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Assessing the safety and pharmacokinetics of the anti-HIV monoclonal antibody CAP256V2LS alone and in combination with VRC07-523LS and PGT121 in South African women: study protocol for the first-in-human CAPRISA 012B phase I clinical trial

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Complete List of Authors:
Mahomed, Sharana; Centre for the Aids Programme of Research in South Africa,
Garrett, Nigel; CAPRISA
Karim, Quarraisha Abdool; CAPRISA; Columbia University, Department of Epidemiology, Mailman School of Public Health
Zuma, Nonhlanhla ; CAPRISA
capparelli, edmund; University of California San Diego
Baxter, Cheryl; CAPRISA
Gengiah, Tanuja; CAPRISA
Archary, Derseree; CAPRISA
Samsunder, Natasha; Centre for the Aids Programme of Research in South Africa
Rose, Nicole; National Institutes of Health
Moore, Penny; CAPRISA; National Institute for Communicable Diseases
Williamson, Carolyn; University of Cape Town, Division of Medical Virology, Institute of Infectious Disease and Molecular Medicine; National Health Laboratory Service
Barouch, Dan; Beth Israel Deaconess Medical Center, Center for Virology and Vaccine Research, Br, Boston, USA
Fast, Patricia; International Aids Vaccine Initiative
Pozzetto, Bruno; Saint-Etienne University Hospital Centre Geriatrics and Internal Medicine Section
Hankins, Catherine; AIGHD, Amsterdam Institute for Global Health and Development
Carlton, Kevin; National Institutes of Health
Ledgerwood, Julie; NIH
Morris, Lynn; National Institute for Communicable Diseases; CAPRISA
Mascola, John; NIH
Abdool Karim, Salim; CAPRISA; Columbia University, Department of Epidemiology, Mailman School of Public Health

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Protocol Paper - CAPRISA 012B

Title

Assessing the safety and pharmacokinetics of the anti-HIV monoclonal antibody CAP256V2LS alone and in combination with VRC07-523LS and PGT121 in South African women: study protocol for the first-in-human CAPRISA 012B phase I clinical trial

Authors

Sharana Mahomed[1], Nigel Garrett[1, 2], Quarraisha Abdool Karim[1,3], Nonhlanhla Yende-Zuma[1], Edmund Capparelli[4], Cheryl Baxter[1], Tanuja N. Gengiah[1], Derseree Archary[1], Natasha Samsunder[1], Nicole A. Doria-Rose[5], Penny L. Moore[1,6], Carolyn Williamson[1, 7, 8], Dan H Barouch[9], Patricia E Fast[10], Bruno Pozzetto[11], Catherine A. Hankins[12], Kevin Carlton[5], Julie E. Ledgerwood[5], Lynn Morris[1,6], John R. Mascola[5], Salim S. Abdool Karim[1,3]

Author affiliations

1. Centre for the AIDS Programme of Research in South Africa (CAPRISA), Durban, South Africa
2. Department of Public Health Medicine, School of Nursing and Public Health, University of KwaZulu-Natal, Durban, South Africa
3. Department of Epidemiology, Mailman School of Public Health, Columba University, New York, USA
4. University of California San Diego, San Diego, California, USA
5. Vaccine Research Center, National Institutes of Health, Bethesda, Maryland, USA
6. National Institute for Communicable Diseases of the National Health Laboratory Services, Johannesburg, South Africa
7. Division of Medical Virology, Institute of Infectious Disease and Molecular Medicine, University of Cape Town, Cape Town, South Africa.
8. National Health Laboratory Services of South Africa
9. Center for Virology and Vaccine Research, Beth Israel Deaconess Medical Center, Boston, USA
10. International AIDS Vaccine Initiative, New York, New York, USA
11. GIMAP (EA3064), University of Saint-Etienne/University of Lyon, Saint-Etienne, France
12. Amsterdam Institute for Global Health and Development

Corresponding Author

Sharana Mahomed

Address: Centre for the AIDS Programme of Research in South Africa (CAPRISA), 3 University Avenue, Durban, South Africa
Telephone number: 031 655 0610
Cell Number: 0813189848
Email address: Sharana.Mahomed@caprisa.org

Abstract: (297/300)

Introduction

New HIV prevention strategies are urgently required. The discovery of broadly neutralizing antibodies (bNAbs) has provided the opportunity to evaluate passive immunisation as a potential prevention strategy and facilitate vaccine development. Since 2014, several bNAbs have been isolated from a clade C infected South African donor, CAPRISA256. One particular bNAb, CAP256-VRC26.25, was found to be extremely potent, with good coverage against clade C viruses, the dominant HIV clade in sub-Saharan Africa. Challenge studies in non-human primates demonstrated that this antibody was fully protective even at extremely low doses. This bNAb was subsequently structurally engineered and the clinical variant is now referred to as CAP256V2LS.

Methods and analysis

CAPRISA 012B is the second of three trials in the CAPRISA 012 bNAb trial programme. It is a first-in-human, phase I study to assess the safety and pharmacokinetics of CAP256V2LS. The study is divided into four groups. Group 1 is a dose escalation of CAP256V2LS administered intravenously to HIV-negative and HIV-positive women. Group 2 is a dose escalation of CAP256V2LS administered subcutaneously, with and without the dispersing agent recombinant human hyaluronidase (rHuPH20) as single or repeat doses in HIV-negative women. Groups 3 and 4 are randomized placebo controlled to assess two (CAP256V2LS + VRC07-523LS; CAP256V2LS + PGT121) and three (CAP256V2LS + VRC07-523LS + PGT121) bNAb combinations administered subcutaneously. Safety will be assessed by the frequency of reactogenicity and adverse events related to study product. Pharmacokinetic disposition of CAP256V2LS alone and in combination with VRC07-523LS and PGT121 will be assessed via dose sub-groups and route of administration.

Ethics and dissemination

The University of KwaZulu-Natal Biomedical Research Ethics Committee and the South African Health Products Regulatory Authority have granted regulatory approval. Trial results

will be disseminated through conference presentations, peer-reviewed publications, and the clinical trial registry.

Registration details

PACTR202003767867253 (Status: Pre-Results)

Strengths and limitations of this study

- This is the first in-human trial to assess the safety and pharmacokinetics of the antibody CAP256V2LS
- The trial will provide new safety, pharmacokinetic, and functional activity data for the administration of CAP256V2LS alone and in combination with VRC07-523LS and/or PGT121.
- The trial will provide new data on the administration of bNAbs via the subcutaneous route compared to the intravenous route.
- This trial is one of the first to assess the use of a recombinant human hyaluronidase (rHuPH20), a subcutaneous dispersing agent, together with an anti-HIV monoclonal antibody.
- Data from this trial could inform the development of a four- or six-monthly injectable HIV prevention technology for young women in sub-Saharan Africa.

Introduction

Despite extensive prevention and treatment efforts, South Africa remains the country worst affected by the HIV-AIDS pandemic (1). Here, young women carry a disproportionately high burden of the disease with a persistently high HIV incidence (2, 3). Insights from universal testing and treatment trials demonstrate that early treatment alone is not sufficient to reduce the number of new infections and achieve epidemic control, but that effective HIV prevention methods are also needed (4). In African women, clinical trials evaluating daily oral tenofovir disoproxil fumarate (TDF) and emtricitabine (TDF/FTC) for pre-exposure prophylaxis demonstrated inconsistent results, most likely owing to varying adherence levels (5). While an effective vaccine remains a major challenge, new HIV prevention strategies are urgently required (6).

The discovery of broadly neutralizing antibodies (bNAbs) has allowed scientists to evaluate passive immunisation as a potential HIV prevention strategy (7, 8). These antibodies are generally recovered from the memory B cells of chronically HIV-infected individuals and

effectively neutralize diverse strains of HIV-1 indicating their breadth of response. Pre-clinical studies have demonstrated that passive immunisation using bNAbs protects rhesus macaques from simian-human immunodeficiency virus (SHIV) infection (9-12). However, there are currently no clinical trial data that show the ability of bNAbs to prevent HIV-1 infection in humans (13).

In 2014 and subsequently, several bNAbs targeting the V2 region of the HIV-1 envelope glycoprotein were isolated from a South African donor participating in the Centre for the AIDS Programme of Research in South Africa (CAPRISA) 002 Acute Infection study (14, 15). This study was established in 2004 in KwaZulu-Natal, South Africa and followed HIV-negative participants for identification of subsequent HIV seroconversion. This participant was infected with a clade C virus and superinfected with a different clade C virus, 15 weeks later (16). One particular bNAb, referred to as CAP256-VRC26.25, was isolated and found to be 10 times more potent than the previously published members of this lineage. Its overall potency (IC_{50} = 0.001 ug/ml) was comparable to, or better than that of existing bNAbs (17).

The exceptional potency of this antibody may be related to the reduced dependence on the N160 glycan, the unique long heavy-chain complementarity-determining region 3 (CDRH3) conformation, or other structural features that have yet to be identified (18-21). Further research using site-directed mutagenesis allowed for the manufacturing of an improved LS version of CAP256-VRC26.25. This mutation increases the binding affinity for the neonatal Fc-receptor (FcRn), resulting in an increased recirculation of functional immunoglobulin G (IgG), thereby increasing plasma half-life (22). *In vivo* studies demonstrated that CAP256-VRC26.25LS was fully protective against a SHIV challenge in monkeys, even at the lowest dose of 0.08mg/kg, with protection achieved at serum antibody concentrations of <0.75 µg/m (23). This was the lowest dose of any bNAb to show protection in monkeys (24).

Furthermore, when tested against a panel of 200 acute infection clade C pseudoviruses, CAP256-VRC26.25 emerged as the most potent member of bNAbs targeting the V2 loop (10, 17). The neutralization profile of CAP256-VRC26.25LS was particularly well suited as a complementary bNAb in combinations with bNAbs targeting other epitopes (17). CAP256-VRC26.25LS was subsequently engineered to prevent proteolytic clipping of the heavy chain through mutation of the lysine at position 100 to an alanine (25). This single amino acid change, K127A, was made in the CDRH3 region to improve manufacturability without altering neutralization potency or breadth. This non-clipped variant of the antibody is referred to as CAP256V2LS.

Pre-clinical studies in rhesus macaques demonstrated that CAP256V2LS has a half-life of 14.3 days when administered via the intravenous (IV) route and 9.9 days when administered via the subcutaneous (SC) route. Toxicology reports showed that CAP256V2LS displayed low poly-specific auto-reactivity to HEp-2 cells and cardiolipin (26). Its high potency and good breadth against clade C HIV viruses and long half-life make CAP256V2LS an excellent candidate for further clinical development. This is particularly important for southern Africa, where clade C is the dominant circulating virus.

The research and development pathway of the CAP256V2LS bNAb led to the CAPRISA 012 clinical trial programme that consists of three trials conducted in South Africa (Figure 1). This programme aims to evaluate the concept of bNAbs as long-acting pre-exposure prophylaxis with two to three SC doses per year to reduce HIV incidence among young women in three trials: CAPRISA 012A, a phase 1 trial of VRC07-523LS and PGT121 in HIV-negative women, previously described in this journal (27), CAPRISA 012B, described here, and CAPRISA 012C, a phase 2 combination bNAb trial in young women.

In CAPRISA 012B, CAP256V2LS will be assessed alone and in combination with VRC07-523LS and/or PGT121. VRC07-523LS targets the HIV-1 Envelope CD4 binding site and PGT121 targets the V3 glycan-dependent epitope region of the HIV Envelope protein. Given the vast genetic diversity of HIV-1, the use of multiple bNAbs may be required to ensure adequate coverage of circulating strains. Recent clinical trial data demonstrate that VRC07-523LS is safe with a half-life of 38 days after IV administration and 33 days after SC administration (28). Preliminary clinical trial data of PGT121 also demonstrate safety with a half-life of 22 days (29).

