the end of IP administration.
   *Defined as post-menopausal, or status post bilateral tubal ligation, or status post bilateral oophorectomy or status post hysterectomy.
   **Acceptable methods are defined as:
   a. IUDs or hormonal contraceptives for at least 30 days prior to using IP. Note: Intravaginal contraceptive rings (e.g., NuvaRing) are not acceptable forms of birth control for this study.
   b. Consistent use of a barrier method, including diaphragms or condoms, for at least 21 days prior to using IP.
   c. Abstinence from vaginal sexual intercourse for at least 21 days prior to using IP.
   d. Exclusively same-sex relationship.
   e. Monogamous relationship with vasectomized partner.

5.1.2 Participant Exclusion Criteria

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

5.1.2.1 Screening Visit(s) Exclusion Criteria:

1. Diagnosis of another vaginal or vulvar condition that may confuse interpretation of response to IP*;
   *For example: erosive lichen planus, desquamative inflammatory vaginitis, or contact dermatitis involving the vulvar epithelium.
2. Concurrent VVC infection with inability to be treated with oral fluconazole;
3. Infectious cause of cervicitis (e.g., *N. gonorrhoeae*, *C. trachomatis*, or *T. vaginalis*) confirmed on physical examination and/or with laboratory testing**+;
   ** Women may be rescreened for eligibility following successful treatment of confounding STI.
   +Results of NAAT testing will be reviewed prior to enrollment.
4. Active genital lesions, including ulcers or vesicles consistent with herpes or warts;
5. Planned ongoing immunosuppressive therapy or systemic antibiotic treatment during the course of the study;
6. History of hypersensitivity, allergy or other contraindication(s) to metronidazole or other CDC-recommended BV treatment used to treat subject;
7. History of hypersensitivity to any TOL-463 formulation components (see IB for product information);
8. Current or untreated cervical intraepithelial neoplasia (CIN) or cervical carcinoma;
9. Currently pregnant or nursing;
10. Any other condition that, in the opinion of the investigator, would interfere with participation in the study;
11. Previous enrollment in the study or at the investigator's discretion.

5.1.2.2 Enrollment Visit Exclusion Criteria:

In addition to confirming all Screening Visit Exclusion Criteria, women who meet any of the following criteria will not be eligible for enrollment in the study.

1. Active menses or significant vaginal bleeding as determined by the study clinician at V1*;
   * Note: women who are menstruating may be reevaluated for study enrollment within the enrollment window.
2. Use of vaginal or systemic antibiotic or antifungal since V0, other than oral metronidazole, CDC-recommended BV treatment, or oral fluconazole, as per protocol;
3. Evidence or suspicion of infectious cause of cervicitis or active genital lesion on pelvic examination at V1;
4. Concurrent VVC infection at V1 with inability to treat with oral fluconazole;

5. Use of any investigational drug within 30 days prior to V1 or planned/anticipated use during study participation.

5.2 Withdrawal from the Study, Discontinuation of Study Product, or Study Termination

5.2.1 Withdrawal from the Study or Discontinuation of the Study Product

5.2.1.1 Reasons for Study Withdrawal

Participants may voluntarily withdraw their consent for further study participation at any time and for any reason without penalty or prejudice to future medical care.

Participants may be required to withdraw from further study participation after discussion with the investigator for the following reasons:

- AE(s) judged to be severe (Grade 3) and related to IP
- SAE(s) judged to be related to IP
- Diagnosis of VVC at any follow-up visits, V2 or V3, or unscheduled visit(s) and cannot be treated with oral fluconazole
- A Pap test result from V0 that was anything other than normal or ASCUS HPV negative; participant is no longer eligible for the study.
- Medical disease or condition, or new clinical finding(s) for which continued participation, in the opinion of the investigator might compromise the safety of the participant, interfere with the participant's successfully completion of the study, or interfere with the evaluation of responses
- Violation of eligibility criteria
- Significant deviation from the treatment plan specified in the protocol (i.e., incorrect administration of IP, failure to attend study visits)
- Participant lost to follow-up
- At the discretion of the investigator

5.2.1.2 Handling of Withdrawals

If a participant withdraws or is withdrawn prior to completion of the study, the reason for this decision must be recorded in the case report forms (CRFs). The investigator will inform the participant that data that has already been collected will be retained and analyzed even if the participant withdraws from this study.

If withdrawal occurs after enrollment, any participant with an ongoing AE will be followed until the AE is resolved or until the participant's condition becomes stable.

Participants who request to withdraw their consent from further study participation after study treatment with IP, will be reminded of the importance of continuing in the study for safety evaluations and will be encouraged to complete the Early Termination Visit if they choose not to complete the remaining study visit(s). The Early Termination Visit procedures are listed in Section 6.3.4. Participants who choose to decline continuation in study participation will no longer be contacted for follow-up.

In the case of participants who fail to appear for a follow-up safety assessment, extensive effort (i.e., three documented contact attempts via phone calls, emails, etc., made on separate occasions) will be made to locate or recall them or at least to determine their health status. Participants who cannot be located after extensive effort will no longer be contacted for follow-up. These efforts will be documented in the participant's records.

If a participant is withdrawn from the study due to an AE that prohibits continued participation in the study, she will be given appropriate care and treatment under medical supervision until the condition has resolved or becomes stable.

Safety and efficacy data will be collected on any participant who is withdrawn from the efficacy endpoint analysis. Withdrawal of participants from the analysis populations is discussed in Section 10.5.1.

5.2.1.3 Discontinuation of Study Product

A participant may discontinue use of the IP as a result of their own decision, due to the investigator's discretion, or due to an AE. All participants who discontinue use of the IP will be encouraged to complete the Early Termination Visit. The Early Termination Visit procedures are listed in Section 6.3.4.

If the participant discontinued use of the IP due to an AE, she will be given appropriate care and treatment under medical supervision until the condition has resolved or becomes stable. If the participant consents, every attempt will be made to follow all AEs through resolution.

5.2.2 Participant Replacement

Participants who sign the Enrollment Informed Consent (EIC), are randomized and receive IP and then subsequently withdraw, or are withdrawn from this study, or are lost to follow-up will not be replaced. Participants who withdraw consent after signing the EIC form and are randomized but have not received IP may be replaced.

5.2.3 Study Termination

Although the study sponsor has every intention of completing the study, the sponsor reserves the right to terminate the study at any time for clinical or administrative reasons. Reasons for termination include, but are not limited to, study closure due to DSMB review and recommendation or at the discretion of DMID.

If the study is prematurely terminated for any reason, the investigator will promptly inform the study participants and assure appropriate therapy or follow-up for the participants, as necessary. The investigator will provide a detailed written explanation of the termination to the IRB/IEC.

6 STUDY PROCEDURES

Study visit information is listed in this section, Section 7, and the Schedule of Events (Appendix A). Further instructions are described in the protocol-specific MOP.

6.1 Visit 0 (Day -15 to -8) Screening

- Potential participants will be provided with a description of the study (purpose and study procedures) and asked to read and sign the Screening Informed Consent form, (SIC). The SIC will be signed prior to performing any study procedures, including any screening procedures.
- Demographic information will be collected from the participant.
- Eligibility criteria will be reviewed with the participant.

At this juncture, if a potential participant is not eligible, information regarding reason for screen failure will be noted and the participant will not have any additional screening procedures completed.

- Complete medical history will be obtained by both interview of participants and review of medical records, if available, to assure eligibility.
- Vaginal symptoms such as odor, itching, irritation, and discharge will be reviewed with the participant.
- Sexual history for the past 30 days will be collected.
- A urine pregnancy test will be performed on all participants of childbearing potential and must be negative prior to administration of oral metronidazole or other CDC-recommend BV treatment induction therapy.
- All concomitant medications taken in the last 30 days will be recorded on the appropriate data collection form (DCF).
- A pelvic examination to include examination of external genitalia will be conducted using a non-lubricated speculum and swabs and samples obtained for analysis of Amsel criteria; KOH and saline microscopy for identification of yeast forms, trichomonas, and clue cells; NAAT for *C. trachomatis, N. gonorrhoeae*, and *T. vaginalis*; BD Max baseline test; future analysis, for participants who consented to future use of their specimens; and Gram staining

for Nugent scoring. Instructions for obtaining vaginal swab specimens for these assessments are outlined in the protocol-specific MOP. Gram stain Nugent scoring will be conducted in the Central Laboratory; all other testing will be conducted locally. A Pap smear should also be done for any participant who meets the following criteria and cannot provide documentation (which can include self-report) of a normal or ASCUS HPV negative Pap test within the prior 3 years: is 21 years of age or older and (a) has not had a hysterectomy or (b) has had a hysterectomy and has a history of CIN2+ in the past 20 years. If a Pap smear is conducted at V0, the results are not required prior to enrollment.

- Vulvovaginal signs such as erythema, edema, excoriation, and discharge will be documented and graded by severity, as applicable.
- A clinical determination for BV will be made based on evaluation of Amsel criteria; the presence of 3/4 criteria is required for a diagnosis of BV at this initial screening.
- BV positive participants meeting all other study criteria will be provided with oral metronidazole, 500 mg twice daily for 7 days or another CDC-recommended BV treatment.
- Participants will be scheduled to return for Visit 1 (Day 1) Enrollment to assess response to metronidazole or other CDC-recommended BV treatment induction therapy and if determined to be a clinical BV cure (by Amsel criteria) and asymptomatic will be invited to review and sign the EIC. If participants are not able to keep the scheduled Visit 1 and exceed the 15-day window, the participant must be re-screened before they can be enrolled.

6.2 Visit 1 (Day 1) Enrollment

- Study personnel will review screening study eligibility criteria to ensure unchanged since the initial screening visit.
- Vaginal symptoms such as odor, itching, irritation, and discharge will be reviewed with the participant.
- Vulvovaginal signs such as erythema, edema, excoriation, and discharge, will be documented and graded by severity, as applicable.
- A pelvic examination to include examination of external genitalia will be conducted using a non-lubricated speculum and swabs and samples obtained for analysis of Amsel criteria; KOH and saline microscopy for identification of yeast forms, trichomonas, and clue cells. Instructions for obtaining vaginal swab specimens for these assessments are outlined in the protocol specific MOP.

- A clinical determination to confirm BV cure, determined by ≤ 2 of 4 Amsel criteria and absence of BV symptoms, are considered eligible for study enrollment. Participants who do not clear BV will be informed that they are no longer eligible. Participants who are BV cured with a secondary VVC infection but appropriate candidates for oral fluconazole treatment can be enrolled. Treatment with oral fluconazole will be per clinic standard of care and provided at no cost to the participant.
- Participants who no longer have BV, will be asked to read and sign the EIC at this time. The EIC form will be signed prior to performing any study procedures.
- Enrollment eligibility criteria will be reviewed with the participant.
- Medical history will be reviewed and updated as appropriate.
- Interim sexual history since the last clinic visit will be collected.
- A urine pregnancy test will be performed on all participants of childbearing potential and must be negative.
- All concomitant medications taken since V0 will be recorded on the appropriate DCF. Previously recorded medications will be updated as appropriate.
- Participants will be registered in Advantage eClinical after being classified as negative for BV and eligible for the study, and randomly assigned to active treatment (TOL-463 Insert) or placebo.
- A targeted physical examination including height and weight will be performed by a qualified study clinician.
- Collect vaginal swabs for Gram staining for Nugent scoring; the BD Max test; and for future analysis, for participants who consented to future use of their specimens. See the protocol-specific MOP for guidance on swab collection.
- Participants with clinical evidence of VVC based on vulvovaginal signs (edema, erythema, and/or excoriation) and symptoms (vulvovaginal itching, burning, and/or irritation) will have a confirmatory vaginal *Candida* culture and treated as appropriate with oral fluconazole. Participants who cannot take oral fluconazole will be terminated from the study.
- Blinded IP based on randomization assignment will be provided to the participant in a prepared kit with other study supplies.

