Title of the study - Bacille Calmette Guerin (BCG) Revaccination of Healthy Adolescents for the Prevention of Mycobacterium Tuberculosis Sustained Infection

NCT number - NCT04152161

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Title Page

Protocol Title: A Randomized, Placebo Controlled, Observer-Blind, Phase IIb Study to Evaluate the Efficacy, Safety, and Immunogenicity of BCG revaccination in Healthy Adolescents for the Prevention of Sustained Infection with *Mycobacterium tuberculosis*

Protocol Number: Gates MRI-TBV01-201

Short Title: BCG Revaccination of Healthy Adolescents for the prevention of *Mycobacterium tuberculosis* sustained infection

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NOTE

This study was paused for screening and randomization by the sponsor, effective 19 March 2020, due to the Coronavirus Disease 2019 (COVID-19) pandemic.
Screening and randomization resumed in July, 2020. Refer to Section 7.4, COVID-19 Contingency Plans for details regarding study pause due to COVID-19.

DOCUMENT HISTORY

Document	Date
Original Version 1 pre-IRB review	14 Feb 2019
Version 2 pre-IRB review	21 Feb 2019
Version 3 pre- IRB review	27 Feb2019
Version 4	25 June 2019 (Version used at Study Start)
Version 5	11 May 2020 (Modified to Version 6 before presented to the IRBs or SAHPRA)
Version 6	23 June 2020
Version 7	21 July 2023

SUMMARY OF CHANGES

A high-level summary of the major changes incorporated into this protocol Version 7 are shown below.

Section Number	Summary of Change	Rationale for Change
Table 1 and Section 3 Objectives and Endpoints	Updated primary and the first secondary efficacy endpoint text to delete IFN-γ concentration cut-off value Replaced "an IFN-γ concentration cut-off value of 0.35 IU/mL" with "positive QFT test results" Added "at least" 118 events to the primary endpoint.	Changed the wording to indicate that the assay is used according to the manufacturer's instructions. The conditions that need to be met for a valid positive result are best described in the package insert. Added to allow for flexibility in the timing of the analysis.
	Moved secondary objective ("To evaluate the efficacy and durability of efficacy, and associated endpoint") to first exploratory objective. Revised the text associated with endpoint for primary QFT conversion **New text:** …at the time of primary endpoint analysis, and after a follow-up of 36- and 48-months post vaccination, using an alternative definition of conversion as a change in IFN-γ concentration from <0.35 IU/mL to ≥4 IU/mL (initial conversion only) **Previous version:** ..based on a QFT IFN-γ concentration cut-off value of 4 IU/mL at the time of primary endpoint analysis, and after a minimum follow-up of 36- and 48-months post vaccination, based on IFN-γ concentration cut-off value of 4 IU/mL (initial conversion only)	The endpoint which includes a threshold of 4 IU/mL for QFT positivity, does not conform to the language in the package insert and we therefore considered declaring this as an exploratory endpoint more appropriate.
	First secondary objective - edited endpoint wording for the efficacy and durability of efficacy against sustained Mtb infection	For clarification

Section Number	Summary of Change	Rationale for Change
	New text: Sustained QFT conversion based on positive QFT test results (initial conversion and QFT-positive at 3- and 6-months PC) with a follow-up of 36- and 48-months post vaccination. **Previous version**: Sustained QFT conversion based on an IFN-γ concentration cut-off value of 0.35 IU/mL (initial conversion and QFT-positive at 3- and 6-months PC) with a minimum follow-up of 36- and 48-months post vaccination	
Section 1.3 SoA and Table 4	The Month 54 visit was deleted from SoA and Table 4, and footnotes updated accordingly. Footnote for HIV testing corrected to add the word "suspected" to HIV testing at screening, and M12, 24, 36, 48, and if TB is ***suspected***. A column for Suspected TB visits was added.	Correction for consistency with text in Section 8.3.6. Suspected TB column added for clarification
Section 4.1 Scientific Rationale for Study Design	Updated wording to reflect changes in endpoint text	For clarification
Section 4.3 End of Study Definition	Changed the end of study definition to remove follow-up at Months 51 and 54 for participants who have their initial QFT seroconversion at Month 48. A participant is considered to have completed the study if he/she completes the final visit at Month 48. The end of the study is defined as the date of the last visit of the last participant in the study or last scheduled procedure shown in the SoA for the last participant in the trial. The primary endpoint is reached when at least 118 participants have converted from a negative to positive QFT result, sustaining positivity for at least 6 months post initial conversion.	The study will end after the Month 48 visit is completed. Clarification for when the primary endpoint is met.

Section Number	Summary of Change	Rationale for Change
Section 7.2. Discontinuation/ Withdrawal from the Study	Removed pregnancy from bulleted list of reasons for investigator-instigated withdrawals and removed pregnancy text Edited text regarding participant withdrawal and required procedures	Pregnancy alone is not a reason for study withdrawal. Post study start pregnancy will lead to cancellation of further blood draws. Clarified text regarding procedures to be performed if participant is not withdrawn.
Section 8.1.1 Evaluation of initial Mtb infection and sustained infection	**New text**: *Mtb* infection is defined for this protocol as QFT conversion from a negative to positive test, as per the manufacturer's QFT-GIT package insert. Sustained *Mtb* infection is defined for this protocol as sustained QFT conversion from a negative to positive test, with initial conversion at any time after a first negative QFT result, post randomization (Day 71 or subsequent visit if Day 71 result is not available), and remaining QFT-positive at 3- and 6-months PC. **Previous text**: *Mtb* infection is defined for this protocol as QFT conversion from a negative to positive test, using the manufacturer's recommended threshold of 0.35 IU/mL, at any time-point. Sustained *Mtb* infection is defined for this protocol as sustained QFT conversion from a negative to positive test, using the manufacturer's recommended IFN-γ concentration cut-off value of 0.35 IU/mL, with initial conversion at any time after Day 71 testing and remaining QFT-positive at 3- and 6-months PC.	Clarification of Mtb infection and sustained infection definitions.
Section 8.2.2 Pregnancy Assessment	Added "and Follow-up" to the subsection title Added text here to indicate that pregnancy after study start does not lead to withdrawal but that blood draws are not performed during pregnancy. Once pregnancy	Stated above.

Section Number	Summary of Change	Rationale for Change
	ends, QFT and HIV blood draws can resume, per PI discretion, but all other blood draws are cancelled.	
Section 8.3.6 Latent TB infection	Updated wording to state that TPT either by referral or offered at site.	To allow for flexibility for sites to refer participants for TPT
Table 9 Populations for Analyses	Revised mITT population description text to include participants who may have missed the Day 71 visit. Revised PP text **New text:** All participants in the mITT population who did not substantially deviate from the protocol procedures. **Previous version:** All participants randomly assigned to study intervention, who received the study intervention, are QFT negative at the Day 71 visit, and did not substantially deviate from the protocol procedures. mITT population definition edited to specify that participants will be analyzed according to the intervention to which they were randomized instead of to the intervention they actually received. PP definition was modified to **"**All participants in the mITT population who did not substantially deviate from the protocol procedures and who received the treatment to which they were randomized."	For clarification
Table 10 Summary of Primary and Secondary Endpoints and Analyses and Section 9.4.1.2 Secondary and Exploratory	Revised text	For consistency to reflect changes made to the Objectives and Endpoints Table

Section Number	Summary of Change	Rationale for Change
Efficacy Endpoint Analyses		
Section 9.4.1.1 Primary Endpoint Analyses	Edited sustained QFT conversion definition to remove "based on an assay threshold of 0.35 IU/mL" Timing of primary efficacy analysis edited to include the option to combine the primary analysis and the Month 36 efficacy analysis if at least 118 events are reached within 6 months before the Month 36 visit is planned to occur.	For consistency with changes made to primary endpoint Timing of primary analysis updated for flexibility
Interim Analysis Section 9.4.6	Removed this IA section and content and moved text regarding IDMC to a new separate subheading: Independent Data Monitoring Committee (IDMC)	The interim analysis was included in the original protocol to allow Gates MRI/BMGF to make the decision as to whether we would run a similar study in other countries. A second revaccination study will not be conducted and therefore, an interim analysis was not required and was not performed.
Section 10.2.6 SAEs, AESIs, Serious ADRs, and SUSAR Reporting	Removed reference to "Immediately Reportable Event Form"	This form will not be used.
Section 10.3	Deleted elective termination of pregnancy from pregnancy complications and specified that AE and SAE reporting periods apply as described in reporting sections.	Clarified text regarding pregnancy complications and reporting of pregnancies.

Refer to Summary of Section 12 for version history and list of changes for previous versions.

Amendment Version 7 Signature Page

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List of Abbreviations

ADR	(serious) adverse drug reaction
AE	adverse event
AESI	adverse event of special interest
ALP	alkaline phosphatase
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BARC SA	Bio Analytical Research Corporation, South Africa
βHCG	beta human chorionic gonadotropin
BCG	Bacille Calmette Guerin
BMI	body mass index
CBC	complete blood count
CD	cluster of differentiation
CFR	Code of Federal Regulations
CFU	Colony-forming units
CoP	correlate of protection
CoR	correlate of risk
COVID-19	Coronavirus Disease 2019
CRF	case report form
CSR	clinical study report
DNA	deoxyribonucleic acid
ELISA	enzyme-linked immunosorbent assay
ELISPOT	enzyme-linked immunospot
GCP	Good Clinical Practices
GMRI	Gates Foundation Medical Research Institute
HIV	human immunodeficiency virus
ICH	International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use
ICS	intracellular cytokine staining
ID	intradermal
IDMC	Independent Data Monitoring Committee
IEC	Independent Ethics Committee
IFN-γ	interferon gamma
IL	interleukin
IRB	Institutional Review Board
IUD	intrauterine device
IVRS	interactive voice response system
IWRS	interactive web response system
LAR	legally acceptable representative
LLN	lower limit of normal
LTBI	latent TB infection
mL	milliliter

mITT	modified intention to treat
Mtb	*Mycobacterium tuberculosis*
NA	not applicable
NK cells	natural killer cells
NTM	non-tuberculous mycobacteria
NTP	(South African) National TB Program
PBMC	peripheral blood mononuclear cells
PC	post conversion
PCR test	polymerase chain reaction test
POD	Prevention of disease
POSI	Prevention of sustained *Mtb* infection
PP	Per protocol
QFT-GIT assay	QuantiFERON-Gold in Tube assay
QFT assay	QuantiFERON®-TB Gold Plus assay
RNA	ribonucleic acid
SAE	serious adverse event
SAHPRA	South African Health Products Regulatory Authority
SAP	Statistical Analysis Plan
SARS-CoV-2	Severe Acute Respiratory Syndrome-Coronavirus-2
SATVI	South African Tuberculosis Vaccine Initiative
SoA	schedule of activities
SUSAR	suspected unexpected serious adverse reaction
TB	Tuberculosis
TNF	tumor necrosis factor
TST	tuberculin skin test
ULN	upper limit of normal
VE	vaccine efficacy
WBC	white blood cell
WHO	World Health Organization
Yr	Year

1. Protocol Summary

1.1. Synopsis

Protocol Title: A Randomized, Placebo Controlled, Observer-Blind Phase IIb Study to Evaluate the Safety, Immunogenicity, and Efficacy of BCG revaccination in Healthy Adolescents for the Prevention of Sustained Infection with *Mycobacterium tuberculosis*

Short Title: BCG Revaccination of Healthy Adolescents for the prevention of *Mycobacterium tuberculosis* sustained infection

Rationale: Recently published results from a Phase 2 study evaluating Bacillus Calmette-Guerin (BCG) revaccination in healthy adolescents in South Africa suggest that BCG revaccination of adolescents 12 to 17 years of age may lead to prevention of sustained *Mycobacterium tuberculosis* (*Mtb*) infection (POSI) over a 24 month period following vaccination, as assessed by sustained QuantiFERON-TB Gold® in-Tube (QFT GIT) conversion (i.e., initial QFT conversion and remaining QFT positive at three and six months post conversion [PC]) (Nemes, 2018).

This current study intends to *i)* confirm that BCG revaccination protects against sustained *Mtb* infection in a larger independent study, *ii)* assess the duration of protection through 48 months post revaccination, *iii)* evaluate BCG revaccination in children ten years of age and above and *iv)* identify/validate biomarkers that correlate with risk for or protection against transient and/or sustained *Mtb* infection, as assessed by QuantiFERON plus (QFT) assay, which is the newer version of the QFT GIT assay.

Table 1: Objectives and Endpoints

Objectives	Endpoints
Primary	
• To demonstrate the efficacy of BCG revaccination against sustained *Mtb* infection versus placebo in previously BCG vaccinated QFT negative, healthy adolescents (event-driven analysis)	• Sustained QFT conversion (at least 118 events) based on positive QFT test results (initial conversion and QFT-positive at 3- and 6-months PC)
Secondary	
• To evaluate the durability of efficacy of BCG revaccination against sustained *Mtb* infection versus placebo in previously BCG vaccinated, QFT negative, healthy adolescents	• Sustained QFT conversion based on positive QFT test results (initial conversion and QFT-positive at 3- and 6-months PC) with a follow-up of 36- and 48-months post vaccination

Objectives	Endpoints
Secondary, continued…	
• To evaluate the safety and reactogenicity of BCG revaccination in previously BCG vaccinated, QFT negative healthy adolescents	• Solicited adverse events (AEs) through 7 days post vaccination • Unsolicited AEs through 28 days post vaccination • All serious adverse events (SAEs) and adverse events of special interest (AESIs) through Month 6 • Serious adverse drug reactions (Serious ADRs) through the end of the study
Exploratory	
• To evaluate the efficacy and durability of efficacy of BCG revaccination against primary *Mtb* infection post vaccination versus placebo in previously BCG vaccinated, QFT negative healthy adolescents	• Primary QFT conversion at the time of primary endpoint analysis, and after a follow-up of 36- and 48-months post vaccination, using an alternative definition of conversion as a change in IFN-γ concentration from <0.35 IU/mL to ≥4 IU/mL (initial conversion only)
• To evaluate the efficacy and durability of efficacy of BCG revaccination against primary and sustained *Mtb* infection post vaccination versus placebo in previously BCG vaccinated, QFT negative healthy adolescents, based on alternate QFT IFN-γ concentration cut-off values	• Primary and sustained QFT conversion at the time of primary endpoint analysis, and after a follow-up of 36- and 48-months post vaccination, based on a definition of primary and sustained conversions using exploratory IFN-γ concentration threshold values
• To assess the immunogenicity of BCG revaccination	• Frequency of *Mtb-, BCG-* and/or non-tuberculous mycobacteria (NTM)-specific cluster of differentiation (CD)4 T cells expressing one or more cytokines, e.g., IFN-γ, tumor necrosis factor (TNF)-α, interleukin (IL)-2 and/or IL-22 by intracellular cytokine staining (ICS)

Objectives	Endpoints
Exploratory, continued…	
• To describe host attributes and host responses to vaccination • To describe host attributes and host responses following QFT conversion • To explore and/or develop candidate correlates of risk (CoRs) and correlates of protection (CoPs) • To detect and describe incident TB disease • To characterize *Mtb* isolates	Endpoints may include: • Genetic markers and sequences • Transcriptomics and gene expression markers • Proteomics • Antibody analyses • Cellular analyses (eg, natural killer cells [NK cells], B cells, T cells, myeloid cells)
• To describe COVID-19 among study participants who present with suspected COVID-19	• Signs and symptoms of illness, by treatment group • SARS-CoV-2 nucleic acid amplification from nasal or oropharyngeal sample
• To describe the serostatus of SARS-CoV-2 infection	• Serological tests for SARS-CoV-2, by treatment group

Refer to Section 7.4 for COVID-19 Pandemic contingency plans.

Overall Design:

- Disclosure Statement: This is a randomized, placebo controlled, observer-blind, phase IIb study with two arms (BCG vaccine and saline placebo). An independent data monitoring committee (IDMC) will be established to oversee the safety of this study.
- Intervention model: Participants will be randomly assigned equally to one of two groups in parallel for the duration of the study.
- Intervention Groups: 2 study groups (BCG group and placebo group) will each receive a single intradermal (ID) injection:
  - BCG group will receive one dose of BCG vaccine (Bacillus Calmette-Guerin SSI, Danish strain 1331, live attenuated 2-8 x 10^5 colony forming units (cfu) in a 0.1mL volume ID injection
  - Placebo group will receive one dose of saline control in a 0.1mL volume ID injection
- Primary Purpose: Intervention (BCG revaccination) is being evaluated for POSI (see Section 2.1 and Section 4.1 for details).
- Masking: Participants, sponsor, investigators, contract research organization (CRO) clinical team, laboratory, and clinical staff are blinded to intervention (BCG vs

placebo) until primary endpoint analyses are performed. BCG vaccine recipients and clinical staff at the sites may inadvertently become unblinded as soon as a BCG lesion develops. Refer to Section 6.3.2 for masking details.

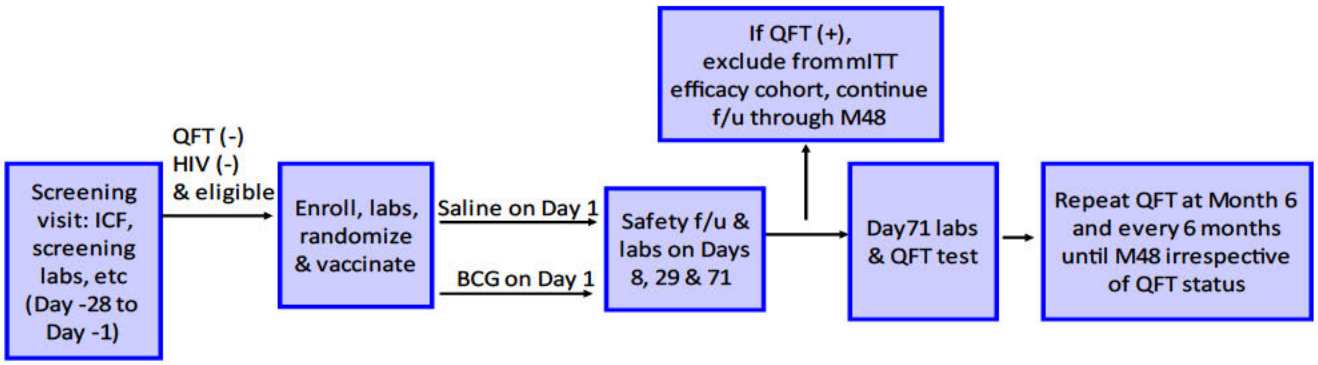
- Number of Participants: Approximately 5625 participants will be screened to enroll and randomize 1800 healthy participants 1:1 to receive either a single dose of BCG vaccine intradermally or a single dose of normal saline placebo intradermally on Day 1. Randomization will be stratified by age strata (10-11 years old, 12-14 years old, and > 14 years old), sex, study site, and school cluster, to account for expected differences in incidence rates. Refer to 6.3.1 for details.
- A subset of 80 participants (the first 80 10-12-year-old participants who are enrolled at the SATVI site) will be included for whole blood ICS analysis to assess frequency of *Mtb-,* BCG- or NTM-specific CD4 T cells expressing one or more cytokines, e.g., IFN-γ, TNF-α, IL-2 and/or IL-22.
- Wash-out period: A 70-day wash-out period will be used to identify participants who may have been infected with *Mtb* just prior to or soon after enrollment. Participants who convert to QFT positive at Day 71 (or if missed or not feasible, at the next feasible visit) will not be asked to return for PC Day 28 and PC Day 84 visits and will be excluded from the primary mITT efficacy analysis but will be followed for safety and efficacy to End-of-Study.
- Total duration of study participation: Each participant will remain in the study for a minimum of 4 years. Enrollment, originally expected to take place over a 1-year period, will be extended as necessary due to COVID-19 pandemic-related restrictions. Refer to 7.4.2 for COVID-19 pandemic contingency plans with regard to participant enrollment, and other details.
- Safety Monitoring: An IDMC will be established to oversee the safety of this study, and will also review and monitor the primary and secondary efficacy results. The IDMC will review unblinded safety data on a regular basis. During active enrollment, the IDMC will meet at least once every three months. Once enrollment is complete, the IDMC will meet at least twice a year. The IDMC review will include solicited and unsolicited AEs, as well as SAEs and AESIs. The IDMC may request additional information, or a pause in recruitment and vaccination, while safety data are being evaluated. The IDMC will make a formal recommendation on the continued enrollment into the trial after each safety review. Each participant will be followed for safety for a minimum of 6 months after vaccination.
- Study sites: Five or more sites in South Africa will participate in this study.
- Participants will be asked to consent to extended passive follow-up for TB disease (using the National TB Programme electronic TB register) for up to 10 years post end of study.

1.2. Schema

Once informed consent and assent have been obtained, healthy participants will be screened for HIV infection and *Mtb* infection.

Approximately 1800 study-eligible HIV-negative, QFT-negative participants will be randomized 1:1 to receive BCG or placebo intradermally. Study procedures are outlined in Figure 1 and details provided in Section 1.3, Schedule of activities (SoA).

Figure 1: Outline of Study Procedures



1.3. Schedule of Activities (SoA)

Table 2: Scheduled Visits and Activities

Study Visit Day (D) or Month (M)	Screen D -28 to D -1	D1	D8[a]	D29[b]	D71[b,c]	M6,12,18, 24,30, 36, 42, 48	Suspected TB Visit	Discon Visit[d]	Notes
Eligibility criteria verification	X	X							Confirm eligibility prior to randomization on D1
Obtain informed consent & assent	X								Re-consent of participants enrolled before study pause
Medical history	X								
Physical examination	X								
Focused physical examination		X	X	X	X	X			Exam if indicated by interval history
Body weight and height measurement	X	X				X			
Body temperature	X	X							
Interval history		X	X	X	X	X			
Vital signs	X	X							
Distribute/review diary cards		X							
Collect diary cards			X						
Distribute memory aid			X						
Collect memory aid				X					
Solicited AEs		X	X						
Unsolicited AEs	X	X	X	X					AEs will be collected from the time informed consent is obtained.

Study Visit Day (D) or Month (M)	Screen D -28 to D -1	D1	D8[a]	D29[b]	D71[b, c]	M6,12,18, 24,30, 36, 42, 48	Suspected TB Visit	Discon Visit[d]	Notes
All serious adverse events (SAEs) and AESI	X	X	X	X	X	X (M6 only)			
Serious ADRs		X	X	X	X	X		X	Collected through the last study visit
Concomitant prescription medications & antipyretic/anti-inflammatory drugs		X	X	X	X				
Immunosuppressive and/or TB medication (curative & preventive)		X	X	X	X	X		X	
Ask about pregnancy		X	X	X	X	X		X	
Injection site examination			X	X	X	X (M6 only)			
TB symptom screen	X			X	X	X		X	
βHCG test all females (mL)	5	X							Serum test at screening Urine test on Day 1, and if pregnancy suspected on subsequent visits
BCG or placebo administration		X							
Urinalysis	X								May be repeated as described in Section 8

Study Visit Day (D) or Month (M)	Screen D -28 to D -1	D1	D8[a]	D29[b]	D71[b, c]	M6,12,18, 24,30, 36, 42, 48	Suspected TB Visit	Discon Visit[d]	Notes
QFT assay (mL)	4				4	4			[e] regarding unevaluable sample
HIV test (mL) with pre- and post-test counselling	3					3	3		At screening, and M12, 24, 36, 48 only, and if TB disease is suspected. [f] regarding unevaluable screening sample
Serum chemistry (mL)	5								Refer to Section 8.2.5 [f] regarding unevaluable screening sample
CBC, differential (mL)	2.5								
Absolute blood cell count (mL)		0.5	0.5	0.5	0.5	0.5			
PAXgene tube for RNA (mL)		2.5	2.5	2.5	2.5	2.5			
Subset only: Whole blood for ICS (mL) for CD4 and CD8 T cells		6		6	6	6 (M6 only)			First 80 10- to 12-year-old participants enrolled at SATVI
PBMC and plasma for CoP & CoR (mL)		34			34	26			
Serum for CoP & CoR (mL) (innate and adaptive)		10	10	10	10	10			
Sputum samples or other appropriate samples for smear microscopy, mycobacterial culture, and GeneXpert							X		
Nasal or oropharyngeal sampling for SARS-CoV-2 diagnostic test	X	X	X	X	X	X			Samples collected only if participant presents with suspected COVID-19

Study Visit Day (D) or Month (M)	Screen D -28 to D -1	D1	D8[a]	D29[b]	D71[b,c]	M6,12,18, 24,30, 36, 42, 48	Suspected TB Visit	Discon Visit[d]	Notes
SARS-CoV-2 serology (mL)	5					5 at M6, M12, M18 and M24			If enrolled before study pause: collect at the next feasible biannual visit (e.g., M6, M12, etc) and biannually thereafter for 24 months
Per visit approximate phlebotomy volume (vol.) (mL) (includes ICS subset)	19.5	53	13	19	57.0	52 at M6 46 at M12			Vol. at M18, 24, 30, 36, 42, 48 = 46 mL per timepoint, excluding SARS
Cumulative approximate vol. (mL) includes ICS subset	19.5	72.5	85.5	104.5	161.5	259.5 Year (Yr)1[g]			Yr 2=351.5mL Yr 3 =443.5mL Yr 4=535.5mL excluding SARS
Cumulative approximate vol. (mL) excludes ICS subset	19.5	66.5	79.5	92.5	143.5	235.5 Yr 1[h]			Yr 2 =327.5mL Yr 3=419.5mL Yr 4 =511.5mL excluding SARS
Cumulative approximate vol. (mL) after study pause (including SARS-CoV-2 serology) includes ICS subset	24.5	77.5	90.5	109.5	166.5	269.5 Yr 1[g]			Yr 2=371.5 mL Yr 3 =463.5mL Yr 4=555.5mL

X indicates procedure to be performed and number indicates blood volume collected (volumes listed are approximate).

[a] Laboratory testing for innate host responses

[b] Laboratory testing for adaptive host responses

[c] QFT test to determine who to exclude from mITT efficacy analysis (end of wash-out period)

[d] A discontinuation (discon) visit will be scheduled for participants who discontinue or withdraw, whenever possible.

[e] QFT sampling: If the blood sample collected for QFT cannot be evaluated (e.g., insufficient volume; damage, hemolytic or lost sample, etc.), collection of a second (repeat) blood sample is permitted to enable evaluation of the primary efficacy endpoint. If the QFT sample was successfully run and has a reported result, including "indeterminate", collection of a second (repeat) blood sample is not permitted.

[f] If a blood sample collected for screening cannot be evaluated (e.g., insufficient volume; damaged, hemolytic or lost sample, etc.) a second (repeat) blood collection is permitted to enable evaluation of study eligibility. The repeat blood collection must occur within the protocol-defined screening visit window. If a screening blood sample was successfully run and has a reported result to determine eligibility, a second (repeat) blood collection is not permitted.

[g] Including subset and includes PC visits Table 3: maximum vol. of whole blood collected through M48 = 637.5mL

[h] Participants not in subset, includes PC visits Table 3: maximum approximate vol. of whole blood collected through M48= 613.5mL

Unscheduled visits should only take place as necessary and data from unscheduled study visits should be captured in the EDC. Refer to Section 8.

Refer to Section 7.4 for COVID-19 Pandemic contingency plans.

Table 3: Schedule of Activities Post QFT Conversion at the Month 6 Visit or Thereafter

Study Visit Day, Post Initial QFT conversion	PC Day 28	PC Day 84
Interval history	X	X
Focused physical examination	X	X
Vaccination-related SAEs	X	X
TB symptom screen	X	X
QFT assay		4
Absolute blood cell count (mL)	0.5	0.5
PAXgene tube for RNA (mL)	2.5	2.5
PBMC and plasma for CoPs & CoRs (mL)	34	34
Serum for CoPs & CoRs (mL)	10	10
Per visit phlebotomy volume (mL)	49	53
Cumulative phlebotomy volume (mL)	49	102

PC = post conversion
X indicates procedure to be performed and number indicates blood volume collected.
Note that participants who become QFT positive will continue to follow procedures in Table 1 through the end of the study
Refer to Section 8 for study procedure details.

Table 4: Study Visit Intervals

Study Visits	Length of Interval	Allowed Interval
Screening	Day -28 to Day -1	Day -28 to Day -1
Day 1	NA	Day 1
Day 8	Day 1 plus **7** days	Day 8 to Day 11
Day 29	Day 1 plus 28 days	Day 29 to Day 35
Day 71	Day 1 plus 70 days	Day 71 to Day 78
Month 6, M12, 18 …M48	Day 1 plus 6 (12, 18, …) calendar months	6 (12, 18, …48) calendar months from Day 1 ±14 days
PC28	Date of QFT conversion plus 28 days	Day 28 to Day 35 post QFT conversion
PC84	Date of QFT conversion plus 84 days	Day 84 to Day 98 post QFT conversion

NA = not applicable
Note that after Day 71, timepoints and intervals will be measured in calendar months.
Refer to Section 7.4 for intervals based on COVID-19 contingency plans.

2. Introduction

Worldwide, tuberculosis (TB) is one of the top 10 causes of death and the leading cause from a single infectious agent. In 2017, TB caused an estimated 1.3 million deaths among HIV-negative people and there were an additional 300,000 deaths from TB among HIV-positive people. It is estimated that 10 million people developed TB disease in 2017: 5.8 million men, 3.2 million women and 1.0 million children: the average case fatality rate of TB is 16% [WHO 2018].

TB is seen in all countries and age groups, but overall, 90% of new cases globally occur in people aged 15 years or above. Thirty high TB burden countries accounted for 87% of the world's cases. The severity of national epidemics varies widely among countries and within countries.