This trial is also one of the first to assess the use of a recombinant human hyaluronidase (rHuPH20) together with an anti-HIV monoclonal antibody. rHuPH20 is the active ingredient of the investigational ENHANZE™ Drug Product (EDP) and optimizes the SC delivery of co-administered therapeutics by depolymerizing hyaluronan in the extracellular matrix of the SC space that normally serves to restrict increased flow volumes. EDP allows co-mixing of antibodies with rHuPH20 at the clinical site. Clinical trials conducted in oncology have demonstrated safety and favourable results with this product (30).

Methods and analysis

Patient and public involvement

The CAPRISA Community Advisory Board (CAB) plays a central role in study planning and recruitment of participants. The CAB includes local community leaders, traditional leaders, leadership of local HIV/AIDS organisations, local health service provider representatives, previous study participants, and HIV-positive local community members. During trial preparation and prior to study start, the study concept is presented to the CAB members for their feedback. Recruitment will include community events, local clinics, street recruitment, and use of snowballing techniques. A recruitment and retention plan will be drawn up at study start and will be reviewed and updated regularly.

Study setting

The CAPRISA 012B trial will be conducted at the CAPRISA eThekwini Clinical Research Site (ECRS). This site is based in a busy commuter area in the city centre of Durban, KwaZulu-Natal, South Africa.

Study population selection

The study population will consist of 66 women, 52 HIV-negative and 14 HIV-positive women who have not yet started antiretroviral therapy (ART). All eligibility criteria must be met, and HIV-positive women will be recruited based on additional inclusion criteria (Table 1).

Study Design

CAPRISA 012B is a first-in-human, phase I study to assess the safety, tolerability, and pharmacokinetics (PK) of CAP256V2LS. The study is divided into four groups (Table 2). Group 1 is a dose escalation of CAP256V2LS administered IV to HIV-negative and HIV-positive women. Group 2 is a dose escalation of CAP256V2LS administered SC with and without rHuPH20 at a single or repeat dose to HIV-negative women. Groups 3 and 4 are randomized placebo controlled to assess two (CAP256V2LS + VRC07-523LS; CAP256V2LS + PGT121) and three (CAP256V2LS + VRC07-523LS + PGT121) bNAb combinations administered SC. Participants will be followed up for 24 weeks after the administration of the last dose of study product/s. HIV positive participants in Groups 1c and 1d will have two additional follow up visits after 12 and 24 months.

Study Objectives

Primary objectives

- To evaluate the safety and tolerability of CAP256V2LS administered
    - SC in HIV-negative women

- IV in HIV-negative and HIV-positive women
- SC in combination with VRC07-523LS and/or PGT121 in HIV-negative women

Secondary objectives

- To characterize the PK profile of CAP256V2LS administered SC as a single dose or as two doses 16 or 24 weeks apart
- To characterize the PK profile of CAP256V2LS administered in combination with VRC07-523LS and/or PGT121
- To characterize the PK profile of CAP256V2LS administered IV as a single dose in HIV-negative and HIV-positive women
- To evaluate the antiviral activity of CAP256V2LS administered IV to HIV-positive women not on ART
- To evaluate the concentrations and functional activity of CAP256V2LS in plasma and genital samples following SC and IV administration
- To determine whether administration of CAP256V2LS induces anti-monoclonal antibody responses
- To assess the acceptability of SC administration of monoclonal antibodies among participants

Primary Endpoints

- Proportion of participants with mild, moderate, and severe reactogenicity events within the first 3 days after IV or SC administration of CAP256V2LS
- Proportion of participants with mild, moderate, and severe adverse events (SAEs) related to the IV or SC administration of CAP256V2LS

Secondary Endpoints

- The difference in the elimination half-life, clearance, volume of distribution, and area under the concentration decay curve among study groups
- Change in plasma HIV-1 RNA levels from baseline
- Changes in the concentration of serum anti-CAP256V2LS titres from baseline
- The difference in the functional activity including IgG/IgA binding responses, cellular immune responses, and antibody function in plasma and mucosal surfaces compared to baseline
- Proportion of participants reporting CAP256V2LS injections to be acceptable as per study questionnaire

Sample size calculation

The main objective of this study is to assess the safety, tolerability, and PK of CAP256V2LS, hence the ability of the study to detect SAEs is key. The probability of detecting no SAE, at least one, or at least two SAEs at a specified true event rate will be calculated. These probabilities highlight the likelihood of the study to detect either rare or common adverse events (AEs). In addition, the 95% confidence interval for the true event rate was calculated. Currently, limited safety data are available to guide the estimation of the true event rate that might be observed in the study. In the absence of available AE rates for CAP256V2LS alone or in combination with other bNAbs, a range of hypothesized event rates to calculate the probability of observing no events, at least one event, or at least two events were used. Among the four participants receiving active product in each of the groups, there is a 34% chance of observing at least one event if the true event rate is 10%. When the true event rate is two or three times higher, this probability rises to 59% and 76% (Table 3).

Since the phase I assessment of the safety of CAP256V2LS administered SC includes eight participants receiving combination of CAP256V2LS and VRC07-523.LS (i.e. Groups 3a and 3b combined), for N=8, there is an 8% chance of observing at least one event, if the true event rate is 1%, but it is as high as 83%, if the true event is 20%. These probabilities are also applicable to the groups that will receive repeat doses (Group 2d and 2f). For the 16 participants receiving CAP256V2LS administered IV in Group 1, there is an 85% chance of observing no events if the event rate is 1%, and less than 1% chance, if the event rate is 30-fold higher. However, if all 56 women receiving CAP256V2LS active product at enrolment are combined, the former probability changes to 57% for no events and remains very low (<1%)-given the event rates of 10%, 20% and 30%. As expected, an increase in sample size, increases the likelihood of detecting rare events.

Study procedures

Informed consent

Prior to screening and enrolment, informed consent will be obtained from every participant in accordance with the South African Good Clinical Practice guidelines. The informed consent procedure will be conducted in either English or isiZulu as per participant preference. If a participant is illiterate, an impartial witness will be present throughout the informed consent procedure to ensure that all questions are answered to the satisfaction of the potential participant. For the purposes of this study, consent for all study related procedures, pharmacogenetic studies, and storage of specimens will be obtained.

Screening

All potential participants will complete the informed consent procedure for screening and provide relevant identification documents. In order to rule out co-enrolment in other intervention trials, potential participants' details will be checked on the Biometric Co-Enrolment Prevention System (BCEPS). The study team will also evaluate and review proof of contraceptive use, such as family planning records. HIV pre-counselling will be performed, and if the participant meets eligibility, the socio-demographic and behavioural questionnaires will be administered. Post-test counselling is provided after disclosure of all HIV results and if the participant is ineligible to enrol into the study, referral to one of several HIV/AIDS care programmes will be facilitated. HIV testing in this study will be conducted by following a study HIV algorithm. At screening, HIV testing will be performed using two rapid antibody testing kits. Any participant with discordant results will be regarded as ineligible and referred to medical care.

A comprehensive medical history and physical examination will be conducted to determine eligibility. All pre-existing conditions and concomitant medication information will be documented. Screening laboratory tests will be conducted as per the schedule of evaluations (SOE) to determine further eligibility. These tests include urinalysis, haematology, blood chemistry tests, liver function tests, testing for sexually transmitted infections including syphilis serology and hepatitis B virus assays. Serum and plasma specimens will also be stored, for further analyses. In addition, a genital specimen using a menstrual cup device will be obtained and stored (31).

Randomisation

An unblinded statistician who is not involved in study conduct, will be responsible for generating the randomization sequence (for groups 1c,1d, 3a to 3c and Group 4a) using SAS version 9.4 software or latest. Participants will be assigned to unique participant identification numbers stratified by HIV status and group number. Sequentially numbered, sealed, opaque envelopes containing the group number and envelope number or treatment code (for use by the unblinded pharmacist only) will be provided to the study coordinator, to be opened once a participant has been deemed eligible and is ready to be enrolled into the study.

The pharmacist will store envelopes in a secured location within the research pharmacy with access restricted to delegated study pharmacists only. The study pharmacist will also receive a randomization list consisting of the unique three-digit envelope number and study

group with the corresponding study drug or placebo and relevant dosage from the unblinded statistician. The envelope number will enable the unblinded pharmacist to assign the correct treatment to the correct participant.

Enrolment

Enrolment will take place within 56 days of screening. After the informed consent is obtained and eligibility criteria are met, the participant will be allocated to a study group (Table 2). At this visit, vital signs will be recorded, a targeted physical examination will be conducted and all laboratory results from the screening visit will be reviewed. For women of childbearing potential, a negative pregnancy test on that day must be obtained prior to product administration. Prior to the infusion/injection, the study team must ensure that the participant is eligible to receive study product. All study product administrations will be completed according to the assigned group and may be via an IV infusion or SC injection. Once safety has been established in the first participants, enrolment into the next groups as well as dose-escalation will take place in a sequential manner, following review at each step of safety data from the preceding groups. For the bNAb combination groups, two separate injections, each containing a single bNAb, will be administered. After receiving study product, all participants will be observed for a minimum of 1 hour after the first product and any repeat product administrations. PK analysis on blood draws will be conducted for both the IV and SC administration groups at timelines outlined in the SOE.

Management of HIV-positive participants enrolled into Group 1c and 1d

All potential volunteers who meet the eligibility criteria for enrolment will receive extensive ART counselling upon entering the study.The benefits of early ART initiation including the universal treatment policy, will be explained. Only participants who, after appropriate counselling, are willing to defer ART initiation will be eligible to enrol. At each study visit the participant's decision to defer ART initiation will be reviewed, and participants are allowed to change their mind and start treatment at any stage during the study. During the study, HIV positive participants will be monitored closely with regular clinical and safety assessments. HIV viral load monitoring and CD4 count measurements will take place as per SOE. Participants will be counselled on reducing the risk of HIV transmission to their sexual partners. Furthermore, a multidisciplinary approach to HIV transmission risk mitigation will be followed as per guidance from previous publications (32).

Specific criteria to initiate ART within 8 weeks of product administration are listed below and have been outlined as per guidance previously described for studies designed for ART interruption (33). Once any of these criteria are met, the participant will receive ART

counselling and will be initiated on ART as per South African guidelines. Criteria to start ART within 8 weeks of product administration include:

- Two consecutive viral loads >10,000 copies/ml after 2 weeks post product administration
- CD4 count <350 cells/mL
- Pregnancy
- If ART is deemed medically necessary
- If requested by the participant at any stage during the study

Safety monitoring

Reactogenicity assessments

After study product administration, the participant will be seen at the clinic for safety assessments on the day of product administration (Day 0), as well as Day 1, Day 2, and Day 3 (4 days in total). Clinicians and/or nurses will assess the product administration site(s) for local reactogenicity on the day of product administration and during the scheduled follow-up visits for all groups. Participants will keep a daily diary of local and systemic symptoms and record their temperature for three days after each product administration. In the event of a missed reactogenicity clinic visit, study staff will review the diary together with the participant once they present to the clinic and will determine the severity of the reactions. For any reactogenicity symptoms that are not resolved within 3 days, clinicians will follow and collect resolution information.

Safety monitoring

Safety monitoring includes internal monitoring by the study team, the protocol safety review team (PSRT), and data safety monitoring board (DSMB), as well as external monitoring including audits. Safety reporting of SAEs, AEs, and other important reportable events will be the responsibility of the entire study team. In addition, the study statistician will prepare routine study safety progress reports, which include reports of AEs experienced by study participants (blinded to treatment assignment), for review by the PSRT. The PSRT will review the clinical safety data on a weekly basis via electronic distribution of reports and will have face-to face meetings as required. The PSRT will be responsible for decisions related to participant safety. In addition, an independent DSMB will meet in-person and/or via teleconference bi-annually and review the study data and study conduct. The DSMB could recommend that the study should proceed as designed, should proceed with design modifications, or should be discontinued. Furthermore, the PI will permit authorized representatives such as external monitors and auditors to inspect the site facilities and records relevant to the study.