- Study protocol requirements and dosing instructions will be reviewed with the participant and a participant memory aid and written study instructions provided. Participants will be counseled to refrain from receptive oral, anal, and vaginal sexual intercourse 24 hours prior to the next study visit, as this can affect the accuracy of vaginal pH measurement.
- Participants will be scheduled to return for follow-up at Visit 2 (Day 29).
- Study staff will contact participants as a reminder for Visit 2 and to discuss adherence to IP and any barriers to IP use.

6.3 Planned Study Visits

6.3.1 Visit 2 (Day 29 [Window: Days 29-35]) Follow-up

- Medical history will be reviewed and updated as appropriate.
- Interim sexual history since the last clinic visit will be collected.
- Vaginal symptoms will be assessed.
- A urine pregnancy test will be performed on all participants of childbearing potential.
- Compliance with the study protocol criteria will be assessed; all used and unused study IP packaging will be collected as relevant and recorded.
- Participant memory aid will be collected and reviewed.
- All concomitant medications taken since V1 will be recorded on the appropriate DCF. Previously recorded medications will be updated as appropriate.
- Study personnel will discuss with participants and assess and record all AE/SAEs. Previously recorded AE/SAEs will be updated as appropriate.
- If clinically indicated, a targeted physical examination will be performed by a qualified study clinician.
- A pelvic examination to include examination of external genitalia will be conducted, and swabs and samples taken for analysis of Amsel criteria; KOH and saline microscopy for identification of yeast forms, trichomonas, and clue cells; Gram staining; the BD Max test; and for future analysis, for participants who consented to future use of their specimens. Instructions for obtaining vaginal swab specimens for these assessments are outlined in the

protocol specific MOP. Gram stain Nugent scoring will be conducted in the Central Laboratory; all other testing will be conducted locally.

- Participants with clinical evidence of VVC based on vulvovaginal signs (edema, erythema, and/or excoriation) and symptoms (vulvovaginal itching, burning, and/or irritation) will have a confirmatory vaginal *Candida* culture and treated as appropriate with oral fluconazole. Participants who cannot take oral fluconazole will be terminated from the study.
- Vulvovaginal signs will be assessed and graded by severity, as applicable.
- A clinical determination for BV recurrence will be made based on evaluation of Amsel criteria. The presence of >2 of 4 Amsel criteria is required for a diagnosis of BV recurrence. Participants with a clinical diagnosis of BV recurrence will complete a satisfaction questionnaire and will be terminated from the study.
- Participants who are negative for BV recurrence as determined by ≤ 2 of 4 Amsel criteria will be scheduled for the next return visit, blinded IP and memory aid will be provided, and protocol requirements and dosing instructions reviewed. Participants will be counseled to refrain from receptive oral, anal, and vaginal sexual intercourse 24 hours prior to the next study visit, as this can affect the accuracy of vaginal pH measurement.
- Study staff will contact participants as a reminder for Visit 3 and to discuss adherence to IP and any barriers to IP use.

6.3.2 Visit 3 (Day 57 [Window: Days 57-63]) Follow-up

- Medical history will be reviewed and updated as appropriate.
- Interim sexual history since the last clinic visit will be collected.
- Vaginal symptoms will be assessed.
- A urine pregnancy test will be performed on all participants of childbearing potential.
- Compliance with the study protocol criteria will be assessed; all used and unused study IP packaging will be collected as relevant and recorded.
- Participant memory aid will be collected and reviewed.
- All concomitant medications taken since V2 will be recorded on the appropriate DCF. Previously recorded medications will be updated as appropriate.

- Study personnel will discuss with participants and assess and record all AE/SAEs. Previously recorded AE/SAEs will be updated as appropriate.
- If clinically indicated, a targeted physical examination will be performed by a qualified study clinician.
- A pelvic examination to include examination of external genitalia will be conducted, and swabs and samples taken for analysis of Amsel criteria; KOH and saline microscopy for identification of yeast forms, trichomonas, and clue cells; Gram staining; the BD Max test; and for future analysis, for participants who consented to future use of their specimens. Instructions for obtaining vaginal swab specimens for these assessments are outlined in the protocol specific MOP. Gram stain Nugent scoring will be conducted in the Central Laboratory; all other testing will be conducted locally.
- Participants with clinical evidence of VVC based on vulvovaginal signs (edema, erythema, and/or excoriation) and symptoms (vulvovaginal itching, burning, and/or irritation) will have a confirmatory vaginal *Candida* culture and treated as appropriate with oral fluconazole. Participants who cannot take oral fluconazole will be terminated from the study.
- Vulvovaginal signs will be assessed and graded by severity, as applicable. A clinical determination for BV recurrence will be made based on evaluation of Amsel criteria. The presence of >2 of 4 Amsel criteria is required for a diagnosis of BV recurrence. Participants with a clinical diagnosis of BV recurrence will complete a satisfaction questionnaire and will be terminated from the study
- Participants who are negative for BV recurrence determined as ≤ 2 of 4 Amsel criteria will be scheduled for the next return visit, blinded IP and memory aid will be provided, and protocol requirements and dosing instructions reviewed. Participants will be counseled to refrain from receptive oral, anal, and vaginal sexual intercourse 24 hours prior to the next study visit, as this can affect the accuracy of vaginal pH measurement.
- Study staff will contact participants as a reminder for Visit 4 and to discuss adherence to IP and any barriers to IP use.

6.3.3 Visit 4 (Day 85 [Window: Days 85-91]) Follow-up (TOC)

- Medical history will be reviewed and updated as appropriate.
- Interim sexual history since the last clinic visit will be collected.
- Vaginal symptoms will be assessed.

- A urine pregnancy test will be performed on all participants of childbearing potential.
- Compliance with the study protocol criteria will be assessed; all used and unused study IP packaging will be collected as relevant and recorded.
- Participant memory aid will be collected and reviewed.
- All concomitant medications taken since V3 will be recorded on the appropriate DCF. Previously recorded medications will be updated as appropriate.
- Study personnel will discuss with participants and assess and record all AE/SAEs. Previously recorded AE/SAEs will be updated as appropriate.
- If clinically indicated, a targeted physical examination will be performed by a qualified study clinician.
- A pelvic examination to include examination of external genitalia will be conducted, and swabs and samples taken for analysis of Amsel criteria; KOH and saline microscopy for identification of yeast forms, trichomonas, and clue cells; Gram staining; BD Max test; and for future analysis, for participants who consented to future use of their specimens. Instructions for obtaining vaginal swab specimens for these assessments are outlined in the protocol specific MOP. Gram stain Nugent scoring will be conducted in the Central Laboratory; all other testing will be conducted locally.
- Participants with clinical evidence of VVC based on vulvovaginal signs (edema, erythema, and/or excoriation) and symptoms (vulvovaginal itching, burning, and/or irritation) will have a confirmatory vaginal *Candida* culture and treated.
- Vulvovaginal signs will be assessed and graded by severity, as applicable. A clinical determination for BV recurrence will be made based on evaluation of Amsel criteria. The presence of >2 of 4 Amsel criteria is required for a diagnosis of BV recurrence.
- Participant will complete a satisfaction questionnaire.

6.3.4 Early Termination Visit

The V4 assessments and DCFs (see Section 6.3.3) must be completed at the early termination visit for participants who withdraw, or are withdrawn, or terminated from the study. If a participant withdraws between scheduled visits, the participant will be asked to return to the clinic to perform the V4 assessments.

6.4 Unscheduled Study Visits

Unscheduled visits may occur at any time during the study. For example, for problems with the study medication or development of intercurrent vaginal symptoms. Any of the following activities will be performed at the discretion of the site PI:

- Medical history will be reviewed and updated as appropriate.
- Interim sexual history since the last clinic visit will be collected.
- Vaginal symptoms will be assessed.
- A urine pregnancy test will be performed on all participants of childbearing potential.
- Compliance with the study protocol criteria will be assessed; any used and unused study IP packaging will be collected as relevant and recorded.
- Participant memory aid will be collected and reviewed.
- All concomitant medications taken since the last study visit will be recorded on the appropriate DCF. Previously recorded medications will be updated as appropriate.
- Study personnel will discuss with participants and assess and record all AEs/SAEs. Previously recorded AEs/SAEs will be updated as appropriate.
- If clinically indicated, a targeted physical examination will be performed by a qualified study clinician.
- A pelvic examination to include examination of external genitalia will be conducted, and swabs and samples taken for analysis of Amsel criteria; KOH and saline microscopy for identification of yeast forms, trichomonas, and clue cells; and Gram staining. Instructions for obtaining vaginal swab specimens for these assessments are outlined in the protocol specific MOP. Gram stain Nugent scoring will be conducted in the Central Laboratory; all other testing will be conducted locally.
- Participants with clinical evidence of VVC based on vulvovaginal signs (edema, erythema, and/or excoriation) and symptoms (vulvovaginal itching, burning, and/or irritation) will have a confirmatory vaginal *Candida* culture and treated as appropriate with oral fluconazole. Participants who cannot take oral fluconazole will be terminated from the study.
- Vulvovaginal signs will be assessed and graded by severity, as applicable. A clinical determination for BV recurrence will be made based on evaluation of Amsel criteria. The

presence of >2 of 4 Amsel criteria is required for a diagnosis of BV recurrence. Participants with a clinical diagnosis of BV recurrence will complete a satisfaction questionnaire and will be terminated from the study.

- Participants who are negative for BV determined as ≤ 2 of 4 Amsel criteria will be scheduled for the next return visit, blinded IP and a memory aid will be provided as needed, and protocol requirements and dosing instructions reviewed.

6.5 Protocol Deviations

A protocol deviation (PD) is any noncompliance with the clinical trial protocol, GCP, or MOP requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions should be developed by the site and implemented promptly

These practices are consistent with ICH E6:

4.5 Compliance with Protocol, Sections 4.5.1, 4.5.2, and 4.5.3

5.1 Quality Assurance and Quality Control, Section 5.1.1

5.20 Noncompliance, Sections 5.20.1, and 5.20.2.

It is the responsibility of the site PI and other study staff to use continuous vigilance to identify and report deviations within 5 working days of identification of the PD, or within 5 working days of the scheduled protocol-required activity. All deviations must be promptly reported to DMID, via the SDCC's electronic study database.

NOTE: Those sites participating in trials with a designated "central unit" will follow the reporting requirements specified in their protocols and MOPs. The "central unit" will be responsible for submission of the PD to TRI/ICON DMID CROMS.

All PDs, as defined above, must be addressed in study participant DCFs. A completed copy of the DMID PD Form must be maintained in the Regulatory File, as well as in the participant's study folder. PDs must be submitted to the central IRB/IEC and to the local IRB/IEC per their guidelines. The site PI and other study staff are responsible for knowing and adhering to their IRB requirements.

7 DESCRIPTION OF CLINICAL AND LABORATORY EVALUATIONS

7.1 Clinical Evaluations

Complete medical history will be obtained by interview of the participants at V0 and will be updated at each clinic visit. At follow-up visits, an interim medical history will be obtained by interview of the participants noting any changes since the previous visit.

Sexual history will be obtained by interview of the participants at V0. Participants will be queried at V1 thru V4 regarding sexual activity since the previous clinic visit.