Burden of disease in South Africa

The national incidence of TB in South Africa is estimated at 322,000 (230,000 to 428,000) new cases in 2017, i.e., an incidence rate of 567 per 100 000 population, one of the highest estimated rates based on the World Health Organization (WHO) Global TB report [WHO 2018]. Cohort studies in the Western Cape province showed that the prevalence of *Mtb* infection increases quickly in adolescence and that about half of the adolescents are latently infected with *Mtb*, with demographic and poverty-related socio-economic factors predicting the risk of TB infection [Mahomed 2011; Nemes 2018]. A recent study, largely enrolled in the Worcester region of the Western Cape, reported QFT-GIT conversion rates of approximately 10% per year [Nemes 2018].

Adolescents who are QFT-GIT positive have an almost three-fold higher incidence of active TB disease in subsequent years, compared to QFT-GIT negative adolescents (0.64 vs 0.22 per 100 person-years) [Mahomed 2011]. However, the highest risk of TB disease occurs in adolescents with recent QFT-GIT conversion. Their TB disease incidence was 8 times higher than the rate in adolescents who had remained QFT-GIT negative (1.46 vs 0.17 per 100 person-years) [Machingaidze 2012]. Therefore, QFT-GIT conversion is an important indicator for the risk to develop active TB.

2.1. Study Rationale

The recently published Aeras Phase 2b study C-040-404 (Aeras 404 study) evaluated the vaccine efficacy (VE) of BCG revaccination in healthy 12 to 17-year-old HIV-negative participants in South Africa against initial QuantiFERON-TB Gold in tube ® (QFT-GIT) conversion, a surrogate for primary *Mtb* infection. The study did not meet its primary endpoint but reported 45% VE against sustained QFT-GIT conversion, i.e., initial QFT-GIT conversion and remaining QFT-GIT positive at 3- and 6-months PC, as a surrogate for sustained latent *Mtb* infection [Nemes 2018].This finding is encouraging as QFT reversion (from QFT-positive to QFT-negative) is thought to be indicative of successful immune control of the initial infection. Treatment of latent TB infection (LTBI) patients with Isoniazid (INH) has been reported to significantly decrease IFN-γ concentrations in the QuantiFERON TB Plus (QFT) assay, suggesting that IFN-γ concentrations may correlate with the bacillary load in LTBI [Petruccioli 2018].

The current study aims to confirm the findings of the previous BCG revaccination study. The study will be conducted at sites in the Western Cape province, where the initial finding of protection was observed, and additional site(s) in other provinces. The study aims to extend the findings of the previous study by modifying three of its aspects: Firstly, the study will enroll adolescents from ten years of age (rather than 12 years of age), aiming to increase the proportion of QFT-negative participants among the population screened for inclusion and to generate safety and immunogenicity data in an age range that could potentially be considered for revaccination of a majority QFT-negative population in high-transmission settings. Secondly, the duration of follow-up will be extended from two years to 4 years in order to assess the duration of protection from sustained infection. Thirdly, biospecimens will be collected 4- and 12-weeks post QFT conversion to characterize potential differences in the immune response between participants who develop sustained conversion versus reversion.

The study described here intends to demonstrate that BCG revaccination of healthy HIV-negative adolescents leads to POSI, as assessed by sustained QFT conversion. Although it remains to be demonstrated that POSI will translate to prevention of disease (POD), it is biologically plausible to argue that successful immune control of the initial *Mtb* infection is likely to reduce the risk of progression to active disease. The goal of this study is to generate the evidence needed to inform whether a POD trial in QFT-negative is justified and necessary.

2.1.1. Rationale for addition of COVID-19 related study objectives

The Government of South Africa declared the COVID-19 pandemic a National Disaster on 15 March 2020 and the Sponsor of this study issued a pause for enrollment on 17 March 2020, effective 19 March 2020.

The Government of South Africa subsequently ordered a lockdown (stay-at-home order) for three weeks on 26 March 2020 and subsequently extended it for an additional two weeks, through the end of April, 2020. Under the lockdown, most trial participants are not able to come to the investigational site for protocol-specified visits.

Since 1 May 2020, South Africa has employed a system of alert levels at the provincial and, in some cases, the district level. Criteria based on the rate of infection as well as health system capacity are used by the National Command Council to determine the alert level.

Enrollment into this study can only resume once the Government, local IRBs, investigators, and the sponsor agree that it is acceptable to resume enrollment. Enrollment may also need to be paused again in the future, depending on the COVID-19 situation at national, province and district level.

Study investigators and study sites have defined infection control measures for COVID-19 that need to be implemented prior to resuming enrollment and are consulting with IRBs to determine the conditions that would allow for enrollment to continue.

The study investigators and study sponsor agreed to add two exploratory COVID-19 endpoints to this study. Firstly, in order to describe COVID-19 among the study population, to differentiate COVID-19 from pulmonary TB, and to minimize risks to participants and staff, a diagnostic test for SARS-CoV-2 (based on nucleic acid amplification) will be performed if a participant presents with suspected COVID-19. A description of the frequency and symptomatology of

COVID-19 by treatment group is of great scientific interest as BCG-induced trained immunity has been hypothesized to potentially offer protection from progression to severe COVID-19. BCG vaccination is currently being tested as a prophylactic intervention against COVID-19 (NCT04327206 and additional studies).

Secondly, to describe the penetrance of SARS-CoV-2 infection in the communities that participate in this study, SARS-CoV-2 serology will be performed once every six months for a duration of 24 months. This will aid our understanding of virus transmission in the communities we work in and may allow a better understanding of the frequency of asymptomatic illness.

2.2. Background

BCG vaccination at birth is part of the South African National Routine Immunization Schedule and prevents severe forms of TB in childhood. BCG revaccination of children is not recommended in the Republic of South Africa but has been practiced in a number of countries. BCG revaccination appears to be safe and well tolerated in adolescents and young adults [Nemes 2018; Pereira 2012; Petruccioli 2018, Rodrigues 2005; Bottiger 1983]. A trial of BCG revaccination in 2,997 fourteen to fifteen-year-old Swedish adolescents reported open vaccination lesions in 4% of vaccines who received the Danish BCG vaccine that will be used in this trial [Bottiger 1983].

Currently there is no consensus with regards to the efficacy or effectiveness of BCG revaccination for the prevention of TB disease. The recent Phase 2b study evaluating BCG revaccination in South Africa suggested that BCG revaccination may confer POSI for at least two years following vaccination. Previous cluster-randomized, open label, uncontrolled trials in Brazil [Petruccioli 2018; Rodrigues 2005; Cunha 2008; Barreto 2011] and a randomized, placebo-controlled trial in Malawi [Fine 1996] did not find statistically significant efficacy in the per protocol population for the prevention of TB disease following BCG revaccination. Differences in populations (no screening for tuberculin skin test [TST] or IFN-γ release assay status), age, geography, climate, force of infection and the prevalence of infection with NTM are being discussed as potential root causes for the observed differences in VE following BCG revaccination. Indeed, 33% (95% CI 4%-53%) VE was observed for the prevention of TB in children who were revaccinated before the age of 11 years at one of two Brazilian sites with lower NTM prevalence [Barreto 2011].

This current study intends to *i)* confirm that BCG revaccination protects against sustained *Mtb* infection in a larger independent study, *ii)* assess the duration of protection through 48 months, *iii)* evaluate BCG revaccination in children 10 years of age and above and *iv)* identify/validate biomarkers that correlate with the risk for or protection against transient and/or sustained *Mtb* infection, as assessed by QFT.

2.3. Benefit/Risk Assessment

The benefits and risks of BCG vaccination with the Danish BCG Vaccine SSI, manufactured by AJ Vaccines, Copenhagen, Denmark, are well documented in the South African package insert. The approved AJ Vaccines package insert serves as Reference Safety Information.

An IDMC will monitor safety data on a regular basis.

With regards to BCG vaccination of older children and adolescents, a recent randomized controlled trial conducted in South Africa reported that BCG revaccination was well tolerated in adolescents 12 to 17 years of age and had a VE against sustained QFT conversion of 45% [Nemes 2018]. A population-based case–control study in the United Kingdom (UK) evaluated protection against TB following primary BCG vaccination of children aged 12–13 years.VE against TB was 51% (95% CI 21, 69%) at 10–15 years after vaccination and 57% (CI 33, 72%) at 15–20 years after vaccination [Mangtani 2017].

With regards to AEs following vaccination, a local reaction is normal after BCG. A small tender red swelling appears at the site of the injection which gradually changes to a small vesicle. An ulcer may appear 2 to 4 weeks after vaccination. The reaction usually subsides within two to five months and in practically all children leaves a superficial scar 2 – 10 mm in diameter. Transient enlargement of the regional lymph nodes (< 1 cm) is normal. Enlargement of axillary lymph nodes may be seen less frequently in the months following immunization.

Suppurative lymphadenitis may occur less frequently. This is a benign condition which heals spontaneously, although often only slowly.

As stated in the BCG vaccine package insert, side-effects include:

Infections and Infestations
Less frequent: Osteomyelitis, Suppurative lymphadenitis, Injection site abscess

Blood and the lymphatic system disorders
Less frequent: Enlargement of axillary lymph nodes > 1 cm may appear following immunization.

Inflammation of gland, sometimes with abscesses and release of fluid from the swellings.

Immune system disorders
Less frequent: Anaphylactic reaction, Allergic reaction

Nervous system disorders
Less frequent: Headache

Skin and subcutaneous tissue disorders
Less frequent: Lupus types of reaction and keloid formation may occur.

Musculoskeletal, connective tissue and bone disorders
Less frequent: Osteitis

General disorders and administrative site conditions
Less frequent: Inadvertent subcutaneous injection produces abscess formation and may lead to ugly retracted scars. Disseminated BCG disease may occur, particularly in immuno-suppressed individuals.
Less frequent: Fever, injection site ulceration, injection site discharge.

Other side effects

Less frequent: Several allergic reactions (such as redness of the face and neck, swelling of the face, throat or neck, skin rash, breathing difficulties and collapse) can occur. These often start very soon after injection.

Less frequent: Infection with the bacteria in the vaccine (*Mycobacterium bovis* BCG) can occur, that can spread through the body, including to the bones. This does not usually happen in people who are otherwise healthy, but it has been reported. These infections need to be treated in a similar way to the treatment of TB.

3. Objectives and Endpoints

Table 5: Objectives and Endpoints

Objectives	Endpoints
Primary	
• To demonstrate the efficacy of BCG revaccination against sustained *Mtb* infection versus placebo in previously BCG vaccinated QFT negative, healthy adolescents (event-driven analysis)	• Sustained QFT conversion (at least 118 events) based on positive QFT test results (initial conversion and QFT-positive at 3- and 6-months PC)
Secondary	
• To evaluate the durability of efficacy of BCG revaccination against sustained *Mtb* infection versus placebo in previously BCG vaccinated, QFT negative, healthy adolescents	• Sustained QFT conversion based on positive QFT test results (initial conversion and QFT-positive at 3- and 6-months PC) with a follow-up of 36- and 48-months post vaccination
• To evaluate the safety and reactogenicity of BCG revaccination in previously BCG vaccinated, QFT negative healthy adolescents	• Solicited adverse events (AEs) through 7 days post vaccination • Unsolicited AEs through 28 days post vaccination • All serious adverse events (SAEs) and adverse events of special interest (AESIs) through Month 6 • Serious adverse drug reactions (Serious ADRs) through the end of the study
Exploratory	
• To evaluate the efficacy and durability of efficacy of BCG revaccination against primary *Mtb* infection post vaccination versus placebo in previously BCG vaccinated, QFT negative healthy adolescents	• Primary QFT conversion at the time of primary endpoint analysis, and after a follow-up of 36- and 48-months post vaccination, using an alternative definition of conversion as a change in IFN-γ concentration from <0.35 IU/mL to ≥4 IU/mL (initial conversion only)
• To evaluate the efficacy and durability of efficacy of BCG revaccination against primary and sustained *Mtb* infection post vaccination versus placebo in previously BCG vaccinated, QFT negative healthy adolescents, based on alternate QFT IFN-γ concentration cut-off values	• Primary and sustained QFT conversion at the time of primary endpoint analysis, and after a follow-up of 36- and 48-months post vaccination, based on a definition of primary and sustained conversions using exploratory IFN-γ concentration threshold values

Objectives	Endpoints
Exploratory, continued…	
• To assess the immunogenicity of BCG revaccination	• Frequency of *Mtb-, BCG-* and/or non-tuberculous mycobacteria (NTM)-specific cluster of differentiation (CD)4 T cells expressing one or more cytokines, e.g., IFN-γ, tumor necrosis factor (TNF)-α, interleukin (IL)-2 and/or IL-22 by intracellular cytokine staining (ICS)
• To describe host attributes and host responses to vaccination • To describe host attributes and host responses following QFT conversion • To explore and/or develop candidate correlates of risk (CoRs) and correlates of protection (CoPs) • To detect and describe incident TB disease • To characterize *Mtb* isolates	Endpoints may include: • Genetic markers and sequences • Transcriptomics and gene expression markers • Proteomics • Antibody analyses • Cellular analyses (eg, natural killer cells [NK cells], B cells, T cells, myeloid cells)
• To describe COVID-19 among study participants who present with suspected COVID-19	• Signs and symptoms of illness, by treatment group • SARS-CoV-2 nucleic acid amplification from nasal or oropharyngeal sample
• To describe the serostatus of SARS-CoV-2 infection	• Serological tests for SARS-CoV-2, by treatment group

Refer to Section 7.4 for COVID-19 Pandemic contingency plans.

4. Study Design

Overall Design:

- Disclosure Statement: This is a randomized, placebo controlled, observer-blind, phase IIb study with two arms (BCG vaccine and saline placebo). An IDMC will be established to oversee the safety of this study.
- Intervention model: Participants will be randomly assigned equally to one of two groups in parallel for the duration of the study.
- Intervention Groups: 2 study groups (BCG group and placebo group) will each receive a single ID injection:
  - BCG group will receive one dose of BCG vaccine (Bacillus Calmette-Guerin SSI, Danish strain 1331, live attenuated 2-8 x 10^5 cfu in a 0.1mL volume ID injection
  - Placebo group will receive one dose of saline control in a 0.1mL volume ID injection
- Primary Purpose: Intervention (BCG revaccination) is being evaluated for POSI (see Section 2.1 and Section 4.1 for details).
- Masking: Participants, sponsor, investigators, CRO clinical team, laboratory, and clinical staff are blinded to intervention (BCG vs placebo) until primary endpoint analyses are performed. BCG vaccine recipients and clinical staff at the sites may inadvertently become unblinded as soon as a BCG lesion develops. Refer to Section 6.3.2 for masking details.
- Number of Participants: Approximately 5625 participants will be screened to enroll and randomize 1800 healthy participants 1:1 to receive either a single dose of BCG vaccine intradermally or a single dose of normal saline placebo intradermally on Day 1. Randomization will be stratified by age strata (10-11 years old, 12-14 years old, and > 14 years old), sex, study site, and school cluster, to account for expected differences in incidence rates. Refer to 6.3.1 for details.
- A subset of 80 participants (the first 80 10-12-year-old participants who are enrolled at the SATVI site) will be included for whole blood ICS analysis to assess frequency of *Mtb-,* BCG- or NTM-specific CD4 T cells expressing one or more cytokines, e.g., IFN-γ, TNF-α, IL-2 and/or IL-22.
- Wash-out period: A 70-day wash-out period will be used to identify participants who may have been infected with *Mtb* just prior to or soon after enrollment. Participants who convert to QFT positive at Day 71 (or if missed or not feasible, at the next feasible visit) will not be asked to return for PC Day 28 and PC Day 84 visits and will be excluded from the primary mITT efficacy analysis but will be followed for safety and efficacy to End-of-Study.
- Total duration of study participation: Each participant will remain in the study for a minimum of 4 years. Enrollment, originally expected to take place over a 1-year

period, will be extended as necessary due to COVID-19 pandemic-related restrictions.

- Refer to Section 7.4 for COVID-19 pandemic contingency plans with regard to participant enrollment, and other details.
- Safety Monitoring: An IDMC will be established to oversee the safety of this study, and will also review and monitor the primary and secondary efficacy results. The IDMC will review unblinded safety data on a regular basis. During active enrollment, the IDMC will meet at least once every three months. Once enrollment is complete, the IDMC will meet at least twice a year. The IDMC review will include solicited and unsolicited AEs, as well as SAEs and AESIs. The IDMC may request additional information, or a pause in recruitment and vaccination, while safety data are being evaluated. The IDMC will make a formal recommendation on the continued enrollment into the trial after each safety review. Each participant will be followed for safety for a minimum of 6 months after vaccination.
- Study sites: Five or more sites in South Africa will participate in this study.
- Participants will be asked to consent to extended passive follow-up for TB disease (using the National TB Programme electronic TB register) for up to 10 years post end of study.

4.1. Scientific Rationale for Study Design

The recently published Aeras 404 study evaluated the VE of BCG revaccination in healthy 12 to 17-year-old HIV-negative participants in South Africa against initial QuantiFERON-TB Gold in tube® (QFT-GIT) conversion, a surrogate for primary *Mtb* infection. The study did not meet its primary endpoint but reported 45% VE against sustained QFT-GIT conversion, i.e., initial QFT-GIT conversion and remaining QFT-GIT positive at 3- and 6-months PC, as a surrogate for sustained latent *Mtb* infection [Nemes 2018]. This finding is encouraging as QFT reversion (from QFT-positive to QFT-negative) is thought to be indicative of successful immune control of the initial infection, and thus POSI could potentially translate into prevention of disease.

The current study is designed to confirm the results of the Aeras 404 study in a larger population by evaluating whether BCG revaccination confers POSI. Eligible clinical trial participants are randomized 1:1 to receive either BCG vaccine or placebo intradermally.

The study design is observer blinded to enable unbiased assessment of adverse events until a BCG lesion develops, thus unblinding the majority of BCG vaccine recipients and the clinical team at the sites. Care will be taken to keep the blind for all other staff, including all laboratory staff and the sponsor staff. Sponsor staff may become aware of individual treatment assignments during the course of data review; however, precautions will be taken to limit exposure of sponsor staff to unblinded data.

Placebo control is used to *a)* allow unbiased safety assessment and *b)* to enable demonstration of vaccine efficacy for POSI.

Participants who convert to QFT-positive at Day 71 will be excluded from the modified intention to treat efficacy analyses and the per protocol immunogenicity analyses as they may have been

infected with *Mtb* prior to or shortly after revaccination. However, these participants will continue study participation with all study procedures through the end of the study.

The primary endpoint in this study, sustained QFT conversion (initial conversion and QFT-positive at both 3- and 6-months PC), is the same as the secondary endpoint in the Aeras 404 study. The intention of our study is to demonstrate VE against POSI with statistical criteria that are typically used for pivotal efficacy trials (ie, 90% power, alpha 2.5%).

The duration of follow-up with semiannual QFT-testing is 4 years, as compared to two years in the Aeras 404 study. The longer follow-up will enable us to assess the duration of protection past 24 months as a secondary objective, and the statistical analysis plan will include duration of protection analyses.

The study intends to enroll adolescents from 10 years of age since the previous trial found that approximately half of the adolescents 12 to 17 years of age are already infected with *Mtb*, as assessed by QFT. Since this intervention aims to prevent sustained infection, it would therefore be meaningful to implement in an age group in which the majority are uninfected.

Another secondary endpoint is to evaluate the efficacy and durability of efficacy of BCG revaccination against primary *Mtb* infection post vaccination using an alternative definition of conversion as a change in IFN-γ concentration from <0.35 IU/mL to ≥4 IU/mL (initial conversion only). In the Aeras 404 study, the QFT-GIT assay was used and a vaccine efficacy of ~45% was observed for this endpoint.

The rationale for this end-point is that QFT conversion above this threshold was associated with an increased risk of tuberculosis disease in infants and adults [Andrews 2017; Winje 2018, a finding that was consistent with predictions from studies in animal models [Andersen 2007]. Therefore, this alternative conversion threshold will be evaluated as a secondary exploratory objective in this trial. The QFT-GIT assay has limitations as a test for *Mtb* infection, due to intra-participant and dynamic variability around the threshold recommended by the manufacturer (Pai, 2012; Mazurek et al, 2010). QFT-GIT assay variability appears maximal in low burden settings with low rates of *Mtb* infection. Findings from high TB burden settings with high risk of *Mtb* infection, such as South Africa, where the rate of QFT-GIT conversion is similar to the expected rate of TST conversion, are in contrast to the pattern observed in serial testing of low risk study populations, where the rate of QFT-GIT conversion has been unexpectedly high compared to TST [Pai 2012; Zwerling 2013].

In this current study the QuantiFERON®-TB-Gold-Plus assay (QFT) will be used, which is the updated version of the QFT-GIT assay.

Optimum use of QFT conversion as a surrogate for *Mtb* infection, and potentially as a correlate of risk for progression to disease, requires further exploration of the effect of alternative QFT conversion thresholds in multiple populations and in high and low burden countries. For example, it has been shown that individuals who had a change in QFT-GIT IFN-γ values from less than 0.2 to greater than 0.7 IU/mL had 10-fold higher tuberculosis incidence rates than those who maintained values less than 0.2 IU/mL over 2 years (P = 0.0003). By contrast, “uncertain” converters, with at least one of serial QFT-GIT results between 0.2 and 0.7 IU/mL, were not at

higher risk than nonconverters (P = 0.229; Nemes, 2018). Therefore, other conversion thresholds will be evaluated as exploratory objectives, similar to Nemes et al.

4.2. Justification for Dose

The BCG vaccine is licensed in South Africa. The dosage used in this study is the same as that of the licensed vaccine.

4.3. End of Study Definition

A participant is considered to have completed the study if he/she completes the final visit at Month 48. The end of the study is defined as the date of the last visit of the last participant in the study or last scheduled procedure shown in the SoA for the last participant in the trial.

The primary endpoint is reached when at least 118 participants have converted from a negative to positive QFT result, sustaining positivity for at least 6 months post initial conversion. The study will continue after the primary endpoint has been reached, and will conclude when all participants have completed the Month 48 visit, or discontinued the study. Participants with an initial conversion at Month 48 will not be evaluated for sustained conversion.

5. Study Population

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

Recruitment

Participants will be recruited from communities and schools with known high burden of TB transmission and disease. Good participatory practices will be followed and site staff will work closely with community organizations, community leaders, parent organizations, teachers and school and Department of Education officials.

Various methods of recruitment may be used, such as community and classroom information sessions, advertising, referrals, word-of mouth, or solicitation through participants previously known to the clinical site.

All recruitment materials will be approved by the appropriate Institutional Review Board/s (IRBs) or Independent Ethics Committee/s (IECs). Interested participants will be invited to participate in the informed consent and assent process.

5.1. Inclusion Criteria

Participants are eligible to be included in the study only if all of the following criteria apply:

Age:

1. ≥ 10 years and ≤ 18 years on Study Day 1

Health and Medical Characteristics:

2. General good health, confirmed by medical history and physical examination
3. Vaccinated with BCG at least 5 years ago, documented through medical history or by presence of healed BCG scar
4. Tests QFT negative at screening
5. For female participants: not pregnant and agrees to avoid pregnancy throughout the first 12 months of the study. Women physically capable of pregnancy must agree to use an acceptable method of avoiding pregnancy during this period. Acceptable methods of avoiding pregnancy include sexual abstinence (not engaging in sexual intercourse), a confirmed sterile partner, or at least 2 contraception methods from the following list: male or female condom, diaphragm, intrauterine devices (IUDs), hormonal contraceptive (oral, injection, transdermal patch, or implant). Refer to Appendix 3, Section 10.3, for details.

Specific Additional Requirements:

6. Agrees to stay in contact with the study site for the duration of the study, provide updated contact information as necessary, and has no current plans to move from the study area for the duration of the study

Informed Consent:

7. Capable of giving signed informed consent/assent as described in Appendix 1, Section 10.1.4 and completes the written informed consent/assent process.

5.2. Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply:

Medical Conditions and History:

8. Acute illness on Study Day 1 (refer to Section 5.4 for additional details). NOTE: This is a temporary exclusion for which the subject may be re-evaluated.
9. Body temperature ≥37.5°C on Study Day 1 (refer to Section 5.4 for additional details). NOTE: This is a temporary exclusion for which the subject may be re-evaluated.
10. History or evidence of any clinically significant disease, including severe eczema and severe asthma, or any acute or chronic illness that might affect the safety, immunogenicity, or efficacy of study vaccine in the opinion of the investigator
11. Any current medical, psychiatric, occupational, or substance abuse problems that, in the opinion of the investigator, will make it unlikely that the participant will comply with the protocol

12. History of autoimmune disease
13. History or evidence of active TB disease
14. History or laboratory evidence of any past or present possible immunodeficiency state including, but not limited to, any laboratory indication of HIV-1 infection
15. History of allergic disease that is likely to be exacerbated by any component of the study vaccine

Prior/Concomitant Therapy

16. History of treatment for active TB disease or history of latent *Mtb* infection
17. Received a TST within 6 months prior to Study Day 1
18. Received immunosuppressive treatment, e.g., chemotherapy, biologics or radiation therapy, or used immunosuppressive medication (daily steroid equivalent of ≥5mg prednisone) within 42 days before Study Day 1. Inhaled and topical corticosteroids are permitted.
19. Received immunoglobulin or blood products within 42 days before Study Day 1
20. Planned administration/administration of a licensed vaccine in the period starting 28 days before and ending 28 days after Study Day 1

Prior/Concurrent Clinical Study Experience

21. Received investigational TB vaccine at any time prior to Study Day 1
22. Received any investigational drug therapy or investigational vaccine within 180 days before Study Day 1, or planned participation in any other clinical trial using investigational product during the study period

Diagnostic assessments

23. Laboratory values from the most recent blood collected prior to randomization outside the normal range that are suggestive of a disease state. Grade 1 abnormalities (as per DAIDS toxicity table version 2.1) do not lead to exclusion if the investigator considers them not clinically significant:
24. Urinalysis abnormality greater than Grade 1 on the Toxicity Scale (with the exception of hematuria in a menstruating female), or urinalysis abnormality judged clinically significant by the investigator

Other Exclusions

25. Shared residence with an individual who is receiving TB treatment or with someone who is known to have incompletely treated TB. E.g., Xpert MTB/RIF assay-positive, PCR-positive, culture-positive, smear-positive TB, or clinically diagnosed unconfirmed TB.
26. Child in Care*
27. Female participants currently pregnant or lactating/nursing; or positive serum pregnancy test during screening or on Day 1, prior to vaccination, or planning a pregnancy within the first 12 months after study intervention. Refer to Appendix 3, Section 10.3 for details.

*Child in Care is defined as a child who is under the care (control or protection) of an agency, organization, institution or entity by the courts, the government body, acting in accordance with powers conferred in them by law or regulations. The definition of a child in care can include a child who is cared for by foster parents or living in a care home or institution, provided that the arrangements falls within the definition above. The definition of a child in care does not include a child who is adopted or has an appointed legal guardian. The definition of a child in care does include a child who is cared for by family members – other than parents – who have not gone through formal legal adoption/guardian processes.

5.3. Lifestyle Considerations

No restrictions are required.

5.4. Screen Failures

Screen failures are defined as participants who consent to participate in the clinical study but are not subsequently randomly assigned to study intervention/entered in the study. A minimal set of screen failure information is required to ensure transparent reporting of screen failure participants to meet the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria, and any AE or SAE from the time of consent.

Screening assessments can be done at any time during the screening interval, except for the written informed consent, which must be completed prior to any screening procedure.

If a participant presents with an acute illness (e.g., elevated temperature, acute respiratory or gastrointestinal illness, UTI) or an abnormal urinalysis (e.g., due to menstruation or urinary tract infection), repeat procedures, with the exception of blood collection (unless allowed as described in section 8), may be performed as long as they are completed within the screening window.

6. Study Intervention

Study intervention is defined as any investigational intervention(s), marketed product(s), placebo, or medical device(s) intended to be administered to a study participant according to the study protocol.

6.1. Study Intervention(s) Administered

Two interventions will be used in this study: participants will receive either BCG vaccine or placebo intradermally. On Study Day 1, study participants will receive their injection after randomization and baseline immunology blood collection and other required assessments are performed.

Participants randomized to the BCG group will receive a single 0.1mL volume of BCG vaccine, administered intradermally in the deltoid region of the upper arm. Participants randomized to the Control group will receive a single 0.1 mL volume of normal saline, administered intradermally in deltoid region of the upper arm.

Table 6: Study Interventions

	BCG licensed Vaccine	**Control**
Intervention Name	BCG vaccine SSI	Normal saline
Unit Dose Strength(s)	2 to 8 x 10^5 cfu	normal saline (0.9% NaCl)
Dosage Volume	0.1 mL, single injection	0.1 mL, single injection
Route of Administration	ID injection	ID injection

6.1.1. BCG Vaccine

The BCG vaccine used in this study, Danish BCG vaccine SSI, is manufactured by AJ Vaccines, Copenhagen, Denmark. The vaccine is registered and distributed in South Africa by the Biovac Institute (15 Alexandra Road, Pinelands, Cape Town), for prevention of TB in children and adults, and is included in their national immunization program. The AJ Vaccine's BCG vaccine is the licensed SSI BCG Danish 1331 vaccine.

The BCG Vaccine SSI is supplied as a freeze-dried (lyophilized) powder containing live attenuated *M. bovis* BCG, Danish BCG strain 1331 and sodium glutamate stabilizer. The vaccine powder, which is in an amber vial, is white and crystalline.

The vaccine is commercially supplied as a pack of 10 vials. Each vial contains 2-8 million colony forming units of BCG, to be reconstituted with 1mL of the diluent, diluted Sauton SSI, to derive 10 adult doses of 0.1mL each.

The diluted Sauton SSI diluent contains magnesium sulfate, heptahydrate, dipotassium phosphate, citric acid monohydrate, L-asparagine monohydrate, ferric ammonium citrate and glycerol 85%, and water for injections.

The diluent is in a clear vial and is a clear, colorless solution without visible particles. The diluent is also supplied by the Biovac Institute in South Africa and commercially supplied as a pack of 10 vials.

6.1.2. Placebo

The placebo control is normal saline, 0.9% NaCl, as commercially available for injection in South Africa. A volume of 0.1mL of normal saline is administered intradermally.