Follow up visits

All enrolled participants in both intervention and control arms will have follow up visits as specified in the SOE. At these visits, behavioural questionnaires will be administered together with HIV risk reduction counselling. Contraception counselling and provision will also take place. Targeted physical examinations will be conducted and all reactogenicity and AEs will be recorded. Laboratory investigations specified in the SOE will be conducted by the CAPRISA Research Laboratories. Where required, sample processing and storage of specimens for potential future testing (blood and vaginal specimens) will also be undertaken. An accredited contract laboratory will perform all safety blood testing and provide a backup laboratory service when required.

Statistical Analysis

Baseline characteristics including demographics and laboratory measurements will be summarized using descriptive statistics either by group or overall. Summaries of the number and percentage of participants experiencing any AE or reactogenicity will be analysed. AEs and SAEs will be coded into the Medical Dictionary for Regulatory Activities (MedDRA) preferred terms. The number and percentages of participants experiencing each specific AE will be tabulated by severity and relationship to study product. For the calculations in these tables, each participant's AE will be counted once under the maximum severity or strongest recorded causal relationship to study product. A complete listing of AEs for each participant will provide details including severity, relationship to study product, onset, duration and outcome. Tolerability evaluation will be mostly descriptive and consist of solicited AEs that occur within 1 hour following study product administration and reasons for any withdrawal or discontinuation based on participant discomfort. This early assessment of tolerability of the monoclonal antibodies will inform which parameters should be solicited or routinely assessed to further characterise the tolerability profile in a larger number of participants. Where appropriate, some of the data will be presented graphically. Analysis will be carried out using either SAS V.9.4 or higher or R.

Pharmacokinetic analysis

Pharmacokinetic disposition of CAP256V2LS alone and in combination with VRC07-523LS and PGT121 will be evaluated in this study and PK will be assessed via dose sub-groups and route of administration. Sixteen participants will receive CAP256V2LS by the IV route. The two lower dose levels will be administered to HIV-negative participants and the highest dose level will be administered to HIV-positive participants. These PK data will serve to characterize dose effects on CAP256V2LS clearance, volumes of distribution, and

elimination half-life. The PK data across Group 1 will also enable a preliminary evaluation of the potential impact of HIV status on CAP256V2LS PK. The PK results from the IV administration groups will be compared to the data from the SC administration groups in order to estimate bioavailability.

A total of 24 participants will receive CAP256V2LS by the SC route, either as a single dose or a repeat dose. A subset of participants will receive CAP256V2LS with SC hyaluronidase to allow the larger doses of antibody to be administered SC. These data will allow concentration profiles, bioavailability, and absorption patterns to be established. The impact of repeat CAP256V2LS SC administration on PK will also be determined. The PK data from the combination groups (Group 3 and 4) will be compared with those from Group 2 to determine if co-administration of VRC07-523LS or PGT121 affects CAP256V2LS PK. In addition, the VRC07-523LS and PGT121 PK results will be compared with findings from PK studies in CAPRISA 012A and reported in the literature, to establish whether CAP256V2LS impacts on the PK of VRC07-523LS and PGT121.

Data management

Data for the CAPRISA 012B study will be collected on case report forms (CRFs) designed specifically to address the protocol requirements. Data will be managed by the CAPRISA Data Management department, using DFdiscover (DF/Net Research, Inc), software specifically designed for clinical trial data management. The site will record data on paper CRFs that will be directly captured onto the DFdiscover system and validated by the data management staff. All source documents will be kept in the participants' study files and medical charts at the clinical research site. All original CRFs and study related documents will be securely stored at the site, during the study and after study completion.

Ethics and dissemination

Regulatory approval has been granted by the University of KwaZulu-Natal (UKZN) Biomedical Research Ethics Committee and by the South African Health Products Regulatory Authority. The study team will disseminate the trial results by sharing with the scientific community at international conferences, through peer-reviewed journal publications, and presentations to the wider community. Trial results will be uploaded onto the UKZN repository and the Pan-African Clinical Trial Registry (PACTR).

Authors' contributions

SSAK and QAK conceived the trial. SSAK, QAK, SM and NG designed the trial. SM and NG wrote the study protocol. EC will conduct the PK simulations and analysis. NYZ performed

sample size calculations and the statistical analysis strategy. KC, NDR, PLM, JM and LM contributed to antibody development. CB, TNG, DA, NS, CW, DHB, JM, JL, CAH, BP and PEF contributed to the planning and conduct of the trial. All authors reviewed the manuscript and consented to publication.

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Competing interest statement

There are no competing interests to declare.

Patient consent for publication

Not required

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Tables

Table 1: Eligibility criteria

Inclusion criteria	Exclusion criteria	Additional inclusion criteria*
• Age 18 to 45 years • Born female • Able and willing to complete the informed consent process • Able to understand the information provided about the study, willing to comply with protocol procedures, and available to attend the study site for the duration of the study • Based on clinical assessment, participants must be in good general health as per opinion of the Principal Investigator (PI) or designee • Haemoglobin > 10g/dl • Neutrophil count within institutional normal range • Platelets within institutional normal range • Creatinine < 1.1 times upper limit of normal • Alanine aminotransferase < 1.25 times upper limit of normal • HIV-negative as per FDA-approved method of detection (for groups with HIV-negative participants only)	• Any clinically significant acute or chronic medical condition that in the opinion of the PI/designee makes the participant unsuitable for participation in the study, or jeopardises the safety or rights of the participant • If planning a pregnancy for the duration of the study, currently pregnant or breastfeeding • Exceeding 95 kilograms in body weight • A history of alcohol or substance use judged by the PI to potentially interfere with participant study compliance • Prior participation in an investigational HIV vaccine trial, except if proof of allocation to the placebo arm is available • Administration of a monoclonal antibody or polyclonal immunoglobulin within 28 days prior to enrolment • Any history of anaphylaxis and related symptoms such as hives, respiratory difficulty, or angioedema	• Confirmed HIV-1 infection prior to enrolment • An HIV viral load of >1000 copies/ml at screening • A CD4 count of ≥ 450 cells/µl at screening • ART naive, and willing to defer treatment (after appropriate counselling) for up to a maximum of 8 weeks after product administration • No major comorbidities or AIDS-defining illness

• Negative pregnancy test • If of reproductive potential, there is evidence of effective contraceptive use and willingness to adhere to effective contraceptive use during the study period • Willing to have blood samples collected, stored, and used for research purposes. • Willing to adhere to reduced risk sexual behaviour during study participation.	• Evidence of autoimmune disease or currently receiving immunosuppressive therapy • Involvement in other concurrent research studies that would interfere with the objectives of this study.	

*Additional eligibility criteria for HIV positive participants in Groups 1c and 1d

Table 2: CAPRISA 012B Group, Dose and Arm Allocation

Group	Participants	Regimen	SA site N=66	Dose (mg/kg)
Group 1: Dose escalation of IV administration of CAP256V2LS				
1a	HIV negative	CAP256V2LS	4	5 mg/kg IV one dose
1b	HIV negative	CAP256V2LS	4	10 mg/kg IV one dose
1c	HIV positive	CAP256V2LS	4/2§	20 mg/kg IV one dose
1d	HIV positive	CAP256V2LS	4/4§	20 mg/kg IV one dose
Group 2: Dose escalation of SC administration of CAP256V2LS				
2a	HIV negative	CAP256V2LS	4	5 mg/kg SC one dose
2b	HIV negative	CAP256V2LS*	4	5 mg/kg SC one dose
2c	HIV negative	CAP256V2LS*	4	10 mg/kg SC one dose
2d	HIV negative	CAP256V2LS*	4	10 mg/kg SC with one repeat dose at 16/24 weeks#
2e	HIV negative	CAP256V2LS*	4	20 mg/kg SC one dose
2f	HIV negative	CAP256V2LS*	4	20 mg/kg SC with one repeat dose at 16/24 weeks#

Group 3: Dose escalation of the two antibody combinations				
3a	HIV negative	CAP256V2LS* + VRC07-523.LS*	4/1[§]	10 mg/kg SC / 10 mg/kg SC one dose
3b	HIV negative	CAP256V2LS* + VRC07-523.LS*	4/1[§]	20 mg/kg SC / 20 mg/kg SC one dose
3c	HIV negative	CAP256V2LS* + PGT121[$]	4/1[§]	20 mg/kg SC / 5 mg/kg SC one dose
Group 4: Three antibody combination				
4a	HIV negative	CAP256V2LS* + PGT121[$] + VRC07-523.LS*	4/1[§]	20 mg/kg SC / 5 mg/kg SC / 20mg/kg SC one dose

*Antibody will be injected with hyaluronidase, so that the antibody dose can be administered as a single SC injection; [#] First two participants will receive two doses 24 weeks apart and the next two participants will receive two doses 16 weeks apart; [$] PGT121 may be replaced with PGT121LS at a higher dose, based on its availability at the time of study initiation, [§] Placebo allocation

Table 3: Probability of observing no events, at least 1 event, or at least 2 events, for a range of hypothetical true event rates

True event rate (%)	Number of participants	0 events	1+ events	2+ events
1	4	0.96	0.04	<0.01
	8	0.92	0.08	<0.01
	12	0.89	0.11	0.01
	16	0.85	0.15	0.01
	24	0.79	0.21	0.02
	56	0.57	0.43	0.11
5	4	0.81	0.19	0.01
	8	0.66	0.34	0.06
	12	0.54	0.46	0.12
	16	0.44	0.56	0.19
	24	0.29	0.71	0.34
	56	0.06	0.94	0.78
10	4	0.66	0.34	0.05
	8	0.43	0.57	0.19
	12	0.28	0.72	0.34
	16	0.19	0.81	0.49
	24	0.08	0.92	0.71

	56	<0.01	>0.99	0.98
20	4	0.41	0.59	0.18
	8	0.17	0.83	0.50
	12	0.07	0.93	0.73
	16	0.03	0.97	0.86
	24	<0.01	>0.99	>0.99
	56	<0.01	>0.99	>0.99
30	4	0.24	0.76	0.35
	8	0.06	0.94	0.74
	12	0.01	0.99	0.91
	16	<0.01	>0.99	0.97
	24	<0.01	>0.99	>0.99
	56	<0.01	>0.99	>0.99