Medications history (concomitant medications) will include a review of all current medications taken 30 days prior to the screening visit and through study completion. Prescription and over-the-counter drugs will be included as well as vitamins, herbs and supplements. Assessment of eligibility will also include a review of all permitted and prohibited medications per the Participant Inclusion and Exclusion Criteria (see Section 5).

A pelvic examination including examination of external genitalia, will be performed at the screening visit, V0. A targeted physical examination including height and weight will be performed at enrollment visit, V1, and if clinically indicated for follow-up visits, and pelvic examination will be performed at the enrollment visit, V1, and at each of the follow-up visits, V2 thru V4. All physical examinations will be performed by a qualified study clinician.

Study IP, memory aids and instructions will be provided to participants at the enrollment visit, V1 and return visits, V2 and V3. Compliance with the study IP will be assessed at all follow-up visits and contacts (V2-V4 and as needed at unscheduled and early termination visits) and recorded.

A pelvic examination and vulvovaginal examination for the evaluation of the vulva, vagina, and cervix will be performed by the site PI or qualified designee. At the screening visit, V0, these examinations will be done for the purpose of determining eligibility. At the enrollment visit, V1, Follow-up visits (V2-V4), or an unscheduled visit these examinations will be done for the purpose of evaluating treatment efficacy and the presence of treatment-emergent AEs (TEAEs).

Pelvic Examination:

During the initial screening visit, V0 pelvic examination, the clinical diagnosis of BV will be based on the presence of 3/4 of the following Amsel criteria:

1. Homogenous vaginal discharge characteristic of BV;
2. A fishy odor of the vaginal discharge with the addition of a drop of 10% KOH (positive whiff test);
3. Clue cells ≥ 20% of the total vaginal squamous epithelial cells on saline microscopy;
4. Vaginal fluid pH >4.5.

Instructions for obtaining and determining the presence of Amsel clinical criteria can be found in the protocol-specific MOP.

During the enrollment visit's, V1 pelvic examination, a clinical determination to confirm BV cure following metronidazole or CDC-recommended BV treatment induction will be determined by ≤ 2 of 4 Amsel criteria and absence of BV symptoms (defined as absence of homogenous discharge and odor).

At all other follow-up visits on suppressive study IP (V2-V4), a clinical determination of BV recurrence will be determined as > 2 of 4 Amsel criteria by pelvic examination.

For the enrollment V1 or follow-up (V2 – V4) visits, participants will be scheduled outside of menses as soon as possible after cessation of menstruation.

At all follow-up visits (V2-V4), a clinical diagnosis of VVC will be based on vulvovaginal signs (edema, erythema, and excoriation) and symptoms (itching, burning, and irritation) and a positive KOH microscopy with identification of yeast forms (hyphae/pseudohyphae) or budding yeasts. A confirmatory Candida culture with speciation will also be performed; Candida culture in the setting of incomplete criteria for the clinical diagnosis may be obtained at the discretion of the study clinician.

7.1.1 Assessment of Concomitant Medications/Treatments other than Study Product

Administration of any medications, therapies, or vaccines will be recorded on the appropriate DCF. Concomitant medications will include all medications taken 30 days prior to the initial screening visit V0, through V4 or early termination, whichever occurs first. Prescription and over-the-counter drugs will be included, as well as herbs, vitamins, and supplements.

Participants who have received study IP and are subsequently diagnosed with a concomitant infection that requires systemic antibiotics will receive treatment according to the local clinic's standard protocols but will be considered non-evaluable for efficacy and terminated from the study. However, participants who have received study IP and are subsequently diagnosed with VVC can remain in the study and be considered evaluable for efficacy if they are appropriate candidates for oral fluconazole therapy.

At the discretion of the site PI, use of new medication should prompt evaluation for the presence of a new diagnosis of chronic medical disease or condition.

Medications that might interfere with the evaluation of the IP should not be used unless absolutely necessary.

Refer to Section 5 (Selection of Participants and Study Enrollment and Withdrawal) for medications that are prohibited for study eligibility and throughout study participation.

7.1.2 Assessment of Participant Compliance with Study Intervention/Investigational Product

The TOL-463 Insert and placebo are inserted by the study participant.

At each follow up visit, V2 through V4, participants will be asked to bring the study IP packaging, unused inserts (if any) and memory aid corresponding to the return visit (V2, V3 or V4) to the study personnel. Study personnel will review adherence to the study IP schedule with the participants, including the participant memory aid, and record this on the appropriate DCF at each follow-up visit and any unscheduled visits. Study personnel will record the number of unused inserts and empty packaging returned at each of the follow-up visits, V2 thru V4. Compliance with the study protocol will be assessed and documented by study personnel at each of the follow-up visits, V2 thru V4, and at any unscheduled visits. Study personnel may discard the participant's memory aid after documenting compliance. Refer to 4.4 for instructions on handling of returned study IP packaging.

7.2 Laboratory Evaluations

7.2.1 Clinical Laboratory Evaluations

At the screening visit, V0, diagnostic laboratory tests for determination of *C. trachomatis*, *N. gonorrhoeae*, and *T. vaginalis* (NAATs) will be performed locally. Vaginal Gram stain Nugent scoring for microbiologic confirmation of clinical findings will be conducted in the Central Laboratory. BD Max baseline testing will be conducted at a certified laboratory. Instructions for obtaining vaginal swab specimens for these assessments are outlined in the protocol-specific MOP**.**

A urine pregnancy test will be performed at all visits on all participants of childbearing potential. Screening and enrollment visit results must be negative and known prior to randomization on V1.

At all scheduled study visits a BD Max test will be conducted at a certified laboratory. At the enrollment and follow-up visits, vaginal Gram stain Nugent scoring for microbiologic confirmation of clinical findings will be conducted in the Central Laboratory. At the follow-up visits participants with clinically diagnosed VVC will have a confirmatory culture for vaginal *Candida* with speciation performed locally.

Nugent Criteria for Determination of BV:

A vaginal swab for microbiologic assessment of BV by Nugent criteria should be performed. The Nugent score utilizes a 0-10 point scale for evaluation of vaginal flora and is based on the weighted sum score of the following three bacterial morphotypes calculated from the slide exam under oil immersion (1000x):

- *Lactobacillus*: Large Gram-negative rods
- *Gardnerella*/*Bacteroides* spp: thin, curved, Gram-variable rods
- *Mobiluncus* spp: thin, curved, Gram-variable rods

The Nugent score is interpreted as follows: 0-3 normal; 4-6 intermediate; and 7-10 BV. For the purposes of this study, a Nugent score of ≥7 will be used to define a microbiologic recurrence of BV for ancillary analysis.

***Candida* Culture for Determination of VVC:**

For participants with a clinical diagnosis of VVC at V1-V4, a vaginal swab should also be taken for confirmation of *Candida* by culture with speciation. See the protocol-specific MOP for further details.

7.2.1.1 Laboratory Specimen Preparation, Handling, and Storage

Specimen preparation, handling, and storage will be done according to local clinic SOPs. The enrolled participants study specimens collected for the BD Max testing and gram stain slides will be stored until the end of the study and then destroyed. Likewise, for enrolled participants, the additional vaginal swab collection and non-albicans *Candida* strain cultures only will be stored for future use. Additional details can be found in the protocol-specific MOP.

7.2.1.2 Laboratory Specimen Shipping

Gram stain and BD Max specimen shipment to the Central Laboratory will occur according to all applicable International Air Transport Association (IATA) requirements. The Central Laboratory address is as follows:

UAB Infectious Diseases Laboratory

703 19th Street South

ZRB 242

Birmingham, AL 35294

Additional details can be found in the protocol-specific MOP.

8 ASSESSMENT OF SAFETY

8.1 Assessing and Recording Safety Parameters

Safety will be monitored throughout the study by targeted physical and pelvic examination; and participant reporting. Safety will be assessed by the frequency and severity of:

1. Serious AEs occurring after randomization and from the time of the IP dose through V4.
2. Non-serious AEs occurring after randomization and from the time of the IP dose through V4.

8.1.1 Adverse Events (AEs)

ICH E6 defines an AE as any untoward medical occurrence in a participant or clinical investigation participant administered a pharmaceutical product regardless of its causal relationship to the study treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of medicinal (investigational) product. The occurrence of an AE may come to the attention of study personnel during study visits and interviews of a study participant presenting for medical care, or upon review by a study monitor.

All AEs not meeting the criteria for "serious adverse events" should be captured on the appropriated CRF. Information to be collected for AEs includes event description, date of onset, clinician's assessment of severity, relationship to IP (assessed only by those with the training and authority to make a diagnosis, or his/her designee), date of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship. All AEs will be followed to adequate resolution.

Any medical condition that is present at the time that the participant is screened will be considered as baseline and not reported as an AE. However, if it deteriorates at any time during the study, it should be recorded as an AE.

All AEs must be graded for severity and relationship to IP.

8.1.1.1 Adverse Events Grading

FDA defines an AE as any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug-related. All AEs (laboratory and clinical symptoms) will be graded for severity and assessed for relationship to IP (see definitions). AEs characterized as intermittent require documentation of onset and duration of each episode. The start and stop date of each reported AE will be recorded on the appropriate DCF and electronic case report form,

(eCRF). Changes in the severity of an AE will be documented to allow an assessment of the duration of the event at each level of intensity.

Severity of Event:

All AEs will be assessed by the clinician using a protocol-defined grading system (refer to Appendix B). For events not included in the protocol-defined grading system, the following guidelines will be used to quantify severity:

- Mild (Grade 1): Events that are usually transient and may require only minimal or no treatment and generally do not interfere with the participant's daily activities.
- Moderate (Grade 2): Events that are usually alleviated with additional specific therapeutic intervention. The event interferes with usual activities of daily living, causing discomfort but poses no significant or permanent risk of harm to the research participant.
- Severe (Grade 3): Events interrupt usual activities of daily living, or significantly affects clinical status, or may require intensive therapeutic intervention. Severe events are usually incapacitating.

Changes in the severity of an AE should be documented to allow an assessment of the duration of the event at each level of intensity to be performed. AEs characterized as intermittent require documentation of onset and duration of each episode.

Relationship to Study Products: The clinician's assessment of the AE's relationship to IP will be done by those with the training and authority to make an assessment, or his/her designee, and the assessment will be part of the documentation process. Whether the AE is related or not, is not a factor in determining what is or is not reported in this trial. If there is any doubt as to whether a clinical observation is an AE, the event should be reported.

In a clinical trial, the study medication must always be suspect. The relationship to study medication will be assessed for AEs using the terms related or not related:

- Related – There is a reasonable possibility that the study product caused the AE. Reasonable possibility means that there is evidence to suggest a causal relationship between the study medication and the AE.
- Not Related – There is not a reasonable possibility that the administration of the study medication caused the event.

8.1.2 Serious Adverse Events (SAEs)

An AE or suspected adverse reaction is considered a SAE if, in the view of either the site PI or sponsor, it results in any of the following outcomes:

- Death,
- a life-threatening adverse event*,
- inpatient hospitalization or prolongation of existing hospitalization,
- a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or
- a congenital anomaly/birth defect.
- Important medical events that may not result in death, be life-threatening, or require hospitalizations may be considered serious when, based upon appropriate medical judgment they may jeopardize the participant and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

* Life-threatening adverse event. An AE is considered "life-threatening" if, in the view of either the site PI or sponsor, its occurrence places the participant at immediate risk of death. It does not include an AE that, had it occurred in a more severe form, might have caused death.