6.1.3. Administration

Administration procedures for the BCG vaccine are found in the package insert. The placebo control will be administered in the same manner. The placebo will be administered intradermally into the deltoid region of the upper arm using standard technique and a fine short needle with a short bevel (25 G/0,50 mm or 26 G/0,45 mm). BCG is slightly opaque compared to the transparent saline placebo.

The unblinded investigational pharmacist will provide BCG and placebo to the clinic as unit-dose syringes with a masking label, which will be identified with the participant identification

number, date and time of dose preparation, and the volume prepared. An investigator (ie, physician) must be present in the clinic at the time of administration.

Before administering the injection, the study intervention administrator must inspect the syringe, checking that the syringe is identified with the correct participant identification number and checking the date and time the dose was prepared.

6.2. Preparation/Handling/Storage/Accountability

Further guidance and information for the preparation, handling, storage and accountability are provided in the Study Reference Manual.

Refer to 6.1.1 for preparing the BCG vaccine for administration. Vaccine will be prepared as per the manufacturer's recommendations by the study pharmacist. All of the participating sites have investigational pharmacies and will use an unblinded pharmacist. Once the vaccine is prepared, the pharmacist will cover the vaccine syringe content with a blinding label so that the clinical staff remains blinded. Vaccine will NOT be administered by the study pharmacist.

Placebo will be prepared as in Section 6.1.2.

BCG must be stored at 2°C to 8°C in a secured location with no access for unauthorized personnel. Reconstituted vaccine may be kept at 2°C to 8°C for up to 6 hours in the original vial. Exposure to light should be kept to a minimum. Any reconstituted vaccine not used within six hours must be discarded.

The saline placebo will be stored in a secured location at 2°C to 8°C in the study pharmacy.

The study pharmacist (or designee) must confirm appropriate temperature conditions have been maintained during transit and during site storage for all study intervention received and any discrepancies are reported and resolved before use of the study intervention. Upon receipt of study vaccine supplies, the unblinded investigational pharmacist must immediately inspect all kits for damage. Any damage or discrepancies from the packing list must be documented and promptly discussed with the sponsor and the study monitor to determine the appropriate action.

Only participants enrolled in the study may receive study intervention and only authorized site staff may supply or administer study intervention. All study intervention must be stored in a secure, environmentally controlled, and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the investigator and authorized site staff.

The principal investigator (PI) is responsible for study intervention accountability, reconciliation, and record maintenance (i.e., receipt, reconciliation, and final disposition records).

Authorization for any unused study vaccine and supplies to be destroyed is the responsibility of the sponsor. Unused supplies will be destroyed according to the facility's SOPs or per local regulations. Any disposal of study vaccine conducted at the clinical site must be documented in the study file.

6.3. Measures to Minimize Bias: Randomization and Blinding

6.3.1. Randomization

Participants will be randomized to one of two interventions (BCG or placebo) based on a randomly-generated sequence of participant identification (identifier) numbers (randomization schedule) using a validated Interactive Voice/Web Response System (IVRS/IWRS).

Before the study is initiated, the telephone number and call-in directions for the IVRS and/or the log in information and instructions for the IWRS will be provided to the study sites.

The randomization schedule will be prepared by a statistician who will not be involved in the analysis of the study in order to maintain the blind of the study team.

The day of randomization for each participant will be Study Day 1. Randomization will be stratified by age strata (10-11 years old, 12-14 years old, and > 14 years old), sex, study site, and school cluster, to account for expected differences in incidence rates.

To minimize risk of study failure by maximizing consistency of study assumptions with available literature data, the overall proportion of patients randomized in the youngest age strata (10-11 years old), and outside the Western Cape Province will be actively monitored and capped at approximately 10% and 20% of the randomized study population, respectively.

6.3.2. Masking

The study will be conducted as an observer blind study until the primary endpoint has been reached and primary endpoint analyses are completed. While the study is blinded, sponsor staff may become aware of individual treatment assignments during the course of data review, however, precautions will be taken to limit exposure of sponsor staff to unblinded data. Following completion of primary endpoint analyses, the study team will be unblinded but laboratory staff will remain blinded until all laboratory studies have been completed.

Only the following people will have access to treatment allocation while the study is blinded:

- Investigational pharmacists preparing the study interventions
- Biostatistician preparing the randomization list
- Biostatistician preparing the IDMC data
- IDMC members
- Unblinded study Monitors.

All unblinded persons must take care to not reveal individual group assignments to any other member of the study team.

Unblinded study personnel must not participate in the evaluation of adverse events. A delegation of authority log will be maintained by the site and will identify the individual(s) authorized to function as the study vaccine manager, i.e., individuals with access to study blinding information.

After primary endpoint analyses are completed – likely more than two years after completion of enrollment– the study will be unblinded to CRO staff and the sponsor to reduce the complexity of statistical analyses for IDMC and other data reviews. Site staff, laboratory staff, and participants will remain blinded.

Because of the unique local reaction to BCG vaccination, the site staff and participant will become aware of study intervention allocation if and when a BCG lesion develops.

6.3.3. Blind Break

The IVRS/IWRS will be programmed with blind-breaking instructions. In addition, instructions on emergency unblinding in case of system outage will be provided. In case of an emergency, the investigator has the sole responsibility for determining if unblinding of a participant's treatment assignment is warranted. Participant safety must always be the first consideration in making such a determination. If the investigator decides that unblinding is warranted, the investigator should make every effort to contact the sponsor prior to unblinding a participant's treatment assignment unless this could delay emergency treatment of the participant. If a participant's treatment assignment is unblinded, during the observer-blind period (prior to completion of primary endpoint analyses), the sponsor must be notified within 24 hours after breaking the blind. The date and reason that the blind was broken must be recorded in the source documentation and case report form (CRF), as applicable.

6.4. Study Intervention Compliance

Participant compliance with study intervention will be recorded on his/her CRF.

6.5. Concomitant Therapy

Any prescription medication, anti-inflammatory drugs and antipyretic drugs*, or vaccine that the participant receives from enrollment through study Day 71 must be recorded along with:

- Reason for use
- Dates of administration including start and end dates
- Dosage information including dose and frequency.

* The use of anti-inflammatory and antipyretic medication should be discouraged during the first 28 study days (Day 1 through Day 28).

Information regarding all immunosuppressant therapy/medications and any TB medication (curative or preventive) will be collected through the last study visit.

The Medical Monitor should be contacted if there are any questions regarding concomitant or prior therapy.

6.6. Dose Modification

There are no dose modification specifications.

All participants will receive either 1 dose (injection) of BCG or 1 dose of placebo as described in Section 6.1.

6.7. Intervention after the End of the Study

There is no intervention planned after the end of the study.

7. Discontinuation of Study Intervention and Participant Discontinuation/Withdrawal and COVID-19 Contingency Plans

Refer to Section 7.4, COVID-19 Contingency Plans for details regarding study pause due to COVID-19.

7.1. Discontinuation of Study Intervention

This is a single dose study. Therefore, there is no plan to discontinue a participant from any additional doses.

7.1.1. Pausing Rules for Study

Any of the below conditions, if identified either by an investigator, the sponsor or the IDMC, will trigger a pause of the enrollment and pause of administration of study intervention until the IDMC has reviewed the safety data and made a recommendation on how to proceed:

1. Anaphylaxis with or without bronchospasm within 4 hours of injection, indicative of an immediate hypersensitivity reaction to the study injection
2. 1% of the safety population and at least two participants experience an SAE judged as related to BCG vaccination by the investigator
3. 20% of participants and at least ten participants experience a Grade 3 or higher event related to BCG vaccination, as judged by the investigator, with the exception of expected local BCG reactions or local injection site reactions that decrease to < Grade 3 within 48 hours.

If an investigator observes that pausing rule 1 or 2 is met, the investigator will inform the sponsor or delegate as soon as possible and within 24 hours of the observation. The sponsor or delegate will notify all study investigators and the IDMC members of the pause in enrollment as soon as possible and within 24 hours of receiving notification of the condition being met. The IDMC members will review all relevant safety data, convene an urgent ad hoc review meeting and make a recommendation to the sponsor with regards to maintaining the pause in enrollment or resuming enrollment.

If, during one of its scheduled or ad-hoc meetings, the IDMC observes that a pausing rule is met, the IDMC Chair or delegate will inform the sponsor as soon as possible and within 24 hours of the identification of the condition being met. If the IDMC has sufficient information to recommend that enrollment resume, this can be communicated to the sponsor at the same time. The sponsor or delegate will notify all study investigators of the pause in enrollment as soon as possible and within 24 hours of receiving notification from the IDMC.

If the sponsor or delegate observes that a pausing is met, the sponsor will notify all study investigators and the IDMC members of the pause in enrollment as soon as possible and within

24 hours. The IDMC members will review all relevant safety data, convene an urgent ad hoc review meeting and make a recommendation to the sponsor with regards to lifting or maintaining the pause in enrollment.

The IDMC may recommend resumption of enrollment with or without changes to the protocol. The final decision to pause or resume study activities will always be the responsibility of the sponsor. All IDMC recommendations will be stored according to the IDMC Charter.

All sponsor decisions will be documented in a memorandum to the study file. The sponsor or delegate is responsible for prompt communication to all relevant study sites of decisions related to pausing or resuming the study activities, including notification to the PI, relevant IRBs/IECs and regulatory authorities.

The clinical sites will be allowed to resume activities only upon receipt of written notification from the sponsor.

7.2. Participant Discontinuation/Withdrawal from the Study

A participant may withdraw (or a participant's parent or guardian (a guardian is a legally acceptable representative [LAR]) may withdraw the participant) from the study at any time or may be withdrawn at any time at the discretion of the investigator for safety, behavioral, compliance, or administrative reasons. This is expected to be uncommon.

If possible, a discontinuation visit should be scheduled for any participant who wishes to discontinue or withdraw from the study. At this visit, topics around subject safety as well as the use of already collected biospecimens will be discussed.

The time and reason for withdrawal should be noted in the space provided for this purpose in the CRF. Participants who are withdrawn because of occurrence of AE should be clearly distinguished from participants who are withdrawn for other reasons. Participants who are withdrawn will be followed up for an AE until the event resolves or stabilizes.

Refer to Appendix 1, Section 10.1.4 regarding the informed consent process.

If a participant withdraws from the study, he/she may request destruction of any samples taken and not tested, and the investigator must document this in the site study records and the CRF.

A participant may, at the investigator's discretion, be withdrawn from the study if any one of the below conditions apply. If not withdrawn, participants with the below conditions will be followed as described in the protocol, including all required blood draws.

- Development of active TB or receipt of curative anti-tuberculous therapy
- Development of autoimmune disease or immunosuppression, or an AE of special interest
- Receipt of investigational drug therapy or investigational vaccine (other than study injection)

No replacements are planned for the withdrawals in the study.

7.3. Lost to Follow-up

A participant will be considered lost to follow-up if he or she repeatedly fails to return for scheduled visits (3 failed visits) and is unable to be contacted by the study site (3 failed attempts per failed visit).

The following actions must be taken if a participant fails to return to the clinic for a required study visit.

The site must attempt to contact the participant and reschedule the missed visit as soon as possible and counsel the participant on the importance of maintaining the assigned visit schedule and ascertain whether or not the participant wishes to and/or should continue in the study.

Before a participant is deemed lost to follow up, the investigator or designee must make every effort to regain contact with the participant (where possible, telephone calls and, if necessary, a home visit by a member of the study team). These contact attempts should be documented in the participant's medical record and the CRF.

Should the participant continue to be unreachable, he/she will be considered lost to follow-up from the study.

7.4. COVID-19 Contingency Plans

7.4.1. Rationale and Plan

The Coronavirus Disease 2019 (COVID-19) pandemic has impacted the world since the beginning of 2020, severely affecting people and societies around the globe.

This study was paused for enrollment by the sponsor, effective 19 March 2020, in order to support social distancing and to help health care professionals address COVID-19.

At the time of the enrollment pause, 349 participants had received investigational product (BCG or placebo) and were on-study.

The Government of South Africa declared a National Disaster on 15 March 2020 and ordered a first lockdown (stay-at-home order) on 26 March 2020. Under the lockdown, most trial participants are not able to come to the investigational site for protocol-specified visits. The sponsor and investigators therefore needed to change study procedures and agreed on telephone contacts as an alternative to study visits for safety assessments, for the duration of the Government restrictions, and/or risks that prevent in-person study visits, as judged by the site PI.

In accordance with the SAHPRA Policy on Conduct of Clinical Trials of Health Products during the Current COVID-19 Pandemic, issued 25 March 2020, this protocol amendment reflects the new and modified processes that were put in place in response to COVID-19. This protocol amendment, therefore, accommodates the changes related to the pandemic that needed to take place in order to continue the study in a manner that minimizes any immediate hazards, and also allows for more flexibility to capture study endpoints, and adds an assessment of SARS-CoV-2 infection and COVID-19 disease.

Changes in study visit procedures, missed visits, and participant discontinuations may lead to missing information (e.g., for protocol-specified procedures). We will capture as much information as possible in the case report form and explain the basis of the missing data.

Approaches used to protect trial participants and to manage study conduct during the COVID-19 pandemic, as well as control measures at study sites, will be documented.

Protocol deviations due to the COVID-19 pandemic will be described in the clinical study report (or in a separate study-specific document) including but not limited to the following:

- Contingency measures implemented to manage study conduct during disruption of the study as a result of COVID-19 control measures.
- A listing of all participants affected by the COVID-19 related study disruption by unique participant number identifier and by investigational site, and a description of how the individual's participation was altered.
- Analyses and corresponding discussions that address the impact of implemented contingency measures (e.g., alternative procedures used to collect critical safety and/or efficacy data) on the safety and efficacy results reported for the study.

Study aspects affected by the study pause due to the COVID-19 pandemic are addressed in the following subsections.

7.4.2. Enrollment of new participants and status of previously enrolled participants

Enrollment was paused by the sponsor effective 19 March 2020 due to COVID-19 and will only resume once IRBs, investigators and the Sponsor agree that it is acceptable to resume study enrollment.

Many of the 349 participants on-study at the time of the lockdown will miss study visits and blood draws. A subset of these participants may not be eligible for inclusion in the primary endpoint analyses. In order to meet the primary study objective, the final sample size will be determined and documented once enrollment has resumed and we can understand the impact of the pause on the ability to address the primary objectives. The total randomized population may increase to a maximum of 2150 participants.

Participants who were screened but not randomized because the study was paused prior to randomization will be offered re-screening when enrollment resumes. Re-screening will also be allowed for any participant who screen-fails due to potential future COVID-19-related restrictions to movement or enrollment.

Participants who are offered re-screening due to COVID-19 will be assigned a new participant identifier (PID), and the new PID will be linked to the previously used PID to avoid double counting of the total number and percent failure of screened participants. Information collected from the re-screening will become the source information to be used to determine eligibility.

Extension of the estimated duration of enrollment (and therefore the overall estimated duration of the trial) will be required since the study was paused.

7.4.3. Study visits for enrolled subjects

The preference for study follow-up is to maintain in person study visits when it is acceptable and feasible to do so. Alternatively, follow-up visits can be performed by phone or another type of remote contact and will be captured in a consistent manner within the CRFs.

When participants are able to return to the site for onsite visits, they may be asked to complete visit assessments and procedures at a time point that is not specified in the protocol, in order to collect missing data and to perform missed procedures (e.g., blood draw or diary card collection). Once in-person visits become feasible, investigators will need to assess what visit the participant should be assigned to, based on guidelines in Table 7.

Refer to Section 7.4.5, regarding impact on mITT cohort for efficacy.

Table 7: Guidance for visits missed due to COVID-19-related restrictions, once in-person study visits become feasible

Screening visit	Re-screening will be allowed if participants were unable to attend the Day 1 study visit due to COVID-19 restrictions or if they were previously screened but not randomized before the study was paused due to the COVID-19 pandemic.
Day 8 visit	If the interval between Day 1 and Day 8 visits is longer than 11 days, the PI will decide whether to invite the participant for a late Day 8 visit or wait until the date of a later scheduled visit (e.g., Day 29, Day 71, or Month 6, etc.). Participants should be seen sooner rather than later, to ensure appropriate safety follow-up. At that visit, investigators will collect data for the missed Day 8 visit, if possible. For example, the diary card should be collected, and data entered into the Day 8 visit CRF form with the actual diary card collection date.
Day 29 visit	If the interval between Day 1 and Day 29 visit is longer than 35 days, the PI will decide whether to invite the participant for a late Day 29 visit or wait until the date of a later scheduled visit (e.g., Day 71, Month 6, etc.). Participants should be seen sooner rather than later, to ensure appropriate safety follow-up. At that visit, investigators will collect data for the missed Day 29 visit, if possible. For example, the memory aid should be collected, and data entered into the Day 29 visit CRF form with the actual memory aid collection date. The Day 29 blood draw should not be performed if the visit occurs after Day 35.
Day 71 visit	If the Day 71 visit was missed, **the visit will be scheduled as soon as possible, in order to obtain the blood sample that determines eligibility of the mITT cohort** (see Section 7.4.5)**.** The Day 71 blood draw should be performed, regardless of when the Day 71 visit occurs.
Month 6 visit and later visits	If the Month 6 visit or later visits are missed, the participant should be seen soon after in-person study visits become feasible. At that visit, investigators will collect data for missed visits, if possible.

7.4.4. Addition of exploratory objectives and endpoints related to COVID-19

The following exploratory objectives were added:

- To describe COVID-19 among study participants who present with suspected COVID-19
- To describe the serostatus of SARS-CoV-2 infection.

7.4.4.1. Serology for COVID-19

In order to describe the penetrance of SARS-CoV-2 infection in the communities that participate in this study, SARS-CoV-2 serology will be performed once every six months for a duration of 24 months. For enrolled participants, serology will be done on a blood sample drawn at the earliest possible study visit (e.g., the Month 6 or Month 12 visit, etc.) and at four subsequent biannual study visits.

For participants who are screened after SAHPRA and ethics committee approval of the Gates MRI-TBV01-201 Amendment Version 5, a blood sample will be collected at the screening visit, and at the Month 6, 12, 18 and Month 24 visits.

7.4.4.2. Diagnostic test for SARS-CoV-2

In order to describe signs and symptoms of COVID-19 among the study population, a diagnostic test for SARS-CoV-2 (based on nucleic acid amplification) will be performed if a participant presents with suspected COVID-19 (e.g., respiratory or febrile illness, anosmia, etc.). Signs and symptoms of illness are described as per investigator assessment (e.g., temperature, respiratory rate, oxygen saturation, medication, etc.).

A nasal or oropharyngeal sample will be obtained (e.g., swab, brush, scraping, or saliva) for the COVID-19 diagnostic test.

7.4.5. Changes to the definition of the mITT cohort for efficacy

Protocol version 4 (prior to COVID-19) defined a 70-day "wash-out" period to identify participants who may have been infected with *Mtb* just prior to or soon after enrollment and stated that participants who convert to QFT positive at Day 71 would be excluded from the primary mITT efficacy analysis.

Because of COVID-19, most of the study participants enrolled less than 70 days prior to the pause on March 19, 2020, missed or will miss their Day 71 visit. The amended protocol, Version 5, defines that participants who missed their Day 71 visit may be eligible for inclusion in the mITT cohort if they test QFT-negative, and are excluded from the mITT cohort if they test QFT-positive, at the first study visit after the COVID-19 related pause is lifted (e.g., late Day 71 visit or Month 6 visit). The mITT analysis will take the duration of follow-up after the QFT-negative test into consideration for the primary analysis.

The impact on data eligibility for certain protocol-defined analyses (e.g., mITT analysis) will be addressed in the Statistical Analysis Plan (SAP). Clear rules for how data obtained during out-of-window study visits will be used in endpoint analyses will be defined in the SAP.

7.4.6. Protocol deviations related to COVID-19

Tracking of all protocol deviations related to COVID-19 will be done proactively by sites and all parties involved. Deviations may include delayed participant visits, missed assessments, etc. Site staff will clearly document all COVID-19-related deviations in detail in participant source documentation, as well as in annotations in the EDC system, to ensure clarity for statistical analyses and clinical study reporting.

7.4.7. Informed consent and re-consent

Participants who were enrolled into the study will be re-consented/assented into the study so that they are aware of the protocol changes.

Newly screened participants will be asked to sign the current consent/assent form which captures applicable protocol changes.

7.4.8. Safety precautions for trial staff after enrollment resumes

Principal Investigators and site personnel have a critical role in the conduct of the study to ensure safety of the trial participants and site staff. The sponsor, clinical research organizations, and principal investigators should adhere to applicable guidelines on measures to control transmission of COVID-19. Investigators and site staff should develop and implement site-specific procedures and processes as appropriate.

7.4.9. References

South African Clinical Research Association (SACRA) Clinical Trials Suggested Business Continuity Plan_COVID-19_Version 1; 16 March 2020.

SAHPRA Policy on Conduct of Clinical Trials of Health Products During the Current COVID-19 Pandemic; 25 March 2020 (adapted from: FDA's Guidance on Conduct of Clinical Trials of Medicinal Products During the COVID-19 Pandemic;18 March 2020).

8. Study Assessments and Procedures

Study procedures and their timing are summarized in the SoA. Protocol waivers or exemptions are not allowed.

Prior to any study procedure, all eligible participants will be assigned a unique participant identifier. This participant identifier will be used throughout the study for participant identification.

Screening for eligibility assessment will occur after informed consent, and as applicable, assent, is obtained. Screening assessments can be done at any time during the period, except for the written informed consent, which must be completed prior to any screening procedure. Eligibility for randomization will be based on the inclusion and exclusion criteria described in Sections 5.1 and 5.2.

Eligibility criteria will be checked during the screening process and prior to vaccine/placebo administration to ensure that all participants enrolled meet all of the inclusion criteria and none

of the exclusion criteria. The investigator will maintain a screening log to record details of all participants screened and to confirm eligibility or record reasons for screening failure, as applicable.

To evaluate eligibility criteria, medical history, physical examination including vital signs, and height and weight will be performed. In addition, safety laboratory tests (hematology, chemistry, and urinalysis) as well as a QFT TB test, HIV test, and a serum pregnancy test (females only) will be performed.

Repeat collection and testing

Note that if a blood sample collected for screening cannot be evaluated (e.g., insufficient volume; damaged, hemolytic or lost sample, etc.) a second (repeat) blood collection is permitted to enable evaluation of study eligibility.

The repeat blood collection must occur within the protocol-defined screening visit window. If a screening blood sample was successfully run and has a reported result to determine eligibility, a second (repeat) blood collection is not permitted.

A second sample for urinalysis may also be repeated if the screening sample is unevaluable or if it is abnormal due to menstruation or suspected urinary tract infection. Urinary tract infection should be resolved prior to the repeat urinalysis and the repeat test must be performed during the screening interval. If repeat urinalysis is abnormal, participant will be a screen failure.

In addition, if the blood sample for QFT collected at any time point after screening is unevaluable, collection of a second (repeat) blood sample is also permitted to enable evaluation of the primary efficacy endpoint. The repeat blood collection must occur within the protocol-defined visit window.

If the QFT sample was successfully run and has a reported result, including “indeterminate”, collection of a second (repeat) blood sample is not permitted.

Details regarding these procedures are provided in the subsections to follow.

If a planned study visit or planned study procedure (e.g., blood draw) cannot be performed because the participant has an acute illness or cannot access the study site (refer to Section 7.4 for COVID-19 related restrictions), the missed visit should be rescheduled as soon as feasible. Unscheduled visits should only take place as necessary and data from unscheduled study visits should be captured in the EDC.

The investigator must document confirmation of eligibility prior to randomization.

8.1. Efficacy Assessments

8.1.1. Evaluation of initial *Mtb* infection and sustained infection

Mtb infection is defined for this protocol as QFT conversion from a negative to positive test, as per the manufacturer’s QFT-GIT package insert.

Sustained *Mtb* infection is defined for this protocol as sustained QFT conversion from a negative to positive test, with initial conversion at any time after a first negative QFT result, post

randomization (Day 71 or subsequent visit if Day 71 result is not available), and remaining QFT-positive at 3- and 6-months PC.

QFT assays will be performed on blood samples collected from all participants at screening, at the Day 71 visit (end of the wash-out period), and every 6 months (counting from Day 1) through Month 48. In addition, participants who have QFT conversion will have one additional QFT assay done three months PC.

An additional exploratory endpoint will be to evaluate alternative threshold values for QFT conversion.

Details on these endpoint cut-off values will be included in the statistical analysis plan.

8.2. Safety Assessments

Planned time points for all safety assessments are provided in the SoA.

Safety outcomes will include:

- Solicited AEs (including local injection site pain, redness, and swelling, and general AEs (headache, fatigue, gastrointestinal symptoms, and fever).
- Unsolicited AEs
- All SAEs
- Vaccination-related SAEs (serious ADRs).

AEs will be collected from the time informed consent is obtained.

Refer to Appendix 2, Section 10.2 for details regarding AEs and SAEs.

Immediate safety concerns should be discussed with the sponsor immediately upon occurrence or awareness.

Adherence to the study protocol, including those specified in the SoA, is essential and required for study conduct.

8.2.1. Physical Examination and Medical History

A medical history and physical examination will be conducted at screening to assess enrollment eligibility. Medical history will include questions regarding acute respiratory illness, febrile illness, and anosmia, e.g., due to SARS-CoV-2, influenza, or other respiratory agents.

Only participants that are considered as healthy by the investigator will be enrolled.

Medical conditions that exist prior to the screening visit will be recorded in the medical history.

Day to day fluctuations in these conditions that do not represent a clinically significant change in the participant's status will not necessarily be reported as AEs.

Physical examination will include, at a minimum, assessment of height and weight, body temperature, and resting vital signs (blood pressure, pulse, and respiratory rate). Vital signs will be measured after at least 5 minutes of rest in a quiet setting without distractions.

Height and body weight will be measured at screening, Day 1, and every six months thereafter.

8.2.1.1. Interim History and Focused Physical Examination

An interim history will be taken on Day 1 and all subsequent visits, as indicated in the SoA. A focused physical examination will be performed if indicated by interval history. Interim medical history will also include questions regarding acute respiratory illness, febrile illness, anosmia, etc, e.g., due to SARS-CoV-2, influenza, or other respiratory agents.

8.2.2. Pregnancy Status Assessment and Follow-Up

A serum βHCG test will be performed at screening, and a urine βHCG test will be performed at Day 1 (prior to enrollment) and any time during the study if a pregnancy is suspected.

During the study, participants will be asked about pregnancy at each time point indicated in the SoA (Section 1.3). If a pregnancy is reported, the investigator should inform the Medical Monitor within 24 hours of learning of the pregnancy and should follow the procedures outlined in Appendix 3, Section 10.3.

If a participant becomes pregnant during the study, she may continue in the study at the investigator's discretion. If she continues study participation, she will be followed for all safety assessments, but no blood will be collected for the study during pregnancy. Once the pregnancy has ended, only blood draws for QFT and HIV tests are allowed.

A participant who becomes pregnant during the study will be followed during the gestation period, and after the delivery to collect information on the health of the participant and the newborn. Details of the pregnancy will be collected (refer to Section 10.3, Collection and Reporting Pregnancy Information). If delivery occurs after the final study visit, the investigator should attempt to maintain contact with the participant to obtain the information.

8.2.3. Pre- and Post-Study Intervention Safety Monitoring

On Day 1, prior to vaccine/placebo administration, vital signs will be taken. Participants will remain under observation for at least 30 minutes after receiving the injection. Allergic reactions to vaccination are possible, therefore, appropriate drugs and medical equipment to treat acute anaphylactic reactions must be immediately available and a physician trained to recognize and treat anaphylaxis must be present in the clinic during vaccination and the post-vaccination monitoring period.

8.2.4. Diary Card and Daily Temperature Monitoring

On Day 1, participants will be given a diary card and receive guidance on how to fill in the card. Participants will also receive a digital thermometer.

The diary card will be used by the study participants or parent or guardian/LAR to record the duration and intensity (Grade 1, 2, 3, or 4) of solicited local and general AEs for 7 days following vaccination. The diary card will also allow for unsolicited AEs to be recorded.

The diary card will be collected and reviewed by the PI (or designee) on Day 8. No changes to the diary card will be permitted; however, any verbally recalled information provided by the participant or parent or guardian/LAR during review of the diary card will be documented in the source document and reported as an AE, as applicable. Any participant-reported Grade 4 event will be assessed by the investigator.

A memory aid will be distributed to the participant at the Day 8 visit and the participant will be instructed to record unsolicited adverse events using the aid. The memory aid will be collected at the Day 29 visit.

8.2.5. Clinical Safety Laboratory Assessments

Safety laboratory assessments will be performed at screening. Refer to the SoA Section Table 2.

Laboratory values from the most recent blood sample collected prior to randomization outside the normal range that are suggestive of a disease state (ie, values greater than Grade 1) will lead to exclusion from study enrollment, with the exception of any grade hematuria in a menstruating female, or a urinalysis abnormality judged not clinically significant by the investigator.

Refer to Appendix 5, Section 10.5 for toxicity table for grading for each laboratory test.

Grade 1 abnormalities will not lead to exclusion if the investigator does not consider them clinically significant.

Clinical safety laboratory parameters that will be evaluated at screening include:

- Hematology: Complete blood count (hemoglobin, platelets and white blood cells) and absolute counts for neutrophils, lymphocytes, and monocytes
- Serum chemistry: ALT, AST, alkaline phosphatase, total bilirubin, creatinine, blood urea nitrogen (BUN)
- Urinalysis: Specific gravity, pH, glucose, protein, blood, ketones, bilirubin, urobilinogen, nitrite, leukocyte esterase by dipstick.

Refer to ***Repeat Collection and Testing*** in Section 8 for unevaluable blood samples.

Abnormal results and findings that make the participant ineligible will be discussed with the participant and the participant will be referred for follow-up care with their healthcare provider if necessary.