References

1. UNAIDS. *UNAIDS data 2017*. Geneva, Switzerland: Joint United Nations Programme on HIV/AIDS (UNAIDS), 2017. http://www.unaids. org/ en/ resources/ documents/ 2017/ 2017_ data_ book (Accessed 01 April 2020).
2. de Oliveira T, Kharsany AB, Gräf T, et al. Transmission networks and risk of HIV infection in KwaZulu-Natal, South Africa: a communitywide phylogenetic study. Lancet HIV. 2017;4:e41–e50.
3. Dellar RC, Dlamini S, Karim QA. Adolescent girls and young women: key populations for HIV epidemic control. Journal of the International AIDS Society. 2015;18:19408.
4. Abdool Karim SS. HIV-1 Epidemic Control—Insights from Test-and-Treat Trials. Mass Medical Soc; 2019; 381:286-288
5. Fang W, Mphoyi BI, Motake DR, Liu J, Li W, Mei F, et al. Efficacy, Adherence and Side Effects of PrEP for HIV-1 Prevention. International Journal of Biology. 2019;11(4).
6. Fauci AS, Marston HD. Ending the HIV–AIDS pandemic—follow the science. New England Journal of Medicine. 2015;373(23):2197-9.
7. Anderson DJ, Politch JA, Vaca GB, Kadasia K, Whaley KJ. Use of Monoclonal Antibodies to Prevent the Sexual Transmission of Human Immunodeficiency Virus Type 1. Current Immunology Reviews. 2019;15(1):123-30.
8. Pegu A, Hessell AJ, Mascola JR, Haigwood NL. Use of broadly neutralizing antibodies for HIV-1 prevention. Immunological reviews. 2017;275(1):296-312.
9. Hessell AJ, Poignard P, Hunter M, Hangartner L, Tehrani DM, Bleeker WK, et al. Effective, low-titer antibody protection against low-dose repeated mucosal SHIV challenge in macaques. Nature medicine. 2009;15(8):951.
10. Walker LM, Huber M, Doores KJ, Falkowska E, Pejchal R, Julien J-P, et al. Broad neutralization coverage of HIV by multiple highly potent antibodies. Nature. 2011;477(7365):466.
11. Moldt B, Rakasz EG, Schultz N, Chan-Hui P-Y, Swiderek K, Weisgrau KL, et al. Highly potent HIV-specific antibody neutralization in vitro translates into effective protection against mucosal SHIV challenge in vivo. Proceedings of the National Academy of Sciences. 2012;109(46):18921-5.
12. Barouch DH, Whitney JB, Moldt B, Klein F, Oliveira TY, Liu J, et al. Therapeutic efficacy of potent neutralizing HIV-1-specific monoclonal antibodies in SHIV-infected rhesus monkeys. Nature. 2013;503(7475):224.
13. Sok D, Burton DR. Recent progress in broadly neutralizing antibodies to HIV. Nature immunology. 2018;19(11):1179.
14. van Loggerenberg F, Mlisana K, Williamson C, Auld SC, Morris L, Gray CM, et al. Establishing a cohort at high risk of HIV infection in South Africa: challenges and experiences of the CAPRISA 002 acute infection study. PloS one. 2008;3(4):e1954.
15. Doria-Rose NA, Schramm CA, Gorman J, Moore PL, Bhiman JN, DeKosky BJ, et al. Developmental pathway for potent V1V2-directed HIV-neutralizing antibodies. Nature. 2014;509(7498):55.
16. Sheward DJ, Marais J, Bekker V, Murrell B, Eren K, Bhiman JN, et al. HIV superinfection drives de novo antibody responses and not neutralization breadth. Cell host & microbe. 2018;24(4):593-9. e3.
17. Wagh K, Bhattacharya T, Williamson C, Robles A, Bayne M, Garrity J, et al. Optimal combinations of broadly neutralizing antibodies for prevention and treatment of HIV-1 clade C infection. PLoS pathogens. 2016;12(3):e1005520.
18. Moore PL, Gray ES, Sheward D, Madiga M, Ranchobe N, Lai Z, et al. Potent and broad neutralization of HIV-1 subtype C by plasma antibodies targeting a quaternary epitope including residues in the V2 loop. Journal of virology. 2011;85(7):3128-41.
19. Bhiman JN, Anthony C, Doria-Rose NA, Karimanzira O, Schramm CA, Khoza T, et al. Viral variants that initiate and drive maturation of V1V2-directed HIV-1 broadly neutralizing antibodies. Nature medicine. 2015;21(11):1332.
20. Doria-Rose NA, Bhiman JN, Roark RS, Schramm CA, Gorman J, Chuang G-Y, et al. New member of the V1V2-directed CAP256-VRC26 lineage that shows increased breadth and exceptional potency. Journal of virology. 2016;90(1):76-91.
21. Gorman J, Chuang G-Y, Lai Y-T, Shen C-H, Boyington JC, Druz A, et al. Structure of Super-Potent Antibody CAP256-VRC26. 25 in Complex with HIV-1 Envelope Reveals a Combined Mode of Trimer-Apex Recognition. Cell Reports. 2020;31(1):107488.
22. Gaudinski MR, Coates EE, Houser KV, Chen GL, Yamshchikov G, Saunders JG, et al. Safety and pharmacokinetics of the Fc-modified HIV-1 human monoclonal antibody VRC01LS: a phase 1 open-label clinical trial in healthy adults. PLoS medicine. 2018;15(1):e1002493.

23. Julg B, Tartaglia LJ, Keele BF, Wagh K, Pegu A, Sok D, et al. Broadly neutralizing antibodies targeting the HIV-1 envelope V2 apex confer protection against a clade C SHIV challenge. Science translational medicine. 2017;9(406):eaal1321.
24. Pegu A, Borate B, Huang Y, Pauthner MG, Hessell AJ, Julg B, et al. A Meta-analysis of Passive Immunization Studies Shows that Serum-Neutralizing Antibody Titer Associates with Protection against SHIV Challenge. Cell Host & Microbe. 2019;26(3):336-46. e3.
25. Ivleva VB, Schneck NA, Gollapudi D, Arnold F, Cooper JW, Lei QP. Investigation of sequence clipping and structural heterogeneity of an HIV broadly neutralizing antibody by a comprehensive LC-MS analysis. Journal of The American Society for Mass Spectrometry. 2018;29(7):1512-23.
26. Investigator's Brochure CAP256V2LS (VRC-HIVMAB0102-00-AB) A Broadly Neutralizing HIV-1 Human Monoclonal Antibody. Version 10 dated 12 February 2020 , Vaccine Research Centre.
27. Mahomed S, Garrett N, Capparelli E, Baxter C, Zuma NY, Gengiah T, et al. Assessing the safety and pharmacokinetics of the monoclonal antibodies, VRC07-523LS and PGT121 in HIV negative women in South Africa: study protocol for the CAPRISA 012A randomised controlled phase I trial. BMJ open. 2019;9(7):e030283.
28. Gaudinski MR, Houser KV, Doria-Rose NA, Chen GL, Rothwell RSS, Berkowitz N, et al. Safety and pharmacokinetics of broadly neutralising human monoclonal antibody VRC07-523LS in healthy adults: a phase 1 dose-escalation clinical trial. The Lancet HIV. 2019;6(10):e667-e79.
29. Stephenson K, Julg B, Ansel J, Walsh S, Tan C, Maxfield L, et al., editors. Therapeutic activity of PGT121 monoclonal antibody in HIV-infected adults. Conference on Retroviruses and Opportunistic Infections; Seattle, Washington; 2019.
30. Locke KW, Maneval DC, LaBarre MJ. ENHANZE® drug delivery technology: a novel approach to subcutaneous administration using recombinant human hyaluronidase PH20. Drug delivery. 2019;26(1):98-106.
31. Archary D, Liebenberg LJ, Werner L, Tulsi S, Majola N, Naicker N, et al. Randomized cross-sectional study to compare HIV-1 specific antibody and cytokine concentrations in female genital secretions obtained by menstrual cup and cervicovaginal lavage. PloS one. 2015;10(7).
32. Peluso MJ, Dee L, Campbell D, Taylor J, Hoh R, Rutishauser RL, et al. A collaborative, multidisciplinary approach to HIV transmission risk mitigation during analytic treatment interruption. Journal of Virus Eradication. 2020;6(1):34.
33. Julg B, Dee L, Ananworanich J, Barouch DH, Bar K, Caskey M, et al. Recommendations for analytical antiretroviral treatment interruptions in HIV research trials—report of a consensus meeting. Lancet HIV. 2019; 6(4), pp.e259-e268.

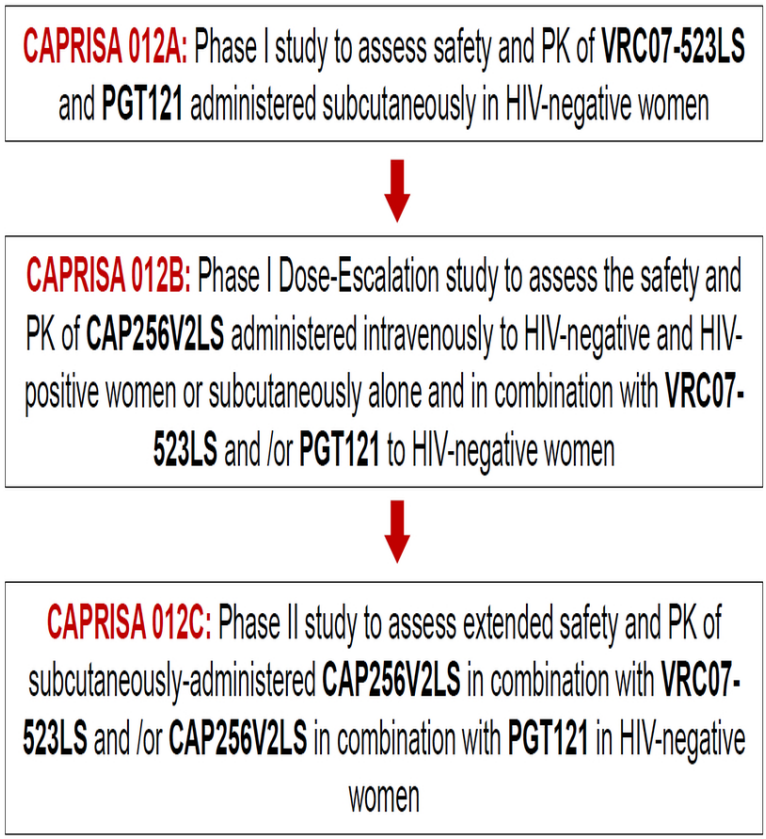

Figure 1: CAPRISA 012 Clinical Trial Programme

SPIRIT

STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	33
	2b	All items from the World Health Organization Trial Registration Data Set	1-13; Registered on the Pan African Clinical Trial Registry (PACTR202003767867253)
Protocol version	3	Date and version identifier	1-35
Funding	4	Sources and types of financial, material, and other support	3
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	2
	5b	Name and contact information for the trial sponsor	1
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	N/A The applicant is the sponsor

	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	26-27
Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	7-12
	6b	Explanation for choice of comparators	22-23
Objectives	7	Specific objectives or hypotheses	12-13
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	13-16
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	16
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	16-17
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	13-15
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	15,20
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	16, 20
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	18-19

Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	13
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	Schedule of Evaluations for each study group
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	27-29
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	17-18
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	13-14,19
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	19, 20, 23
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	23
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	23
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	23

Methods: Data collection, management, and analysis			
Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	29-30
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	20,26,27,32
Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	30-31
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	29-30
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	29-30
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	N/A
Methods: Monitoring			
Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	26-27
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	15,16,26,27
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	24-26
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	27

Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	31
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	31
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	31
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	31
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	33
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	2-3
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	33
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	18
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	33
	31b	Authorship eligibility guidelines and any intended use of professional writers	N/A
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	33
Appendices			

Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	31
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	21

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated.

BMJ Open

Assessing the safety and pharmacokinetics of the anti-HIV monoclonal antibody CAP256V2LS alone and in combination with VRC07-523LS and PGT121 in South African women: study protocol for the first-in-human CAPRISA 012B phase I clinical trial

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Secondary Subject Heading:	Infectious diseases, Epidemiology, Immunology (including allergy)
Keywords:	INFECTIOUS DISEASES, HIV & AIDS < INFECTIOUS DISEASES,

	Microbiology < PATHOLOGY, Public health < INFECTIOUS DISEASES, Epidemiology < INFECTIOUS DISEASES

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Protocol Paper - CAPRISA 012B

Title

Assessing the safety and pharmacokinetics of the anti-HIV monoclonal antibody CAP256V2LS alone and in combination with VRC07-523LS and PGT121 in South African women: study protocol for the first-in-human CAPRISA 012B phase I clinical trial

Authors

Sharana Mahomed[1], Nigel Garrett[1, 2], Quarraisha Abdool Karim[1,3], Nonhlanhla Yende-Zuma[1], Edmund Capparelli[4], Cheryl Baxter[1], Tanuja N. Gengiah[1], Derseree Archary[1], Natasha Samsunder[1], Nicole A. Doria-Rose[5], Penny L. Moore[1,6], Carolyn Williamson[1, 7, 8], Dan H Barouch[9], Patricia E Fast[10], Bruno Pozzetto[11], Catherine A. Hankins[12], Kevin Carlton[5], Julie E. Ledgerwood[5], Lynn Morris[1,6], John R. Mascola[5], Salim S. Abdool Karim[1,3]

Author affiliations

1. Centre for the AIDS Programme of Research in South Africa (CAPRISA), Durban, South Africa
2. Department of Public Health Medicine, School of Nursing and Public Health, University of KwaZulu-Natal, Durban, South Africa
3. Department of Epidemiology, Mailman School of Public Health, Columba University, New York, USA
4. University of California San Diego, San Diego, California, USA
5. Vaccine Research Center, National Institutes of Health, Bethesda, Maryland, USA
6. National Institute for Communicable Diseases of the National Health Laboratory Services, Johannesburg, South Africa
7. Division of Medical Virology, Institute of Infectious Disease and Molecular Medicine, University of Cape Town, Cape Town, South Africa.
8. National Health Laboratory Services of South Africa
9. Center for Virology and Vaccine Research, Beth Israel Deaconess Medical Center, Boston, USA
10. International AIDS Vaccine Initiative, New York, New York, USA
11. GIMAP (EA3064), University of Saint-Etienne/University of Lyon, Saint-Etienne, France
12. Amsterdam Institute for Global Health and Development

Corresponding Author

Sharana Mahomed

Address: Centre for the AIDS Programme of Research in South Africa (CAPRISA), 3 University Avenue, Durban, South Africa
Telephone number: 031 655 0610
Cell Number: 0813189848
Email address: Sharana.Mahomed@caprisa.org

Abstract: (297/300)

Introduction

New HIV prevention strategies are urgently required. The discovery of broadly neutralizing antibodies (bNAbs) has provided the opportunity to evaluate passive immunisation as a potential prevention strategy and facilitate vaccine development. Since 2014, several bNAbs have been isolated from a clade C infected South African donor, CAPRISA256. One particular bNAb, CAP256-VRC26.25, was found to be extremely potent, with good coverage against clade C viruses, the dominant HIV clade in sub-Saharan Africa. Challenge studies in non-human primates demonstrated that this antibody was fully protective even at extremely low doses. This bNAb was subsequently structurally engineered and the clinical variant is now referred to as CAP256V2LS.