All SAEs will be:

- Assessed for severity and relationship to study product and alternate etiology (if not related to study product) by a licensed study physician or by the Institution as the site PI or Sub Investigator, (SubI).
- Recorded on the appropriate SAE DCF and eCRF.
- Followed through resolution by a licensed study physician.
- Reviewed and evaluated by DMID, an Independent Safety Monitor (ISM) (as deemed necessary), the DSMB (periodic review unless related), and the IRB/IEC.

8.2 Specification of Safety Parameters

Safety specifications are noted in Section 8.1

8.3 Reporting Procedures

8.3.1 Reporting Serious Adverse Events

SAEs will be followed until resolution even if this extends beyond the study-reporting period. Resolution of an SAE is defined as the return to pretreatment status or stabilization of the condition with the expectation that it will remain chronic.

Any AE that meets a protocol-defined serious criterion must be submitted immediately (within 24 hours of site awareness) on an SAE form to the DMID Pharmacovigilance Group, at the following address:

DMID Pharmacovigilance Group

Clinical Research Operations and Management Support (CROMS)

6500 Rock Spring Dr. Suite 650

Bethesda, MD 20817, USA

SAE Hot Line: 1-800-537-9979 (US) or 1-301-897-1709 (outside US)

SAE FAX Number: 1-800-275-7619 (US) or 1-301-897-1710 (outside US)

SAE Email Address: PVG@dmidcroms.com

In addition to the SAE form, select SAE data fields must also be entered into the Advantage eClinical® system. Please see the protocol-specific MOP for details regarding this procedure.

Other supporting documentation of the event may be requested by the DMID Pharmacovigilance Group and should be provided as soon as possible.

The site will send a copy of the SAE report(s) to the ISM (as deemed necessary) when they are provided to the DMID Pharmacovigilance Group. The DMID Medical Monitor and DMID Clinical Project Manager will be notified of the SAE by the DMID Pharmacovigilance Group. The DMID Medical Monitor will review and assess the SAE for regulatory reporting and potential impact on study participant safety and protocol conduct.

At any time after completion of the study, if the site PI or appropriate SubI becomes aware of an SAE that is suspected to be related to study medication, the site PI or appropriate SubI will report the event to the DMID Pharmacovigilance Group.

8.3.2 Regulatory Reporting for Studies Conducted Under DMID-Sponsored IND

Following notification from the site PI or appropriate SubI, DMID, as the IND sponsor, will report any suspected unexpected SAE. DMID will report an AE as a suspected unexpected AE only if there is evidence to suggest a causal relationship between the study intervention and the AE. DMID will submit an IND safety report to the FDA and will notify all participating site PIs (i.e., all PIs to whom the sponsor is providing drug under its IND or under any PI's IND) of potential serious risks from clinical studies or any other source, as soon as possible. DMID will report to the FDA any unexpected fatal or life-threatening suspected adverse reaction as soon as possible, but in no case later than 7 calendar days after the sponsor's initial receipt of the information. If the event is not fatal or life-threatening the IND safety report will be submitted within 15 calendar days after the sponsor determines that the information qualifies for reporting as specified in 21 CFR Part 312.32. Relevant follow up information to an IND safety report will be submitted as soon as the information is available. Upon request from FDA, DMID will submit to the FDA any additional data or information that the agency deems necessary, as soon as possible, but in no case later than 15 calendar days after receiving the request.

All SAEs designated as "not related" to study product(s), will be reported to the FDA at least annually in a summary format.

8.3.3 Reporting of Pregnancy

Participants of child-bearing potential will be counseled to continue using acceptable forms of birth control during the study period. If a participant becomes pregnant during the study, dosing will be discontinued immediately, and early termination assessments will be performed. A pregnancy reporting form will be completed for any study participant who becomes pregnant through seven days after the last IP dose. The site will maintain contact with pregnant study participants to obtain pregnancy outcome information. The pregnant participant will be followed until delivery or until the end of pregnancy (in the case of miscarriage or pregnancy termination). Infants born to these study participants will also be monitored for SAEs (congenital anomalies or other birth defects) and other complications for up to two months after birth. Pregnancy reporting forms will include collecting data on the following information:

- prior maternal history including congenital abnormalities or pregnancy complications;
- estimated and actual date of delivery or end of pregnancy;
- pregnancy outcome (live birth, stillbirth, miscarriage or elective termination);

- mode of delivery;
- maternal complications; and
- neonatal complications (i.e., lethal or nonlethal congenital abnormality).

Pregnancies occurring in study participants will be reported via Advantage eClinical on the Pregnancy Report form.

8.4 Type and Duration of Follow-up of Participants after Adverse Events

AEs and SAEs will be followed from the time of study treatment through resolution even if this extends beyond the study reporting period, V4. Resolution of an AE/SAE is defined as the return to pretreatment status or stabilization of the condition with the expectation that it will remain chronic.

Follow-up procedures, evaluations, and outcomes will be recorded on the appropriate DCF.

8.5 Procedures to be Followed in the Event of Abnormal Laboratory Test Values or Abnormal Clinical Findings

The site PI or appropriate SubI is responsible for reporting all AEs/SAEs that are observed or reported during the study, regardless of their relationship to study product. AEs/SAEs, abnormal clinical laboratory values, or abnormal clinical findings will be documented, reported, and followed appropriately.

8.6 Halting Rules

Further enrollment will be halted for DSMB review/recommendation if any of the following are reported.

If one or more participants experience an SAE judged by an investigator to be related to study IP

If two or more participants experience an AE that is judged to be Grade 3 and related to study IP

An overall pattern of symptomatic, clinical, or laboratory events that the DMID Medical Monitor or DSMB consider associated with study drug and that may appear minor in terms of individual events, but that may collectively represent a serious potential concern for safety

If any of the halting rules are met, the study will not continue with the remaining enrollments or study treatments without a review by and recommendation from the DSMB to proceed.

DMID retains the authority to suspend additional enrollment and administration of study product during the entire study, as applicable.

8.7 Safety Oversight (ISM plus DSMB)

8.7.1 Independent Safety Monitor (ISM)

An ISM at each clinical research site will oversee the safety of research participants at that site and will provide independent written evaluation of SAEs and related Grade 3 AEs to the DMID Clinical Project Manager and DMID Medical Monitor. The ISM will serve as an independent consultant for the site PI on participant-related issues. The ISM will communicate with the site PI and study PIs to resolve any issues.

8.7.2 Data and Safety Monitoring Board (DSMB)

Safety oversight will be conducted by a DSMB that is an independent group of experts that monitors participant safety and advises DMID. The DSMB members will be separate and independent of study personnel participating in this study and should not have scientific, financial or other conflict of interest related to this study. The DSMB will consist of members with appropriate expertise to contribute to the interpretation of the data from this trial.

The DSMB will review study progress and participant clinical and safety data at the following time points:

- An initial organizational meeting will be held prior to the study starting and enrollment of participants.
- At specified times during the course of study as defined in the DSMB Charter.
- Ad hoc meeting convened to address a specific safety concern such as when a halting rule is met or for immediate concerns regarding observations during the study.
- A final closeout review will be held within six months after the database is locked to review all cumulative safety data.

The DSMB will operate under the rules of a DMID-approved charter that will be written at the organizational meeting of the DSMB. Procedures for DSMB reviews/meetings will be defined in the charter. Reports may include enrollment and demographic information, medical history, concomitant medications, physical assessments, clinical laboratory values, dosing compliance, and solicited and unsolicited AE/SAEs. As an outcome of each review/meeting, the DSMB will make a recommendation as to the advisability of proceeding with the study administrations (as applicable), and to continue, modify, or terminate the study.

DMID or the DSMB chair may convene the DSMB on an ad hoc basis according to protocol criteria or if there are immediate concerns regarding observations during the course of the study. The DMID Medical Monitor is empowered to stop enrollment and study treatment if AEs that meet the halting criteria are reported. The DMID Medical Monitor and the ISM (as deemed necessary) will be responsible for reviewing SAEs in real time. The DSMB will review SAEs on a regular basis and ad hoc during the study.

9 HUMAN SUBJECTS PROTECTION

9.1 Institutional Review Board

Each site PI will obtain IRB approval for this protocol to be conducted at the research site(s) and send supporting documentation to the DMID before initiating recruitment of participants. The investigator will submit applicable information to the IRB on which it relies for the review, to conduct the review in accordance with 45 CFR 46, ICH E6 GCP, and as applicable, 21 CFR 56 (Institutional Review Boards) and 21 CFR 50 (Protection of Human Subjects), other federal, state, and local regulations. The IRB must be registered with OHRP as applicable to the research. DMID must receive the documentation that verifies IRB approval for this protocol, informed consent documents (optional pre-screen ICF, SIC and EIC), and upon request any recruitment material and handouts or surveys intended for the participants, prior to the recruitment, screening, and enrollment of participants.

Should amendments to the protocol or consent forms be required, the amendments will be approved by the sponsor and provided to the site PI for IRB submission. IRB review and approval will occur at least annually throughout the enrollment and follow-up of participants and may cease if annual review is no longer required by applicable regulations and the IRB. The PI will notify the IRB of deviations from the protocol and reportable SAEs, as applicable to the IRB policy.

Each institution engaged in this research will hold a current FWA issued by OHRP for federally funded research. The FWA number will be provided to DMID.

A single IRB of record will be accountable for compliance with regulatory requirements for this multi-centered study, at participating sites. A Reliance Agreement, also called an IRB Authorization Agreement will be required to be signed by institutions involved in the study and permits the local IRB to cede review to the single IRB. The IRB Authorization Agreement will set forth the specific responsibilities of the IRB and each participating site. Participating sites will then rely on the IRB of record to satisfy the regulatory requirements relevant to the IRB review. The participating sites will maintain essential required documentation of IRB reviews, approvals, and correspondence, and must provide copies of any agreements and essential documentation to the DMID or regulatory authorities upon request.

9.2 Informed Consent Process

Informed consent is a process that is initiated prior to an individual agreeing to participate in a trial and continuing throughout the individual's trial participation. There will be two consent forms and an optional pre-screen consent form: SIC for the screening procedures (determination of eligibility confirmed by initial clinical BV diagnosis and then subsequent confirmation of BV cleared post induction treatment); an EIC for the full study; and an optional pre-screen consent form to aid in initial BV diagnosis. At the enrollment visit, V1, upon confirmation of BV cure and confirmation of eligibility, the participant will then need to be consented using the EIC prior to continuation. Participants will receive a concise and focused presentation of key information about the clinical trial, verbally and with the written consent forms. The explanation will be organized and presented in lay terminology and language that facilitates understanding why the individual might or might not want to participate.

The site PI, or designated study staff will describe the protocol to potential participants face-to-face. The key information about the purpose of the study, the procedures and experimental aspects of the study, risks and discomforts, any expected benefits to the participant, and alternative treatment will be presented to the participant.

Participants will also receive an explanation that the trial involves research, and a detailed summary of the proposed study procedures and study interventions/medications. This will include aspects of the trial that are experimental, the probability for random assignment to Tol-463 Insert vs. placebo, any expected benefits, all possible risks (including a statement that the study medication may involve risks to the participant or to the embryo or fetus, if the participant may become pregnant, that are currently unforeseeable), the expected duration of the participant's participation in the trial, alternative procedures that may be available and the important potential benefits and risks of these available alternative procedures.

Participants will be informed that they will be notified in a timely manner if information becomes available that may be relevant to their willingness to continue participation in the trial. Participants will receive an explanation as to whether any compensation and any medical treatments are available if injury occurs, and, if so, what they consist of, or where further information may be obtained. Participants will be informed of the anticipated financial expenses, if any, to the participant for participating in the screening and enrollment visits, as well as any anticipated prorated payments, if any, to the participant for participating in the trial. They will be informed of whom to contact (e.g., the investigator) for answers to any questions relating to the research project.