All screening laboratory specimens will be processed according to laboratory SOPs available from the clinical laboratory(ies) designated for the study. Information about the laboratory(ies), including any instructions for performing and interpreting specific tests, will be maintained in the investigator's study files.

The investigator must review the laboratory report, document this review, and record any clinically relevant changes occurring during the study in the AE section of the CRF. The laboratory reports must be filed with the source documents. Clinically significant abnormal laboratory findings are those which are not associated with the underlying disease, unless judged by the investigator to be more severe than expected for the participant's condition.

All protocol-required laboratory assessments must be conducted in accordance with the laboratory manual and the SoA.

If laboratory values from non-protocol specified laboratory assessments require a change in participant management or are considered clinically significant by the investigator (eg, AE or SAE), then the results must be recorded in the CRF.

All protocol-required safety laboratory tests will be performed by the central laboratory, Bio Analytical Research Corporation South Africa (BARC SA or similar).

Additional tests may be performed at any time during the study as determined necessary by the investigator or required by local regulations.

8.2.6. Injection Site Examination

A local reaction is normal after BCG. Site of injection examinations will be performed from Day 8 through Month 6 to evaluate the observed local reactions. As stated in the BCG SSI vaccine package insert (South Africa):

- A small tender red swelling appears at the site of the injection which gradually changes to a small vesicle. An ulcer may appear 2 to 4 weeks after vaccination. An adherent dressing is not recommended.
- The reaction usually subsides within two to five months and in practically all children leaves a superficial scar 2 – 10 mm in diameter.
- Transient enlargement of the regional lymph nodes (< 1 cm) is normal.
- Enlargement of axillary lymph nodes may be seen less frequently in the months following immunisation. Suppurative lymphadenitis may occur less frequently. This is a benign condition which heals spontaneously, although often only slowly.

The presence and maximum diameter of injection site reactions will be recorded as follows:

- Redness, swelling, or induration ≤20 mm is considered as anticipated.
- Redness, swelling, or induration >20 mm is reported as an AE.
- Papule, vesicle, pustule, ulcer, or scar ≤5 mm is considered as anticipated.
- Papule, vesicle, pustule, ulcer, or scar >5 mm is reported as an AE.

The presence and maximum diameter of homolateral axillary lymph nodes larger than 1 cm will be recorded.

8.2.7. Participant Follow-Up

Participants will be instructed to contact a study team member to report new or worsening AEs, as well as new diagnoses, and to come to the study clinic if medical attention is needed.

For emergencies and other unscheduled visits to a medical facility other than the study clinic, medical records will, to the extent possible, be obtained by the investigator.

For participants who attend school, study visits will be scheduled outside of school hours whenever possible. Participants will be asked about the occurrence of AEs and SAEs, concomitant prescription medications/vaccinations, and change in general health status and any other change in status that may affect the participant's participation, as indicated in the SoA.

An HIV blood sample will be collected for HIV testing at screening and again yearly, with pre- and post-test HIV counseling. HIV counseling will be performed by study staff. In addition, any participant suspected of having TB disease (see Section 8.3.6) will be tested for HIV at the time of diagnosis or as soon as possible following suspicion of TB disease.

All deviations from protocol procedures, evaluations, and/or visits will be documented.

8.3. Adverse Events and Serious Adverse Events

The definitions of AEs, SAEs, serious ADRs, SUSARs, and adverse events of special interest (AESIs) can be found in Appendix 2, Section 10.2.1, and 10.2.2, 10.2.3, 10.2.4, respectively.

AEs will be reported by the participant (or, as appropriate, the participant's legally authorized representative). Biochemistry and/or hematology findings may also qualify as AEs if the investigator considers them as such.

SAEs that the investigator assessed as related to vaccination will be collected throughout the study.

Study nurses and physicians are responsible for collecting and documenting information and events that would potentially meet the definition of an AE. However only the investigator (study physician) is responsible for assessment, including assignment of causality and intensity, reporting and management of all AEs. The investigator is responsible for following up AEs that are serious, considered related to the study intervention or study procedures, or that caused the participant to discontinue the study (Section 7).

8.3.1. Time Period and Frequency for Collecting AE and SAE Information

Type of Event	Collection Time Period
All solicited AEs	Day 1 through Day 7 (inclusive)
Unsolicited AEs	Screening through Day 28 (inclusive)*
All SAEs and AESIs	Screening through Month 6*
Serious ADRs	Day 1 through the end of the study

*Refer to the SoA, Section 1.3 for collection of AEs from time of signed consent.

Distribution and collection days for diary cards are shown in the SoA (Section 1.3).

Medical conditions that begin before the start of study intervention but after obtaining informed consent will be recorded on the Medical History/Current Medical Conditions section of the CRF, not the AE section.

All SAEs and AESIs will be reported to the sponsor or designee within 24 hours, as indicated in Appendix 2, Section 10.2.6. The investigator will submit any updated SAE data to the sponsor or designees within 24 hours of it being available.

Investigators are not obligated to actively seek AEs or SAEs after conclusion of study participation. However, if the investigator learns of any SAE, including a death, at any time after a participant has been discharged from the study, and he/she considers the event to be related to the study intervention or study participation, the investigator must notify the sponsor or designee within the same 24 hours timeline.

8.3.2. Method of Detecting AEs and SAEs

The methods of recording and follow-up of AEs and SAEs are provided in detail in Appendix 2, Section 10.2.5, which includes assessments of intensity, causality, expectedness and outcome of AEs and SAE.

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading questions are the preferred method to inquire about AE occurrences.

8.3.3. Follow-up of AEs

After the initial AE/SAE report, the investigator is required to proactively follow each participant at subsequent visits/contacts. All SAEs will be followed until resolution, stabilization, or the participant is lost to follow-up (as defined in Section 7.3). Refer to Appendix 2, Section 10.2.5.

8.3.3.1. AE Intensity

AEs will be classified by the investigator as mild (Grade 1, not interfering with normal daily activities), moderate (Grade 2, interfering with normal daily activities), severe (Grade 3, preventing normal daily activities) or potentially life-threatening (Grade 4). Refer to Appendix 2, Section 10.2.5.1 and Appendix 5 for details. Note that a body temperature between 37.5°C and 37.9°C, can be captured as an AE, at the PI's discretion.

8.3.3.2. AE Causality

All AEs will be evaluated by the PI or medically qualified designee (ie, investigator, study physician) to assess the relationship between study intervention and AE. Careful medical judgement should be exercised to determine the level of causal relationship between an AE and the study intervention. The causality will be assessed as related or not related.

The sponsor or designee will have the opportunity to confirm the seriousness and case causality based on the clinical judgement of the Medical Monitor and sponsor designee. If a serious adverse event is considered unrelated by the investigator but the sponsor believes that there is a reasonable possibility that the event is related, the sponsor will upgrade the case to a 'related' status. The sponsor or designee will never downgrade a case from serious to non-serious, or related to not related.

Refer to Appendix 2, Section 10.2.5.2 for details. Expectedness of AEs is defined in Appendix 2, Section 10.2.5.3.

8.3.3.3. AE Resolution

All AEs/SAEs must be followed until resolution. Refer to Appendix 2, Section 10.2.5.4 for details.

8.3.4. Regulatory Reporting Requirements for SAEs

Refer to Appendix 2, Section 10.2.6 for details regarding SAE reporting.

For the purposes of regulatory reporting, this study will be conducted like a post-marketing study, as the product used is a marketed product approved in the country.

Prompt notification by the investigator to the sponsor of a SAE is essential so that legal obligations and ethical responsibilities towards the safety of participants and the safety of a study intervention under clinical investigation are met.

The sponsor or delegate has a legal responsibility to notify both the local regulatory authority and potentially other regulatory agencies about the safety of the study intervention under clinical investigation. The sponsor will comply with country-specific regulatory requirements relating to safety reporting to the regulatory authority, IRBs/IECs, and investigators.

All fatal and life-threatening SUSARs are to be reported to South African Health Products Regulatory Authority (SAHPRA) within 7 calendar days after first knowledge with a complete report to be submitted within an additional 8 calendar days. SUSARs that are not fatal or life threatening need to be reported to SAHPRA no later than 15 calendar days after first knowledge. An investigator who receives a SUSAR report or other specific safety information (e.g., summary or listing of SAEs) from the sponsor will review and file it. A tracker of all such reports received from the CRO will be maintained by the site. The PI will notify the IRB/IEC, if appropriate and according to local requirements. IRB/IEC submissions will be conducted per the local IRB/IEC SOP.

The sponsor will prepare, distribute and submit SUSAR reports according to local regulatory requirements. The CRO, on behalf of the sponsor will submit the SUSAR reports to SAHPRA.

Refer to Appendix 2, Section 10.2.6 for details regarding SAE reporting.

8.3.5. Death Events

Any untoward medical occurrence resulting in death is reported as an SAE. The cause of death will be appropriately documented in the SAE report form and supporting evidence will be provided.

8.3.6. Latent TB infection and TB disease

Participants in this trial will be managed according to current national and/or international guidelines, in accordance with ethical standards. A recent international guideline (*Latent tuberculosis infection. Updated and consolidated guidelines for programmatic management.* [WHO, 2018b]) recommends that in countries with a high TB incidence, children aged ≥ 5 years, adolescents and adults who are household contacts of people with bacteriologically confirmed pulmonary TB who are found not to have active TB may be given preventive therapy (*Conditional recommendation, low-quality evidence. New recommendation*). Household contact is used in this context as a proxy for possible *Mtb* infection, and associated risk for progression to TB disease, in countries where testing for infection is not routine.

This trial will test adolescent participants for sustained *Mtb* infection using the QFT assay. Since it would be illogical to offer preventive therapy based on household contact status when serial QFT results are available, all participants with sustained QFT conversion will either be referred for TB preventive therapy (TPT) regardless of whether or not they are household contacts of a confirmed TB case, or be offered TPT by the site. Participants with transient QFT conversion, i.e., participants who's initial QFT positive result reverts to negative within 6 months, will not be referred for preventive therapy.

All participants who are QFT positive (from screening throughout study period) will be educated on the symptoms of TB and the benefits of early diagnosis and treatment.

Active surveillance for signs and symptoms compatible with incident TB disease, e.g., cough, fever, fatigue or night sweats, for longer than two weeks, or loss of weight / insufficient weight gain or growth, will continue throughout the study.

Participants with suspected TB disease will be tested for HIV infection and will be asked to provide two sputum samples or other appropriate samples for smear microscopy, mycobacterial culture, and GeneXpert.

Participants with a microbiological, radiological, and/or clinical diagnosis of TB disease will be referred to the NTP for TB treatment which is provided free of charge by the NTP.

A bacteriologically confirmed case of active TB disease will be defined as clinical evidence of TB with positive *Mtb* mycobacteria growth indicator tube culture, or GeneXpert *MTB*/rifampicin (Cepheid, USA), from sputum or an extra-pulmonary site.

A clinical case of active TB disease will be defined as clinical and/or radiographic evidence of active TB disease that is not bacteriologically confirmed.

8.4. Treatment of Overdose

If a dosing error occurs, the investigator, safety physician and medical monitor need to be informed as soon as possible.

8.5. Pharmacokinetics

PK parameters are not evaluated in this vaccine study.

8.6. Pharmacodynamics

Pharmacodynamic parameters (other than characterization of the immune response to vaccination) are not evaluated in this vaccine study.

8.7. Genetics

Germline genetic information may be assessed to aid in the interpretation of study results related to the host response to BCG vaccination and to *Mtb* infection. Germline genetic analyses may include HLA typing, Fcγ receptor and NK cell genotyping, and assessment of other genetic information that may influence vaccine-induced and infection-induced responses, as well as susceptibility to infectious diseases. Germline genetic information will not be used for analyses related to paternity, ancestry or hereditary diseases. See Appendix 4, Section 10.4 regarding information for use and analysis of DNA samples.

8.8. Biomarkers

Collection and analyses of blood samples for biomarker research and identification of potential correlates of risk for and correlates of protection from sustained *Mtb* infection is an essential part of this study. Analytic approaches for the exploratory endpoints below will be informed by best practice and the most recent advances in biomarker discovery and systems biology.

Exploratory analyses of host characteristics, as well as host responses following vaccination, that correlate with the risk of transient and/or sustained QFT conversion will be conducted. Specific objectives and hypotheses related to these analyses will be documented and reported in a separate scientific and statistical analysis plan, prior to full unblinding of data associated with these exploratory analyses.

Exploratory endpoints may include proteomics, antibody analyses and cellular analyses (eg, NK cells, B cells, T cells), as well as candidate CoP and CoR identified in other studies.

PBMC, plasma, serum and whole blood samples will be collected at time points likely to show peak responses for cellular, humoral, and transcriptomic responses, respectively. Refer to SoA, Section 1.3 for specific time points and volumes for collection of samples. If an effect of vaccination is seen on infection with *Mtb*, immunologic assays may be conducted to search for correlates of risk or protection. Functional assays such as mycobacterial growth inhibition may also be conducted.

Transcriptome studies may be conducted using RNA sequencing or comparable techniques, which facilitate the simultaneous measurement of the relative abundances of thousands of RNA species resulting in a transcriptome profile for each blood sample. This will enable the evaluation of changes in transcriptome profiles that may correlate with biological response relating the prevention of initial and sustained QFT conversion.

Biomarkers studies using DNA-sequencing technologies such as Assay for Transposase-Accessible Chromatin with high throughput Sequencing (ATAC-seq) may be used to identify accessible chromatin sequences from small cell numbers, e.g., PBMCs, to generate epigenomic profiles of immune cells. Histone modification and other epigenetic modifications that are potentially affected by vaccination and/or *Mtb* infection may be analyzed. These genetic and

epigenetic studies aim to identify correlates of risk and correlates of protection for *Mtb* infection, for sustained *Mtb* infection, and TB disease. If any CoRs or CoPs are identified in other studies, validity of such correlates may be tested using data from this this study.

Plasma and/or serum may be analyzed for soluble immune mediators by quantification using multiplex assays.

Samples will be stored according to local regulations at a facility selected by the sponsor.

Collected samples will be used for protocol related biomarker research. In addition, only with the explicit and optional consent of study participants, samples or partial sample volumes that remain once a protocol-defined assay has been completed, may be stored and used for purposes other than protocol-related endpoints. Such "future research studies" would include assay development, assay quality control and development of methods related to tuberculosis research and TB vaccine research, including studies that pertain to the improvement, development and quality assurance of the lab tests described in this protocol. Such future research is subject to laws and regulations of South Africa and is subject to IRB/IEC approval.

Refer to Appendix 4, Section 10.4 regarding use/analysis of DNA.

8.8.1. Immunological Assessments

Immunological outcomes may include ICS on PBMC and whole blood, phenotyping of cell subsets by flow cytometry, transcriptomics and genetic or epigenetics studies using RNA and/or DNA extracted from whole blood or white blood cells, and quantification of soluble immune mediators by multiplex assays.

The assessment of immune responses to vaccine may include the frequency of *Mtb*-, BCG- and/or NTM-specific CD4 T cells positive for markers such as IFN-γ, TNF-α, IL-2 and/or IL-22. Responses will be measured by flow cytometry in the ICS assay.

Antibody titers against BCG may also be measured using ELISA, multiplex assays or functional assays (eg, mycobacterial killing assays etc).

8.8.2. Subset Assessment

For a subset of the first 80 10- to 12-year-old participants enrolled at the SATVI site, ICS will be performed directly *ex vivo* after stimulation of whole blood. Site staff will use the Study Reference Manual (provided separately) for further instructions and additional information on specimen collection and processing. All blood volumes in the table are approximate.

Note that the type of assays performed will be based on the most current information available at the time of analysis.

8.9. Health Economics

This section is not applicable.

9. Statistical Considerations

9.1. Statistical Hypotheses

Primary: Revaccination with BCG reduces the risk of sustained *Mtb* infection versus placebo in previously BCG vaccinated, QFT negative healthy adolescents.

Secondary:

- Revaccination with BCG reduces the risk of sustained *Mtb* infection versus placebo in previously BCG vaccinated, QFT negative healthy adolescents, with VE durability that lasts at least 36 months in all participants.

- Revaccination with BCG reduces the risk of sustained *Mtb* infection versus placebo in previously BCG vaccinated, QFT negative healthy adolescents, with VE durability that lasts 48 months in all participants.

9.2. Sample Size Determination

The primary analysis will be triggered when the number of required events to achieve 90% power with a 1-sided alpha of 2.5% has been accrued. If the true vaccine efficacy is 45%, at least 118 sustained QFT conversion events are required for the primary analysis. Additional sample size considerations were based on the following assumptions: It will take one year to fully enroll the study, 7% of enrolled participants will be ineligible for the mITT efficacy population due to a positive QFT assay result at Day 71, an expected 8-10% per year will convert to QFT positive in the placebo arm, 70% of initial conversions are expected to remain sustained, 5% on average loss to follow-up per year and 45% VE.

The proposed sample size was determined based on targeting relatively high probability (~80%) of obtaining 118 sustained conversion events within 3.5 years of study start. The probability of obtaining 118 sustained conversion events within 3 and 3.5 years of study start are shown in Table 8 for sample sizes ranging from 600 to 800 in the mITT efficacy population. Due to COVID-19 related restrictions, it will take longer to complete enrollment and to reach the number of events needed for analyses. After accounting for the expected 7% expected to be ineligible for the mITT efficacy population due to a positive QFT assay result at Day 71, the table shows that randomizing approximately 800 participants to each treatment group is sufficient to detect a true VE of at least 45% under the specified assumptions, resulting in ~80% probability to accrue 118 events within 3.5 years of study initiation when the attack rate is as low as 8%. Since it is expected that approximately 12% of the population enrolled will be 10- or 11-year-old adolescents, where there is limited information on the attack rate and potential VE, a further inflation of the required 800 participants per group by 12% resulted in a final proposed sample size of 900 participants per group, or 1800 participants total.

Approximately 6720 participants will be screened to enroll and randomize up to 2150 participants.

Table 8: Sample Size Requirements to Ensure 75%-80% Probability of Performing the Primary Analysis within 3.5 years

N/grp mITT Efficacy Population	Attack rate = 10%		Attack rate = 8%		N/grp Randomized*	N/grp (+12%)**	Final N (Total)
	Prob (analysis in ≤ 3 yrs)	Prob (analysis in ≤ 3.5 yrs)	Prob (analysis in ≤ 3 yrs)	Prob (analysis in ≤ 3.5 yrs)			
600	4.3	75.4	< 0.1	5.4	646	723	1446
650	21.9	94.0	0.2	20.8	699	783	1566
700	50.4	99.4	0.8	52.7	753	844	1688
750	**77.0**	**> 99.9**	**5.3**	**80.2**	**807**	**904**	**1808**
800	93.0	> 99.9	20.5	94.5	861	964	1928

Assumes equal randomization (randomization ratio 1:1)
Probability analysis occurs within 3 and 3.5 years (yrs) based on 2000 simulations, assuming 1 yr accrual, 70% of primary infection remain sustained, 5% annual drop-out rate
* Assumes 7% removed from those randomized due to QFT + result at Day 71
** Assumes additional 12% of participants will be 10 or 11year olds, where we have limited information on attack rate and potential VE

9.3. Populations for Analyses

Analysis populations are shown in Table 9.

Table 9: Populations for Analyses

Population	Description
Modified intention to treat (mITT) efficacy population	All participants randomly assigned to study intervention, who received the study intervention and are QFT negative at the Day 71 visit, or if the Day 71 study visit was missed, or the QFT result was not available, the first study visit post Day 71 for which a QFT result is available. Participants will be analyzed according to the intervention to which they were randomized.
Intention to treat (ITT) efficacy population	All participants randomly assigned to study intervention and who received the study intervention. Participants will be analyzed according to the intervention to which they were randomized.
Per Protocol (PP) efficacy population	All participants in the mITT population who did not substantially deviate from the protocol procedures and who received the treatment to which they were randomized Participants who substantially deviated will be identified prior to database lock and unblinding.
Safety	All participants randomly assigned to study intervention and who received the study intervention. Participants will be analyzed according to the intervention they actually received.

9.4. Statistical Analyses

A detailed statistical analysis plan (SAP) centered on primary and secondary endpoints will be developed and finalized prior to initiation of enrollment and will further describe the participant populations to be included in each analysis, details of the statistical methods, including procedures for accounting for missing, unused, and spurious data. Some exploratory analyses may be included in this SAP. Analyses are summarized in Table 10.

Exploratory endpoint analyses, in particular those centered on immunogenicity and exploratory biomarker analyses, will be described in a separate exploratory scientific and statistical analysis plan (exploratory SSAP). Specific objectives and hypotheses related to exploratory biomarker analyses will be documented in this exploratory SSAP prior to full data unblinding and analysis.

Results from the exploratory analyses may be reported in a separate results memo and included as an addendum to the clinical study report (CSR).

Table 10: Summary of Primary and Secondary Endpoints and Analyses

Endpoint	Statistical Analysis
Primary:	
Sustained QFT conversion based on positive QFT test results (initial conversion and QFT-positive at 3- and 6-months PC)	The primary analysis will be triggered when at least 118 sustained *Mtb* infection events in the mITT efficacy population are accrued. A log-rank test, stratified by sex and age group (10-11 years old, 12-14 years old, and > 14 years old), will be used to evaluate differences in the distributions of event times between the BCG vaccination and placebo groups. The primary hypothesis will be met if the 1-sided p-value associated with evaluating whether the event free distribution is longer in the BCG vaccination group relative the placebo group is less than or equal to 0.025. Hazard ratios and 95% confidence intervals will be estimated using a stratified Cox proportional hazards regression model.
Secondary:	
Sustained QFT conversion based on positive QFT test results (initial conversion and QFT-positive at 3- and 6-months PC) with a follow-up of 36- and 48-months post vaccination	Two durability analyses will be performed, the first after all participants have at least 36 months of follow-up and the second after all participants have 48 months of follow-up. Log-rank tests, stratified by sex and age group (10-11 years old, 12-14 years old, and > 14 years old), will be used to evaluate differences in the distributions of event times between the BCG vaccination and placebo groups. The secondary hypotheses will be met if the 1-sided p-value associated with evaluating whether the event free distribution is longer in the BCG vaccination group relative the placebo group is less than or equal to 0.025. Hazard ratios and 95% confidence intervals will be estimated using stratified Cox proportional hazards regression models.

Endpoint	Statistical Analysis
Secondary, continued	
Safety endpoints post vaccination Solicited AEs through 7 days Unsolicited AEs through 28 days All SAEs and AESIs through Month 6 Serious ADRs through the end of the study	The incidence and 95% confidence intervals of AEs will be summarized by treatment group, overall, and by grade, intensity, and relatedness. The incidence and 95% confidence intervals of SAEs will be summarized by treatment group. Summary statistics will be generated for raw laboratory safety tests, as well as for changes from baseline, as deemed clinically appropriate.

9.4.1. Efficacy Analyses

9.4.1.1. Primary Endpoint Analyses

The primary efficacy outcome measure is sustained QFT conversion, defined as an initial positive QFT test, and sustained positive tests at both 3- and 6-months after primary conversion.

To assess the vaccine efficacy on this primary endpoint, an event driven analysis will be utilized.

The primary analysis will occur when at least 118 sustained *Mtb* infection events occur in the mITT efficacy population. If at least 118 sustained QFT conversion events are reached within a 6- month period prior to all participants completing 36 months of follow up, then the primary analysis and the Month 36 secondary endpoint analysis may be combined into a single analysis to occur after all participants have completed the Month 36 visit.

A log-rank test, stratified by sex and age group (10-11 years old, 12-14 years old, and > 14 years old), will be used to evaluate differences in the distributions of event times between the BCG vaccination and placebo groups. The primary hypothesis will be met if the 1-sided p-value associated with evaluating whether the event free distribution is longer in the BCG vaccination group relative the placebo group is less than or equal to 0.025. Hazard ratios and 95% confidence intervals will be estimated using a Cox proportional hazards regression model, adjusting for sex and age group.

9.4.1.2. Secondary and Exploratory Efficacy Endpoint Analyses

To address the secondary objectives (sustained QFT conversion based on positive QFT test results with a follow-up of 36- and 48-months post vaccination), two analyses will be performed to assess the durability of efficacy against sustained *Mtb* infection post vaccination. The first analysis will be performed subsequent to the initial primary analysis after all participants have at least 36 months of follow-up, and the second analysis will be performed subsequent to the initial and 36-month analysis, after all participants have 48 months of follow-up. However, as noted above, depending on the timing of the sustained conversion events, the 36-month analysis may be combined with the primary analysis.

Again, stratified log-rank tests will be used to evaluate differences in the distributions of event times between the BCG vaccination and placebo groups; and hazard ratios and 95% confidence intervals will be estimated from Cox regression models, adjusted for sex and age group.

If the one-sided p-value after all participants have 36 months of follow-up is less than or equal to 0.025, durability after all participants have 48 months of follow-up will be assessed.

As such, durability after all participants have reached 36- and 48-months of follow-up will only be concluded if statistically significant differences in event times between the BCG vaccination and placebo arms were met at all prior time points evaluated. Failure to meet the primary or 36-month durability hypothesis will not result in a formal early stopping of the study.

Exploratory analyses will be performed similarly using an alternative definition of conversion as a change in IFN-γ concentration from <0.35 IU/mL to ≥4 IU/mL.

Further similar exploratory analyses will be performed for various primary and sustained *Mtb* infection post vaccination endpoints, conversion defined by various exploratory QFT IFN-γ concentration thresholds (other than the package-insert defined 0.35 IU/mL value in the QFT assay). In addition, similar analyses will be performed using the ITT and PP efficacy populations to assess the robustness of results.

9.4.1.2.1. Multiplicity

To account for multiple testing, a step-down approach will be utilized to preserve the overall Type I error at a one-sided 2.5% for the primary and first secondary hypotheses. Specifically, if the one-sided p-value associated with the primary hypothesis of BCG vaccine efficacy relative to placebo is less than or equal to 0.025, the durability of the vaccine efficacy after all participants have 36 months of follow-up will be formally assessed.

9.4.2. Safety Analyses

The incidence and 95% confidence intervals of solicited AEs through 7 days post vaccination, unsolicited AEs through 28 days post vaccination, all SAEs and AESIs through 6 months post vaccination, and related SAEs throughout the entire study duration will be summarized by treatment group.

For all presentations of adverse events, additional summaries based on reporting period of AEs following each study vaccination may also be presented.

The number and percentage of participants with AEs will be summarized by Medical Dictionary for Regulatory Activities (MedDRA) system organ class (SOC) and preferred term (PT). Additional summaries will present the number and percentage of participants with adverse events by intensity and by relationship to study vaccine; each participant will be counted once per preferred term at the greatest intensity or most related state recorded for that term.

Separate summaries of the number and percentage of participants with solicited AEs will also be presented. Solicited AEs will also be summarized by intensity and relationship to study vaccine; each participant will be counted once per preferred term at the greatest intensity or most related state recorded for that term.

Summary statistics will be generated for raw laboratory safety test results, as well as for changes from baseline, as deemed clinically appropriate.

Depending on the safety parameter, the difference from baseline will either be computed on the original scale (raw change from baseline) or on the log scale and back-transformed for reporting (percent change from baseline).

Summary statistics for continuous parameters will be presented by treatment regimen for all pre- and post-vaccination assessments and for change from pre-vaccination to post-vaccination assessments.

9.4.3. Immunogenicity and Exploratory Biomarker Analyses

9.4.3.1. Immune Response by ICS

To evaluate the immunogenicity of BCG revaccination, frequency of CD4 T cells positive for one or more cytokines, e.g., IFNγ, IL-2, IL-22, and/or TNF-α by ICS, will be summarized by treatment group and timepoint.

Median DMSO-subtracted cytokine responses and associated 95% confidence intervals (CI) or other descriptive statistics as appropriate will be used to summarize percentage T cell responses. Summaries of T cell response will be presented by T cell type (CD4 and CD8) and by stimulation antigen. Summaries will include immune responses at pre- and post-vaccination immunology time points, and change from pre-vaccination to post-vaccination time points.

For the first 80 10-12-year-old participants enrolled at the SATVI site, whole blood ICS will be conducted as an immunologic endpoint and will measure the frequencies and patterns of CD4 and CD8 T cells expressing one or more cytokines following stimulation of whole blood with viable BCG from the vaccine vial. Analysis of these results will be performed as outlined above.

9.4.3.2. Other Exploratory Analyses

The SARS-CoV-2 serostatus will be summarized overall, as well as by time and treatment group. Among participants who present with suspected COVID-19, the proportion of participants with positive nucleic acid amplification will be summarized overall and by treatment group. Signs and symptoms of disease will be described for each participant.

Further exploratory analyses related to host characteristics and responses to vaccination and or *Mtb* infection will also be examined. Specific objectives and hypotheses related to these analyses will be documented and reported in a separate scientific and statistical analysis plan prior to full unblinding and analysis of data associated with these exploratory analyses.

9.4.4. Subgroup Analyses

The consistency of results with respect to the primary and secondary objectives will be examined within various subgroup populations (eg, site, sex, and age group). Details will be provided in the SAP.

9.4.5. Demographic and Compliance Analyses

Demographic parameters (age, sex, and race/ethnicity) and other baseline characteristics will be summarized by treatment group for all participants in the safety population.

Listings of randomized participants with protocol deviations will be presented by treatment group.

9.4.6. Independent Data Monitoring Committee (IDMC)

The IDMC will be established to oversee the safety of this study and to review results from the primary data analysis (after at least 118 sustained *Mtb* infection events occur in the mITT efficacy population), and the secondary analysis (after all participants have 36 months of follow-up). Refer to Sections 9.4.1.1 and 9.4.1.2, for primary and secondary endpoint analyses, respectively.

The independent IDMC will operate according to a charter. The IDMC structure, participants and other details will be provided in the charter. The charter will be available prior to study start.

The IDMC will review unblinded safety data during regular scheduled safety review meetings as well as the outcome of the primary and secondary analyses. The IDMC may request additional information, or a pause in recruitment and vaccination, while safety data are being evaluated.