Methods and analysis

CAPRISA 012B is the second of three trials in the CAPRISA 012 bNAb trial programme. It is a first-in-human, phase I study to assess the safety and pharmacokinetics of CAP256V2LS. The study is divided into four groups. Group 1 is a dose escalation of CAP256V2LS administered intravenously (IV) to HIV-negative and HIV-positive women. Group 2 is a dose escalation of CAP256V2LS administered subcutaneously (SC), with and without the dispersing agent recombinant human hyaluronidase (rHuPH20) as single or repeat doses in HIV-negative women. Groups 3 and 4 are randomized placebo controlled to assess two (CAP256V2LS + VRC07-523LS; CAP256V2LS + PGT121) and three (CAP256V2LS + VRC07-523LS + PGT121) bNAb combinations administered SC to HIV-negative women. Safety will be assessed by the frequency of reactogenicity and adverse events related to study product. Pharmacokinetic disposition of CAP256V2LS alone and in combination with VRC07-523LS and PGT121 will be assessed via dose sub-groups and route of administration.

Ethics and dissemination

The University of KwaZulu-Natal Biomedical Research Ethics Committee (BREC) and the South African Health Products Regulatory Authority (SAHPRA) have granted regulatory

approval (Trial reference numbers: BREC00000857/2019 and SAHPRA 20200123). Trial results will be disseminated through conference presentations, peer-reviewed publications, and the clinical trial registry.

Registration details

PACTR202003767867253 (Status: Pre-Results)

Strengths and limitations of this study

- This is the first in-human trial to assess the safety and pharmacokinetics of the monoclonal antibody CAP256V2LS.
- The trial investigates the administration of CAP256V2LS in combination with two potent antibodies, VRC07-523LS and PGT121.
- The trial assesses the subcutaneous administration of monoclonal antibodies for HIV prevention.
- The study evaluates the use of a dispersing agent, recombinant human hyaluronidase (rHuPH20), together with antibodies against HIV.
- The study is not powered to show efficacy of CAP256V2LS against HIV.

Introduction

Despite extensive prevention and treatment efforts, South Africa remains the country worst affected by the HIV-AIDS pandemic (1). Here, young women carry a disproportionately high burden of the disease with a persistently high HIV incidence (2, 3). Insights from universal testing and treatment trials demonstrate that early treatment alone is not sufficient to reduce the number of new infections and achieve epidemic control, but that effective HIV prevention methods are also needed (4). In African women, clinical trials evaluating daily oral tenofovir disoproxil fumarate (TDF) and emtricitabine (TDF/FTC) for pre-exposure prophylaxis demonstrated inconsistent results, most likely owing to varying adherence levels (5). While an effective vaccine remains a major challenge, new HIV prevention strategies are urgently required (6).

The discovery of broadly neutralizing antibodies (bNAbs) has allowed scientists to evaluate passive immunisation as a potential HIV prevention strategy (7, 8). These antibodies are generally recovered from the memory B cells of chronically HIV-infected individuals and effectively neutralize diverse strains of HIV-1 indicating their breadth of response. Pre-clinical studies have demonstrated that passive immunisation using bNAbs protects rhesus

macaques from simian-human immunodeficiency virus (SHIV) infection (9-12). However, there are currently no clinical trial data that show the ability of bNAbs to prevent HIV-1 infection in humans (13).

In 2014 and subsequently, several bNAbs targeting the V2 region of the HIV-1 envelope glycoprotein were isolated from a South African donor participating in the Centre for the AIDS Programme of Research in South Africa (CAPRISA) 002 Acute Infection study (14, 15). This study was established in 2004 in KwaZulu-Natal, South Africa and followed HIV-negative participants for identification of subsequent HIV seroconversion. This participant was infected with a clade C virus and superinfected with a different clade C virus, 15 weeks later (16). One particular bNAb, referred to as CAP256-VRC26.25, was isolated and found to be 10 times more potent than the previously published members of this lineage. Its overall potency (IC_{50} = 0.001 ug/ml) was comparable to, or better than that of existing bNAbs (17).

The exceptional potency of this antibody may be related to the reduced dependence on the N160 glycan, the unique long heavy-chain complementarity-determining region 3 (CDRH3) conformation, or other structural features that have yet to be identified (18-21). Further research using site-directed mutagenesis allowed for the manufacturing of an improved LS version of CAP256-VRC26.25. This mutation increases the binding affinity for the neonatal Fc-receptor (FcRn), resulting in an increased recirculation of functional immunoglobulin G (IgG), thereby increasing plasma half-life (22). *In vivo* studies demonstrated that CAP256-VRC26.25LS was fully protective against a SHIV challenge in monkeys, even at the lowest dose of 0.08mg/kg, with protection achieved at serum antibody concentrations of <0.75 µg/m (23). This was the lowest dose of any bNAb to show protection in monkeys (24).

Furthermore, when tested against a panel of 200 acute infection clade C pseudoviruses, CAP256-VRC26.25 emerged as the most potent member of bNAbs targeting the V2 loop (10, 17). The neutralization profile of CAP256-VRC26.25LS was particularly well suited as a complementary bNAb in combinations with bNAbs targeting other epitopes (17). CAP256-VRC26.25LS was subsequently engineered to prevent proteolytic clipping of the heavy chain through mutation of the lysine at position 100 to an alanine (25). This single amino acid change, K127A, was made in the CDRH3 region to improve manufacturability without altering neutralization potency or breadth. This non-clipped variant of the antibody is referred to as CAP256V2LS.

Pre-clinical studies in rhesus macaques demonstrated that CAP256V2LS has a half-life of 14.3 days when administered via the intravenous (IV) route and 9.9 days when administered

via the subcutaneous (SC) route. Toxicology reports showed that CAP256V2LS displayed low poly-specific auto-reactivity to HEp-2 cells and cardiolipin (26). In non-human primate (NHP) studies, non-pathological implications such as anti-cardiolipin activity were associated with the administration of anti-Env monoclonal antibodies (mAbs). Although these mAbs have polyspecific reactivities to host antigens (27) the immune response of NHPs to therapeutic mAbs is not considered to be predictive of the human response. This is due to the differences at species level. Thus, the ability to compare relative immunogenicity of mAbs in NHPs and humans is low (28). Its high potency and good breadth against clade C HIV viruses and long half-life make CAP256V2LS an excellent candidate for further clinical development. This is particularly important for southern Africa, where clade C is the dominant circulating virus.

The research and development pathway of the CAP256V2LS bNAb led to the CAPRISA 012 clinical trial programme that consists of three trials conducted in South Africa (Figure 1). This programme aims to evaluate the concept of bNAbs as long-acting pre-exposure prophylaxis with two to three subcutaneously (SC) doses per year to reduce HIV incidence among young women in three trials: CAPRISA 012A, a phase 1 trial of VRC07-523LS and PGT121 in HIV-negative women, previously described in this journal (29), CAPRISA 012B, described here, and CAPRISA 012C, a phase 2 combination bNAb trial in young women.

In CAPRISA 012B, CAP256V2LS will be assessed alone and in combination with VRC07-523LS and/or PGT121. VRC07-523LS targets the HIV-1 Envelope CD4 binding site and PGT121 targets the V3 glycan-dependent epitope region of the HIV Envelope protein. Given the vast genetic diversity of HIV-1, the use of multiple bNAbs may be required to ensure adequate coverage of circulating strains. Recent clinical trial data demonstrate that VRC07-523LS is safe with a half-life of 38 days after IV administration and 33 days after SC administration (30). Preliminary clinical trial data of PGT121 also demonstrate safety with a half-life of 22 days (31).

This trial is also one of the first to assess the use of a recombinant human hyaluronidase (rHuPH20) together with an anti-HIV monoclonal antibody (32). rHuPH20 is the active ingredient of the investigational ENHANZE™ Drug Product (EDP) and optimizes the SC delivery of co-administered therapeutics by depolymerizing hyaluronan in the extracellular matrix of the SC space that normally serves to restrict increased flow volumes. EDP allows co-mixing of antibodies with rHuPH20 at the clinical site. Clinical trials conducted in oncology have demonstrated safety and favourable results with this product (33).

SC administration of rHuPH20 was well tolerated in healthy participants, participants with diabetes, rheumatoid arthritis, cancer and dehydration. SC administrations of rHuPH20 alone or in combination with morphine, ceftriaxone, ondansetron, insulin, adalimumab, IgG and hydration fluids was also well-tolerated (34, 35). Most adverse events reported were mild, transient injection site reactions, including erythema, pruritus, tenderness, induration, and paraesthesia. Moderate injection site reactions, occurring less frequently, include burning, erythema, pain, and numbness. Mild-to-moderate headache was also reported (33). Local tissue changes induced by rHuPH20 are reversible within 24-48 hours after administration, without inflammatory or histological changes. Co-administration demonstrated beneficial effects such as improved absorption, increased bioavailability and decreased PK variability (33, 36). rHuPH20 is currently co-formulated with two approved anticancer therapies, trastuzumab and rituximab.

Methods and analysis

Patient and public involvement

The CAPRISA Community Advisory Board (CAB) plays a central role in study planning and recruitment of participants. The CAB includes local community leaders, traditional leaders, leadership of local HIV/AIDS organisations, local health service provider representatives, previous study participants, and HIV-positive local community members. During trial preparation and prior to study start, the study concept is presented to the CAB members for their feedback. Recruitment will include community events, local clinics, street recruitment, and use of snowballing techniques. A recruitment and retention plan will be drawn up at study start and will be reviewed and updated regularly.

Study setting

The CAPRISA 012B trial will be conducted at the CAPRISA eThekwini Clinical Research Site (ECRS). This site is based in a busy commuter area in the city centre of Durban, KwaZulu-Natal, South Africa.

Study population selection

The study population will consist of 66 women, 52 HIV-negative and 14 HIV-positive women who have not yet started antiretroviral therapy (ART). All eligibility criteria must be met, and HIV-positive women will be recruited based on additional inclusion criteria (Table 1).

Study Design

CAPRISA 012B is a first-in-human, phase I study to assess the safety, tolerability, and pharmacokinetics (PK) of CAP256V2LS. The study is divided into four groups (Table 2). Group 1 is a dose escalation of CAP256V2LS administered IV to HIV-negative and HIV-positive women. Group 2 is a dose escalation of CAP256V2LS administered SC with and without rHuPH20 at a single or repeat dose to HIV-negative women. Groups 3 and 4 are randomized placebo controlled to assess two (CAP256V2LS + VRC07-523LS; CAP256V2LS + PGT121) and three (CAP256V2LS + VRC07-523LS + PGT121) bNAb combinations administered SC to HIV-negative women. Participants will be followed up for 24 weeks after the administration of the last dose of study product/s. HIV positive participants in Groups 1c and 1d will have two additional follow up visits after 12 and 24 months.