Information will also include the foreseeable circumstances and/or reasons under which the participant's participation in the trial may be terminated. The participants will be informed that participation is voluntary and that they are free to withdraw from the study for any reason at any time without penalty or loss of benefits to which the participant is otherwise entitled.

The extent of the confidentiality of the participants' records will be defined, and participants will be informed that applicable data protection legislation will be followed. Participants will be informed that the monitor(s), auditors(s), IRB, NIAID, and regulatory authority(ies) will be granted direct access to the participant's original medical records for verification of clinical trial procedures and/or data without violating the confidentiality of the participant, to the extent permitted by the applicable laws and regulations, and that, by signing a written informed consent form (ICF), the participant is authorizing such access.

Participants will be informed that records identifying the participant will be kept confidential, and, to the extent permitted by the applicable laws and/or regulations, will not be made publicly available and, if the results of the trial are published, the participant's identity will remain confidential. Participants will be informed whether private information collected from this research and/or specimens will be used for additional research, even if identifiers are removed.

Participants will be allowed sufficient time to consider participation in this research trial and have the opportunity to discuss this trial with their family, friends or legally authorized representative, or think about it prior to agreeing to participate.

The SIC, EIC, and optional pre-screen consent forms will be IRB-approved, and participants will be asked to read and review the SIC form and if applicable, the EIC or pre-screen consent form. Participants must sign the SIC form prior to starting any screening study procedures being done specifically for this trial. Likewise, if applicable participants must sign the EIC form prior to starting any study procedures.

Once signed, a copy of all consent forms will be given to the participant for their records. The participant(s) may withdraw consent at any time throughout the course of the trial. The rights and welfare of the participant(s) will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

Study staff may employ IRB approved recruitment efforts prior to obtaining the participant's consent; however, before conducting protocol-specific screening activities the SIC must be signed. Site clinical staff may pre-screen via chart review and refer potential participants to the study staff.

New information will be communicated by the site PI to participants who sign the EIC form to participate in this trial in accordance with IRB requirements. The EIC form will be updated and participants will be re-consented per IRB requirements, if necessary. Participants will be given a copy of all EIC forms that they sign.

9.3 Consent for Future Use of Stored Specimens and Data

Both the screening and enrolled participant populations will be consented to potentially have extra vaginal swab specimens collected and stored for future research use only. Additionally, for participants who consent, any non-albicans *Candida* strain cultures will be stored for future research use only. Participants who do not consent to have vaginal swab specimens stored for future use can still participate in the study. If the participant consents, an extra swab will be collected at V0 and if the participant is enrolled and consents, one extra swab will be collected at all scheduled study visits. Storage of samples is optional and not a requirement for the study. If the participant consents to storage and future use of specimens, specimens will be stored appropriately at a secure UAB facility.

Participants will not be contacted with the results of these future research studies. Future testing on specimens will only occur to the extent authorized in each study site's SIC and EIC (approved by the single IRB) or as otherwise authorized under applicable law and after review and approval by DMID, the single IRB, and the IRB of the researcher requesting the specimens.

There will be no direct benefit to the participant from allowing the specimens to be stored and used for future purposes. However, the results may provide information that will help in the diagnosis or treatment of future patients. Refer to section 2.3.1 for potential participant risks when obtaining the vaginal swab specimen(s) for future use. During this study, specimens saved for future use will be identified only by the participant number which will allow linkage of the specimens to study data but not to any personal identifiers. Since participant study records are securely maintained by study staff, there is minimal risk of breach of confidentiality. At the end of the study, the future use specimens will be stored in a secure location and unlinked from any information that could connect them with identifiable data from this study.

The participant's future use specimen will be kept indefinitely until it is used up or destroyed. It may be used to develop new tests or products. For example, the specimens could be used to test a new method for detecting vaginal infections, or to study why BV can cause pregnancy problems, or why BV can increase women's risk of HIV infection. In some instances, these may have potential commercial value. No genetic testing or whole genetic sequencing will be performed on the samples.

If a participant decides at any time that she does not want the specimen stored for future research, she must contact the study staff who will then notify the laboratory/specimen archive. The laboratory staff will then mark the specimens by adding a "Destroy" label and they will be destroyed at the end of this study or removed from storage and destroyed.

9.4 Exclusion of Women, Minorities, and Children (Special Populations)

This trial will be inclusive of female adults who meet the Participant Inclusion/Exclusion Criteria, regardless of religion or ethnic background. Should the outcome of this trial be deemed acceptable, additional trials may be initiated in other populations.

Persons who are unable to fully participate in the study due to an inability to provide informed consent will not be enrolled, for example non-English speakers, children, illiterate or non-writing individuals. This exclusion is to ensure participants who cannot fully understand the implications of study participation due to language or cognitive barriers are not recruited.

9.5 Participant Confidentiality

Participants will have code numbers and will not be identified by name. Participant confidentiality is strictly held in trust by the participating site PIs, their study staff, and the sponsor(s) and their agents. This confidentiality includes the study protocol, documentation, investigation data, participant's clinical information, testing of biological specimens, and all other information generated during participation in the study. No information concerning the study, or the data generated from the study will be released to any unauthorized third party without prior written approval of the DMID. Authorized third parties include OHRP or other government agencies as part of their duties, the FDA, single IRB, NIH, study staff, study monitors, Toltec, and designees. Participant confidentiality will be maintained when study results are published or discussed in conferences.

All information provided by the sponsor and all data and information generated by the participating site as part of the trial (other than a participant's medical records) will be kept confidential by the site PI and other study personnel to the extent permitted by law. This information and data will not be used by the site PI or other study personnel for any purpose other than conducting the trial. These restrictions do not apply to: (1) information that becomes publicly available through no fault of the site PI or other study personnel; (2) information that is necessary to disclose in confidence to an IRB solely for the evaluation of the trial; (3) information that is necessary to disclose in order to provide appropriate medical care to a study participant; or (4) study results that may be published as described in Section 15. If the

confidentiality provisions in the executed site contracts for the conduct of this trial are inconsistent with this statement, the site contract's confidentiality provisions shall apply rather than this statement.

The study monitor, or other authorized representatives of the sponsor, or governmental regulatory agencies such as the FDA may inspect all documents and records required to be maintained by the site PI. This includes, but is not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The participating sites will permit access to such records.

9.6 Certificate of Confidentiality

To protect privacy, we have received a Certificate of Confidentiality (CoC). With this CoC, the participating sites cannot be forced to release information that may identify the research participant, even by a court subpoena, in any federal, state, or local civil, criminal, administrative, legislative, or other proceedings. The participating sites will use the CoC to resist any demands for information that would identify the participant, except as explained below.

The CoC cannot be used to resist a demand for information from personnel of the United States Government that is used for auditing or evaluation of federally funded projects, like this study, or for information that must be released in order to meet the requirements of the FDA.

A CoC does not prevent the participant from voluntarily releasing information about themselves or their involvement in this research. If any person or agency obtains a written consent to receive research information, then the researchers may not use the Certificate to withhold that information.

The CoC does not prevent the participating sites from reporting without the participant's consent, information that would identify the participant as a participant in the study regarding matters that must be legally reported including: child and elder abuse, sexual abuse, or wanting to harm themselves or others.

The release of individual private information or specimens for other research will only occur if consent was obtained from the individual to whom the information, document, or biospecimen pertains, or for the purposes of other research that is in compliance with applicable Federal regulations governing the protection of human participants in research.

9.7 Costs, Participant Compensation, and Research Related Injuries

There is no cost to participants for taking part in this trial.

Procedures and treatment for clinical care may be billed to the participant, participant's insurance or third party. Participants may be compensated for their participation in this trial. Compensation will be in accordance with the local IRB's policies and procedures, and subject to IRB approval.

If it is determined by the site PI that an injury occurred to a participant as a direct result of the tests or treatments that are done for this trial, then referrals to appropriate health care facilities will be provided to the participant. Study staff will try to reduce, control, and treat any complications from this trial. Immediate medical treatment may be provided by the participating site. No financial compensation will be provided to the participant by the NIAID, NIH, or by the participating site for any injury suffered due to participation in this trial.

10 STATISTICAL CONSIDERATIONS

10.1 Study Hypotheses

The primary objective of the study is to evaluate the clinical efficacy of a twice-weekly application of the TOL-463 vaginal insert for the suppression of BV following successful induction with oral metronidazole or other CDC-recommended BV treatment in women with a history of recurrent BV. Clinical efficacy (suppression) is defined by clinical cure (see Section 3.3.1). There is one planned hypothesis test with respect to the primary outcome measure. The hypothesis test compares the proportion of participants with recurrent BV by V4 between treatment arms. The null hypothesis for the comparison is that there is no difference in cure rates between treatments, with a two-sided alternative. The test will be conducted using the Pearson Chi-Square test at the 5% two-sided significance level.

10.2 Sample Size Considerations

As detailed in Section 10.5, efficacy analyses will be performed in an evaluable subset of the enrolled population; this subset was used to derive the study's sample size. Assuming a 40% BV recurrence rate among women receiving placebo and a 50% reduction in recurrence among women treated with TOL-463 (i.e. 20% recurrence rate among TOL-463 recipients) and a 1:1 allocation ratio between treatment arms, a sample size of 200 evaluable participants will provide at least 85% power using the Chi-Square Test. Assuming 80% of enrolled participants will be evaluable for the primary analysis, the enrollment target is set to 250 to achieve 200 evaluable participants.

10.3 Treatment Assignment Procedures

10.3.1 Randomization Procedures

Participants will be randomized 1:1 to receive one of two treatments: TOL-463 vaginal inserts or Placebo vaginal inserts.

The list of randomized treatment assignments will be prepared by statisticians at the SDCC and included in the enrollment module of the Emmes' Internet Data Entry System (IDES). Advantage eClinical® will assign each participant to a blinded treatment number from the list after demographic and eligibility data have been entered. Each site will have a supply of blinded study drug kits pre-labeled with treatment numbers, each containing sufficient doses to treat a

participant for twelve weeks. The packaged study drug kits contain three, separate four-week kits to be dispensed to the participant at Enrollment V1 and Follow-ups V2 and V3 as applicable. Once a participant is assigned a treatment number, the corresponding kit will be distributed to the participant.

Instructions for using the enrollment module are included in the IDES User's Guide. Manual back-up randomization procedures are provided in the MOP for use in case the site temporarily loses Internet access, or the online enrollment system is unavailable.

The trial will use a site-stratified, permuted, blocked randomization scheme. Permuted blocked randomization is used to avoid the potential for serious imbalance in the number of participants assigned to each group, which can occur in simple randomization procedures.

10.3.2 Masking Procedures

Participants, the study staff who dispense study drug and perform study assessments after study drug administration, data entry personnel at the sites, and laboratory personnel will be blinded to treatment assignment. The DSMB may receive efficacy data in aggregate and presented by treatment group, as specified in the DSMB charter. The DSMB may request to be completely unblinded to individual study drug assignments, as needed, to adequately assess SAEs. Refer to the MOP for unblinding procedures.

Study drug will be prepared at Fisher BioServices in blinded numbered kits. All kits will look identical and contain an identical number of inserts. Emmes will provide an unblinded list identifying the treatment identity (TOL-463 or placebo) in each kit to the Fisher staff responsible for preparing study drug kits. Clinicians, investigators, and all blinded staff will not have access to this list until after the trial has ended.

10.4 Planned Interim Analyses

10.4.1 Interim Safety Review

The study will be monitored to determine if any of the safety halting rules described in Section 8.6 are met. The DSMB will meet and review safety data and enrollment data at specified times during the course of study and at a study closeout meeting, as defined in the Section 8.7.2 and the DSMB Charter.