During active enrollment, the IDMC will meet at least once every 3 months and ad hoc as necessary. Once enrollment is complete, the IDMC will meet at least twice a year and ad hoc as necessary. The IDMC review will include solicited and unsolicited AEs, as well as SAEs. All procedures associated with this review, including objectives, data handling, and elements to be included for review will be documented.

The IDMC may request additional information, or a pause in recruitment and vaccination, while safety data are being evaluated. The IDMC will make a formal recommendation on the continued enrollment into the trial after each safety review.

The IDMC charter will provide meeting information and other details.

If study vaccine administration is paused by the Medical Monitor or the PI, the IDMC will convene ad hoc.

The recommendations of the IDMC, along with the sponsor's decision, will be communicated to the investigators and the IRBs/IECs and the national regulatory authorities as required. The sponsor or its designee agrees to abide by any directives issued by the national regulatory authority or the IRB/IEC.

9.4.7. National Regulatory Authority

SAHPRA receives all expedited safety reports for clinical trials and has the authority to terminate, suspend or require changes to a clinical trial.

10. Supporting Documentation and Operational Considerations

10.1. Appendix 1: Regulatory, Ethical, and Study Oversight Considerations

10.1.1. Regulatory and Ethical Considerations

This study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences (CIOMS) International Ethical Guidelines
- Applicable International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use (ICH) Good Clinical Practice (GCP) Guidelines
- Applicable laws and regulations
- The protocol, protocol amendments, ICF, and other relevant documents (eg, diary cards) must be submitted to an IRB/IEC by the investigator and reviewed and approved by the IRB/IEC before the study is initiated.
- Any amendments to the protocol will require IRB/IEC approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.

10.1.2. Study Oversight

The study sponsor, the institution through which the research is performed and all members of the PI's clinical team and the national regulatory authority share responsibility for ensuring the safety of participants in this trial.

The PI will be responsible for the following:

- Providing written summaries of the status of the study to the IRB/IEC annually or more frequently, in accordance with the requirements, policies, and procedures established by the IRB/IEC
- Notifying the IRB/IEC of SAEs or other significant safety findings as required by IRB/IEC procedures
- Providing oversight of the conduct of the study at the site and adherence to requirements of ICH and GCP guidelines, SAHPRA Regulations, the IRB/IEC, and all other applicable country and local regulations
- Closely monitoring study participants and taking whatever measures necessary to ensure their safety. The PI may delay an individual's study vaccine administration or pause study vaccine administration altogether if the investigator is concerned that the study vaccine might place a participant or participants at significant risk. Where specified, the responsibilities of the PI may be delegated to a medically qualified

team member (designee). The investigator determines intensity and causality with respect to the study vaccine for each AE.

The sponsor has an institutional responsibility to ensure participant safety and is ultimately accountable for safety oversight. Medical monitors and the IDMC play an important role in this regard and support the sponsor.

The Medical Monitor is the sponsor's representative and is a physician. The Medical Monitor:

- reviews the safety of the product for protocols in a specific region and, in consultation with the sponsor, determines expectedness of AEs.
- is responsible for safety oversight in-country and plays an important role in the reporting of serious ADRs, SAEs, pregnancies, and other important safety information, as described in the protocol
- in consultation with the sponsor, may assess the intensity and causality for AEs and may upgrade the degree of intensity and causality determined by the PI or designee

The Medical Monitor, like the PI, will be blinded until primary endpoint analyses are completed, unless emergency unblinding is required.

The Institutional Review Board or Ethics Committee has institutional responsibility for the safety of research participants. The Institutional Review Board or Ethics Committee has the authority to terminate, suspend or require changes to a clinical trial.

The national regulatory authority, SAHPRA, has the authority to terminate, suspend or require changes to a clinical trial.

10.1.3. Financial Disclosure

Investigators and sub-investigators will provide the sponsor with sufficient, accurate financial information as requested to allow the sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are responsible for providing information on financial interests during the course of the study and for 1 year after completion of the study.

10.1.4. Informed Consent Process

Written informed consent and assent will be obtained prior to conducting any study-related procedures.

Participants must be informed that their participation is voluntary. The PI or designee will explain the study to the participant and, for participants who are minors, to his/her parent or guardian/LAR, and answer all questions regarding the study. The PI or designee will conduct the consent and assent discussions on an individual basis with each participant and parent or guardian/LAR. Adequate time will be allowed for all questions to be addressed. Potential participants will be interviewed to ensure that they meet all entry criteria relating to history.

10.1.4.1. Informed Consent forms

Informed consent for study participation

Participants or parent or guardian/LAR will be required to sign a statement of informed consent that meets the requirements of 21 CFR 50, local regulations, ICH guidelines, Health Insurance Portability and Accountability Act (HIPAA) requirements, where applicable, and the IRB/IEC or study center.

The informed consent form will be obtained by the use of a written informed consent form (ICF) approved by the IRB/IEC and signed and dated by the parent or guardian/LAR of participants less than 18 years of age. Consent for HIV testing and genetic testing will also be obtained prior to enrollment.

Participants 18 years of age at the time of consent will also be asked to sign a consent form.

Similarly, informed assent from participants who are 10 to 17 years of age will be obtained by the use of a written assent form approved by the IRB/IEC and signed and dated by the participant at the time of assent.

Other consents will be presented to the participant and parent or guardian/LAR which are optional and if not signed, would not exclude the participant from the study:

- Consent for storage and testing of samples for research not described in the protocol.
- Consent to participant in Extended Follow-up for the National TB Registry.

A copy of the signed consent and assent forms shall be given to the participant and parent or guardian/LAR prior to conducting any study-related procedures.

The medical record must include a statement that written informed consent was obtained before the participant was enrolled in the study and the date the written consent was obtained. The authorized person obtaining the informed consent must also sign the ICF.

If there is a change to the ICF during the conduct of the study, participants must be re-consented to the most current version of the ICF. Participants who become 18 years of age during the study (those who previously signed an assent form) will be asked to sign an informed consent form following their 18th birthday.

Any withdrawal of consent for sample testing will be documented in the CRF.

If a participant cannot be randomized on the intended day of vaccination (eg, if elevated temperature) her/she is not required to sign another ICF, as long as re-screening and vaccination occur within the protocol-defined window.

Laboratory assays for primary and secondary endpoints will be carried out in South Africa. Participants will be informed that some of the assays for exploratory endpoints may be carried out in laboratories outside South Africa.

The ICF will contain a separate section that addresses the use of remaining mandatory samples for research not described in the protocol, e.g., assay development and assay quality control. A separate signature will be required to document a participant's agreement to allow any remaining specimens to be used for research not described in the protocol.

Participants will be told that they are free to refuse to participate and may withdraw their consent at any time and for any reason during the storage period.

All participants will be asked to give specific consent to allow the sponsor (or contract partner) to use the samples for future research. Any sample testing will be in line with the consent of the participant and/or parent or guardian/LAR.

Consent will also be requested for extended passive follow-up on the TB registry after end of study.

10.1.5. Data Protection

Participants will be assigned a unique identifier by the sponsor. Any participant record or dataset that is transferred to the sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred to the sponsor.

The participant must be informed that his/her study-related data will be used by the sponsor in accordance with local data protection law. The level of data disclosure must also be explained to the participant.

The participant must be informed that his/her medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the sponsor, by appropriate IRB/IEC members, and by inspectors from regulatory authorities.

10.1.6. Dissemination of Clinical Study Data

Study information from this protocol will be posted on publicly available clinical trials registers (clinicaltrials.gov and the South African registry [sanctr.gov.za]) before enrollment of participants begins.

The final study report will include all available safety data, immunogenicity data, clinical assessments, and concomitant medications through the final study visit. The database will be locked prior to preparation of the final study report when all of the above data have been entered, reviewed, and all queries related to the data have been addressed. Modifications or additions to the analyses will be included in the relevant statistical analysis plan. Any decisions to deviate from the planned analyses described in the protocol and in the statistical analysis plan will be described in detail in the final study report.

The final clinical study report will be reviewed and approved by the sponsor signatory and the PI.

Summaries of the results of the study will also be posted on the same website.

10.1.7. Data Quality Assurance

All participant data relating to the study will be recorded on a printed CRF or, by an electronic CRF using an Electronic Data Capture (EDC) system, unless transmitted to the sponsor or designee electronically (eg, laboratory data). The investigator is responsible for verifying that data entries are accurate and correct by physically signing or electronically signing the CRF.

The investigator must maintain accurate documentation (Appendix 1, Section 10.1.8) that supports the information entered in the CRF.

The study will be monitored regularly by the sponsor or its designee throughout the study period. The investigator must permit study-related monitoring, audits, IRB/IEC review, and regulatory agency inspections and provide direct access to source data documents.

Monitoring details describing strategy (eg, risk-based initiatives in operations and quality such as Risk Management and Mitigation Strategies and Analytical Risk-Based Monitoring), methods, responsibilities and requirements, including handling of noncompliance issues and monitoring techniques (central, remote, or on-site monitoring) are provided in the Monitoring Plan.

The sponsor or designee is responsible for the data management of this study including quality checking of the data.

The sponsor assumes accountability for actions delegated to other individuals (e.g., CROs).

Study monitors will perform ongoing source data verification to confirm that data entered into the CRF by authorized site personnel are accurate, complete, and verifiable from source documents; that the safety and rights of participants are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.

Records and documents, including signed ICFs, pertaining to the conduct of this study must be retained by the investigator for 10 years after study completion unless local regulations or institutional policies require a longer retention period. No records may be destroyed during the retention period without the written approval of the sponsor. No records may be transferred to another location or party without written notification to the sponsor.

10.1.8. Source Documents

Source documentation consists of existing medical records and/or study records developed and maintained by the investigator. Source documents provide evidence for the existence of the participant and substantiate the integrity of the data collected.

Data recorded on source documents will be transcribed onto CRFs either paper, or electronically using an EDC system.

Data entered in the CRF that are transcribed from source documents must be consistent with the source documents or the discrepancies must be explained. The investigator may need to request previous medical records or transfer records, depending on the study.

For the purpose of monitoring and auditing the study, source documentation will consist of existing medical records and/or study records developed and maintained by the investigator.

10.1.9. Study and Site Closure

The sponsor designee reserves the right to close the study site(s) or terminate the study at any time for any reason at the sole discretion of the sponsor. Study sites will be closed upon study completion. A study site is considered closed when all required documents and study supplies have been collected and a study-site closure visit has been performed.

The investigator may initiate study-site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the sponsor or investigator may include but are not limited to:

- Failure of the investigator to comply with the protocol, the requirements of the IRB/IEC or local health authorities, the sponsor's procedures, or GCP guidelines
- Inadequate recruitment of participants by the investigator
- Discontinuation of further study intervention development

At the discretion of the sponsor, all materials and supplies provided to the investigator will be returned or disposed of in compliance with local regulatory requirements upon authorization from the sponsor, upon study completion. The investigator or designated clinical site staff will notify the IRB/IEC when the study has been completed.

10.1.10. Publication Policy

The results of this study may be published or presented at scientific meetings. If this is foreseen, the investigator agrees to submit all manuscripts or abstracts to the sponsor before submission. This allows the sponsor to protect proprietary information and to provide comments.

The sponsor will comply with the requirements for publication of study results. In accordance with standard editorial and ethical practice, the sponsor will generally support publication of multicenter studies only in their entirety and not as individual site data. In this case, a coordinating investigator will be designated by mutual agreement.

Authorship will be determined by mutual agreement and in line with International Committee of Medical Journal Editors authorship requirements.

10.2. Appendix 2: Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting

10.2.1. Definition of AE

AE Definition
• An AE is any untoward medical occurrence in a patient or clinical study participant, temporally associated with the use of study intervention, whether or not considered related to the study intervention. • NOTE: An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of study intervention.

Events Meeting the AE Definition
• Any abnormal laboratory test results (hematology, clinical chemistry, or urinalysis) or other safety assessments (e.g., ECG, radiological scans, vital signs), including those that worsen from baseline, considered clinically significant in the medical and scientific judgment of the investigator. • Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition. • New conditions detected or diagnosed after study intervention administration even though it may have been present (date of first sign or symptom) before the start of the study. • Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction. • Signs, symptoms, or the clinical sequelae of a suspected overdose of either study intervention or a concomitant medication. Overdose per se will not be reported as an AE/SAE unless it is an intentional overdose taken with possible suicidal/self-harming intent. Such overdoses should be reported regardless of sequelae. "Lack of efficacy" per se will not be reported as an AE or SAE. Such instances will be captured in the efficacy assessments.

Events <u>NOT</u> Meeting the AE Definition
• Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE. • Situations in which an untoward medical occurrence did not occur (social and/or convenience admission to a hospital). • Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

10.2.2. Definition of SAE

If an event is not an AE per definition above, then it cannot be an SAE even if serious conditions are met.

A SAE is defined as any untoward medical occurrence that, at any dose:
a. Results in death
b. Is life-threatening The term 'life-threatening' in the definition of 'serious' refers to an event in which the participant was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.
c. Requires inpatient hospitalization or prolongation of existing hospitalization In general, hospitalization signifies that the participant has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether "hospitalization" occurred or was necessary, the AE should be considered serious. Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.
d. Results in persistent disability/incapacity • The term disability means a substantial disruption of a person's ability to conduct normal life functions. • This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, and accidental trauma (eg, sprained ankle) which may interfere with or prevent everyday life functions but do not constitute a substantial disruption.
e. Is a congenital anomaly/birth defect
f. Other situations: • Medical or scientific judgment should be exercised in deciding whether SAE reporting is appropriate in other situations such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the participant or may require medical or surgical intervention to prevent one of the other outcomes listed in the above definition. These events should usually be considered serious. Examples of such events include invasive or malignant cancers, intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias or

convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

10.2.3. Definition of Serious ADR and SUSAR

When an adverse event is judged to be serious and related to an investigational product, it is a Serious Adverse Drug Reaction. A serious unexpected suspected adverse drug reaction is referred to as a SUSAR. Refer to Section 10.2.6 for the different reporting requirements.

10.2.4. Definition of AESI

Adverse events of special interest (AESIs) are adverse events that the sponsor wants to monitor carefully. The following AEs will be collected and reported as AESIs:

Immune system disorders:

Anaphylactic reaction

General disorders:

Disseminated BCG disease

Infections and Infestations:

Osteomyelitis

Suppurative lymphadenitis

Injection site abscess

Skin and subcutaneous tissue disorders:

Injection site lupus vulgaris

Injection site keloid formation

Bone disorders: Osteitis.

10.2.5. Recording and Follow-Up of AE and/or SAE

AE and SAE Recording

- Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the participant is the preferred method to inquire about AE occurrences.
- When an AE/SAE occurs, it is the responsibility of the investigator to review all documentation (eg, hospital progress notes, laboratory reports, and diagnostics reports) related to the event.
- The investigator will then record all relevant AE/SAE information in the CRF.
- AEs will be reported on the AE CRF using a recognized medical term or diagnosis that accurately reflects the event.

- AE evaluations will be reviewed by the PI or a medically qualified delegate. AE CRF pages are to be completed by members of the study team designated in writing by the PI. The onset and resolution dates of an AE and action taken in response to the AE will be documented.
- After the initial AE/SAE report, the investigator is required to proactively follow each participant at subsequent visits/contacts. All SAEs, and non-serious AESIs will be followed until resolution, stabilization, the event is otherwise explained, or the participant is lost to follow-up.
- It is **not** acceptable for the investigator to send photocopies of the participant's medical records to the sponsor in lieu of completion of the AE/SAE CRF page.
- There may be instances when copies of medical records for certain cases are requested by the Medical Monitor, the IDMC or the sponsor. In this case, all participant identifiers, with the exception of the participant number, will be redacted on the copies of the medical records before submission.
- The investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.
- SAEs will be assessed for intensity and causal relationship to the study product.

Follow-up of AEs and SAEs and Resolution

- The investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated to elucidate the nature and/or causality of the AE or SAE as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.
- If a participant dies during participation in the study or during a recognized follow-up period, the investigator will provide the sponsor with a copy of any post-mortem findings including histopathology, if available.
- New or updated information will be recorded in the originally completed CRF.
- The investigator will submit any updated SAE data to the sponsor within 24 hours of receipt of the information.
- The onset and resolution dates of the event and medical care taken in response to the event will be documented.
- AEs will be considered resolved when the condition returns to normal or returns to the participant's baseline status as established on Study Day 1, or when the condition has stabilized with the expectation that it will remain chronic.

- If the event has not resolved by the final study visit, it will be documented as "ongoing" on the CRF, however, follow-up of the SAE must continue until resolved or the condition has stabilized. Information recorded on the CRF must be substantiated in the source documents.
- The resolution date to be recorded on the CRF is the last date on which the participant experienced the AE.

10.2.5.1. Assessment of Intensity

The investigator will make an assessment of intensity for each AE reported during the study and assign it to 1 of 4 categories, as defined in Appendix 5 Section 10.5:

Note that a body temperature between 37.5°C and 37.9°C**,** can be captured as an AE, at the PI's discretion.

- **Mild** symptoms causing no or minimal interference with usual social and functional activities with intervention not indicated
- **Moderate** symptoms causing greater than minimal interference with usual social and functional activities with intervention indicated
- **Severe** symptoms causing inability to perform usual social and functional activities with intervention or hospitalization indicated
- **Potentially life-threatening** symptoms causing inability to perform basic self-care functions with intervention indicated to prevent permanent impairment, persistent disability, or death

  An event is defined as 'serious' when it meets at least 1 of the predefined outcomes as described in the definition of an SAE, NOT when it is rated as severe.

10.2.5.2. Assessment of Causality

- The investigator is obligated to assess the relationship between study intervention and each occurrence of each AE/SAE.
- An AE/SAE is considered related to study intervention if there is a reasonable possibility that the study intervention contributed to the AE.
- Not-related means there is no reasonable possibility that the AE is causally related to administration of the study intervention. There are other more likely causes for the AE.
- The investigator will use clinical judgment to determine the relationship.
- Alternative causes, such as underlying disease(s), concomitant therapy, and other risk factors, as well as the temporal relationship of the event to study intervention administration will be considered and investigated.

- For each AE/SAE, the investigator **must** document in the medical notes that he/she has reviewed the AE/SAE and has provided an assessment of causality.
- There may be situations in which an SAE has occurred and the investigator has minimal information to include in the initial report. However, **it is very important that the investigator always makes an assessment of causality before the initial transmission of the SAE data.**
- The investigator may change his/her opinion of causality in light of follow-up information and send a SAE follow-up report with the updated causality assessment.
- The causality assessment is one of the criteria used when determining regulatory reporting requirements.

10.2.5.3. Assessment of Expectedness

Expected adverse events are adverse events consistent with the reference safety information provided by the sponsor (i.e., the approved AJ Vaccines BCG SSI package insert). The sponsor determines expectedness.

10.2.5.4. Assessment of Outcome

The outcome of each SAE must be reported to the sponsor. For analysis purposes, the outcome for serious adverse events will be determined on the final study visit.

Outcome of all AEs will be classified as one of the following:

•Resolved

•Resolved with sequelae

•Ongoing

•Death.

10.2.6. SAEs, AESIs, Serious ADRs, and SUSAR Reporting

Reporting to sponsor delegate's (CRO Safety Team) via the Electronic Data Collection Tool

- The primary mechanism for reporting an SAE or AESI by the investigator to the sponsor or delegate will be the electronic data collection tool.
- The site will enter the SAE or AESI data into the electronic system as soon as it is identified.
- All SAEs (related and unrelated) and AESIs are reported to the sponsor and to the CRO Safety team throughout he first six months after study intervention administration.
- Serious ADRs are reported to the sponsor and to the CRO Safety team for the entire study period. Serious ADRs are reported even after the trial is over, if the sponsor, Medical Monitor or PI become aware of them.

- The investigator must not wait to collect additional information to fully document the event before notifying the CRO Safety team of an SAE or AESI. The initial notification should include the following:
    - Protocol number and name and contact number of the investigator
    - Participant ID number (and initials and date of birth, if available)
    - Date participant received study vaccine
    - SAE term and date of event onset
    - Current status of SAE.
- The investigator is responsible for expedited safety report submission to the Sponsor delegate and the Sponsor delegate reports to SAHPRA within specific time periods of being notified of the event. Therefore, it is important that the investigator submit additional information requested as soon as it becomes available.
- All fatal and life-threatening SUSARs are to be reported to SAHPRA with 7 calendar days after first knowledge with a complete report to be submitted within an additional 8 calendar days. Other SUSARs that are not fatal or life threatening need to be reported to SAHPRA no later than 15 calendar days after first knowledge. The sponsor will notify the IDMC of all SUSARs within the same timelines the report is sent to investigators, health authorities, IRB/IEC and other relevant parties. Any follow-up report will be sent with the same timelines.
- If the electronic system is unavailable, the site may use the paper SAE data collection tool (see next section) instead of the EDC, in order to report the event within 24 hours of becoming aware.
- After the study is completed at a given site, the electronic data collection tool, if used, will be taken off-line to prevent the entry of new data or changes to existing data.
- If a site receives a report of a new SAE from a study participant or receives updated data on a previously reported SAE after the electronic data collection tool has been taken off-line, then the site can report this information on a paper SAE form (see next section).
- Contacts for SAE reporting and for all safety personnel are contained in the Team Contact List which will be stored on site in the Site Regulatory Binder and maintained by the study sponsor.
- Refer to Section 1.3 for the reporting schema.

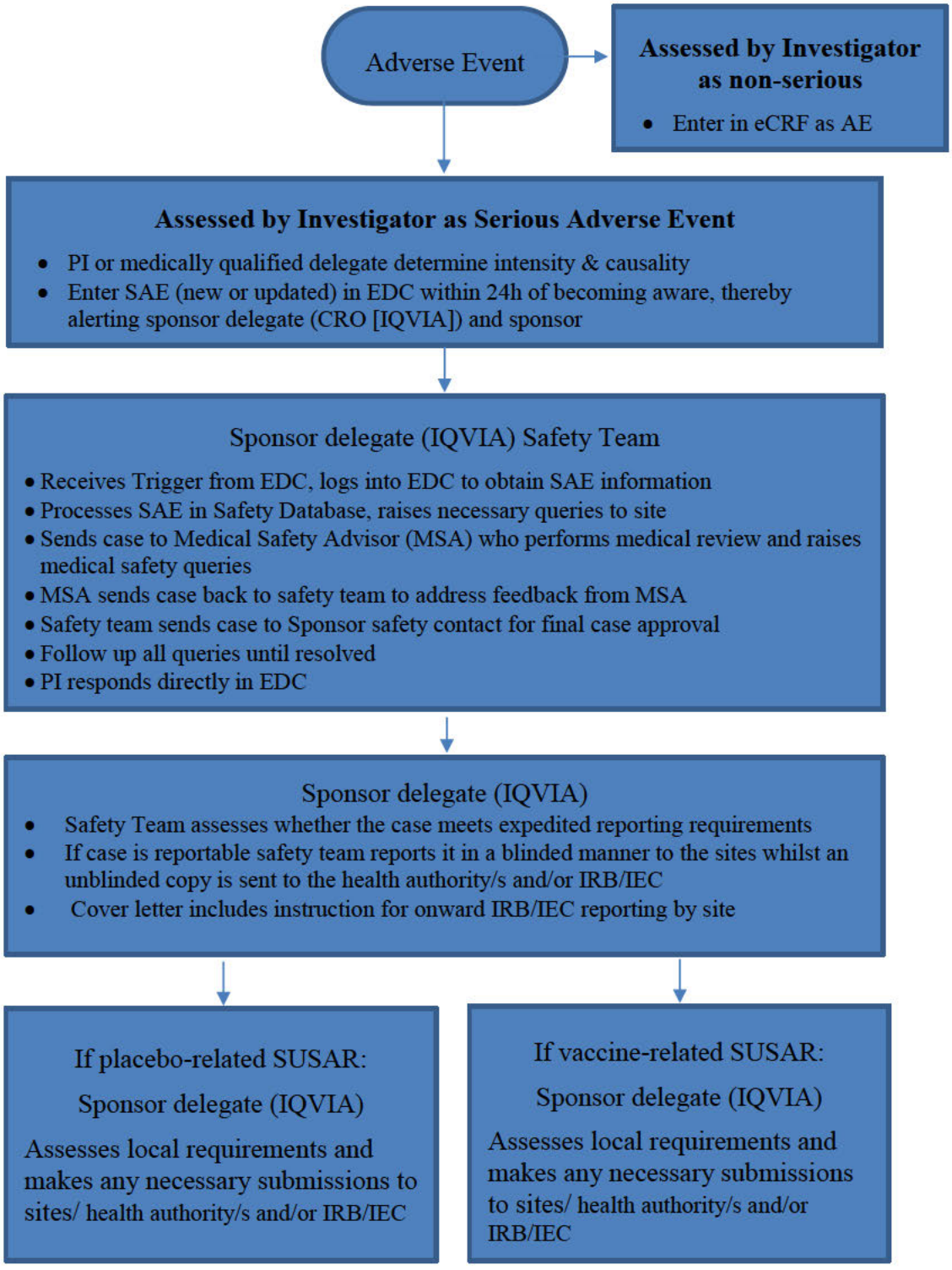
SAE Reporting via Paper CRF

- If the CRF cannot be completed, the Supplemental SAE Report (paper form) should be completed by the PI or his/her designee, and scanned and emailed, or faxed to the CRO Safety Team. The investigator is responsible for ensuring an adequate transmission of the fax and will store the distribution confirmation in the study file.
- In rare circumstances and in the absence of facsimile equipment, notification by telephone is acceptable with a copy of the SAE report form sent by overnight mail or courier service.
- Initial notification via telephone does not replace the need for the investigator to complete and sign the SAE CRF pages within the designated reporting time frames.

Other Events Requiring Immediate Reporting

- The investigator must report the following events by scanning and emailing, ***eCRF entry***, or faxing the appropriate form to the CRO Safety Team within 24 hours of becoming aware of the event:
  - Withdrawal of consent during the study for medical reasons
  - Emergency unblinding
  - Protocol violation affecting the safety of a participant or involving the vaccination process
  - Any event that, in the opinion of the investigator, precludes further administration of the study vaccine
  - Pregnancy (Pregnancy Notification Form)

Figure 2: SAE and SUSAR Reporting Scheme



See Section 10.2.6 under *Reporting to sponsor delegate's (CRO Safety Team) via the Electronic Data Collection Tool* for required reporting intervals.

10.3. Appendix 3: Contraceptive Guidance and Collection of Pregnancy Information

Definitions:

Female of Childbearing Potential (FOCBP)
A female is considered fertile following menarche. If fertility is uncertain (eg, amenorrhea in adolescents) and a menstrual cycle cannot be confirmed before first dose of study intervention, additional evaluation should be considered. If in doubt, the participant should be considered fertile.

Women in the following categories are not considered FOCBP

1. Premenarchal
2. Documented hysterectomy, bilateral salpingectomy or bilateral oophorectomy
    - For individuals with permanent infertility due to an alternate medical cause other than the above, (e.g., Mullerian agenesis, androgen insensitivity), investigator discretion should be applied to determining study entry.

    Note: Documentation can come from the site personnel's: review of the participant's medical records, medical examination, or medical history interview.

Contraception Guidance:

Women physically capable of pregnancy must agree to use an acceptable method of avoiding pregnancy **for one year after study intervention**.

Acceptable methods of avoiding pregnancy include:

- sexual abstinence (not engaging in sexual intercourse)
- a confirmed sterile partner

OR at least 2 of the below contraceptive methods:

- hormonal contraceptives (oral, injection, transdermal patch, or implant),
- IUD
- male or female condom
- diaphragm.

Collection and Reporting Pregnancy Information

- The investigator will collect pregnancy information on any female participant who becomes pregnant while participating in this study. Information will be recorded on the appropriate form and submitted to the sponsor within 24 hours of learning of a participant's pregnancy.

- The health status of the mother and child, the date of delivery, and the child's sex, birth weight and parity should be recorded and be reported to the Medical Monitor after delivery. If delivery occurs before the last scheduled study visit, the participant should continue to be followed to determine the outcome of the pregnancy, and for SAEs through the final study visit unless withdrawal of consent has occurred. If delivery occurs after the final study visit, the investigator should attempt to maintain contact with the participant to obtain information after delivery.

- The investigator will collect follow-up information on the participant and the neonate and the information will be forwarded to the sponsor. Generally, follow-up will not be required for longer than 6 weeks beyond the estimated delivery date. Any termination of pregnancy will be reported, regardless of fetal status (presence or absence of anomalies) or indication for the procedure.
- While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication will be reported as an AE or SAE. Abnormal pregnancy outcomes (eg, spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy) are considered SAEs. AE and SAE reporting periods apply as specified in Section 8.3.1.
- Any post-study pregnancy-related SAE considered related to the study intervention by the investigator will be reported to the sponsor. While the investigator is not obligated to actively seek this information in former study participants, he or she may learn of an SAE through spontaneous reporting.

10.4. Appendix 4: Genetics

Use/Analysis of DNA

- Genetic variation may impact a participant's response to study intervention, susceptibility to infection, progression to disease and severity of disease. Variable response to study intervention may be due to genetic determinants that impact vaccine responses. Therefore, where local regulations and IRB/IEC allow, a blood sample may be used for genetic analyses, from participants with a valid consent for genetic analyses only.
- Germline genetic analyses may include HLA typing, Fcγ receptor and NK cell genotyping, and assessment of other genetic information that may influence vaccine-induced and infection-induced responses, as well as susceptibility to infectious diseases. Germline genetic information will not be used for analyses related to paternity, ancestry or hereditary diseases.
- The samples may be analyzed as part of a multi-study assessment of genetic factors involved in the response to study intervention or study interventions of this class to understand study disease or related conditions.
- The results of genetic analyses may be reported in the CSR or in a separate study summary.
- The sponsor will store the DNA samples in a secure storage space with adequate measures to protect confidentiality.