Study Objectives

Primary objectives

- To evaluate the safety and tolerability of CAP256V2LS administered
  - SC in HIV-negative women
  - IV in HIV-negative and HIV-positive women
  - SC in combination with VRC07-523LS and/or PGT121 in HIV-negative women

Secondary objectives

- To characterize the PK profile of CAP256V2LS administered SC as a single dose or as two doses 16 or 24 weeks apart
- To characterize the PK profile of CAP256V2LS administered SC in combination with VRC07-523LS and/or PGT121
- To characterize the PK profile of CAP256V2LS administered IV as a single dose in HIV-negative and HIV-positive women
- To evaluate the antiviral activity of CAP256V2LS administered IV to HIV-positive women not on ART
- To evaluate the concentrations and functional activity of CAP256V2LS in plasma and genital samples following SC and IV administration
- To determine whether administration of CAP256V2LS induces anti-monoclonal antibody responses

- To assess the acceptability of SC administration of monoclonal antibodies among participants

Primary Endpoints

- Proportion of participants with mild, moderate, and severe reactogenicity events within the first 3 days after IV or SC administration of CAP256V2LS
- Proportion of participants with mild, moderate, and severe adverse events as well as serious adverse events (SAEs) related to the IV or SC administration of CAP256V2LS

Secondary Endpoints

- The difference in the elimination half-life, clearance, volume of distribution, and area under the concentration decay curve of CAP256V2LS mAb among study groups
- Change in plasma HIV-1 RNA levels from baseline (only for groups 1c and 1d).
- Changes in the concentration of serum anti-CAP256V2LS titres from baseline
- The difference in the functional activity including IgG/IgA binding responses, cellular immune responses, and antibody function in plasma and mucosal surfaces compared to baseline
- Proportion of participants reporting CAP256V2LS injections to be acceptable as per study questionnaire

Sample size calculation

The main objective of this study is to assess the safety, tolerability, and PK of CAP256V2LS, hence the ability of the study to detect SAEs is key. The probability of detecting no SAE, at least one, or at least two SAEs at a specified true event rate will be calculated. These probabilities highlight the likelihood of the study to detect either rare or common adverse events (AEs). In addition, the 95% confidence interval for the true event rate was calculated. Currently, limited safety data are available to guide the estimation of the true event rate that might be observed in the study. In the absence of available AE rates for CAP256V2LS alone or in combination with other bNAbs, a range of hypothesized event rates to calculate the probability of observing no events, at least one event, or at least two events were used. Among the four participants receiving active product in each of the groups, there is a 34% chance of observing at least one event if the true event rate is 10%. When the true event rate is two or three times higher, this probability rises to 59% and 76% (Table 3).

Since the phase I assessment of the safety of CAP256V2LS administered SC includes eight participants receiving combination of CAP256V2LS and VRC07-523.LS (i.e. Groups 3a and 3b combined), for N=8, there is an 8% chance of observing at least one event, if the true event rate is 1%, but it is as high as 83%, if the true event is 20%. These probabilities are also applicable to the groups that will receive repeat doses (Group 2d and 2f). For the 16 participants receiving CAP256V2LS administered IV in Group 1, there is an 85% chance of observing no events if the event rate is 1%, and less than 1% chance, if the event rate is 30-fold higher. However, if all 56 women receiving CAP256V2LS active product at enrolment are combined, the former probability changes to 57% for no events and remains very low (<1%)-given the event rates of 10%, 20% and 30%. As expected, an increase in sample size, increases the likelihood of detecting rare events.

Study procedures

Informed consent

Prior to screening and enrolment, informed consent will be obtained from every participant in accordance with the South African Good Clinical Practice guidelines. The informed consent procedure will be conducted in either English or isiZulu as per participant preference. If a participant is illiterate, an impartial witness will be present throughout the informed consent procedure to ensure that all questions are answered to the satisfaction of the potential participant. For the purposes of this study, consent for all study related procedures, pharmacogenetic studies, and storage of specimens will be obtained.

Screening

All potential participants will complete the informed consent procedure for screening and provide relevant identification documents. In order to rule out co-enrolment in other intervention trials, potential participants' details will be checked on the Biometric Co-Enrolment Prevention System (BCEPS). The study team will also evaluate and review proof of contraceptive use, such as family planning records. HIV pre-counselling will be performed, and if the participant meets eligibility, the socio-demographic and behavioural questionnaires will be administered. Post-test counselling is provided after disclosure of all HIV results and if the participant is ineligible to enrol into the study, referral to one of several HIV/AIDS care programmes will be facilitated. HIV testing in this study will be conducted by following a study HIV algorithm. At screening, HIV testing will be performed using two rapid antibody testing kits. Any participant with discordant results will be regarded as ineligible and referred to medical care.

A comprehensive medical history and physical examination will be conducted to determine eligibility. All pre-existing conditions and concomitant medication information will be documented. Screening laboratory tests will be conducted as per the schedule of evaluations (SOE) to determine further eligibility. These tests include urinalysis, haematology, blood chemistry tests, liver function tests, testing for sexually transmitted infections including syphilis serology and hepatitis B virus assays. Serum and plasma specimens will also be stored, for further analyses. In addition, a genital specimen using a menstrual cup device will be obtained and stored (37).

Randomisation

An unblinded statistician who is not involved in study conduct, will be responsible for generating the randomization sequence (for groups 1c,1d, 3a to 3c and Group 4a) using SAS version 9.4 software or latest. Participants will be assigned to unique participant identification numbers stratified by HIV status and group number. Sequentially numbered, sealed, opaque envelopes containing the group number and envelope number or treatment code (for use by the unblinded pharmacist only) will be provided to the study coordinator, to be opened once a participant has been deemed eligible and is ready to be enrolled into the study.

The pharmacist will store envelopes in a secured location within the research pharmacy with access restricted to delegated study pharmacists only. The study pharmacist will also receive a randomization list consisting of the unique three-digit envelope number and study group with the corresponding study drug or placebo and relevant dosage from the unblinded statistician. The envelope number will enable the unblinded pharmacist to assign the correct treatment to the correct participant.

Enrolment

Enrolment will take place within 56 days of screening. After the informed consent is obtained and eligibility criteria are met, the participant will be allocated to a study group (Table 2). At this visit, vital signs will be recorded, a targeted physical examination will be conducted and all laboratory results from the screening visit will be reviewed. For women of childbearing potential, a negative pregnancy test on that day must be obtained prior to product administration. Prior to the infusion/injection, the study team must ensure that the participant is eligible to receive study product. All study product administrations will be completed according to the assigned group and may be via an IV infusion or SC injection. Once safety has been established in the first participants, enrolment into the next groups as well as dose-

escalation will take place in a sequential manner, following review at each step of safety data from the preceding groups. For the bNAb combination groups, two separate injections, each containing a single bNAb, will be administered. After receiving study product, all participants will be observed for a minimum of 1 hour after the first product and any repeat product administrations. PK analysis on blood draws will be conducted for both the IV and SC administration groups at timelines outlined in the SOE.

Management of HIV-positive participants enrolled into Group 1c and 1d

All potential volunteers who meet the eligibility criteria for enrolment will receive extensive ART counselling upon entering the study.The benefits of early ART initiation including the universal treatment policy, will be explained. Only participants who, after appropriate counselling, are willing to defer ART initiation will be eligible to enrol. At each study visit the participant's decision to defer ART initiation will be reviewed, and participants are allowed to change their mind and start treatment at any stage during the study. During the study, HIV positive participants will be monitored closely with regular clinical and safety assessments. HIV viral load monitoring and CD4 count measurements will take place as per SOE. Participants will be counselled on reducing the risk of HIV transmission to their sexual partners. Furthermore, a multidisciplinary approach to HIV transmission risk mitigation will be followed as per guidance from previous publications (38).

Specific criteria to initiate ART within 8 weeks of product administration are listed below and have been outlined as per guidance previously described for studies designed for ART interruption (39). Once any of these criteria are met, the participant will receive ART counselling and will be initiated on ART as per South African guidelines. Criteria to start ART within 8 weeks of product administration include:

- Two consecutive viral loads >10,000 copies/ml after 2 weeks post product administration
- CD4 count <350 cells/mL
- Pregnancy
- If ART is deemed medically necessary
- If requested by the participant at any stage during the study

Safety monitoring

Reactogenicity assessments

Reactogenicity events are 12 common infusion/injection-related signs and symptoms. These are a subset of adverse events and have specific reporting requirements. Reactogenicity signs and symptoms are solicited from the start of the infusion/injection through the 3- day

post infusion/injection reactogenicity period. Reactogenicity events may be infusion/injection site reactions (infusion/injection related erythema/redness or induration/swelling), local symptoms (pain, tenderness) or systemic signs or symptoms (increased body temperature, malaise and/or fatigue, myalgia, headache, chills, arthralgia, nausea, and vomiting). After study product administration, the participant will be seen at the clinic for safety assessments on the day of product administration (Day 0), as well as Day 1, Day 2, and Day 3 (4 days in total). Clinicians and/or nurses will assess the product administration site(s) for local reactogenicity on the day of product administration and during the scheduled follow-up visits for all groups. Participants will keep a daily diary of local and systemic symptoms and record their temperature for three days after each product administration. In the event of a missed reactogenicity clinic visit, study staff will review the diary together with the participant once they present to the clinic and will determine the severity of the reactions. For any reactogenicity symptoms that are not resolved within 3 days, clinicians will follow and collect resolution information.

Safety monitoring

Safety monitoring includes internal monitoring by the study team, the protocol safety review team (PSRT), and data safety monitoring board (DSMB), as well as external monitoring including audits. Safety reporting of SAEs, AEs, and other important reportable events will be the responsibility of the entire study team. In addition, the study statistician will prepare routine study safety progress reports, which include reports of AEs experienced by study participants (blinded to treatment assignment), for review by the PSRT. The PSRT will review the clinical safety data on a weekly basis via electronic distribution of reports and will have face-to face meetings as required. The PSRT will be responsible for decisions related to participant safety. In addition, an independent DSMB will meet in-person and/or via teleconference semi-annually and review the study data and study conduct. The DSMB could recommend that the study should proceed as designed, should proceed with design modifications, or should be discontinued. Furthermore, the PI will permit authorized representatives such as external monitors and auditors to inspect the site facilities and records relevant to the study.

Follow up visits

All enrolled participants in both intervention and control arms will have follow up visits as specified in the SOE. At these visits, behavioural questionnaires will be administered together with HIV risk reduction counselling. Contraception counselling and provision will also take place. Targeted physical examinations will be conducted and all reactogenicity and AEs will be recorded. Laboratory investigations specified in the SOE will be conducted by

the CAPRISA Research Laboratories. Where required, sample processing and storage of specimens for potential future testing (blood and vaginal specimens) will also be undertaken. An accredited contract laboratory will perform all safety blood testing and provide a backup laboratory service when required.

Statistical Analysis

Baseline characteristics including demographics and laboratory measurements will be summarized using descriptive statistics either by group or overall. Summaries of the number and percentage of participants experiencing any AE or reactogenicity will be analysed. AEs and SAEs will be coded into the Medical Dictionary for Regulatory Activities (MedDRA) preferred terms. The number and percentages of participants experiencing each specific AE will be tabulated by severity and relationship to study product. For the calculations in these tables, each participant's AE will be counted once under the maximum severity or strongest recorded causal relationship to study product. A complete listing of AEs for each participant will provide details including severity, relationship to study product, onset, duration and outcome. Tolerability evaluation will be mostly descriptive and consist of solicited AEs that occur within 1 hour following study product administration and reasons for any withdrawal or discontinuation based on participant discomfort. This early assessment of tolerability of the monoclonal antibodies will inform which parameters should be solicited or routinely assessed to further characterise the tolerability profile in a larger number of participants. Where appropriate, some of the data will be presented graphically. Analysis will be carried out using either SAS V.9.4 or higher or R.