10.4.2 Interim Efficacy Review

There are no planned interim efficacy analyses for this study.

10.5 Final Analysis Plan

A separate statistical analysis plan, (SAP) document will be generated which will contain the details of the analyses. This section outlines the major components of the analyses.

10.5.1 Analysis Populations

Safety Population: This analysis population includes all randomized participants who received at least one dose of study treatment.

Modified Intent-to-Treat (mITT) Population: This analysis population includes all randomized participants who are evaluable.

The mITT population will be used as the primary efficacy analysis population.

Per-Protocol (PP) Population: This analysis population includes all randomized participants who met all inclusion/exclusion criteria throughout the study, are evaluable (defined below), complied with the assigned IP, and returned to the study site for the TOC visit within the specified window (Visit 4 [Window: Day 85-91]) or were followed up to their diagnosis of recurrent BV.

Evaluable is defined as follows:

- Negative STI tests taken at baseline (*C. trachomatis*, *N. gonorrhoeae*, or *T. vaginalis*);
- For participants 21 years of age or older, documentation of or self-report of a normal or ASCUS HPV negative Pap test prior to screening, or a Pap smear performed at screening/enrollment with normal result

A participant is compliant with the study treatment if she misses ≤ 2 non-consecutive doses of IP per month, as assessed by participant memory aid and returned IP and packaging at each follow up visit (V2, V3, V4).

In the unlikely event of an error in randomization or IP administration (i.e., incorrect treatment), participants will be grouped by the treatment they actually received in safety analyses but will be grouped by their intended randomized assignment in efficacy analyses.

10.5.2 Baseline Characteristics

Baseline and demographic characteristics will be summarized overall and by treatment. For both continuous and categorical variables, appropriate summary statistics will be applied. For continuous variables, descriptive statistics will include the number of non-missing values, mean,

standard deviation, median, minimum, and maximum. For categorical variables, descriptive statistics will include counts and percentages per category.

10.5.3 Safety Analysis Plan

Safety evaluations will be based on the incidence, severity, and type of AEs, SAEs, clinically significant physical and pelvic examination findings as detailed in Section 9. In addition, the incidence of culture confirmed secondary VVC will be evaluated. Safety variables will be tabulated and presented for all participants in the safety population, grouped by type of infection and treatment.

AEs and SAEs will be coded by MedDRA® for preferred term and system organ class. The rate and exact 95% confidence intervals (CIs) of AEs in aggregate, as well as by MedDRA® categories, will be computed. The number of SAEs is likely to be small in this study and will be reported by a detailed listing showing the type, MedDRA® coding, relevant dates (IP administration dates and AE onset and resolution dates), severity, relatedness, and outcome for each event.

The occurrence of culture confirmed secondary VVC following initiation of study treatment and through the final study visit will be summarized in aggregate and by treatment group.

10.5.4 Efficacy Analysis Plan

10.5.4.1 Primary Efficacy Analysis

The primary efficacy analysis estimates the clinical efficacy rate of TOL-463. At each follow-up visit, the following Amsel criteria will be assessed:

a. Positive KOH whiff test
b. Homogeneous discharge characteristic of BV
c. Clue cells ≥ 20% of vaginal squamous epithelial cells
d. Vaginal pH > 4.5

For the primary analysis, a participant is deemed to experience a recurrence of BV at a follow-up visit if >2 of the above criteria are met. A participant has clinical suppression of BV if she meets two or fewer of the criteria at each observed follow-up visit through V4.

The proportion of participants with recurrent BV up to and including V4 will be summarized overall and by treatment group along with two-sided 95% Wilson CIs. The primary analysis will be performed in the mITT analysis population. Secondary analyses of the primary outcome measure will be performed in the PP analysis population. Recurrence status of participants who

are withdrawn or drop out early before an observed recurrence will be imputed; multiple imputation methods may be explored, and details will be provided in the SAP document.

In addition, the rate of recurrence, which considers the amount of observed follow-up time for each participant, will be estimated within each treatment group along with corresponding 95% CIs. Any additional sensitivity and/or corroborative analyses to be performed will be described in the separate SAP document.

10.5.4.2 Secondary Efficacy Analyses

The secondary efficacy analyses are briefly described below. Secondary efficacy analyses will be performed in the mITT and PP analysis populations, where appropriate. Details of these analyses and any sensitivity analyses will be specified in the separate SAP document.

For the analysis of Secondary Objective 1, the median time to BV recurrence will be estimated by treatment group using Kaplan-Meier estimates. For participants who experience BV recurrence by V4, the time of BV recurrence will be defined as the midpoint between the earliest time point of a positive BV diagnosis (post-baseline) and the previous observed time-point. For participants without BV recurrence at or before V4, the BV recurrence endpoint will be coded as censored, with time to recurrence defined as the elapsed time from the first dose of IP, to the earliest of: V4 time-point, the last observed time-point prior to loss to follow-up, or the time-point at which the participant became ineligible for the particular analysis population.

For the analysis of Secondary Objective 2, the proportion of participants experiencing vaginal symptoms as assessed by the participant via the daily memory aid will be estimated overall, by symptom category, and by treatment group.

For the analysis of Secondary Objective 3, responses to the satisfaction questionnaire will be summarized overall and by treatment group. Categorical responses will be summarized using contingency tables and/or odds ratios, while continuous responses will be summarized using measures of central tendency such as the mean or median, and measures of dispersion such as the standard deviation or interquartile range.

10.5.4.3 Exploratory and Other Efficacy Analyses

Descriptions of the analyses to assess the exploratory objective as well as other analyses that will support the primary and secondary efficacy analyses will be provided in the separate SAP document.

Examples of additional analyses that will support the primary and secondary efficacy analyses will include, but not limited to, summaries of Nugent scores by visit and treatment group, and explorations of recurrences by demographic and other baseline characteristics.

11 SOURCE DOCUMENTS AND ACCESS TO SOURCE DATA/DOCUMENTS

Each participating site will maintain appropriate medical and research records for this trial, in compliance with ICH E6, Section 4.9 and regulatory and institutional requirements for the protection of confidentiality of participants. Each participating site will permit authorized representatives of the DMID, its designees, and appropriate regulatory agencies to examine (and when required by applicable law, to copy) clinical study records for the purposes of quality assurance (QA) reviews, audits, monitoring, and evaluation of the study safety and progress. These representatives will be permitted access to all source data and source documents, which include, but are not limited to, hospital records, clinical and office charts, laboratory notes, memoranda, participants' evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, microfiches, photographic negatives, microfilm or magnetic media, x-rays, and participant files and records kept at the pharmacy, at the laboratories, and medico-technical departments involved in the clinical trial. Source data are all information, original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. DCF's will be provided by the SDCC.

12 QUALITY CONTROL AND QUALITY ASSURANCE

Following a written DMID-accepted site quality management plan, each participating site is responsible for conducting routine QA and quality control (QC) activities to internally monitor study progress and protocol compliance. The site PI will provide direct access to all study-related sites, source data/DCFs, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities. The site PI will ensure all study personnel are appropriately trained and applicable documentations are maintained on site.

The SDCC will implement QC procedures beginning with the data entry system and generate data QC checks that will be run on the database. Any missing data or data anomalies will be communicated to the participating site(s) for clarification and resolution.

13 DATA HANDLING AND RECORD KEEPING

13.1 Data Management Responsibilities

The site PI is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported.

DCFs will be derived from the eCRFs and provided by the SDCC to record and maintain data for each participant screened and enrolled in the study. All DCFs should be completed in a neat, legible manner to ensure accurate interpretation of data. Black or blue ink is required to ensure clarity of reproduced copies. When making a change or correction, cross out the original entry with a single line and initial and date the change. Do not erase, overwrite, or use correction fluid or tap on the original.

Data reported in the eCRF derived from the source DCFs should be consistent or the discrepancies should be explained.

The sponsor and/or its designee will provide guidance to the site PIs and other study personnel on making corrections to the DCFs and eCRF.

13.2 Data Coordinating Center/Biostatistician Responsibilities

All DCFs and laboratory reports must be reviewed by the clinical team and data entry personnel, who will ensure that they are accurate and complete. AEs must be recorded on the appropriate DCF, assessed for severity and relationship, and reviewed by the site PI or his/her designee.

Data collection is the responsibility of the study personnel at the participating clinical study site under the supervision of the site PI. During the study, the site PI must maintain complete and accurate documentation for the study.

The SDCC for this study will be responsible for data management, quality review, analysis, and reporting of the study data.

13.3 Data Capture Methods

Clinical data (including, but not limited to, AE/SAEs, concomitant medications, medical history, physical assessments, and clinical laboratory values) will be collected on DCFs by study personnel then entered into eCRFs via a 21 CFR Part 11-compliant internet data entry system, Advantage eClinical, provided by the SDCC. The data system includes password protection and

internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the DCFs completed by the study personnel.

13.4 Types of Data

Data for this trial will include clinical, safety, and outcome measures.

13.5 Study Records Retention

Study records and reports including, but not limited to, eCRFs, source documents, ICFs, laboratory test results, and IP inventory records will be retained for 2 years after a marketing application is approved for the IP for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for the IP, until 2 years after the investigation is discontinued and the FDA has been notified. These documents will be retained for a longer period, however, if required by local regulations. SICs and EICs for future use will be maintained as long as the sample/specimen exists.

No records will be destroyed without the written consent of the sponsor. It is the responsibility of the sponsor to inform the site PI when these documents no longer need to be retained. The participating sites must contact DMID for authorization prior to the destruction of any study records.

14 CLINICAL MONITORING

Site monitoring is conducted to ensure that the human subject protections study and laboratory procedures, study intervention administration, and data collection processes are of high quality and meet sponsor, ICH/GCP guidelines and applicable regulations, and that this trial is conducted in accordance with the protocol, protocol-specific MOP and applicable sponsor SOPs. A separate monitoring plan document should be developed to describe who will conduct the monitoring, at what frequency monitoring will be done, and what level of detail monitoring will be conducted. Most DMID sponsored studies are monitored by TRI/ICON DMID CROMS, and the clinical monitoring plan is written jointly by DMID and TRI/ICON. Preference is given to a separate monitoring plan to be agreed upon with the Office of Clinical Research Affairs (OCRA), which will describe protocol-specific items to be monitored. The monitoring plan must include the number of participant charts to be reviewed, which/what proportion of data fields and what will be monitored, who will be responsible for conducting the monitoring visits, and who will be responsible for ensuring that monitoring findings are addressed.

Site visits will be made at standard intervals as defined by the monitoring plan and may be made more frequently as directed by DMID. Monitoring visits will include, but are not limited to, review of regulatory files, accountability records, eCRFs, informed consent forms (SIC and EIC), medical and laboratory reports, and protocol and GCP compliance. Site monitors will have access to each participating site, study personnel, and all study documentation according to the DMID-approved monitoring plan. Study monitors will meet with site PIs to discuss any problems and actions to be taken and will document site visit findings and discussions.

15 PUBLICATION POLICY

Following completion of the study, the lead PI is expected to publish the results of this research in a scientific journal. All investigators funded by the NIH must submit or have submitted for them to the National Library of Medicine's PubMed Central (http://www.ncbi.nlm.nih.gov/pmc/) an electronic version of their final, peer-reviewed manuscripts upon acceptance for publication, to be made publicly available no later than 12 months after the official date of publication. The NIH Public Access Policy ensures the public has access to the published results of NIH funded research. It requires investigators to submit final peer-reviewed journal manuscripts that arise from NIH funds to the digital archive PubMed Central upon acceptance for publication. Further, the policy stipulates that these papers must be accessible to the public on PubMed Central no later than 12 months after publication.