10.5. Appendix 5: Toxicity Table

Modified from Division of AIDS Table for Grading the Intensity (severity) of Adult and Pediatric Adverse Events Version 2.1, July 2017

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
ESTIMATING INTENSITY GRADE				
Clinical adverse event NOT identified elsewhere in the Grading Table	Mild symptoms causing no or minimal interference with usual social & functional activities with intervention not indicated	Moderate symptoms causing greater than minimal interference with usual social & functional activities with intervention indicated	Severe symptoms causing inability to perform usual social & functional activities, with intervention or hospitalization indicated	Potentially life-threatening symptoms causing inability to perform basic self-care functions, with intervention indicated to prevent permanent impairment, persistent disability, or death

Cardiovascular

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Arrhythmia (by ECG or physical examination) *Specify type, if applicable*	No symptoms AND No intervention indicated	No symptoms AND Non-urgent intervention indicated	Non-life-threatening symptoms AND Non-urgent intervention indicated	Life-threatening arrhythmia OR Urgent intervention indicated
Blood Pressure Abnormalities[1] ***Hypertension*** *(with the lowest reading taken after repeat testing during a visit)* *≥ 18 years of age*	140 to < 160 mmHg systolic OR 90 to < 100 mmHg diastolic	≥ 160 to < 180 mmHg systolic OR ≥ 100 to < 110 mmHg diastolic	≥ 180 mmHg systolic OR ≥ 110 mmHg diastolic	Life-threatening consequences in a participant not previously diagnosed with hypertension (eg, malignant hypertension) OR Hospitalization indicated
< 18 years of age	> 120/80 mmHg	≥ 95th to < 99th percentile + 5 mmHg adjusted for age, height, and sex (systolic and/or diastolic)	≥ 99th percentile + 5 mmHg adjusted for age, height, and sex (systolic and/or diastolic)	Life-threatening consequences in a participant not previously diagnosed with hypertension (eg, malignant hypertension) OR Hospitalization indicated
Hypotension	No symptoms	Symptoms corrected with oral fluid replacement	Symptoms AND IV fluids indicated	Shock requiring use of vasopressors or mechanical assistance to maintain blood pressure

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Cardiac Ischemia or Infarction *Report only one*	NA	NA	New symptoms with ischemia (stable angina) OR New testing consistent with ischemia	Unstable angina OR Acute myocardial infarction
Heart Failure	No symptoms AND Laboratory or cardiac imaging abnormalities	Symptoms with mild to moderate activity or exertion	Symptoms at rest or with minimal activity or exertion (eg, hypoxemia) OR Intervention indicated (eg,oxygen)	Life-threatening consequences OR Urgent intervention indicated (eg, vasoactive medications, ventricular assist device, heart transplant)
Hemorrhage (with significant acute blood loss)	NA	Symptoms AND No transfusion indicated	Symptoms AND Transfusion of ≤ 2 units packed RBCs indicated	Life-threatening hypotension OR Transfusion of > 2 units packed RBCs (for children, packed RBCs > 10 cc/kg) indicated
Prolonged PR Interval or AV Block *Report only one* *> 16 years of age*	PR interval 0.21 to < 0.25 seconds	PR interval ≥ 0.25 seconds OR Type I 2nd degree AV block	Type II 2nd degree AV block OR Ventricular pause ≥ 3.0 seconds	Complete AV block
≤ 16 years of age	1st degree AV block (PR interval > normal for age and rate)	Type I 2nd degree AV block	Type II 2nd degree AV block OR Ventricular pause ≥ 3.0 seconds	Complete AV block
Prolonged QTc interval Interval [2]	0.45 to 0.47 seconds	> 0.47 to 0.50 seconds	> 0.50 seconds OR ≥ 0.06 seconds above baseline	Life-threatening consequences (eg, Torsade de pointes, other associated serious ventricular dysrhythmia)
Thrombosis or Embolism *Report only one*	NA	Symptoms AND No intervention indicated	Symptoms AND Intervention indicated	Life-threatening embolic event (eg, pulmonary embolism, thrombus)

Blood pressure norms for children < 18 years of age can be found in: Expert Panel on Integrated Guidelines for Cardiovascular Health and Risk Reduction in Children and Adolescents. Pediatrics 2011;128;S213; originally published online November 14, 2011; DOI: 10.1542/peds.2009-2107C.

[2] As per Bazett's formula.

Dermatologic

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Alopecia (scalp only)	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing greater than minimal interference with usual social & functional activities	NA	NA
Bruising	Localized to one area	Localized to more than one area	Generalized	NA
Cellulitis	NA	Non-parenteral treatment indicated (eg, oral antibiotics, antifungals, antivirals)	IV treatment indicated (eg, IV antibiotics, antifungals, antivirals)	Life-threatening consequences (eg, sepsis, tissue necrosis)
Hyperpigmentation	Slight or localized causing no or minimal interference with usual social & functional activities	Marked or generalized causing greater than minimal interference with usual social & functional activities	NA	NA
Hypopigmentation	Slight or localized causing no or minimal interference with usual social & functional activities	Marked or generalized causing greater than minimal interference with usual social & functional activities	NA	NA
Petechiae	Localized to one area	Localized to more than one area	Generalized	NA
Pruritus [1] (without skin lesions)	Itching causing no or minimal interference with usual social & functional activities	Itching causing greater than minimal interference with usual social & functional activities	Itching causing inability to perform usual social & functional activities	NA

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Rash *Specify type, if applicable*	Localized rash	Diffuse rash OR Target lesions	Diffuse rash AND Vesicles or limited number of bullae or superficial ulcerations of mucous membrane limited to one site	Extensive or generalized bullous lesions OR Ulceration of mucous membrane involving two or more distinct mucosal sites OR Stevens-Johnson syndrome OR Toxic epidermal necrolysis

[1] For pruritus associated with injections or infusions, see the *Site Reactions to Injections and Infusions* section

Endocrine and Metabolic

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Diabetes Mellitus	Controlled without medication	Controlled with medication OR Modification of current medication regimen	Uncontrolled despite treatment modification OR Hospitalization for immediate glucose control indicated	Life-threatening consequences (eg, ketoacidosis, hyperosmolar nonketotic coma, end organ failure)
Gynecomastia	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing pain with greater than minimal interference with usual social & functional activities	Disfiguring changes AND Symptoms requiring intervention or causing inability to perform usual social & functional activities	NA

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Hyperthyroidism	No symptoms AND Abnormal laboratory value	Symptoms causing greater than minimal interference with usual social & functional activities OR Thyroid suppression therapy indicated	Symptoms causing inability to perform usual social & functional activities OR Uncontrolled despite treatment modification	Life-threatening consequences (eg, thyroid storm)
Hypothyroidism	No symptoms AND Abnormal laboratory value	Symptoms causing greater than minimal interference with usual social & functional activities OR Thyroid replacement therapy indicated	Symptoms causing inability to perform usual social & functional activities OR Uncontrolled despite treatment modification	Life-threatening consequences (eg, myxedema coma)
Lipoatrophy[1]	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing greater than minimal interference with usual social & functional activities	Disfiguring changes	NA
Lipohypertrophy[2]	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual	Obvious on visual inspection AND Causing greater than minimal interference with usual social & functional activities	Disfiguring changes	NA

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
	social & functional activities			

Definition: A disorder characterized by fat loss in the face, extremities, and buttocks.
[2] Definition: A disorder characterized by abnormal fat accumulation on the back of the neck, breasts, and abdomen.

Gastrointestinal

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Anorexia	Loss of appetite without decreased oral intake	Loss of appetite associated with decreased oral intake without significant weight loss	Loss of appetite associated with significant weight loss	Life-threatening consequences OR Aggressive intervention indicated (eg, tube feeding, total parenteral nutrition)
Ascites	No symptoms	Symptoms AND Intervention indicated (eg, diuretics, therapeutic paracentesis)	Symptoms recur or persist despite intervention	Life-threatening consequences
Bloating or Distension *Report only one*	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	NA

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Cholecystitis	NA	Symptoms AND Medical intervention indicated	Radiologic, endoscopic, or operative intervention indicated	Life-threatening consequences (eg, sepsis, perforation)
Constipation	NA	Persistent constipation requiring regular use of dietary modifications, laxatives, or enemas	Obstipation with manual evacuation indicated	Life-threatening consequences (eg, obstruction)
Diarrhea *≥ 1 year of age*	Transient or intermittent episodes of unformed stools OR Increase of ≤ 3 stools over baseline per 24- hr period	Persistent episodes of unformed to watery stools OR Increase of 4 to 6 stools over baseline per 24-hour period	Increase of ≥ 7 stools per 24-hour period OR IV fluid replacement indicated	Life-threatening consequences (eg, hypotensive shock)
Dysphagia or Odynophagia *Report only one and specify location*	Symptoms but able to eat usual diet	Symptoms causing altered dietary intake with no intervention indicated	Symptoms causing severely altered dietary intake with intervention indicated	Life-threatening reduction in oral intake
Gastrointestinal Bleeding	Not requiring intervention other than iron supplement	Endoscopic intervention indicated	Transfusion indicated	Life-threatening consequences (eg, hypotensive shock)
Mucositis or Stomatitis *Report only one and specify location*	Mucosal erythema	Patchy pseudomembranes or ulcerations	Confluent pseudomembranes or ulcerations OR Mucosal bleeding with minor trauma	Life-threatening consequences (eg, aspiration, choking) OR Tissue necrosis OR Diffuse spontaneous mucosal bleeding

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Nausea	Transient (< 24 hours) or intermittent AND No or minimal interference with oral intake	Persistent nausea resulting in decreased oral intake for 24 to 48 hours	Persistent nausea resulting in minimal oral intake for > 48 hours OR Rehydration indicated (eg, IV fluids)	Life-threatening consequences (eg, hypotensive shock)
Pancreatitis	NA	Symptoms with hospitalization not indicated	Symptoms with hospitalization indicated	Life-threatening consequences (eg, circulatory failure, hemorrhage, sepsis)
Perforation (colon or rectum)	NA	NA	Intervention indicated	Life-threatening consequences
Proctitis	Rectal discomfort with no intervention indicated	Symptoms causing greater than minimal interference with usual social & functional activities OR Medical intervention indicated	Symptoms causing inability to perform usual social & functional activities OR Operative intervention indicated	Life-threatening consequences (eg, perforation)
Rectal Discharge	Visible discharge	Discharge requiring the use of pads	NA	NA
Vomiting	Transient or intermittent AND No or minimal interference with oral intake	Frequent episodes with no or mild dehydration	Persistent vomiting resulting in orthostatic hypotension OR Aggressive rehydration indicated (eg, IV fluids)	Life-threatening consequences (eg, hypotensive shock)

Musculoskeletal

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Arthralgia	Joint pain causing no or minimal interference with usual social & functional activities	Joint pain causing greater than minimal interference with usual social & functional activities	Joint pain causing inability to perform usual social & functional activities	Disabling joint pain causing inability to perform basic self-care functions
Arthritis	Stiffness or joint swelling causing no or minimal interference with usual social & functional activities	Stiffness or joint swelling causing greater than minimal interference with usual social & functional activities	Stiffness or joint swelling causing inability to perform usual social & functional activities	Disabling joint stiffness or swelling causing inability to perform basic self-care functions
Myalgia (generalized)	Muscle pain causing no or minimal interference with usual social & functional activities	Muscle pain causing greater than minimal interference with usual social & functional activities	Muscle pain causing inability to perform usual social & functional activities	Disabling muscle pain causing inability to perform basic self-care functions
Osteonecrosis	NA	No symptoms but with radiographic findings AND No operative intervention indicated	Bone pain with radiographic findings OR Operative intervention indicated	Disabling bone pain with radiographic findings causing inability to perform basic self-care functions
Osteopenia[1] *< 30 years of age*	BMD z-score -2 to -1	NA	NA	NA

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Osteoporosis[1] *< 30 years of age*	NA	BMD z-score < -2	Pathologic fracture (eg, compression fracture causing loss of vertebral height)	Pathologic fracture causing life-threatening consequences

[1] BMD t and z scores can be found in: Kanis JA on behalf of the World Health Organization Scientific Group (2007). Assessment of osteoporosis at the primary health-care level. Technical Report. World Health Organization Collaborating Centre for Metabolic Bone Diseases, University of Sheffield, UK. 2007: Printed by the University of Sheffield.

Neurologic

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Acute CNS Ischemia	NA	NA	Transient ischemic attack	Cerebral vascular accident (eg, stroke with neurological deficit)
Altered Mental Status (for Dementia, see *Cognitive, Behavioral, or Attentional Disturbance* below)	Changes causing no or minimal interference with usual social & functional activities	Mild lethargy or somnolence causing greater than minimal interference with usual social & functional activities	Confusion, memory impairment, lethargy, or somnolence causing inability to perform usual social & functional activities	Delirium OR Obtundation OR Coma
Ataxia	Symptoms causing no or minimal interference with usual social & functional activities OR No symptoms with ataxia detected on examination	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Disabling symptoms causing inability to perform basic self-care functions
Cognitive, Behavioral, or Attentional Disturbance (includes dementia and attention deficit disorder) *Specify type, if applicable*	Disability causing no or minimal interference with usual social & functional activities OR Specialized resources not indicated	Disability causing greater than minimal interference with usual social & functional activities OR Specialized resources on part time basis indicated	Disability causing inability to perform usual social & functional activities OR Specialized resources on a fulltime basis indicated	Disability causing inability to perform basic self-care functions OR Institutionalization indicated

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Developmental Delay *< 18 years of age* *Specify type, if applicable*	Mild developmental delay, either motor or cognitive, as determined by comparison with a developmental screening tool appropriate for the setting	Moderate developmental delay, either motor or cognitive, as determined by comparison with a developmental screening tool appropriate for the setting	Severe developmental delay, either motor or cognitive, as determined by comparison with a developmental screening tool appropriate for the setting	Developmental regression, either motor or cognitive, as determined by comparison with a developmental screening tool appropriate for the setting
Headache	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Symptoms causing inability to perform basic self-care functions OR Hospitalization indicated OR Headache with significant impairment of alertness or other neurologic function
Neuromuscular Weakness (includes myopathy and neuropathy) *Specify type, if applicable*	Minimal muscle weakness causing no or minimal interference with usual social & functional activities OR No symptoms with decreased strength on examination	Muscle weakness causing greater than minimal interference with usual social & functional activities	Muscle weakness causing inability to perform usual social & functional activities	Disabling muscle weakness causing inability to perform basic self-care functions OR Respiratory muscle weakness impairing ventilation

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Neurosensory Alteration (includes paresthesia and painful neuropathy) *Specify type, if applicable*	Minimal paresthesia causing no or minimal interference with usual social & functional activities OR No symptoms with sensory alteration on examination	Sensory alteration or paresthesia causing greater than minimal interference with usual social & functional activities	Sensory alteration or paresthesia causing inability to perform usual social & functional activities	Disabling sensory alteration or paresthesia causing inability to perform basic self-care functions
Seizures ***New Onset Seizure*** *≥ 18 years of age*	NA	NA	1 to 3 seizures	Prolonged and repetitive seizures (eg, status epilepticus) OR Difficult to control (eg, refractory epilepsy)
< 18 years of age (includes new or preexisting febrile seizures)	Seizure lasting < 5 minutes with < 24 hours postictal state	Seizure lasting 5 to <20 minutes with < 24 hours postictal state	Seizure lasting ≥ 20 minutes OR > 24 hours postictal state	Prolonged and repetitive seizures (eg, status epilepticus) OR Difficult to control (eg, refractory epilepsy)
Pre-existing Seizure	NA	Increased frequency from previous level of control without change in seizure character	Change in seizure character either in duration or quality (eg, intensity or focality)	Prolonged and repetitive seizures (eg, status epilepticus) OR Difficult to control (eg, refractory epilepsy)

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Syncope	Near syncope without loss of consciousness (eg, pre-syncope)	Loss of consciousness with no intervention indicated	Loss of consciousness AND Hospitalization or intervention required	NA

Pregnancy, Puerperium, and Perinatal

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Stillbirth (report using mother's participant ID) *Report only one*	NA	NA	Fetal death occurring at ≥ 20 weeks gestation	NA
Preterm Birth (report using mother's participant ID)	Live birth at 34 to < 37 weeks gestational age	Live birth at 28 to < 34 weeks gestational age	Live birth at 24 to < 28 weeks gestational age	Live birth at < 24 weeks gestational age
Spontaneous Abortion or Miscarriage[1] (report using mother's participant ID) *Report only one*	Chemical pregnancy	Uncomplicated spontaneous abortion or miscarriage	Complicated spontaneous abortion or miscarriage	NA

[1] A pregnancy loss occurring at < 20 weeks gestational age

Psychiatric

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Insomnia	Mild difficulty falling asleep, staying asleep, or waking up early causing no or minimal interference with usual social & functional activities	Moderate difficulty falling asleep, staying asleep, or waking up early causing more than minimal interference with usual social & functional activities	Severe difficulty falling asleep, staying asleep, or waking up early causing inability to perform usual social & functional activities requiring intervention or hospitalization	NA
Psychiatric Disorders (includes anxiety, depression, mania, and psychosis) *Specify disorder*	Symptoms with intervention not indicated OR Behavior causing no or minimal interference with usual social & functional activities	Symptoms with intervention indicated OR Behavior causing greater than minimal interference with usual social & functional activities	Symptoms with hospitalization indicated OR Behavior causing inability to perform usual social & functional activities	Threatens harm to self or others OR Acute psychosis OR Behavior causing inability to perform basic self-care functions
Suicidal Ideation or Attempt *Report only one*	Preoccupied with thoughts of death AND No wish to kill oneself	Preoccupied with thoughts of death AND Wish to kill oneself with no specific plan or intent	Thoughts of killing oneself with partial or complete plans but no attempt to do so OR Hospitalization indicated	Suicide attempted

Respiratory

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Acute Bronchospasm	Forced expiratory volume in 1 second or peak flow reduced to ≥70 to <80% OR Mild symptoms with intervention not indicated	Forced expiratory volume in 1 second or peak flow 50 to <70% OR Symptoms with intervention indicated OR Symptoms causing greater than minimal interference with usual social & functional activities	Forced expiratory volume in 1 second or peak flow 25 to <50% OR Symptoms causing inability to perform usual social & functional activities	Forced expiratory volume in 1 second or peak flow < 25% OR Life-threatening respiratory or hemodynamic compromise OR Intubation
Dyspnea or Respiratory Distress *Report only one*	Dyspnea on exertion with no or minimal interference with usual social & functional activities OR Wheezing OR Minimal increase in respiratory rate for age	Dyspnea on exertion causing greater than minimal interference with usual social & functional activities OR Nasal flaring OR Intercostal retractions OR Pulse oximetry 90 to < 95%	Dyspnea at rest causing inability to perform usual social & functional activities OR Pulse oximetry < 90%	Respiratory failure with ventilator support indicated (eg, CPAP, BPAP, intubation)

Sensory

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Hearing Loss *≥ 12 years of age*	NA	Hearing aid or intervention not indicated	Hearing aid or intervention indicated	Profound bilateral hearing loss (> 80 dB at 2 kHz and above) OR Non-serviceable hearing (ie, >50 dB audiogram and <50% speech discrimination)
< 12 years of age (based on a 1, 2, 3, 4, 6 and 8 kHz audiogram)	> 20 dB hearing loss at ≤ 4 kHz	> 20 dB hearing loss at > 4 kHz	> 20 dB hearing loss at ≥ 3 kHz in one ear with additional speech language related services indicated (where available) OR Hearing loss sufficient to indicate therapeutic intervention, including hearing aids	Audiologic indication for cochlear implant and additional speech/language related services indicated (where available)
Tinnitus	Symptoms causing no or minimal interference with usual social & functional activities with intervention not indicated	Symptoms causing greater than minimal interference with usual social & functional activities with intervention indicated	Symptoms causing inability to perform usual social & functional activities	NA
Uveitis	No symptoms AND Detectable on examination	Anterior uveitis with symptoms OR Medical intervention indicated	Posterior or panuveitis OR Operative intervention indicated	Disabling visual loss in affected eye(s)

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Vertigo	Vertigo causing no or minimal interference with usual social & functional activities	Vertigo causing greater than minimal interference with usual social & functional activities	Vertigo causing inability to perform usual social & functional activities	Disabling vertigo causing inability to perform basic self-care functions
Visual Changes (assessed from baseline)	Visual changes causing no or minimal interference with usual social & functional activities	Visual changes causing greater than minimal interference with usual social & functional activities	Visual changes causing inability to perform usual social & functional activities	Disabling visual loss in affected eye(s)

Systemic

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Acute Allergic Reaction	Localized urticaria (wheals) with no medical intervention indicated	Localized urticaria with intervention indicated OR Mild angioedema with no intervention indicated	Generalized urticaria OR Angioedema with intervention indicated OR Symptoms of mild bronchospasm	Acute anaphylaxis OR Life-threatening bronchospasm OR Laryngeal edema
Chills	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	NA
Cytokine Release Syndrome[1]	Mild signs and symptoms AND Therapy (ie, antibody infusion) interruption not indicated	Therapy (ie, antibody infusion) interruption indicated AND Responds promptly to symptomatic treatment OR Prophylactic medications indicated for ≤ 24 hours	Prolonged severe signs and symptoms OR Recurrence of symptoms following initial improvement	Life-threatening consequences (eg, requiring pressor or ventilator support)

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Fatigue or Malaise *Report only one*	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Incapacitating symptoms of fatigue or malaise causing inability to perform basic self-care functions
Fever (non-axillary temperatures only)	38.0 to <38.6°C or 100.4 to <101.5°F	≥ 38.6 to <39.3°C or ≥101.5 to <102.7°F	≥39.3 to < 40.0°C or ≥ 102.7 to <104.0°F	≥40.0°C or ≥104.0°F
Pain[2] (not associated with study agent injections and not specified elsewhere) *Specify location*	Pain causing no or minimal interference with usual social & functional activities	Pain causing greater than minimal interference with usual social & functional activities	Pain causing inability to perform usual social & functional activities	Disabling pain causing inability to perform basic self-care functions OR Hospitalization indicated
Serum Sickness[3]	Mild signs and symptoms	Moderate signs and symptoms AND Intervention indicated (eg, antihistamines)	Severe signs and symptoms AND Higher level intervention indicated (eg, steroids or IV fluids)	Life-threatening consequences (eg, requiring pressor or ventilator support)
Underweight[4] *>5 to 19 years of age*	WHO BMI z-score < -1 to -2	WHO BMI z-score < -2 to -3	WHO BMI z-score < -3	WHO BMI z-score < -3 with life-threatening consequences
Unintentional Weight Loss (excludes postpartum weight loss)	NA	5 to < 9% loss in body weight from baseline	≥ 9 to < 20% loss in body weight from baseline	≥ 20% loss in body weight from baseline OR Aggressive intervention indicated (eg, tube feeding, total parenteral nutrition)

[1] Definition: A disorder characterized by nausea, headache, tachycardia, hypotension, rash, and/or shortness of breath.
[2] For pain associated with injections or infusions, see the *Site Reactions to Injections and Infusions* section
[3] Definition: A disorder characterized by fever, arthralgia, myalgia, skin eruptions, lymphadenopathy, marked discomfort, and/or dyspnea.
[4] WHO reference tables may be accessed by clicking the desired age range or by accessing the following URLs:
http://www.who.int/growthref/who2007_bmi_for_age/en/ for participants > 5 to 19 years of age and http://www.who.int/childgrowth/standards/chart_catalogue/en/ for those ≤ 5 years of age

Urinary

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Urinary Tract Obstruction	NA	Signs or symptoms of urinary tract obstruction without hydronephrosis or renal dysfunction	Signs or symptoms of urinary tract obstruction with hydronephrosis or renal dysfunction	Obstruction causing life-threatening consequences

Site Reactions to Injections and Infusions

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Injection Site Pain or Tenderness *Report only one*	Pain or tenderness causing no or minimal limitation of use of limb	Pain or tenderness causing greater than minimal limitation of use of limb	Pain or tenderness causing inability to perform usual social & functional activities	Pain or tenderness causing inability to perform basic self-care function OR Hospitalization indicated
Injection Site Erythema or Redness[1] *Report only one > 15 years of age*	2.1 to < 5 cm in diameter	≥5 to <10 cm in max diameter or or ≥25 to < 100 cm2 surface area OR Symptoms causing greater than minimal interference with usual social & functional activities	≥10 cm in diameter OR ≥100 cm^2 surface area OR Phlebitis OR Sterile abscess OR Symptoms causing inability to perform usual social & functional activities	Potentially life-threatening consequences (eg, abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue) OR Hospitalization indicated

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Injection Site Erythema or Redness[1] *Report only one* *≤ 15 years of age*	2.1 cm to ≤ 2.5 cm in diameter	> 2.5 cm in diameter with < 50% surface area of the extremity segment involved (eg, upper arm or thigh)	≥ 50% surface area of the extremity segment involved (eg, upper arm or thigh) OR Phlebitis OR Sterile abscess	Potentially life-threatening consequences (eg, abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue) OR Hospitalization indicated
Injection Site Induration or Swelling *Report only one* *> 15 years of age*	Same as for **Injection Site Erythema or Redness,** *> 15 years of age*	Same as for **Injection Site Erythema or Redness,** *> 15 years of age*	Same as for **Injection Site Erythema or Redness,** *> 15 years of age*	Same as for **Injection Site Erythema or Redness,** *> 15 years of age*
≤ 15 years of age	Same as for **Injection Site Erythema or Redness,** *≤ 15 years of age*	Same as for **Injection Site Erythema or Redness,** *≤ 15 years of age*	Same as for **Injection Site Erythema or Redness,** *≤ 15 years of age*	Same as for **Injection Site Erythema or Redness,** *≤ 15 years of age*
Injection Site Pruritus	Itching localized to the injection site that is relieved spontaneously or in < 48 hours of treatment	Itching beyond the injection site that is not generalized OR Itching localized to the injection site requiring ≥ 48 hours treatment	Generalized itching causing inability to perform usual social & functional activities	NA

[1] Injection Site Erythema or Redness should be evaluated and graded using the greatest single diameter or measured surface area.

Laboratory Values* Chemistries

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Acidosis	NA	pH ≥ 7.3 to < LLN	pH < 7.3 without life-threatening consequences	pH < 7.3 with life-threatening consequences
Albumin, Low (g/dL; *g/L*)	3.0 to < LLN *30 to < LLN*	≥ 2.0 to < 3.0 *≥ 20 to < 30*	< 2.0 *< 20*	NA
Alkaline Phosphatase, High	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Alkalosis	NA	pH > ULN to ≤ 7.5	pH > 7.5 without life-threatening consequences	pH > 7.5 with life-threatening consequences
ALT or SGPT, High *Report only one*	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Amylase (Pancreatic) or Amylase (Total), High *Report only one*	1.1 to < 1.5 x ULN	1.5 to < 3.0 x ULN	3.0 to < 5.0 x ULN	≥ 5.0 x ULN
AST or SGOT, High *Report only one*	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Bicarbonate, Low (mEq/L; *mmol/L*)	16.0 to < LLN *16.0 to < LLN*	11.0 to < 16.0 *11.0 to < 16.0*	8.0 to < 11.0 *8.0 to < 11.0*	< 8.0 *< 8.0*
Bilirubin ***Direct Bilirubin[1], High > 28 days of age***	NA	NA	> ULN with other signs and symptoms of hepatotoxicity.	> ULN with life-threatening consequences (eg, signs and symptoms of liver failure)
Total Bilirubin, High > 28 days of age	1.1 to < 1.6 x ULN	1.6 to < 2.6 x ULN	2.6 to < 5.0 x ULN	≥ 5.0 x ULN

*Reminder: An asymptomatic abnormal laboratory finding without an accompanying AE should not be reported to DAIDS in an expedited time frame unless it meets protocol-specific reporting requirements.

[1] Direct bilirubin > 1.5 mg/dL in a participant < 28 days of age should be graded as grade 2, if < 10% of the total bilirubin.