Pharmacokinetic analysis

Pharmacokinetic disposition of CAP256V2LS alone and in combination with VRC07-523LS and PGT121 will be evaluated in this study and PK will be assessed via dose sub-groups and route of administration. Sixteen participants will receive CAP256V2LS by the IV route. The two lower dose levels will be administered to HIV-negative participants and the highest dose level will be administered to HIV-positive participants. These PK data will serve to characterize dose effects on CAP256V2LS clearance, volumes of distribution, and elimination half-life. The PK data across Group 1 will also enable a preliminary evaluation of the potential impact of HIV status on CAP256V2LS PK. The PK results from the IV administration groups will be compared to the data from the SC administration groups in order to estimate bioavailability.

A total of 24 participants will receive CAP256V2LS by the SC route, either as a single dose or a repeat dose. A subset of participants will receive CAP256V2LS with SC hyaluronidase

to allow the larger doses of antibody to be administered SC. These data will allow concentration profiles, bioavailability, and absorption patterns to be established. The impact of repeat CAP256V2LS SC administration on PK will also be determined. The PK data from the combination groups (Group 3 and 4) will be compared with those from Group 2 to determine if co-administration of VRC07-523LS or PGT121 affects CAP256V2LS PK. In addition, the VRC07-523LS and PGT121 PK results will be compared with findings from PK studies in CAPRISA 012A and reported in the literature, to establish whether CAP256V2LS impacts on the PK of VRC07-523LS and PGT121.

Data management

Data for the CAPRISA 012B study will be collected on case report forms (CRFs) designed specifically to address the protocol requirements. Data will be managed by the CAPRISA Data Management department, using DFdiscover (DF/Net Research, Inc), software specifically designed for clinical trial data management. The site will record data on paper CRFs that will be directly captured onto the DFdiscover system and validated by the data management staff. All source documents will be kept in the participants' study files and medical charts at the clinical research site. All original CRFs and study related documents will be securely stored at the site, during the study and after study completion.

Ethics and dissemination

Regulatory approval has been granted by the University of KwaZulu-Natal (UKZN) Biomedical Research Ethics Committee and by the South African Health Products Regulatory Authority (Trial reference numbers: BREC00000857/2019 and SAHPRA 20200123). The study team will disseminate the trial results by sharing with the scientific community at international conferences, through peer-reviewed journal publications, and presentations to the wider community. Trial results will be uploaded onto the UKZN repository and the Pan-African Clinical Trial Registry (PACTR).

Authors' contributions

SSAK and QAK conceived the trial. SSAK, QAK, SM and NG designed the trial. SM and NG wrote the study protocol. EC will conduct the PK simulations and analysis. NYZ performed sample size calculations and the statistical analysis strategy. KC, NDR, PLM, JM and LM contributed to antibody development. CB, TNG, DA, NS, CW, DHB, JM, JL, CAH, BP and PEF contributed to the planning and conduct of the trial. All authors reviewed the manuscript and consented to publication.

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Competing interest statement

There are no competing interests to declare.

Patient consent for publication

Not required

Figure Legends

Figure 1: CAPRISA 012 clinical trial programme

Tables

Table 1: Eligibility criteria

Inclusion criteria	Exclusion criteria	Additional inclusion criteria*
• Age 18 to 45 years • Born female • Able and willing to complete the informed consent process • Able to understand the information provided about the study, willing to comply with protocol procedures, and available to attend the study site for the duration of the study • Based on clinical assessment, participants must be in good general health as per opinion of the Principal Investigator (PI) or designee • Haemoglobin > 10g/dl • Neutrophil count within institutional normal range • Platelets within institutional normal range • Creatinine < 1.1 times upper limit of normal • Alanine aminotransferase < 1.25 times upper limit of normal • HIV-negative as per FDA-approved method of detection (for groups with HIV-negative participants only) • Negative pregnancy test	• Any clinically significant acute or chronic medical condition that in the opinion of the PI/designee makes the participant unsuitable for participation in the study, or jeopardises the safety or rights of the participant • Any history of anaphylaxis and related symptoms such as hives, respiratory difficulty, or angioedema • Evidence of autoimmune disease or currently receiving immunosuppressive therapy • If planning a pregnancy for the duration of the study, currently pregnant or breastfeeding • Exceeding 95 kilograms in body weight (due to limitations related to SC antibody administration) • A history of alcohol or substance use judged by the PI to potentially interfere with participant study compliance • Prior participation in an investigational HIV vaccine trial, except if proof of allocation to the placebo arm is available • Administration of a monoclonal antibody or polyclonal	• Confirmed HIV-1 infection prior to enrolment • An HIV viral load of >1000 copies/ml at screening • A CD4 count of ≥ 450 cells/µl at screening • ART naive, and willing to defer treatment (after appropriate counselling) for up to a maximum of 8 weeks after product administration • No major comorbidities or AIDS-defining illness

• If of reproductive potential, there is evidence of effective contraceptive use and willingness to adhere to effective contraceptive use during the study period • Willing to have blood samples collected, stored, and used for research purposes. • Willing to adhere to reduced risk sexual behaviour during study participation.	immunoglobulin within 28 days prior to enrolment • Involvement in other concurrent research studies that would interfere with the objectives of this study.	

*Additional eligibility criteria for HIV positive participants in Groups 1c and 1d

Table 2: CAPRISA 012B Group, Dose and Arm Allocation

Group	Participants	Regimen	SA site N=66	Dose (mg/kg)
Group 1: Dose escalation of IV administration of CAP256V2LS				
1a	HIV negative	CAP256V2LS	4	5 mg/kg IV one dose
1b	HIV negative	CAP256V2LS	4	10 mg/kg IV one dose
1c	HIV positive	CAP256V2LS	4/2§	20 mg/kg IV one dose
1d	HIV positive	CAP256V2LS	4/4§	20 mg/kg IV one dose
Group 2: Dose escalation of SC administration of CAP256V2LS				
2a	HIV negative	CAP256V2LS	4	5 mg/kg SC one dose
2b	HIV negative	CAP256V2LS*	4	5 mg/kg SC one dose
2c	HIV negative	CAP256V2LS*	4	10 mg/kg SC one dose
2d	HIV negative	CAP256V2LS*	4	10 mg/kg SC with one repeat dose at 16/24 weeks#
2e	HIV negative	CAP256V2LS*	4	20 mg/kg SC one dose
2f	HIV negative	CAP256V2LS*	4	20 mg/kg SC with one repeat dose at 16/24 weeks#
Group 3: Dose escalation of the two antibody combinations				

3a	HIV negative	CAP256V2LS* + VRC07-523.LS*	4/1[§]	10 mg/kg SC / 10 mg/kg SC one dose
3b	HIV negative	CAP256V2LS* + VRC07-523.LS*	4/1[§]	20 mg/kg SC / 20 mg/kg SC one dose
3c	HIV negative	CAP256V2LS* + PGT121[$]	4/1[§]	20 mg/kg SC / 5 mg/kg SC one dose
Group 4: Three antibody combination				
4a	HIV negative	CAP256V2LS* + PGT121[$] + VRC07-523.LS*	4/1[§]	20 mg/kg SC / 5 mg/kg SC / 20mg/kg SC one dose

*Antibody will be injected with hyaluronidase, so that the antibody dose can be administered as a single SC injection; [#] First two participants will receive two doses 24 weeks apart and the next two participants will receive two doses 16 weeks apart; [$] PGT121 may be replaced with PGT121LS at a higher dose, based on its availability at the time of study initiation, [§] Placebo allocation

Table 3: Probability of observing no events, at least 1 event, or at least 2 events, for a range of hypothetical true event rates

True event rate (%)	Number of participants	0 events	1+ events	2+ events
1	4	0.96	0.04	<0.01
	8	0.92	0.08	<0.01
	12	0.89	0.11	0.01
	16	0.85	0.15	0.01
	24	0.79	0.21	0.02
	56	0.57	0.43	0.11
5	4	0.81	0.19	0.01
	8	0.66	0.34	0.06
	12	0.54	0.46	0.12
	16	0.44	0.56	0.19
	24	0.29	0.71	0.34
	56	0.06	0.94	0.78
10	4	0.66	0.34	0.05
	8	0.43	0.57	0.19
	12	0.28	0.72	0.34
	16	0.19	0.81	0.49
	24	0.08	0.92	0.71
	56	<0.01	>0.99	0.98

20	4	0.41	0.59	0.18
	8	0.17	0.83	0.50
	12	0.07	0.93	0.73
	16	0.03	0.97	0.86
	24	<0.01	>0.99	>0.99
	56	<0.01	>0.99	>0.99
30	4	0.24	0.76	0.35
	8	0.06	0.94	0.74
	12	0.01	0.99	0.91
	16	<0.01	>0.99	0.97
	24	<0.01	>0.99	>0.99
	56	<0.01	>0.99	>0.99

References

1. HIV/AIDS JUNPo. Geneva: UNAIDS; 2018 [cited 2019]. 2018.
2. De Oliveira T, Kharsany AB, Gräf T, Cawood C, Khanyile D, Grobler A, et al. Transmission networks and risk of HIV infection in KwaZulu-Natal, South Africa: a community-wide phylogenetic study. The lancet HIV. 2017;4(1):e41-e50.
3. Dellar RC, Dlamini S, Karim QA. Adolescent girls and young women: key populations for HIV epidemic control. Journal of the International AIDS Society. 2015;18:19408.
4. Abdool Karim SS. HIV-1 Epidemic Control—Insights from Test-and-Treat Trials. Mass Medical Soc; 2019.
5. Fang W, Mphoyi BI, Motake DR, Liu J, Li W, Mei F, et al. Efficacy, Adherence and Side Effects of PrEP for HIV-1 Prevention. International Journal of Biology. 2019;11(4).
6. Fauci AS, Marston HD. Ending the HIV–AIDS pandemic—follow the science. New England Journal of Medicine. 2015;373(23):2197-9.
7. Anderson DJ, Politch JA, Vaca GB, Kadasia K, Whaley KJ. Use of Monoclonal Antibodies to Prevent the Sexual Transmission of Human Immunodeficiency Virus Type 1. Current Immunology Reviews. 2019;15(1):123-30.
8. Pegu A, Hessell AJ, Mascola JR, Haigwood NL. Use of broadly neutralizing antibodies for HIV-1 prevention. Immunological reviews. 2017;275(1):296-312.
9. Hessell AJ, Poignard P, Hunter M, Hangartner L, Tehrani DM, Bleeker WK, et al. Effective, low-titer antibody protection against low-dose repeated mucosal SHIV challenge in macaques. Nature medicine. 2009;15(8):951.
10. Walker LM, Huber M, Doores KJ, Falkowska E, Pejchal R, Julien J-P, et al. Broad neutralization coverage of HIV by multiple highly potent antibodies. Nature. 2011;477(7365):466.
11. Moldt B, Rakasz EG, Schultz N, Chan-Hui P-Y, Swiderek K, Weisgrau KL, et al. Highly potent HIV-specific antibody neutralization in vitro translates into effective protection against mucosal SHIV challenge in vivo. Proceedings of the National Academy of Sciences. 2012;109(46):18921-5.
12. Barouch DH, Whitney JB, Moldt B, Klein F, Oliveira TY, Liu J, et al. Therapeutic efficacy of potent neutralizing HIV-1-specific monoclonal antibodies in SHIV-infected rhesus monkeys. Nature. 2013;503(7475):224.
13. Sok D, Burton DR. Recent progress in broadly neutralizing antibodies to HIV. Nature immunology. 2018;19(11):1179.
14. van Loggerenberg F, Mlisana K, Williamson C, Auld SC, Morris L, Gray CM, et al. Establishing a cohort at high risk of HIV infection in South Africa: challenges and experiences of the CAPRISA 002 acute infection study. PloS one. 2008;3(4):e1954.
15. Doria-Rose NA, Schramm CA, Gorman J, Moore PL, Bhiman JN, DeKosky BJ, et al. Developmental pathway for potent V1V2-directed HIV-neutralizing antibodies. Nature. 2014;509(7498):55.
16. Sheward DJ, Marais J, Bekker V, Murrell B, Eren K, Bhiman JN, et al. HIV superinfection drives de novo antibody responses and not neutralization breadth. Cell host & microbe. 2018;24(4):593-9. e3.
17. Wagh K, Bhattacharya T, Williamson C, Robles A, Bayne M, Garrity J, et al. Optimal combinations of broadly neutralizing antibodies for prevention and treatment of HIV-1 clade C infection. PLoS pathogens. 2016;12(3):e1005520.
18. Moore PL, Gray ES, Sheward D, Madiga M, Ranchobe N, Lai Z, et al. Potent and broad neutralization of HIV-1 subtype C by plasma antibodies targeting a quaternary epitope including residues in the V2 loop. Journal of virology. 2011;85(7):3128-41.
19. Bhiman JN, Anthony C, Doria-Rose NA, Karimanzira O, Schramm CA, Khoza T, et al. Viral variants that initiate and drive maturation of V1V2-directed HIV-1 broadly neutralizing antibodies. Nature medicine. 2015;21(11):1332.
20. Doria-Rose NA, Bhiman JN, Roark RS, Schramm CA, Gorman J, Chuang G-Y, et al. New member of the V1V2-directed CAP256-VRC26 lineage that shows increased breadth and exceptional potency. Journal of virology. 2016;90(1):76-91.
21. Gorman J, Chuang G-Y, Lai Y-T, Shen C-H, Boyington JC, Druz A, et al. Structure of Super-Potent Antibody CAP256-VRC26. 25 in Complex with HIV-1 Envelope Reveals a Combined Mode of Trimer-Apex Recognition. Cell Reports. 2020;31(1):107488.
22. Gaudinski MR, Coates EE, Houser KV, Chen GL, Yamshchikov G, Saunders JG, et al. Safety and pharmacokinetics of the Fc-modified HIV-1 human monoclonal antibody VRC01LS: a phase 1 open-label clinical trial in healthy adults. PLoS medicine. 2018;15(1):e1002493.