Refer to:

- NIH Public Access Policy, http://publicaccess.nih.gov/
- NIH Office of Extramural Research (OER) Grants and Funding, http://grants.nih.gov/grants/oer.htm

As of January 2018, all clinical trials supported by the NIH must be registered on ClinicalTrials.gov, no later than 21 days after the enrollment of the first participant. Results of all clinical trials supported by the NIH, generally, need to be submitted no later than 12 months following the primary completion date. A delay of up to 2 years is available for trials that meet certain criteria and have applied for certification of delayed posting.

As part of the result posting a copy of this protocol (and its amendments) and a copy of the SAP will be posted on ClincialTrials.gov.

For this trial the responsible party is NIAID which will register the trial and post results.

The responsible party does not plan to request certification of delayed posting.

Refer to:

- Public Law 110-85, Section 801, Clinical Trial Databases
- 42CFR11
- NIH NOT-OD-16-149

16 LITERATURE REFERENCES

1. Muzny, C. and J. Schwebke, *Biofilms: an underappreciated mechanism of treatment failure and recurrence in vaginal infections.* Clin Infect Dis, 2015. **61**(4): p. 601-606.
2. *Diflucan [package insert]. New York, NY: Pfizer.* 2018.
3. *Nuvessa [package insert]. Irvine, CA: Allergan.* 2016.
4. Marrazzo, J., et al., *Bacterial Vaginosis: Identifying Research Gaps Proceedings of a Workshop Sponsored by DHHS/NIH/NIAID November 19-20, 2008.* Sex Transm Dis, 2010. **37**(12): p. 732-744.
5. Aguin, T. and J. Sobel, *Vulvovaginal Candidiasis in Pregnancy.* Curr Infect Dis Rep, 2015. **17**(6): p. 462.
6. van de Wijgert, J., et al., *Bacterial vaginosis and vaginal yeast, but not vaginal cleansing, increase HIV-1 acquisition in African women.* J Acquir Immune Defic Syndr, 2008. **48**(2): p. 203-210.
7. Bradshaw, C. and R. Brotman, *Making inroads into improving treatment of bacterial vaginosis – striving for long-term cure.* BMC Infect Dis, 2015. **15**(1): p. 292.
8. Allsworth, J. and J. Peipert, *Prevalence of bacterial vaginosis: 2001-2004 National Health and Nutrition Examination Survey data.* Obstet Gynecol, 2007. **109**(1): p. 114-120.
9. Sobel, J., et al., *Suppressive antibacterial therapy with 0.75% metronidazole vaginal gel to prevent recurrent bacterial vaginosis.* Am J Obstet Gynecol, 2006. **194**(5): p. 1283-1289.
10. *CDC Sexually Transmitted Diseases Treatment Guidelines.* MMWR, 2015. **64**(3): p. 1-138.
11. Nyirjesy, P., *Management of persistent vaginitis.* Obstet Gynecol, 2014. **124**(5): p. 1135-1146.
12. Reichman, O., R. Akins, and J. Sobel, *Boric acid addition to suppressive antimicrobial therapy for recurrent bacterial vaginosis.* Sex Transm Dis, 2009. **36**(11): p. 732-734.

17 APPENDICES

Schedule of Events

Female Genital Grading Table for Physical Examination Findings

Appendix A. Schedule of Events

Table 17-1: Schedule of Events

Procedures	**Visit 0/V0** Screening (Day -15 to -8)	**Visit 1/V1** Enrollment (Day 1)	**Visit 2/V2** Follow-up (Days 29-35)	**Visit 3/V3** Follow-up (Days 57-63)	**Visit 4/V4/TOC** (Days 85-91)	**Unscheduled Visit**	**Early Termination Visit**
Signed consent form[1]	X	X					
Review of Inclusion/Exclusion criteria	X	X					
Collection of Demographics	X						
Review of Medical History[2, 3]	X	X	X	X	X	(X)	X
Review of Sexual History[4]	X	X	X	X	X	(X)	X
Urine Pregnancy Test[12]	X	X	X	X	X	(X)	X
Review of Concomitant Medications[5]	X	X	X	X	X	(X)	X
Pelvic Examination[3,6, 13]	X	X	X	X	X	(X)	X
Clinical assessment of Vulvovaginal signs	X	X	X	X	X	(X)	X

Procedures	**Visit 0/V0** Screening (Day -15 to -8)	**Visit 1/V1** Enrollment (Day 1)	**Visit 2/V2** Follow-up (Days 29-35)	**Visit 3/V3** Follow-up (Days 57-63)	**Visit 4/V4/TOC** (Days 85-91)	**Unscheduled Visit**	**Early Termination Visit**
STI Screen (*C. trachomatis, N. gonorrhoeae*, and *T. vaginalis)*[7]	X						
Clinical Determination of presence or absence of BV (Amsel Criteria)	X	X	X	X	X	(X)	X
Gram stain evaluation/Nugent score for BV	X	X	X	X	X	(X)	X
BD Max Test	X	X	X	X	X		X
Future Analysis Specimen Collection[11]	(X)	(X)	(X)	(X)	(X)		(X)
Dispense Oral Metronidazole (or other CDC-recommended BV treatment)	X						
Randomization		X					
Targeted Physical Examination[8]		X	(X)	(X)	(X)	(X)	(X)
IP Dispensation, Participant Instructions		X	X	X		(X)	
Participant Memory Aid[9]		X	X	X	X	(X)	X
Reminder contact (for Visits 1, 2, and 3)		X	X	X			
Assessment of IP compliance (via review of Memory Aid and IP return)			X	X	X	(X)	X

Procedures	**Visit 0/V0** Screening (Day -15 to -8)	**Visit 1/V1** Enrollment (Day 1)	**Visit 2/V2** Follow-up (Days 29-35)	**Visit 3/V3** Follow-up (Days 57-63)	**Visit 4/V4/TOC** (Days 85-91)	**Unscheduled Visit**	**Early Termination Visit**
Participant Assessment of Vaginal Symptoms	X	X	X	X	X	(X)	X
Candida Culture[10]		(X)	(X)	(X)	(X)	(X)	(X)
Assessment of AEs/SAEs			X	X	X	(X)	X
Participant satisfaction questionnaire			(X)	(X)	X		X

(X) – As indicated/appropriate.

1. At V0 the participant is signing the SIC Form. At V1 the participant will then sign the EIC Form.
2. At V0 collect complete medical symptom history; at subsequent visits, review and update medical symptom history as appropriate.
3. A Pap smear should be done at V0 for any participant who meets the following criteria and cannot provide documentation (which can include self-report) of a normal or ASCUS HPV negative Pap test within the prior 3 years: is 21 years of age or older and (a) has not had a hysterectomy or (b) has had a hysterectomy and has a history of CIN grade 2+ (CIN2+) in the past 20 years. If a Pap smear is conducted at V0, the results are not required prior to enrollment.
4. At V0, collect sexual history for the past 30 days; at subsequent visits, collect interim sexual history since the last visit.
5. At V0, collect concomitant medications taken in the last 30 days; at subsequent visits, collect concomitant medications taken since the last visit.
6. At V1, one pelvic exam is performed and if the participant is enrolled and randomized additional vaginal swabs are collected.
7. Participants who test positive for STIs may be reconsidered for eligibility once the treatment course is complete.

8. At the enrollment visit, V1, this will include weight and height measurements. At all follow-up visits, a targeted physical exam is only performed when clinically indicated.
9. After randomization, the participant will be provided with the Participant Memory Aid for the first 4 weeks of the study. At each of the follow-up visits, V2-V4, the Memory Aid will be reviewed/collected, and a new Memory Aid for the next 4 weeks will be provided.
10. At the enrollment visit, V1, and all follow-up visits V2-V4, participants with clinical evidence of VVC and symptoms will have a confirmatory *Candida* culture performed and treated as appropriate with oral fluconazole.
11. A Future Analysis Specimen Collection is only performed for participants who consented to future use of their specimens.
12. A urine pregnancy test is only required for participants of childbearing potential.
13. The pelvic exam includes KOH and saline microscopy for identification of yeast forms, trichomonas and clue cells.

Appendix B. Toxicity Grading Table

Note: the following table is adapted from the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events, Addendum 1: Female Genital Grading Table for Use in Microbicide Studies (V1.0, November 2007)

Table 17-2: Toxicity Grading Table

INDIVIDUAL SIGNS/SYMPTOMS				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD**	**GRADE 2 MODERATE**	**GRADE 3 SEVERE**
GENERAL				
Odor	No complaint	Mild-moderate unpleasant	Severe unpleasant odor	NA
PAIN AND TENDERNESS (Specify Area: Vulvar/Perineum, Vagina, Cervix (including cervical motion tenderness), Uterus, Adnexae, Pelvic/Lower Abdominal, or Ovulatory) ***Note – if both pain and tenderness are present, only report the one with the most severe grade**				
Pain* 1	None	Pain causing no or minimal interference with usual social & functional activities	Pain causing greater than minimal interference with usual social & functional activities or the need for non-narcotic medication	Pain causing inability to perform usual social & functional activities or the need for narcotic medication OR hospitalization (other than emergency room visit) indicated
Tenderness* 1	None	Mild tenderness	Moderate tenderness	Severe tenderness

Dyspareunia (pain with sexual activity)	None	Pain causing no or minimal interference with sexual function	Pain causing greater than minimal interference with sexual function	NA
Dysmenorrhea/cramping with menses	None	Pain causing no or minimal interference with usual social & functional activities	Pain causing greater than minimal interference with usual social & functional activities or the need for non-narcotic medication	Pain causing inability to perform usual social or functional activities or the need for narcotic medication

[1] If pain or tenderness is included in the grading of another category (e.g., PID), it should not be graded again in the pain or tenderness category

INDIVIDUAL SIGNS/SYMPTOMS				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD**	**GRADE 2 MODERATE**	**GRADE 3 SEVERE**
GENITOURINARY SIGNS/SYMPTOMS – VULVA				
Vulvar/vaginal itching	None	Itching causing no, mild, or moderate interference with usual social & functional activities	Itching causing inability to perform usual social & functional activities; may require intervention such as antihistamine or bathing to provide relief	NA
Vulvar edema	None	Mild, non-pitting edema	Moderate, 1-2+ pitting edema	3+ pitting edema, severe enough to require urinary drainage, or weeping edema ± skin breakdown
Vulvar erythema	None	Erythema covering < 50% of vulvar surface	Erythema covering ≥ 50% of vulvar surface	NA
Vulvar lesions (findings seen only by colposcopy should not be included here)	Normal variants including skin tags, moles, scars, etc.	Blisters, ulcerations, or pustules - no treatment indicated	Blisters, ulcerations or pustules, with treatment indicated	Severe epithelial disruption with hospitalization indicated (other than emergency room visit)

Vulvar rash	None	Rash covering < 50% of vulvar surface	Rash covering ≥ 50% of vulvar surface	Severe epithelial disruption with hospitalization indicated (other than emergency room visit)
Bartholin's or Skene's gland	No findings	Cyst with no inflammation	Cyst or abscess with outpatient intervention indicated	Cyst or abscess with hospitalization indicated, (other than emergency room visit) including necrotizing fasciitis from Bartholin's abscess
GENITOURINARY SIGNS/SYMPTOMS – VAGINA **** Note – if vaginal discharge is present both by history and on examination, only report the one with the most severe grade**				
Vaginal edema	None	Mild-moderate engorgement	Loss of ruggae and friability	NA
Vaginal erythema	None	Erythema covering < 50% of vaginal surface	Erythema covering ≥ 50% of vaginal surface	NA
Vaginal dryness	No complaint	Dryness causing no or minimal interference with usual sexual, social, & functional activities	Dryness causing greater than minimal interference with usual sexual, social, & functional activities	NA

INDIVIDUAL SIGNS/SYMPTOMS				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD**	**GRADE 2 MODERATE**	**GRADE 3 SEVERE**
Vaginal discharge by participant report **	Participant's usual amount of discharge, regardless of color or quantity	Mild-moderate increase in amount above participant baseline - no sanitary protection	Profuse increase in discharge requiring pad use or other hygienic intervention	NA
Vaginal discharge as observed by clinician ** (red or brown discharge should be reported under bleeding, not discharge)	Slight amount of discharge, any color	Mild-moderate increase in amount	Significant increase in amount with pooling in vagina on examination	NA
Vaginal abrasions or lacerations (including probable applicator injuries)	None	Superficial disruptions and disruptions extending through the mucosa with minimal impact on life	Large disruptions extending through the mucosa or large superficial disruptions, hospitalization not indicated	Large disruptions extending through the mucosa or large superficial disruptions including lacerations, hospitalization indicated (other than emergency room visit)
Vaginal lesions (findings seen only by colposcopy should not be included here)	Normal variants including skin tags, moles, scars, etc.	Blisters, ulcerations, or pustules, no treatment indicated	Blisters, ulcerations, or pustules with treatment indicated	Severe epithelial disruption requiring hospitalization (other than emergency room visit)

Vaginal and Cervical masses (polyps, myomas, or possible malignancy)	None or normal variants such as Nabothian cyst or Gartner duct cyst	Polyp or myoma or undiagnosed mass without symptoms	Polyp, myoma, or undiagnosed mass causing mild symptoms, e.g., bleeding/pain not requiring more than mild analgesia	Polyp, myoma, or undiagnosed mass causing severe symptoms, e.g., bleeding/pain affecting bladder and bowel function, or visible cervical cancer
GENITOURINARY SIGNS/SYMPTOMS – CERVIX				
Cervical edema and friability	None	Edema without friability	Friable cervix	NA
Cervical erythema	None	Erythema covering < 50% of cervix	Erythema covering ≥ 50% of cervix	NA
Cervical discharge	White or clear discharge	Small amount of purulent discharge at os	Purulent discharge extending onto cervix or vagina	NA
Visible cervical lesions (findings seen only by colposcopy should not be included here)	Normal variants including skin tags, moles, scars, etc.	Blisters, ulcerations, or pustules, no treatment indicated	Blisters, ulcerations, or pustules with treatment indicated	NA

INDIVIDUAL SIGNS/SYMPTOMS				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD**	**GRADE 2 MODERATE**	**GRADE 3 SEVERE**
GENITOURINARY SIGNS/SYMPTOMS – UTERUS				
Uterine masses/enlargement based on bimanual examination	Normal to 8 week size, no palpable myomas	Enlarged uterus and mild symptoms, e.g., bleeding/pain requiring mild analgesics	Enlarged uterus/myoma with moderate pain or symptoms, e.g., bleeding	Mass causing severe bleeding/pain or with impact on bowel/bladder function or requires transfusion or surgery
Polyp, submucosal fibroid, or thickened endometrium detected by transvaginal ultrasound (new or increasing in size from prior exam)	None or unchanged/reduced in size from prior exam	New myomas < 6 cm diameter (single or multiple) or diameter increased < 6 cm since prior exam	New myomas ≥ 6 cm diameter (single or multiple) or diameter increased ≥ 6 cm since prior exam	Hospitalization (other than emergency room visit) and/or surgery indicated
GENITOURINARY SIGNS/SYMPTOMS – ADNEXA				
Not pregnancy- or infection-related adnexal masses based on bimanual exam (use if no ultrasound done; if ultrasound done, use ultrasound categories below)	None, ≤ 4 cm, normal size ovary	> 4 cm with minimal or no symptoms	> 4 cm with severe symptoms, e.g., pain, but hospitalization not indicated (see footnote #1)	> 4 cm with severe symptoms, e.g., pain and hospitalization indicated (other than emergency room visit) (see footnote #1)

Hydrosalpinx based on ultrasound	None	Asymptomatic, suspected hydrosalpinx	Hydrosalpinx with pain, but without evidence of infection or ectopic pregnancy	Signs/symptoms of infection with hospitalization (other than emergency room visit) and/or surgery indicated
Adnexal mass based on ultrasound	None	Simple cyst, asymptomatic	Simple cyst, symptomatic	Malignant mass suspicious for malignancy

INDIVIDUAL SIGNS/SYMPTOMS				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD**	**GRADE 2 MODERATE**	**GRADE 3 SEVERE**
GENITOURINARY SIGNS/SYMPTOMS – ABDOMEN				
Abdominal mass not palpable on pelvic exam of unknown diagnosis	None or known (pre-existing) mass unchanged in size	New mass or increased size of known mass requiring mild analgesia with minimal impact	New mass or increased size of known mass with moderate symptoms	Mass causing severe bleeding/ pain with impact on bladder/bowel function or with hospitalization (other than emergency room visit) indicated or malignancy
GENITOURINARY SIGNS/SYMPTOMS – URINARY TRACT				
Urinary frequency	None	Up to 2 times participant's normal frequency	> 2 times participant's normal frequency	NA
Dysuria	None	Superficial only	Deep ± superficial	Inability to void due to pain
Hematuria	None	Microscopic, no intervention indicated (beyond evaluation for infection)	Gross blood in urine or medical intervention/ evaluation indicated (beyond evaluation for infection)	Persistent bleeding with transfusion, hospitalization (other than emergency room visit) or intervention indicated to obtain hemostasis (endoscopy, interventional radiology, or operative)

COMPOSITE SIGNS/SYMPTOMS (Use instead of individual categories if 2 or more signs/symptoms are present)				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD (Use if all signs/ symptoms would individually be Grade 0 or 1)**	**GRADE 2 MODERATE (Use if one or more signs/symptoms would individually be Grade 2 and all others Grade 0 or 1)**	**GRADE 3 SEVERE (Use if one or more signs/symptoms would individually be Grade 3)**
NO ORGANISM IDENTIFIED BUT INADEQUATE TESTING PERFORMED				
Vulvovaginitis (combinations of pain, itching, erythema, edema, rash, tenderness, or discharge)	None	Mild signs/symptoms	Moderate signs/symptoms	Severe signs/ symptoms
Cervicitis (combinations of dyspareunia, erythema, edema, tenderness, and discharge)	None	Mild signs/ symptoms	Moderate signs/ symptoms	Severe signs/ symptoms

PID (if Gonorrhea or Chlamydia identified use that category)	None	NA	Cervicitis with mild uterine tenderness, ± mild cervical motion tenderness, no signs of peritoneal	More diffuse tenderness, any signs of peritoneal irritation, or indications for hospitalization (other than emergency room visit) including tubo-ovarian abscess
NO ORGANISM IDENTIFIED AFTER APPROPRIATE TESTING PERFORMED				
Vulvovaginitis (combinations of pain, itching, erythema, edema, rash, tenderness, or discharge)	None	Mild signs/ symptoms	Moderate signs/ symptoms	Severe signs/ symptoms
Cervicitis (combinations of dyspareunia, erythema, edema, tenderness, and discharge)	None	Mild signs/ symptoms	Moderate signs/ symptoms	Severe signs/ symptoms
PID (if Gonorrhea or Chlamydia identified use that category)	None	NA	Cervicitis with mild uterine tenderness, ± mild cervical motion tenderness, no signs of peritoneal irritation	More diffuse tenderness, any signs of peritoneal irritation, or indications for hospitalization (other than emergency room visit) including tubo-ovarian abscess

INFECTIONS AND DYSPLASIA				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD**	**GRADE 2 MODERATE**	**GRADE 3 SEVERE**
GENITOURINARY INFECTIONS				
Genital herpes	No lesions	Characteristic ulcerative or vesicular lesions confirmed by culture, PCR, Tzanck prep or other diagnostic test of lesion or previous type-specific serology, covering < 25% of vulva, vagina, or cervix	Same criteria as mild but covering 25-50% of vulvar, vaginal, or cervical surface	Same criteria as mild but covering > 50% of vulvar, vaginal, or cervical surface and/or symptoms of significant systemic involvement, e.g., encephalitis, hepatitis
Candida	Absence of symptoms regardless of candida test results	Positive culture, wet mount, or other laboratory test for yeast, with mild symptoms	Positive culture, wet mount, or other laboratory test for yeast, with moderate to severe symptoms	NA
Trichomonas	Negative	NA	Positive wet mount, culture, PCR or other licensed test, excluding pap smear, showing *T. vaginalis,* regardless of symptoms	NA

Chlamydia	Negative	NA	Positive culture or other diagnostic test for Chlamydia, asymptomatic or with mild uterine or cervical motion tenderness (no signs of peritoneal irritation)	Positive test for Chlamydia with abdominal or uterine or adnexal tenderness on examination, with or without adnexal mass, diffuse tenderness, any signs of peritoneal irritation, or indications for hospitalization (other than emergency room visit) including tubo-ovarian abscess

INFECTIONS AND DYSPLASIA				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD**	**GRADE 2 MODERATE**	**GRADE 3 SEVERE**
Gonorrhea	Negative	NA	Positive culture or other diagnostic test for Gonorrhea, asymptomatic or with mild uterine or cervical motion tenderness (no signs of peritoneal irritation)	Positive test for Gonorrhea with abdominal or uterine or adnexal tenderness on examination, with or without adnexal mass, diffuse tenderness, any signs of peritoneal irritation, or indications for hospitalization (other than emergency room visit) including tubo-ovarian abscess or disseminated gonococcal infection
Urinary tract infection (by urinalysis and urine culture)	Negative	5-10 WBC/hpf on urinalysis with a negative culture per protocol definition (with or without symptoms)	> 10 WBC/hpf on urinalysis OR a positive culture per protocol definition (with or without symptoms)	Pyelonephritis or sepsis (septicemia) due to urinary tract infection

Syphilis	Negative treponemal or non-treponemal test or both positive with known treatment and stable titers (< 4 fold increase)	NA	Syphilis diagnosed by a positive treponemal test along with a positive non-treponemal test and no previous treatment or a four- fold rise in titer on the non-treponemal test after previous treatment regardless of symptoms or non-oral lesions positive by darkfield exam for treponemes	Syphilis in the presence of neurologic symptoms or a positive CSF VDRL or FTA-ABS
GENITAL DYSPLASIA				
Condyloma (specify site: cervical, vaginal, vulvar, perianal)	None	Condylomata causing no or mild interference with daily function	Condylomata causing moderate interference with daily function	Condylomata causing severe interference with daily function, secondary infection, or hospitalization (other than emergency room visit) indicated

INFECTIONS AND DYSPLASIA				
PARAMETER	**GRADE 0 NORMAL**	**GRADE 1 MILD**	**GRADE 2 MODERATE**	**GRADE 3 SEVERE**
Intraepithelial Neoplasia by biopsy (VIN, CIN, VAIN)	None	Intraepithelial Neoplasia 1 (IN1)	Intraepithelial Neoplasia 2 (IN2)	Carcinoma in situ (CIS) or Invasive carcinoma
Pap (use this category only if treatment performed without diagnostic testing, otherwise use biopsy category above)	nl PAP	ASCUS or LSIL	HSIL	Carcinoma in situ or carcinoma