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Calcium, High (mg/dL; *mmol/L*) *≥ 7 days of age*	10.6 to < 11.5 *2.65 to < 2.88*	11.5 to < 12.5 *2.88 to < 3.13*	12.5 to < 13.5 *3.13 to < 3.38*	≥ 13.5 ≥ 3.38
Calcium (Ionized), High (mg/dL; *mmol/L*)	> ULN to < 6.0 *> ULN to < 1.5*	6.0 to < 6.4 *1.5 to < 1.6*	6.4 to < 7.2 *1.6 to < 1.8*	≥ 7.2 *≥ 1.8*
Calcium, Low (mg/dL; *mmol/L*) *≥ 7 days of age*	7.8 to < 8.4 *1.95 to < 2.10*	7.0 to < 7.8 *1.75 to < 1.95*	6.1 to < 7.0 *1.53 to < 1.75*	< 6.1 *< 1.53*
Calcium (Ionized), Low (mg/dL; *mmol/L*)	< LLN to 4.0 *< LLN to 1.0*	3.6 to < 4.0 *0.9 to < 1.0*	3.2 to < 3.6 *0.8 to < 0.9*	< 3.2 *< 0.8*
Cardiac Troponin I, High	NA	NA	NA	Levels consistent with myocardial infarction or unstable angina as defined by the laboratory
Creatine Kinase, High	3 to < 6 x ULN	6 to < 10x ULN	10 to < 20 x ULN	≥ 20 x ULN
Creatinine, High **Report only one*	1.1 to 1.3 x ULN	> 1.3 to 1.8 x ULN OR Increase to 1.3 to < 1.5 x participant's baseline	> 1.8 to < 3.5 x ULN OR Increase to 1.5 to < 2.0 x participant's baseline	≥3.5 x ULN OR Increase of ≥ 2.0 x participant's baseline

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Creatinine Clearance[1] or eGFR, Low **Report only one*	NA	< 90 to 60 ml/min or ml/min/1.73 m^2 OR 10 to < 30% decrease from participant's baseline	< 60 to 30 ml/min or ml/min/1.73 m^2 OR 30 to < 50% decrease from participant's baseline	< 30 ml/min or ml/min/1.73 m^2 OR ≥ decrease from participant's baseline or dialysis needed
Glucose (mg/dL; *mmol/L*) ***Fasting, High***	110 to 125 *6.11 to < 6.95*	> 125 to 250 *6.95 to < 13.89*	> 250 to 500 *13.89 to < 27.75*	≥ 500 *≥ 27.75*
Nonfasting, High	116 to 160 *6.44 to < 8.89*	> 160 to 250 *8.89 to < 13.89*	> 250 to 500 *13.89 to < 27.75*	≥ 500 *≥ 27.75*
Glucose, Low (mg/dL; *mmol/L*) *≥ 1 month of age*	55 to 64 *3.05 to <3.55*	40 to < 55 *2.22 to < 3.05*	30 to < 40 *1.67 to < 2.22*	< 30 *< 1.67*
Lactate, High	ULN to < 2.0 x ULN without acidosis	≥ 2.0 x ULN without acidosis	Increased lactate with pH < 7.3 without life-threatening consequences	Increased lactate with pH < 7.3 with life-threatening consequences
Lipase, High	1.1 to < 1.5 x ULN	1.5 to < 3.0 x ULN	3.0 to < 5.0 x ULN	≥ 5.0 x ULN
Lipid Disorders (mg/dL; *mmol/L*) ***Cholesterol, Fasting, High*** *≥ 18 years of age*	200 to < 240 *5.18 to < 6.19*	240 to < 300 *6.19 to < 7.77*	≥ 300 *≥ 7.77*	NA
< 18 years of age	170 to < 200 *4.40 to < 5.15*	200 to < 300 *5.15 to < 7.77*	≥ 300 *≥ 7.77*	NA
LDL, Fasting, High *≥ 18 years of age*	130 to < 160 *3.37 to < 4.12*	160 to < 190 *4.12 to < 4.90*	≥ 190 *≥ 4.90*	NA

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
> 2 to < 18 years of age	110 to < 130 *2.85 to < 3.34*	130 to < 190 *3.34 to < 4.90*	≥ 190 *≥ 4.90*	NA
Triglycerides, Fasting, High	150 to 300 *1.71 to 3.42*	>300 to 500 *>3.42 to 5.7*	>500 to < 1,000 *>5.7 to 11.4*	> 1,000 *> 11.4*
Magnesium [2], Low (mEq/L; *mmol/L*)	1.2 to < 1.4 *0.60 to < 0.70*	0.9 to < 1.2 *0.45 to < 0.60*	0.6 to < 0.9 *0.30 to < 0.45*	< 0.6 *< 0.30*
Phosphate, Low (mg/dL; *mmol/L*) *> 14 years of age*	2.0 to < LLN *0.65 to < LLN*	1.4 to < 2.0 *0.45 to < 0.65*	1.0 to < 1.4 *0.32 to < 0.45*	< 1.0 *< 0.32*
1 to 14 years of age	3.0 to < 3.5 *0.97 to < 1.13*	2.5 to < 3.0 *0.81 to < 0.97*	1.5 to < 2.5 *0.48 to < 0.81*	< 1.5 *< 0.48*
Potassium, High (mEq/L; *mmol/L*)	5.6 to < 6.0 *5.6 to < 6.0*	6.0 to < 6.5 *6.0 to < 6.5*	6.5 to < 7.0 *6.5 to < 7.0*	≥ 7.0 *≥ 7.0*
Potassium, Low (mEq/L; *mmol/L*)	3.0 to < 3.4 *3.0 to < 3.4*	2.5 to < 3.0 *2.5 to < 3.0*	2.0 to < 2.5 *2.0 to < 2.5*	< 2.0 *< 2.0*
Sodium, High (mEq/L;*mmol/L*)	146 to < 150 *146 to < 150*	150 to < 154 *150 to < 154*	154 to < 160 *154 to < 160*	≥ 160 *≥ 160*
Sodium, Low (mEq/L;*mmol/L*)	130 to < 135 *130 to < 135*	125 to < 130 *125 to < 130*	121 to < 125 *121 to < 125*	≤ 120 *≤ 120*
Uric Acid, High (mg/dL; *mmol/L*)	7.5 to < 10.0 *0.45 to < 0.59*	10.0 to < 12.0 *0.59 to < 0.71*	12.0 to < 15.0 *0.71 to < 0.89*	≥ 15.0 *≥ 0.89*

Creatinine clearance- Use the applicable formula (ie, Cockcroft-Gault in mL/min or Schwartz, MDRD, CKD-Epi in mL/min/1.73m^2). Sites should choose the method defined in their study and when not specified, use the method most relevant to the study population.
*Reminder: Choose the method that selects for the higher grade.

[2] To convert a magnesium value from mg/dL to mmol/L, laboratories should multiply by 0.4114.

Hematology

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Absolute CD4 Count, Low (cell/mm^3; *cells/L*)				
> 5 years of age (not HIV infected)	300 to < 400 *300 to < 400*	200 to < 300 *200 to < 300*	100 to < 200 *100 to < 200*	< 100 *< 100*
Absolute Lymphocyte Count, Low (cell/mm^3; *cells/L*)				
> 5 years of age (not HIV infected)	600 to < 650 *0.600 x 10^9 to < 0.650 x 10^9*	500 to < 600 *0.500 x 10^9 to < 0.600 x 10^9*	350 to < 500 *0.350 x 10^9 to < 0.500 x 10^9*	< 350 *< 0.350 x 10^9*
Absolute Neutrophil Count (ANC), Low (cells/mm^3; *cells/L*)				
> 7 days of age	800 to 1,000 *0.800 x 10^9 to 1.000 x 10^9*	600 to 799 *0.600 x 10^9 to 0.799 x 10^9*	400 to 599 *0.400 x 10^9 to 0.599 x 10^9*	< 400 *< 0.400 x 10^9*
Fibrinogen, Decreased (mg/dL; *g/L*)	100 to < 200 *1.00 to < 2.00* OR 0.75 to < 1.00 x LLN	75 to < 100 *0.75 to < 1.00* OR ≥ 0.50 to < 0.75 x LLN	50 to < 75 *0.50 to < 0.75* OR 0.25 to < 0.50 x LLN	< 50 *< 0.50* OR < 0.25 x LLN OR Associated with gross bleeding
Hemoglobin[1], Low (g/dL; *mmol/L*)[2]				
≥ 13 years of age (male)	10.0 to 10.9 *6.19 to 6.76*	9.0 to < 10.0 *5.57 to < 6.19*	7.0 to < 9.0 *4.34 to < 5.57*	< 7.0 *< 4.34*
≥ 13 years of age (female)	9.5 to 10.4 *5.88 to 6.48*	8.5 to < 9.5 *5.25 to < 5.88*	6.5 to < 8.5 *4.03 to < 5.25*	< 6.5 *< 4.03*
57 days of age to < 13 years of age (male and female)	9.5 to 10.4 *5.88 to 6.48*	8.5 to < 9.5 *5.25 to < 5.88*	6.5 to < 8.5 *4.03 to < 5.25*	< 6.5 *< 4.03*

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
INR, High (not on anticoagulation therapy)	1.1 to < 1.5 x ULN	1.5 to < 2.0 x ULN	2.0 to < 3.0 x ULN	≥ 3.0 x ULN
Methemoglobin (%hemoglobin)	5.0 to < 10.0%	10.0 to < 15.0%	15.0 to < 20.0%	≥ 20.0%
PTT, High (not on anticoagulation therapy)	1.1 to < 1.66 x ULN	1.66 to < 2.33 x ULN	2.33 to < 3.00 x ULN	≥ 3.00 x ULN
Platelets, Decreased (cells/mm^3; *cells/L*)	100,000 to < 125,000 *100.000 x 10^9 to < 125.000 x 10^9*	50,000 to < 100,000 *50.000 x 10^9 to < 100.000 x 10^9*	25,000 to < 50,000 *25.000 x 10^9 to < 50.000 x 10^9*	< 25,000 *< 25.000 x 10^9*
PT, High (not on anticoagulation therapy	1.1 to < 1.25 x ULN	1.25 to < 1.50 x ULN	1.50 to < 3.00 x ULN	≥ 3.00 x ULN
WBC, Decreased (cells/mm^3; *cells/L*) *> 7 days of age*	2,000 to 2,499 *2.000 x 10^9 to 2.499 x 10^9*	1,500 to 1,999 *1.500 x 10^9 to 1.999 x 10^9*	1,000 to 1,499 *1.000 x 10^9 to 1.499 x 10^9*	< 1,000 *< 1.000 x 10^9*

[1] Male and female sex are defined as sex at birth. For transgender participants ≥13 years of age who have been on hormone therapy for more than 6 consecutive months, grade hemoglobin based on the gender with which they identify (ie, a transgender female should be graded using the female sex at birth hemoglobin laboratory values).

[2] The most commonly used conversion factor to convert g/dL to mmol/L is 0.6206. For grading hemoglobin results obtained by an analytic method with a conversion factor other than 0.6206, the result must be converted to g/dL using appropriate conversion factor for the particular laboratory

Urinalysis

Parameter	Grade 1 Mild	Grade 2 Moderate	Grade 3 Severe	Grade 4 Potentially Life-Threatening
Glycosuria (random collection tested by dipstick)	Trace to 1+ or ≤ 250 mg	2+ or > 250 to ≤ 500 mg	> 2+ or > 500 mg	NA
Hematuria (not to be reported based on dipstick findings or on blood believed to be of menstrual origin)	6 to < 10 RBCs per high power field	≥ 10 RBCs per high power field	Gross, with or without clots OR With RBC casts OR Intervention indicated	Life-threatening consequences
Proteinuria (random collection tested by dipstick)	1+	2+	3+ or higher	NA

11. References

Andersen P, Doherty T, Pai M, et al. The prognosis of latent tuberculosis: can disease be predicted? TRENDS in Mol Med. 2007;13(5):175-82.

Andrews J, Nemes E, Tameris M, et al. Serial QuantiFERON testing and tuberculosis disease risk among young children: an observational cohort study. Lancet Respir Med. 2017;4:282-290. doi: 10.1016/S2213-2600(17)30060-7.

Bottiger M, de Verdier C, Lind A, et al. A comparative study of Danish (Statens Seruminstitut), Glaxo and Behringwerke vaccines--revaccination of schoolchildren. J Biological Standardization 1983;11:1-12.

Barreto ML, Pereira SM, Pilger D, et al. Evidence of an effect of BCG revaccination on incidence of tuberculosis in school-aged children in Brazil: second report of the BCG-REVAC cluster-randomised trial. Vaccine. 2011;29(31):4875-7. doi: 10.1016/j. PMID 21616115

Cunha SS, Alexander N, Barreto ML, et al. BCG Revaccination Does Not Protect Against Leprosy in the Brazilian Amazon: A Cluster Randomised Trial. PLoS Negl Trop Dis. 2008;2(2):e167. doi:10.1371/journal.pntd. PMID 18270542

Fine P, Clayton, D, Colston M, et al (Karonga Prevention Trial Group). Randomised controlled trial of single BCG, repeated BCG, or combined BCG and killed Mycobacterium leprae vaccine for prevention of leprosy and tuberculosis in Malawi. Lancet. 1996;348(9019);17-24. DOI:https://doi.org/10.1016/S0140-6736(96)02166-6. PMID 8691924

Machingaidze S, Verver S, Mulenga H, et al. Predictive value of recent QuantiFERON conversion for tuberculosis disease in adolescents. Am J Respir Crit Care Med 2012;186:1051-6.

Mahomed H, Hawkridge T, Verver S, et al. The tuberculin skin test versus QuantiFERON TB Gold in predicting tuberculosis disease in an adolescent cohort study in South Africa. PLoS One 2011;6(3):e17984.

Mangtani, P; Nguipdop-Djomo, P; Keogh, et al. The duration of protection of school-aged BCG vaccination in England: a population-based case-control study. Inter. J of epidemiol 2017;47(1):193-201. ISSN 0300-5771 DOI: https://doi.org/10.1093/ije/dyx141

Mazurek G, Jereb J, Vernon A, et. al.Updated Guidelines for Using Interferon Gamma Release Assays to Detect Mycobacterium tuberculosis Infection --- United States, 2010 Recommendations and Reports. MMWR 2010;59(RR05):1-25.

Nemes E, H. Geldenhuys, V. Rozot, KT, et al. Prevention of M. tuberculosis Infection with H4:IC31 Vaccine or BCG Revaccination. N Engl J Med 2018;379:138-49.

Pai M. Serial testing with TB interferon-gamma release assays: toward a nuanced understanding. Chest 2012;142:1366-1367.

Pereira SM, Barreto ML, Pilger D, Cruz AA, Sant'Anna C, et al. Effectiveness and cost effectiveness of first BCG vaccination against tuberculosis in school-age children without previous tuberculin test (BCG-REVAC trial): a cluster-randomised trial. Lancet Infect Dis 2012;12:300-306.

Petruccioli E, Chiacchio T, Vanini V. Effect of therapy on Quantiferon-Plus response in patients with active and latent tuberculosis infection. Sci Rep. 2018;8(1):15626.

Rodrigues LC, Pereira SM, Cunha SS, Genser B, et al. Effect of BCG revaccination on incidence of tuberculosis in school-aged children in Brazil: the BCG-REVAC cluster-randomized trial. Lancet 2005;366:1290-95. PMID 16214599.

WHO. Global Tuberculosis Report 2018. Geneva:World Health Organization, 2018.

WHO. Latent tuberculosis infection. Updated and consolidated guidelines for programmatic management. World Health Organization. 2018b. WHO/CDS/TB/2018.4. https://www.who.int/tb/publications/2018/latent-tuberculosis-infection/en/

Winje B, White R, Syre H, et al. Stratification by interferon-γ release assay level predicts risk of incident TB. Thorax. 2018;Apr 5. pii: thoraxjnl-2017-211147. doi: 10.1136/thoraxjnl-2017-211147.

Zwerling A, Benedetti A, Cojocariu M, McIntosh F, Pietrangelo F, et al. Repeat IGRA testing in Canadian health workers: conversions or unexplained variability? PLoS One. 2013;8:e54748.

12. Version History

Versions prior to study start

Pre-IRB review:

- Version 1: 14 Feb 2019
- Version 2: 21-Feb-2019: Minor change from Version 1: added grade 4 to Sections 8.2.4 and 8.3.3.1 to align with Appendix 2)
- Version 3: 27-Feb-2019: minor change from Version 2: Randomization will be stratified by age strata (10-11 years old, 12-14 years old, and > 14 years old), sex, study site, and school **cluster (SATVI site only)**~~(if participants are students) or area of residence for non-students)~~, to account for expected differences in incidence rates

Post IRB reviews:

- Version 4: 25 June 2019: Changes from Version 3 include (new text indicated here by bold, italics):
    - Throughout protocol: Revised unblinding to occur after completion of primary endpoint analyses.
    - SoA and Section 8.2.1: Added that height and body weight will be measured at Day 1 and every six months thereafter
    - SoA and Section 8.2.6: Removed Day 1 injection site examination (edited: Site of injection examinations will be performed ***from Day 8*** through Month 6).
    - Section 4.1, Scientific Rationale for Study Design: Added text for clarity regarding blinding: Care will be taken to keep the blind for all other staff, including ***all*** laboratory staff and the sponsor staff. ***Sponsor staff may become aware of individual treatment assignments during the course of data review, however, precautions will be taken to limit exposure of sponsor staff to unblinded data.***
    - Section 5.2: Clarified exclusion criterion #25 by adding text: Shared residence with an individual who is receiving TB treatment or with someone who is known to have incompletely treated TB, e.g., ***PCR-positive***, culture-positive, smear-positive, ***or clinically diagnosed unconfirmed TB***.
    - Section 5.2: Clarified exclusion criterion #26 by adding text to clarify child in care definition – ***Child in care is*** defined as …

      The definition of a child in care does include a child cared for by family members - other than parents - who have not gone through formal legal adoption/guardian processes.
    - Section 6.1.3, Administration: Edited for clarity regarding blinding: Administration procedures for the BCG vaccine are found in the package insert.

The placebo control will be administered in the same manner ~~in this observer blind study~~… The unblinded investigational pharmacist will provide BCG and placebo to the clinic as unit-dose syringes ***with a masking label***, which will be identified with… . Before administering the injection, the study intervention administrator must inspect the syringe, ~~and vaccine volume,~~ checking that the syringe….

- Section 6.2, Preparation/Handling/Storage/Accountability., Clarified as follows: Vaccine will be prepared ~~and administered~~ as per the manufacturer's recommendations by the study pharmacist from multi-dose vials ***and*** dispensed according to the package insert.

  The saline placebo will be stored in a secured location ~~at room temperature or~~ 2°C to 8°C in the study pharmacy.

- Section 6.3.2, Masking text was updated to reflect unblinding and to reflect that sponsor staff may also become aware of individual treatment assignments during the course of data review. by adding text: The study will be ***conducted as an*** observer blind study ***until the primary endpoint has been reached and primary endpoint analyses are completed***. ***While the study is blinded, sponsor staff may become aware of individual treatment assignments during the course of data review, however, precautions will be taken to limit exposure of sponsor staff to unblinded data. Following completion of primary endpoint analyses, the study team will be unblinded but laboratory staff will remain blinded until all laboratory studies have been completed.*** . ~~meaning neither the clinical study team, laboratory staff, sponsor, CRO staff, nor participants will know the treatment allocation (BCG or placebo) administered on Day 1.~~

  Added this sentence to indicate approximate timeframe for completion of primary endpoint analyses: ***After primary endpoint analyses are completed – likely more than two years after completion of enrollment - the study will be unblinded to reduce the complexity of statistical analyses for IDMC and other data reviews.***

- Section 6.3.3. Blind Break: edited sentence for clarity by adding text: If a participant's treatment assignment is unblinded, ***during the observer-blind period (prior to completion of primary endpoint analyses)***, the sponsor must be notified within 24 hours after breaking the blind.

- ~~o~~ Section 8.1.1. Evaluation of infection: deleted text since it does not represent the processing of the QFT samples as agreed with our central lab, BARC: ~~The QFT assay will be run in a central laboratory. The initial incubations will be performed at the clinical sites using a well-defined procedure. QFT tubes will be centrifuged at the site after incubation, plasma will be transferred to vials and frozen before being shipped to the central laboratory.~~

- Section 8.2.5, Clinical Safety Lab Assessments: Removed analysis of red blood cells, eosinophils and basophils and removed testing at local laboratories (all safety analyses will be performed at the central laboratory). Edited the text: If laboratory values from non-protocol specified laboratory assessments ~~performed~~

~~at the institution's local laboratory~~ require a change in participant management or are considered clinically significant by the investigator (eg, AE or SAE), then the results must be recorded in the CRF.

All ***protocol-required*** safety laboratory tests will be performed by the central laboratory, Bio Analytical Research Corporation South Africa (BARC SA or similar). ~~Local laboratory results are only required in the event that the central laboratory results are not available in time for either study intervention administration and/or response evaluation. If a local sample is required, it is important that the sample for central analysis is obtained at the same time. Additionally, if the local laboratory results are used to make either a study intervention decision or response evaluation, the results must be entered into the CRF.~~

- Section 8.2.6, Site of Injection site examination: reworded for clarity: ~~These reactions will be recorded as expected AEs~~. ***The presence and maximum diameter of injection site swelling, vesicle, ulcer and/or scar will be recorded.The presence and maximum diameter of homolateral axillary lymph nodes larger than 1cm will be recorded.***
- Section 8.2.7, Participant follow-up, edited text for clarity: An HIV blood sample will be collected for HIV testing at screening and again yearly, with pre-and post-test HIV counseling. HIV counseling will be performed by study staff. In addition, any participant diagnosed with TB disease (see Section 8.3.6) will be tested for HIV ~~8 weeks after the~~ ***at the time of*** diagnosis ***or as soon as possible following diagnosis of TB disease***.
- Section 8.3.1, Table Time period for Collecting AE and SAE information: deleted row injection site examination since that is not applicable here.
- Section 8.8, Biomarkers: edited for clarity: Specific objectives and hypotheses related to these analyses will be documented and reported in a separate scientific and statistical analysis plan, prior to ~~data~~ ***full*** unblinding ***of data associated with these exploratory analyses***.
- Section 9.4.3.2 Other Exploratory Analyses: Specific objectives and hypotheses related to these analyses will be documented and reported in a separate scientific and statistical analysis plan prior to ~~data~~ ***full*** unblinding and analysis ***of data associated with these exploratory analyses***.
- Section 10.1.2 Study Oversight: Edited for clarity: The Medical Monitor, like the PI, will be blinded ***until primary endpoint analyses are completed***, unless emergency unblinding is required.
- Section 10.1.6, Dissemination of Clinical Study Data: Edited for clarity based on change to unblinding The database will be locked prior to ~~unblinding and~~ preparation of the final study report when all of the above data have been entered, reviewed, and all queries related to the data have been addressed.

- Section 10.2.6 SAEs, AESIs, and Serious ADR Reporting, Other Events Requiring Immediate Reporting: The investigator must report the following events by scanning and emailing, ***eCRF entry***, or faxing the appropriate form to the CRO Safety Team within 24 hours of becoming aware of the event
- Section 10.5 Toxicity table row **Cardiac Troponin I, as defined by ~~local~~ laboratory.**

Version 5, 11 May 2020

Updated protocol to address changes following March 2020 study pause due to COVID-19 pandemic to allow for flexibility in visits and other COVID-19 related issues.

Changes from Amendment 4 (first version approved by IRB) to Amendment 5 by section

New text indicated with *bold italics*

1. Title page and signature page: Amendment Version 4 changed to Version 5

   Added text ***NOTE: This study was paused for screening and randomization by the sponsor, effective 19 March 2020, due to the Coronavirus Disease 2019 (COVID-19) pandemic. Refer to Section 7.4, COVID-19 Contingency Plans for details regarding study pause due to COVID-19.***

2. List of abbreviations added ***COVID-19, PCR test, SARS-CoV-2, SUSAR,*** and corrected GMRI from Gates ~~Foundation~~ to ***Bill and Melinda Gates***
3. Synopsis 1.1 and Section 3: edited as follows:
   Rationale (synopsis only): edited text : …post conversion ***[PCJ***) (Nemes, 2018).
   Then throughout document, replaced post conversion with PC.
   Added two COVID-19 exploratory objectives and 3 endpoints as shown:

Objectives	Endpoints
• ***To describe COVID-19 among study participants who present with suspected COVID-19*** • ***To describe the serostatus of SARS-CoV-2 infection***	• ***Signs and symptoms of illness, by treatment group*** • ***SARS-CoV-2 nucleic acid amplification from nasal or oropharyngeal sample*** • ***Serological tests for SARS-CoV-2 by treatment group***

4. Added text below table: ***Refer to Section 7.4 for COVID-19 Pandemic contingency plans.***
5. Synopsis 1.1 and Section 4 Study design-Overall design

   - Total duration of study participation: …Enrollment ~~is~~ ***originally*** expected to take place over a 1-year period***, will be extended as necessary due to COVID-19 pandemic-related restrictions***.

- Wash-out period: A 70-day wash-out period will be used to identify participants who may have been infected with *Mtb* just prior to or soon after enrollment. Participants who convert to QFT positive at Day 71 ***(or if missed or not feasible, at the next feasible visit) will not be asked to return for post conversion (PC) Day 28 and PC Day 84 visits and*** will be excluded from the primary mITT efficacy analysis but will be followed for safety and efficacy to End-of-Study.
- Primary Purpose: Intervention (BCG revaccination) is being evaluated for POSI (see ***Section 2.1 and Section 4.1*** ~~Section 3~~ for details for details).
- Added: ***Refer to Section 7.4.2 for COVID-19 pandemic contingency plans with regard to participant enrollment, and other details.***
- Study sites~~: 2~~ **5** or more sites in South Africa will participate in this study.

6. Section 1.3 SoA

Extended screening period Day -28 to Day -7 to Day -28 to Day -1.

Added note in row regarding consent: ***Re-consent of participants enrolled before study pause***

Added to screening visit: Body weight and height, unsolicited AEs, All SAEs and AESI

Changed: Site of injection examination to ***Injection site*** examination

Added rows to table for sampling for SARS-CoV-2 diagnostic test and SARS-CoV-2 serology as shown below

7. Updated blood volumes in SOA to reflect new endpt collections – additional 5 mL at each visit M6, 12 18 and 24 =20 additional mL for study

Added text under Refer to Section 7.4 for COVID-19 Pandemic contingency plans.

Table 11: Scheduled Visits and Activities

Study Visit Day (D) or Month (M)	Screen D -28 to D ~~-7~~ -1	D1	D8 [a]	D29 [b]	D71 [b,c]	M6,12,18, 24 30, 36, 42, 48 (&54 [d])	Discon Visit [e]	Notes
Body weight and height measurement	*X*	X				X		
Unsolicited AEs	*X*	X	X	X				***AEs will be collected from the time informed consent is obtained.***
All serious adverse events (SAEs) and AESI	*X*	X	X	X	X	X (M6 only)		
Serious ADRs		X	X	X	X	X	X	Collected through the last study visit
Nasal or oropharyngeal sampling for SARS-CoV-2 diagnostic test	*X*	*X*	*X*	*X*	*X*	*X*		***Samples collected only if participant presents with***

Study Visit Day (D) or Month (M)	Screen D -28 to D ~~-7~~ -1	D1	D8[a]	D29[b]	D71[b,c]	M6,12,18, 24 30, 36, 42, 48 (&54[d])	Discon Visit[e]	Notes
								suspected COVID-19
SARS-CoV-2 serology (mL)	***5***					***5 at M6, M12, M18 and M24***		***If enrolled before study pause: collect at the next feasible biannual visit (e.g., M6, M12, etc) and biannually thereafter to Month 24***
Cumulative vol. (mL) after study pause (including SARS-COV-2 serology) includes subset	24.5	77.5	90.5	109.5	166.5	269.5 Yr 1[f]		Yr 2=371.5 mL Yr 3 =463.5 mL Yr 4=555.5 mL

added ***excluding SARS*** in the last column for rows

Per visit phlebotomy volume (vol.) (mL) (includes subset)
Cumulative vol. (mL) includes subset
Cumulative vol. (mL) excludes subset

Added sentences below SoA:

Unscheduled visits should only take place as necessary and data from unscheduled study visits should be captured in the EDC. Refer to Section 8.

Refer to Section 7.4 for COVID-19 Pandemic contingency plans.

8. Table 3 title edited: Table 3: Schedule of Activities Post QFT Conversion ***at the Month 6 visit or thereafter*** and removed PC from first column heading: Study Visit Day, Post Initial QFT conversion (PC) .Added footnote: ***PC = post conversion***
9. Table 4 study visit intervals as follows:

Table 12: Study Visit Intervals

Study Visits	Length of Interval	Allowed Interval
Screening	Day -28 to Day ~~-7~~ ***-1***	Day -28 to Day ~~-7~~ ***-1***
Day 8	Day 1 plus **7** days	Day ~~**7 8 through**~~ ***to*** Day ~~**10**~~ ***11***
Day 29	Day 1 plus 28 days	Day~~**28**~~ **29** to Day 35
Day 71	Day 1 plus 70 days	Day ~~**70**~~***71*** to Day ~~**77**~~ ***78***

Added: Refer to Section 7.4 for intervals based on COVID-19 contingency plans.

10. Section 2.1 Study Rationale: edited sentence: The study will be conducted at ~~2 or more~~ sites in the Western Cape province, where the initial finding of protection was observed, and ~~one or more~~ additional site(s) ~~outside~~ in other provinces.

11. Section 2.1.1 added section and text

2.1.1 Rationale for addition of COVID-19 related study objectives

The Government of South Africa declared the COVID-19 pandemic a National Disaster on 15 March 2020 and the Sponsor of this study issued a pause for enrollment on 17 March 2020, effective 19 March 2020.

The Government of South Africa subsequently ordered a lockdown (stay-at-home order) for three weeks on 26 March 2020 and subsequently extended it for an additional two weeks, through the end of April, 2020. Under the lockdown, most trial participants are not able to come to the investigational site for protocol-specified visits.

Since 1 May 2020, South Africa has employed a system of alert levels at the provincial and, in some cases, the district level. Criteria based on the rate of infection as well as health system capacity are used by the National Command Council to determine the alert level.

Enrollment into this study can only resume once the Government, local IRBs, investigators and the sponsor agree that it is acceptable to resume enrollment. Enrollment may also need to be paused again in the future, depending on the COVID-19 situation at national, province and district level.

Study investigators and study sites have defined infection control measures for COVID-19 that need to be implemented prior to resuming enrollment and are consulting with IRBs to determine the conditions that would allow for enrollment to continue.

The study investigators and study sponsor agreed to add two exploratory COVID-19 endpoints to this study. Firstly, in order to describe COVID-19 among the study population, to differentiate COVID-19 from pulmonary TB, and to minimize risks to participants and staff, a diagnostic test for SARS-CoV-2 (based on nucleic acid amplification) will be performed if a participant presents with suspected COVID-19. A description of the frequency and symptomatology of COVID-19 by treatment group is of great scientific interest as BCG-induced trained immunity has been hypothesized to potentially offer protection from progression to severe COVID-19. BCG vaccination is currently being tested as a prophylactic intervention against COVID-19 (NCT04327206 and additional studies).

Secondly, to describe the penetrance of SARS-CoV-2 infection in the communities that participate in this study, SARS-CoV-2 serology will be performed once every six months for a duration of 24 months. This will aid our understanding of virus transmission in the communities we work in and may allow a better understanding of the frequency of asymptomatic illness.

12. Section 4 and 1.1: Study sites: ***Five*** ~~Two~~ or more sites in South Africa will participate in this study.
13. Section 4.1 Scientific Rationale for Study Design

Participants who convert to QFT-positive at Day ~~70~~ ***71*** will be excluded from the modified intention to treat efficacy analyses and the per protocol immunogenicity analyses as they may have been infected with Mtb prior to or shortly after revaccination.

14. Section 5.2 Exclusion Criteria

Medical Conditions and History:

8.Acute illness on Study Day 1 ***(refer to Section 5.4 regarding rescreening)***

Other Exclusions

25. Shared residence with an individual who is receiving TB treatment or with someone who is known to have incompletely treated TB. e.g., ***Xpert MTB/RIF assay-positive***, PCR-positive, culture-positive, smear-positive TB, or clinically diagnosed unconfirmed TB.

15. Section 5.4 Screen failures

Minimal information includes demography, screen failure details, eligibility criteria,~~-~~***and any AE or SAE from the time of consent.***

Rescreening is permitted only if a participant presents with an acute illness (e.g., elevated temperature, ***acute respiratory or gastrointestinal illness, UTI) or an abnormal urine analysis due to menstruation. or urinary tract infection),*** and meets all other inclusion/exclusion criteria and is rescreened within the originally defined ***interval*** ~~screening window~~ (Section 1.3).

~~Rescreened~~ **P**~~p~~articipants ***who are rescreened within the allowed interval,*** *should* be assigned the same participant number as for the initial screening. Refer to Appendix 1, Section 10.1.4 regarding consent process for rescreened subjects. ***Refer to Section 7.4.2 for rescreening due to COVID-19.***

16. Section 6.2 Preparation/Handling/Storage/Accountability

Vaccine will be prepared as per the manufacturer's recommendations by the study pharmacist. ~~from multi-dose vials and dispensed according to the package insert.~~ ***All of the participating sites have investigational pharmacies and will use an unblinded pharmacist. Once the vaccine is prepared, the pharmacist will cover the vaccine syringe content with a blinding label so that the clinical staff remains blinded. Vaccine will NOT be administered by the study pharmacist.***

BCG must be stored at 2°C to 8°C in a secured location with no access for unauthorized personnel. Reconstituted vaccine may be kept at 2°C to 8°C for up to 6 hours ***in the original vial.***

17. Section 6.3 Randomization

To minimize risk of study failure by maximizing consistency of study assumptions with available literature data, the overall proportion of patients randomized in the youngest age strata (10-11 years old) and outside the Western Cape Province will be actively monitored

and capped at ***approximately 10% and 20% of the randomized study population, respectively.*** ~~***180*** subjects and 360 subjects, respectively~~.

18. Section 7 Discontinuation of Study Intervention and Participant Discontinuation/Withdrawal

Added: ***Refer to Section 7.4, COVID-19 Contingency Plans for details regarding study pause due to COVID-19.***

19. Section 7.1.1 Pausing Rules for Study

- 2. 1% of the safety population and at least two participants experience an SAE judged as related to ***BCG*** vaccination by the investigator
- 3. 20% of participants and at least ten participants experience a Grade 3 or higher event related to ***BCG vaccination*** ~~study vaccine~~, as judged by the investigator, with the exception of expected local BCG reactions or local injection site reactions that decrease to < Grade 3 within 48 hours.

20. Added Section 7.4 COVID-19 Contingency Plans

COVID-19 Contingency Plans

7.4.1 Rationale and Plan

The Coronavirus Disease 2019 (COVID-19) pandemic has impacted the world since the beginning of 2020, severely affecting people and societies around the globe.

This study was paused for enrollment by the sponsor, effective 19 March 2020, in order to support social distancing and to help health care professionals address COVID-19.

At the time of the enrollment pause, 349 participants had received investigational product (BCG or placebo) and were on-study.

The Government of South Africa declared a National Disaster on 15 March 2020 and ordered a first lockdown (stay-at-home order) on 26 March 2020. Under the lockdown, most trial participants are not able to come to the investigational site for protocol-specified visits. The sponsor and investigators therefore needed to change study procedures and agreed on telephone contacts as an alternative to study visits for safety assessments, for the duration of the Government restrictions, and/or risks that prevent in-person study visits, as judged by the site PI.

In accordance with the SAHPRA Policy on Conduct of Clinical Trials of Health Products During the Current COVID-19 Pandemic, issued 25 March 2020, this protocol amendment reflects the new and modified processes that were put in place in response to COVID-19. This protocol amendment, therefore, accommodates the changes related to the pandemic that needed to take place in order to continue the study in a manner that minimizes any immediate hazards, and also allows for more flexibility to capture study endpoints, and adds an assessment of SARS-CoV-2 infection and COVID-19 disease.

Changes in study visit procedures, missed visits, and participant discontinuations may lead to missing information (e.g., for protocol-specified procedures). We will capture as much information as possible in the case report form and explain the basis of the missing data.

Approaches used to protect trial participants and to manage study conduct during the COVID-19 pandemic, as well as control measures at study sites, will be documented.

Protocol deviations due to the COVID-19 pandemic will be described in the clinical study report (or in a separate study-specific document) including but not limited to the following:

- ***Contingency measures implemented to manage study conduct during disruption of the study as a result of COVID-19 control measures.***
- ***A listing of all participants affected by the COVID-19 related study disruption by unique participant number identifier and by investigational site, and a description of how the individual's participation was altered.***
- ***Analyses and corresponding discussions that address the impact of implemented contingency measures (e.g., alternative procedures used to collect critical safety and/or efficacy data) on the safety and efficacy results reported for the study.***

Study aspects affected by the study pause due to the COVID-19 pandemic are addressed in the following subsections.

7.4.2 Enrollment of new participants and status of previously enrolled participants

Enrollment was paused by the sponsor effective 19 March 2020 due to COVID-19 and will only resume once IRBs, investigators and the Sponsor agree that it is acceptable to resume study enrollment.

Many of the 349 participants on-study at the time of the lockdown will miss study visits and blood draws. A subset of these participants may not be eligible for inclusion in the primary endpoint analyses. In order to meet the primary study objective, the final sample size will be determined and documented once enrollment has resumed and we can understand the impact of the pause on the ability to address the primary objectives. The total randomized population may increase to a maximum of 2150 participants.

Participants who were screened but not randomized because the study was paused prior to randomization will be offered re-screening when enrollment resumes. Re-screening will also be allowed for any participant who screen-fails due to potential future COVID-19-related restrictions to movement or enrollment.

Participants who are offered re-screening due to COVID-19 will be assigned a new participant identifier (PID), and the new PID will be linked to the previously used PID to avoid double counting of the total number and percent failure of screened participants. Information collected from the re-screening will become the source information to be used to determine eligibility.

Extension of the estimated duration of enrollment (and therefore the overall estimated duration of the trial) will be required since the study was paused.

7.4.3 Study visits for enrolled subjects

The preference for study follow-up is to maintain in person study visits when it is acceptable and feasible to do so. Alternatively, follow-up visits can be performed by phone or another type of remote contact and will be captured in a consistent manner within the CRFs.

When participants are able to return to the site for onsite visits, they may be asked to complete visit assessments and procedures at a time point that is not specified in the protocol, in order to collect missing data and to perform missed procedures (e.g., blood draw or diary card collection). Once in-person visits become feasible, investigators will need to assess what visit the participant should be assigned to, based on guidelines in Table 7.

Refer to Section 7.4.5 regarding impact on mITT cohort for efficacy.

Table 6: Guidance for visits missed due to COVID-19-related restrictions, once in-person study visits become feasible

Screening visit	***Re-screening will be allowed if participants were unable to attend the Day 1 study visit due to COVID-19 restrictions or if they were previously screened but not randomized before the study was paused due to the COVID-19 pandemic.***
Day 8 visit	***If the interval between Day 1 and Day 8 visits is longer than 11 days, the PI will decide whether to invite the participant for a late Day 8 visit or wait until the date of a later***

	scheduled visit (e.g., Day 29, Day 71, or Month 6, etc.). Participants should be seen sooner rather than later, to ensure appropriate safety follow-up. ***At that visit, investigators will collect data for the missed Day 8 visit, if possible. For example, the diary card should be collected and data entered into the Day 8 visit CRF form with the actual diary card collection date.***
Day 29 visit	***If the interval between Day 1 and Day 29 visit is longer than 35 days, the PI will decide whether to invite the participant for a late Day 29 visit or wait until the date of a later scheduled visit (e.g., Day 71, Month 6, etc.). Participants should be seen sooner rather than later, to ensure appropriate safety follow-up.*** ***At that visit, investigators will collect data for the missed Day 29 visit, if possible. For example, the memory aid should be collected and data entered into the Day 29 visit CRF form with the actual memory aid collection date. The Day 29 blood draw should not be performed if the visit occurs after Day 35.***
Day 71 visit	***If the Day 71 visit was missed, the visit will be scheduled as soon as possible, in order to obtain the blood sample that determines eligibility of the mITT cohort (see Section 7.4.5). The Day 71 blood draw should be performed, regardless of when the Day 71 visit occurs.***
Month 6 visit and later visits	***If the Month 6 visit or later visits are missed, the participant should be seen soon after in-person study visits become feasible. At that visit, investigators will collect data for missed visits, if possible.***

7.4.4 Addition of exploratory objectives and endpoints related to COVID-19

The following exploratory objectives were added:

- ***To describe COVID-19 among study participants who present with suspected COVID-19***
- ***To describe the serostatus of SARS-CoV-2 infection.***

7.4.4.1 Serology for COVID-19

In order to describe the penetrance of SARS-CoV-2 infection in the communities that participate in this study, SARS-CoV-2 serology will be performed once every six months for a duration of 24 months. For enrolled participants, serology will be done on a blood sample drawn at the earliest possible study visit (e.g., the Month 6 or Month 12 visit, etc.) and at four subsequent biannual study visits.

For participants who are screened after SAHPRA and ethics committee approval of the Gates MRI-TBV01-201 Amendment Version 5, a blood sample will be collected at the screening visit, and at the Month 6, 12, 18 and Month 24 visits.

7.4.4.2.Diagnostic test for SARS-CoV-2

In order to describe signs and symptoms of COVID-19 among the study population, a diagnostic test for SARS-CoV-2 (based on nucleic acid amplification) will be performed if a participant presents with suspected COVID-19 (e.g., respiratory or febrile illness, anosmia, etc.). Signs and symptoms of illness are described as per investigator assessment (e.g., temperature, respiratory rate, oxygen saturation, medication, etc.).

A nasal or oropharyngeal sample will be obtained (e.g., swab, brush, scraping, or saliva) for the COVID-19 diagnostic test.

7.4.5 Changes to the definition of the mITT cohort for efficacy

Protocol version 4 (prior to COVID-19) defined a 70-day "wash-out" period to identify participants who may have been infected with Mtb just prior to or soon after enrollment and stated that participants who convert to QFT positive at Day 71 would be excluded from the primary mITT efficacy analysis.

Because of COVID-19, most of the study participants enrolled less than 70 days prior to the pause on March 19, 2020, missed or will miss their Day 71 visit. The amended protocol, Version 5, defines that participants who missed their Day 71 visit may be eligible for inclusion in the mITT cohort if they test QFT-negative, and are excluded from the mITT cohort if they test QFT-positive, at the first study visit after the COVID-19 related pause is lifted (e.g., late Day 71 visit or Month 6 visit). The mITT analysis will take the duration of follow-up after the QFT-negative test into consideration for the primary analysis.

The impact on data eligibility for certain protocol-defined analyses (e.g., mITT analysis) will be addressed in the Statistical Analysis Plan (SAP). Clear rules for how data obtained during out-of-window study visits will be used in endpoint analyses will be defined in the SAP.

7.4.6 Protocol deviations related to COVID-19

Tracking of all protocol deviations related to COVID-19 will be done proactively by sites and all parties involved. Deviations may include delayed participant visits, missed assessments, etc. Site staff will clearly document all COVID-19-related deviations in detail in participant source documentation, as well as in annotations in the EDC system, to ensure clarity for statistical analyses and clinical study reporting.

7.4.7 Informed consent and re-consent

Participants who were enrolled into the study will be re-consented/assented into the study so that they are aware of the protocol changes.

Newly screened participants, will be asked to sign the current consent/assent form which captures applicable protocol changes.

7.4.8 Safety precautions for trial staff after enrollment resumes

Principal Investigators and site personnel have a critical role in the conduct of the study to ensure safety of the trial participants and site staff. The sponsor, clinical research organizations, and principal investigators should adhere to applicable guidelines on measures to control transmission of COVID-19. Investigators and site staff should develop and implement site-specific procedures and processes as appropriate.

7.4.9 References

South African Clinical Research Association (SACRA) Clinical Trials Suggested Business Continuity Plan_COVID-19_Version 1; 16 March 2020.

SAHPRA Policy on Conduct of Clinical Trials of Health Products During the Current COVID-19 Pandemic; 25 March 2020 (adapted from: FDA's Guidance on Conduct of Clinical Trials of Medicinal Products During the COVID-19 Pandemic;18 March 2020).

21. Section 8: Study Assessments and Procedures

To evaluate eligibility criteria, ~~and a~~ medical history, ~~and~~ physical examination including vital signs***, and height and weight*** will be performed.

Added: ***If a planned study visit or planned study procedure (e.g., blood draw) cannot be performed because the participant has an acute illness or cannot access the study site (refer to Section 7.4 for COVID-19 related restrictions), the missed visit should be rescheduled as soon as feasible. Unscheduled visits should only take place as necessary and data from unscheduled study visits should be captured in the EDC.***

22. Section 8.1.1 Evaluation of initial Mtb infection and sustained infection

QFT assays will be performed on blood samples collected from all participants at screening, ~~on~~ ***at the*** Day 71 ***visit*** (end of the wash-out period), and every 6 months (counting from Day 1) through Month 48.

23. 8.2 Safety Assessments

Added: ***AEs will be collected from the time informed consent is obtained.***

24. 8.2.1 Physical Examination and Medical History

Added: ***Medical history will include questions regarding acute respiratory illness, febrile illness, and anosmia, e.g., due to SARS-CoV-2, influenza or other respiratory agents.***

~~All~~ ***Medical*** conditions that exist prior to ***the screening visit*** ~~administration of study intervention~~ will be recorded in the medical history.

Added: Height and body weight will be measured at ***screening*****,** Day 1, and every six months thereafter.

25. Section 8.2.1.1 Interim History and Focused Physical Examination

Added sentence: ***Interim medical history will also include questions regarding acute respiratory illness, febrile illness, anosmia, etc, e.g., due to SARS-CoV-2, influenza or other respiratory agents.***

26. Section 8.2.2 Pregnancy Status Assessment

If a pregnancy occurs after study start, ~~D~~***d***etails of ***the pregnancy*** ~~all pregnancies~~ will be collected (***refer to Section 10.3, Collection and Reporting Pregnancy Information).*** ~~*after* the start of study intervention and until the end of the study~~***. If delivery occurs after the final study visit, the investigator should attempt to maintain contact with the participant to obtain the information.***

27. Section 8.2.4 Diary Card and Daily Temperature Monitoring

On Day 1, participants will be given a diary card and receive guidance on how to fill in the card. Participants will also receive a digital thermometer.

The diary card will be used by the study participants/ ***or parent or guardian/LAR*** to record the duration and intensity (Grade 1, 2, 3, or 4) of solicited local and general AEs for 7 days following vaccination. The diary card will also allow for unsolicited AEs to be recorded.

The diary card will be collected and reviewed by the PI (or designee) on Day 8. No changes to the diary card will be permitted; however, any verbally recalled information provided by the participant or parent or guardian/LAR during review of the diary card will be documented in the source document and reported as an AE***, as applicable. Any participant-reported Grade 4 event will be assessed by the investigator.***

28. Section 8.2.5 Clinical Safety Laboratory Assessments

Added: Safety laboratory assessments will be performed at screening. ***Refer to the SoA Section Table 2.***

29. Section 8.2.6 Title of section changed from Site of Injection Examination to ***Injection Site*** ~~of~~ Examination

Added: ***The presence and maximum diameter of injection site reactions will be recorded as follows:***

- ***Redness, swelling, or induration ≤20 mm is considered as anticipated.***
- ***Redness, swelling, or induration >20 mm is reported as an AE.***
- ***Papule, vesicle, pustule, ulcer, or scar ≤5 mm is considered as anticipated.***
- ***Papule, vesicle, pustule, ulcer, or scar >5 mm is reported as an AE.***

This text was formatted in header style and was changed to normal text: The presence and maximum diameter of homolateral axillary lymph nodes larger than 1cm will be recorded.

30. Section 8.3 added SUSARs to first sentence.. The definitions of AEs, SAEs, serious ADRs, ***SUSARs***, and adverse events of special interest …

31. Section 8.3.1 Time Period and Frequency for Collecting AE and SAE Information

Unsolicited AEs	***Screening*** through Day 28 (inclusive)*****
All SAEs and AESIs	***Screening*** through Month 6*****

****Refer to the SoA Section 1.3 for collection of AEs from time of signed consent.***

32. Section 8.3.3.1 AE Intensity

Added: Section 10.2.5.1 ***and Appendix 5*** for details. ***Note that a body temperature between 37.5°C and 37.9°C, can be captured as an AE, at the PI's discretion.***

33. Section 8.3.4 Regulatory Reporting Requirements for SAEs

Changed serious ADR to SUSAR

All fatal and life-threatening serious ~~ADR~~ ***SUSARs*** are to be reported to South African Health Products Regulatory Authority (SAHPRA) within 7 calendar days after first knowledge with a complete report to be submitted within an additional 8 calendar days. ~~Serious ADRs~~ ***SUSARs*** that are not fatal or life threatening need to be reported to SAHPRA no later than 15 calendar days after first knowledge. An investigator who receives a ~~Serious ADRs~~ ***SUSAR*** report or other specific safety information (e.g., summary or listing of SAEs) from the sponsor will review and file it. A tracker of all such reports received from the CRO will be maintained by the site. The PI will notify the IRB/IEC, if appropriate and according to local requirements. IRB/IEC submissions will be conducted per the local IRB/IEC SOP.

The sponsor will prepare, distribute and submit ~~Serious ADRs~~ ***SUSAR*** reports according to local regulatory requirements. The CRO, on behalf of the sponsor will submit the ~~Serious ADRs~~ ***SUSAR*** reports to SAHPRA.

34. Section 8.3.6 Latent TB infection: edited as follows

Participants in this trial will be managed according to ***current*** national and/or international guidelines, in accordance with ethical standards. ~~The South African National TB Programme (NTP) does not currently recommend preventive therapy for HIV-uninfected adults and children older than 5 years of age with asymptomatic (latent) *Mtb* infection, due to the high risk of reinfection after completion of preventive therapy. However, a~~ ***A*** recent international guideline (*Latent tuberculosis infection. Updated and consolidated guidelines for programmatic…*

35. Section 8.4 Treatment of Overdose

~~Not applicable~~ ***If a dosing error occurs, the investigator, safety physician and medical monitor need to be informed as soon as possible.***

36. Section 9.2 Sample Size Determination

Added ***Due to COVID-19 related restrictions, it will take longer to complete enrollment and to reach the number of events needed for analyses.***

~~Approximately 5625 participants will be screened to enroll and randomize 1800 healthy participants 1:1 to either intervention arm.~~***Approximately 6720 participants will be screened to enroll and randomize up to 2150 participants.***

Table 6 re-numbered to Table ***7***.

37. Section 9.3 Table 7 re-numbered to Table ***8***.

Table ***8***: under Description edited text for mITT and per Protocol: All participants randomly assigned to study intervention, who received the study intervention and are QFT negative ***at the*** Day 71 ***visit.***

38. Section 9.4 Statistical Analyses

Table 8 re-numbered to Table ***9***.

39. Section 9.4.3.2 Other Exploratory Analyses

Added

The SARS-CoV-2 serostatus will be summarized overall, as well as by time and treatment group. Among participants who present with suspected COVID-19, the proportion of participants with positive nucleic acid amplification will be summarized overall and by treatment group. Signs and symptoms of disease will be described for each participant.

40. Section 10.2.3. edited section title: Definition of ~~Serious Adverse Drug Reaction (~~Serious ADR ***and SUSAR***

Edited text: When an adverse event is judged to be serious and related to an investigational product, it is a serious ADR ~~and is subject to expedited reporting based on the parameters of this study~~. ***A serious unexpected suspected adverse drug reaction is referred to as a SUSAR. Refer to Section 10.2.6 for the different reporting requirements.***

41. Section 10.2.5.1 Assessment of Intensity

Added: ***Note that a body temperature between 37.5°C and 37.9°C, can be captured as an AE, at the PI's discretion.***

42. Section 10.2.6.SAEs, AESIs, ~~and~~ Serious ADR, and SUSAR Reporting

- All fatal and life-threatening ~~adverse drug reactions (ADRs)~~ ***SUSARs*** are to be reported to SAHPRA with 7 calendar days after first knowledge with a complete report to be submitted within an additional 8 calendar days. ~~Serious ADRs~~ ***Other SUSARs*** that are not fatal or life threatening need to be reported to SAHPRA no later than 15 calendar days after first knowledge. The sponsor will notify the IDMC of all ~~Serious ADRs~~ ***SUSARs*** within the same timelines the report is sent to investigators, health authorities, IRB/IEC and other relevant parties.

43. Figure 2 title edited: SAE and ~~Serious ADR~~ ***SUSAR*** Reporting Scheme and same change in bottom 2 boxes in figure.
44. Section 10.3 Appendix 3 Contraceptive Guidance and Collection of Pregnancy Information Edited:
    • The health status of the mother and child, the date of delivery, and the child's sex, birth weight and ~~multi~~parity should be recorded and be reported to the Medical Monitor after delivery.

45. Section 10.5 Appendix 5: Toxicity Table

Site Reactions to Injections and Infusions

Previous version:

~~Parameter~~	~~Grade 1 Mild~~	~~Grade 2 Moderate~~	~~Grade 3 Severe~~	~~Grade 4 Potentially Life-Threatening~~
~~Injection Site Erythema or Redness[1]~~ ~~*Report only one*~~ ~~*> 15 years of age*~~	~~2.5 to < 5 cm in diameter~~ ~~OR~~ ~~6.25 to < 25 cm^2 surface area~~ ~~AND~~ ~~Symptoms causing no or minimal interference with usual social & functional activities~~	~~≥5 to <10 cm in diameter or ≥25 to < 100 cm^2 surface area~~ ~~OR~~ ~~Symptoms~~ ~~causing greater than minimal interference with usual social & functional activities~~	~~≥10 cm in diameter~~ ~~OR~~ ~~≥100 cm^2 surface area~~ ~~OR~~ ~~Ulceration~~ ~~OR~~ ~~Secondary infection~~ ~~OR~~ ~~Phlebitis~~ ~~OR~~ ~~Sterile abscess~~ ~~OR~~ ~~Drainage~~ ~~OR~~ ~~Symptoms causing inability to perform usual social & functional activities~~	~~Potentially life-threatening consequences (eg, abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue)~~
~~Injection Site Erythema or Redness[1]~~ ~~*Report only one*~~ ~~*≤ 15 years of age*~~	~~≤ 2.5 cm in diameter~~	~~> 2.5 cm in diameter with < 50% surface area of the extremity segment involved (eg, upper arm or thigh)~~	~~≥ 50% surface area of the extremity segment involved (eg, upper arm or thigh)~~ ~~OR~~ ~~Ulceration~~ ~~OR~~ ~~Secondary infection~~ ~~OR~~ ~~Phlebitis~~ ~~OR~~ ~~Sterile abscess~~ ~~OR~~ ~~Drainage~~	~~Potentially life-threatening consequences (eg, abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue)~~

New version

Injection Site Erythema or Redness[1] *Report only one* *> 15 years of age*	2.1 to < 5 cm in diameter	≥5 to <10 cm in max diameter or ≥ 25 to < 100 cm^2 surface area OR Symptoms causing greater than minimal interference with usual social & functional activities	≥10 cm in diameter OR ≥100 cm^2 surface area OR Phlebitis OR Sterile abscess OR Symptoms causing inability to perform usual social & functional activities	Potentially life-threatening consequences (eg, abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue) OR Hospitalization indicated
Injection Site Erythema or Redness[1] *Report only one* *≤ 15 years of age*	2.1cm to ≤ 2.5 cm in diameter	> 2.5 cm in diameter with < 50% surface area of the extremity segment involved (eg, upper arm or thigh)	≥ 50% surface area of the extremity segment involved (eg, upper arm or thigh) OR Phlebitis OR Sterile abscess	Potentially life-threatening consequences (eg, abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue) OR Hospitalization indicated

Note: Minor typos were corrected throughout.

Note that Version 5 was modified to Version 6 before it was presented to the IRBs or SAHPRA.

Version 6, 23 June 2020 – This version was used for study initiation.

Updated protocol Version 5 11 May 2020 to allow for repeat blood samples for screening laboratory tests due to unevaluable samples to enable evaluation of study eligibility. Moreover, text was modified to also allow for re-sampling for QFT testing at any timepoint, if the first sample was unevaluable.

Other edits were made for clarity or to update information.

Changes from Amendment 5 to Amendment 6 by Section

New text indicated with *bold italics*

Bill and Melinda Gates changed to Bill ***&*** Melinda Gates.

Address changed from 245 Main Street …MA 02142 changed to ***One Kendall Square, Building 600, Suite 6-301,*** …MA 021***39***

Changes to the Schedule of events (SoA) Table

Body temperature row was added since it will be taken at both visits as stated elsewhere in the protocol. Notes were added to screening urinalysis, QFT, HIV testing, safety labs, and SARS-CoV-2 serology to clarify procedure for unevaluable participants.

Table 2 Scheduled Visits and Activities

Study Visit Day (D) or Month (M)	**Screen D -28 to D -1**	**D1**	**Notes**
Body temperature	***X***	***X***	
Urinalysis	X		***May be repeated as described in Section 8.***
QFT assay (mL)	4		***[f] regarding unevaluable sample***
HIV test (mL) with pre- and post-test counselling	3		At screening, and M12, 24, 36, 48, and if diagnosed with active TB disease. ***[g] regarding unevaluable screening sample***
Serum chemistry (mL)	5		***[g] Regarding unevaluable screening sample***
CBC, differential (mL)	2.5		
SARS-CoV-2 serology (mL)	5		If enrolled before study pause: collect at the next feasible biannual visit (e.g., M6, M12, etc) and biannually thereafter ~~to Month 24~~ ***for 24 months***

[f] QFT sampling: If the blood sample collected for QFT cannot be evaluated (e.g., insufficient volume; damage, hemolytic or lost sample, etc.), collection of a second (repeat) blood sample is permitted to enable evaluation of the primary efficacy endpoint. If the QFT sample was successfully run and has a reported result, including "indeterminate", collection of a second (repeat) blood sample is not permitted.

[g] If a blood sample collected for screening cannot be evaluated (e.g., insufficient volume; damaged, hemolytic or lost sample, etc.) a second (repeat) blood collection is permitted to enable evaluation of study eligibility. The repeat blood collection must occur within the protocol-defined screening visit window. If a screening blood sample was successfully run and has a reported result to determine eligibility, a second (repeat) blood collection is not permitted.

[f] changed to [h] and [g] changed to [i]

Section 5.2 Exclusion criteria:

8. Acute illness on Study Day 1 (refer to Section 5.4 ~~regarding rescreening~~ ***for additional details). NOTE: This is a temporary exclusion for which the subject may be re-evaluated***.
9. Body temperature ≥37.5°C on Study Day 1 (***refer to Section 5.4 for additional details). NOTE: This is a temporary exclusion for which the subject may be re-evaluated.***

Section 5.4 Screen Failures

~~Rescreening is permitted only if a participant presents with an acute illness (e.g., elevated temperature, acute respiratory or gastrointestinal illness, UTI) or an abnormal urine analysis (e.g., due to menstruation or urinary tract infection), and meets all other inclusion/exclusion criteria and is rescreened within the originally defined interval (Section 1.3).~~

~~Participants who are rescreened within the allowed interval, should be assigned the same participant number as for the initial screening. Refer to Appendix 1, Section 10.1.4 regarding consent process for rescreened subjects. Refer to Section 7.4.2 for rescreening due to COVID-19.~~

Screening assessments can be done at any time during the screening interval, except for the written informed consent, which must be completed prior to any screening procedure.

If a participant presents with an acute illness (e.g., elevated temperature, acute respiratory or gastrointestinal illness, UTI) or an abnormal urinalysis (e.g., due to menstruation or urinary tract infection), repeat procedures, with the exception of blood collection (unless allowed as described in section 8), may be performed as long as they are completed within the screening window.

Figure 1 Outline of study procedures- screening timepoints corrected to be consistent with SoA table (Day -28 to ~~Day -7~~ corrected to Day -28 to Day ***-1***)

Section 7. Edited the title: Discontinuation of Study Intervention and Participant Discontinuation/Withdrawal ***and COVID-19 Contingency Plans***

Section 8 Study Assessments and Procedures

Added: ***Screening assessments can be done at any time during the period, except for the written informed consent, which must be completed prior to any screening procedure.***

Repeat collection and testing

Note that if a blood sample collected for screening cannot be evaluated (e.g., insufficient volume; damaged, hemolytic, or lost sample, etc.) a second (repeat) blood collection is permitted to enable evaluation of study eligibility.

The repeat blood collection must occur within the protocol-defined screening visit window. If a screening blood sample was successfully run and has a reported result to determine eligibility, a second (repeat) blood collection is not permitted.

A second sample for urinalysis may also be repeated if the screening sample is unevaluable or if it is abnormal due to menstruation or suspected urinary tract infection. Urinary tract

infection should be resolved prior to the repeat urinalysis and the repeat test must be performed during the screening interval. If repeat urinalysis is abnormal, participant will be a screen failure.

In addition, if the blood sample for QFT collected at any time point after screening is unevaluable, collection of a second (repeat) blood sample is also permitted to enable evaluation of the primary efficacy endpoint. The repeat blood collection must occur within the protocol-defined visit window.

If the QFT sample was successfully run and has a reported result, including "indeterminate", collection of a second (repeat) blood sample is not permitted.

Section 8.2.5: Clinical Safety Laboratory Assessments: Added ***Refer to Repeat Collection and Testing in Section 8 for unevaluable blood samples.***

Version 7: 21 July 2023

Refer to high level summary of changes on page 4.

All changes, including minor changes, are provided in the track changes version.