23. Julg B, Tartaglia LJ, Keele BF, Wagh K, Pegu A, Sok D, et al. Broadly neutralizing antibodies targeting the HIV-1 envelope V2 apex confer protection against a clade C SHIV challenge. Science translational medicine. 2017;9(406):eaal1321.
24. Pegu A, Borate B, Huang Y, Pauthner MG, Hessell AJ, Julg B, et al. A Meta-analysis of Passive Immunization Studies Shows that Serum-Neutralizing Antibody Titer Associates with Protection against SHIV Challenge. Cell Host & Microbe. 2019;26(3):336-46. e3.
25. Ivleva VB, Schneck NA, Gollapudi D, Arnold F, Cooper JW, Lei QP. Investigation of sequence clipping and structural heterogeneity of an HIV broadly neutralizing antibody by a comprehensive LC-MS analysis. Journal of The American Society for Mass Spectrometry. 2018;29(7):1512-23.
26. Centre VR. INVESTIGATOR'S BROCHURE CAP256V2LS (VRC-HIVMAB0102-00-AB) A Broadly Neutralizing HIV-1 Human Monoclonal Antibody. Version 10 12 February 2020
27. Haynes BF, Moody MA, Verkoczy L, Kelsoe G, Alam SM. Antibody polyspecificity and neutralization of HIV-1: a hypothesis. Human antibodies. 2005;14(3-4):59-67.
28. van Meer PJ, Kooijman M, Brinks V, Gispen-de Wied CC, Silva-Lima B, Moors EH, et al., editors. Immunogenicity of mAbs in non-human primates during nonclinical safety assessment. MAbs; 2013: Taylor & Francis.
29. Mahomed S, Garrett N, Capparelli E, Baxter C, Zuma NY, Gengiah T, et al. Assessing the safety and pharmacokinetics of the monoclonal antibodies, VRC07-523LS and PGT121 in HIV negative women in South Africa: study protocol for the CAPRISA 012A randomised controlled phase I trial. BMJ open. 2019;9(7):e030283.
30. Gaudinski MR, Houser KV, Doria-Rose NA, Chen GL, Rothwell RSS, Berkowitz N, et al. Safety and pharmacokinetics of broadly neutralising human monoclonal antibody VRC07-523LS in healthy adults: a phase 1 dose-escalation clinical trial. The Lancet HIV. 2019;6(10):e667-e79.
31. Stephenson K, Julg B, Ansel J, Walsh S, Tan C, Maxfield L, et al., editors. Therapeutic activity of PGT121 monoclonal antibody in HIV-infected adults. Conference on Retroviruses and Opportunistic Infections; Seattle, Washington; 2019.
32. Mahomed S, Garrett N, Baxter C, Abdool Karim Q, Abdool Karim SS. Clinical trials of broadly neutralizing monoclonal antibodies for HIV Prevention: A Review. The Journal of Infectious Diseases. 2020.
33. Locke KW, Maneval DC, LaBarre MJ. ENHANZE® drug delivery technology: a novel approach to subcutaneous administration using recombinant human hyaluronidase PH20. Drug delivery. 2019;26(1):98-106.
34. Morcos PN, Zhang X, McIntyre C, Bittner B, Rowell L, Hussain Z. Pharmacokinetics and pharmacodynamics of single subcutaneous doses of tocilizumab administered with or without rHuPH20. International journal of clinical pharmacology and therapeutics. 2013;51(7):537-48.
35. Morrow L, Muchmore DB, Ludington EA, Vaughn DE, Hompesch M. Reduction in intrasubject variability in the pharmacokinetic response to insulin after subcutaneous co-administration with recombinant human hyaluronidase in healthy volunteers. Diabetes technology & therapeutics. 2011;13(10):1039-45.
36. Shpilberg O, Jackisch C. Subcutaneous administration of rituximab (MabThera) and trastuzumab (Herceptin) using hyaluronidase. British journal of cancer. 2013;109(6):1556-61.
37. Archary D, Liebenberg LJ, Werner L, Tulsi S, Majola N, Naicker N, et al. Randomized cross-sectional study to compare HIV-1 specific antibody and cytokine concentrations in female genital secretions obtained by menstrual cup and cervicovaginal lavage. PloS one. 2015;10(7).
38. Peluso MJ, Dee L, Campbell D, Taylor J, Hoh R, Rutishauser RL, et al. A collaborative, multidisciplinary approach to HIV transmission risk mitigation during analytic treatment interruption. Journal of Virus Eradication. 2020;6(1):34.
39. Julg B, Dee L, Ananworanich J, Barouch DH, Bar K, Caskey M, et al. Recommendations for analytical antiretroviral treatment interruptions in HIV research trials—report of a consensus meeting. The Lancet HIV. 2019.

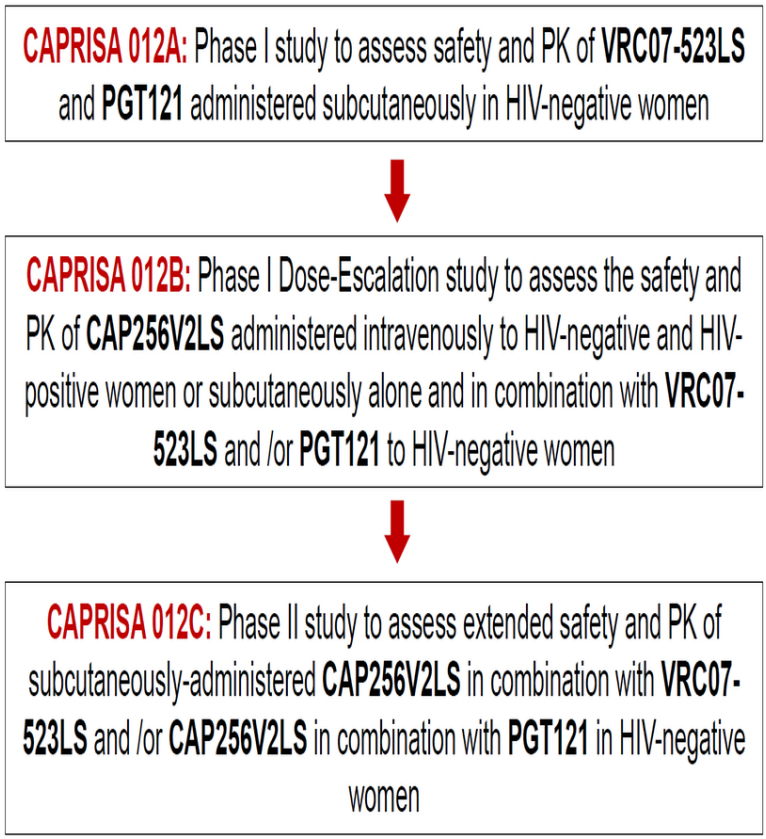

Figure 1: CAPRISA 012 Clinical Trial Programme

SPIRIT
STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

Section/item	Item No	Description	Addressed on page number
Administrative information			
Title	1	Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym	1
Trial registration	2a	Trial identifier and registry name. If not yet registered, name of intended registry	33
	2b	All items from the World Health Organization Trial Registration Data Set	1-13; Registered on the Pan African Clinical Trial Registry (PACTR202003767867253)
Protocol version	3	Date and version identifier	1-35
Funding	4	Sources and types of financial, material, and other support	3
Roles and responsibilities	5a	Names, affiliations, and roles of protocol contributors	2
	5b	Name and contact information for the trial sponsor	1
	5c	Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities	N/A The applicant is the sponsor

	5d	Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee)	26-27
Introduction			
Background and rationale	6a	Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention	7-12
	6b	Explanation for choice of comparators	22-23
Objectives	7	Specific objectives or hypotheses	12-13
Trial design	8	Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)	13-16
Methods: Participants, interventions, and outcomes			
Study setting	9	Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained	16
Eligibility criteria	10	Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)	16-17
Interventions	11a	Interventions for each group with sufficient detail to allow replication, including how and when they will be administered	13-15
	11b	Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)	15,20
	11c	Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)	16, 20
	11d	Relevant concomitant care and interventions that are permitted or prohibited during the trial	18-19

Outcomes	12	Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended	13
Participant timeline	13	Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)	Schedule of Evaluations for each study group
Sample size	14	Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations	27-29
Recruitment	15	Strategies for achieving adequate participant enrolment to reach target sample size	17-18
Methods: Assignment of interventions (for controlled trials)			
Allocation:			
Sequence generation	16a	Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions	13-14,19
Allocation concealment mechanism	16b	Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned	19, 20, 23
Implementation	16c	Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions	23
Blinding (masking)	17a	Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how	23
	17b	If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial	23

Methods: Data collection, management, and analysis

Data collection methods	18a	Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol	29-30
	18b	Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols	20,26,27,32
Data management	19	Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol	30-31
Statistical methods	20a	Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol	29-30
	20b	Methods for any additional analyses (eg, subgroup and adjusted analyses)	29-30
	20c	Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation)	N/A

Methods: Monitoring

Data monitoring	21a	Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed	26-27
	21b	Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial	15,16,26,27
Harms	22	Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct	24-26
Auditing	23	Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor	27

Ethics and dissemination			
Research ethics approval	24	Plans for seeking research ethics committee/institutional review board (REC/IRB) approval	31
Protocol amendments	25	Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators)	31
Consent or assent	26a	Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32)	31
	26b	Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable	31
Confidentiality	27	How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial	33
Declaration of interests	28	Financial and other competing interests for principal investigators for the overall trial and each study site	2-3
Access to data	29	Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators	33
Ancillary and post-trial care	30	Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation	18
Dissemination policy	31a	Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions	33
	31b	Authorship eligibility guidelines and any intended use of professional writers	N/A
	31c	Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code	33
Appendices			

Informed consent materials	32	Model consent form and other related documentation given to participants and authorised surrogates	31
Biological specimens	33	Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable	21

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated.