

CLINICAL STUDY PROTOCOL

HIPRA-HH-4

A PHASE IIB/III, OPEN LABEL, SINGLE ARM, MULTI-CENTRE, TRIAL TO ASSESS THE IMMUNOGENICITY AND SAFETY OF AN ADDITIONAL DOSE VACCINATION WITH A RECOMBINANT PROTEIN RBD FUSION HETERODIMER CANDIDATE (PHH-1V) AGAINST SARS-COV-2, IN ADULTS WITH PRE-EXISTING IMMUNOSUPPRESSIVE CONDITIONS VACCINATED AGAINST COVID-19.

Sponsor: HIPRA SCIENTIFIC, S.L.U.
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SIGNATURE PAGE

Protocol Title: A Phase IIb/III, open label, single arm, multi-centre, trial to assess the immunogenicity and safety of an additional dose vaccination with a recombinant protein RBD fusion heterodimer candidate (PHH-1V) against SARS-CoV-2, in adults with pre-existing immunosuppressive conditions vaccinated against COVID-19.

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INVESTIGATOR STATEMENT

I understand that all documentation provided to me by HIPRA SCIENTIFIC S.L.U. or its designated representative(s) concerning this study that has not been published previously will be kept in the strictest confidence. This documentation includes the study protocol, Investigator's brochure, case report forms, and other scientific data.

This study will not commence without the prior written approval of a properly constituted Independent Ethics Committee. No changes will be made to the study protocol without the prior written approval of HIPRA SCIENTIFIC S.L.U. and the Independent Ethics Committee, except when necessary to eliminate an immediate hazard to the patient.

I have read, understood, and agree to abide by all the conditions and instructions contained in this protocol.

Investigator Name

Investigator Signature

Date

Investigational site or name of institution and location (printed)

CLINICAL STUDY SYNOPSIS

Protocol code	HIPRA-HH-4.	
Title of the Study	A phase IIb/III, open label, single arm, multi-centre, trial to assess the immunogenicity and safety of an additional dose vaccination with a recombinant protein RBD fusion heterodimer candidate (PHH-1V) against SARS-CoV-2, in adults with pre-existing immunosuppressive conditions vaccinated against COVID-19	
EudraCT No.	2022-000785-18.	
Clinical Phase	IIb/III	
Sites / countries	Phase IIb/III: This Phase of the clinical study will be conducted at 6 sites located in Spain and Turkey.	
Study period	In Spain, each participant will be followed for approximately 52 weeks (365 days) after the administration of the vaccination on Day 0. In Turkey, each participant will be followed for approximately 26 weeks (182 days) after the administration of the vaccination on Day 0.	
Primary Objectives and Endpoints	Objective/s	Endpoint/s
	1. To determine immunogenicity measured by pseudovirus neutralization against Omicron BA1 at Baseline and at Day 14 after administration of HIPRA's vaccine (PHH-1V). 2. To determine immunogenicity measured by live virus neutralization for the	1.1 Neutralization titer against Omicron BA1 measured as inhibitory concentration 50 (IC50) by a pseudovirion-based neutralization assay (PBNA) and reported as reciprocal concentration for each individual sample and geometric mean titer (GMT) for descriptive statistics analysis at Baseline and at Day 14. 1.2 The geometric mean fold rise (GMFR) in neutralizing antibody titer from baseline to Day 14. 2.1 Neutralization titer against Omicron measured as a ID50 (the reciprocal dilution inhibiting 50% of the infection) by a live virus assay (VNA) to measure

	HIV cohort against Omicron at Baseline and Day 14 after administration of HIPRA's vaccine (PHH-1V).	cytopathic effect in Vero E6 cells at Baseline and Day 14 after PHH-1V administration.
Secondary Objectives and Endpoints	1. To determine and compare the changes of the immunogenicity measured by pseudovirus neutralization against Omicron, D614G, Beta and any other relevant Variants of Concern (VOC) in the epidemiologic moment at Days baseline, 14, 91 and 182, and 365 only for Spanish participants, after administration of HIPRA's vaccine (PHH-1V). 2. To determine and compare the changes of the immunogenicity measured by live virus neutralization for the HIV cohort against Omicron at days 91, 182 and 365 after administration of HIPRA's vaccine (PHH-1V). 3. To determine and compare the changes of the immunogenicity measures by live virus	1.1 Neutralization titer against Omicron, D614G, Beta strains, and any other relevant VOC in the epidemiologic moment, measured as inhibitory concentration 50 (IC50) by a pseudovirion-based neutralization assay (PBNA) and reported as reciprocal concentration for each individual sample and geometric mean titer (GMT) for descriptive statistics analysis at Days Baseline, 14, 91 and 182 for all participants, and 365 only for Spanish participants. 2.1 Neutralization titer against Omicron measured as ID50 (the reciprocal dilution inhibiting 50% of the infection) by a live virus assay (VNA) to measure cytopathic effect in Vero E6 cells at Days 91, 182 and 365 after PHH-1V administration. 3.1 Neutralization titer against D614G and any other relevant VOC in the epidemiologic moment, measured as a ID50 (the reciprocal dilution inhibiting 50% of the infection) by a live virus assay (VNA) to measure cytopathic effect in

	neutralization for the HIV cohort against D614G and any other relevant VOC in the epidemiologic moment at days Baseline, 14, 91 182 and 365 after administration of PHH-1V.	Vero E6 cells at Days Baseline, 14, 91 182 and 365 after PHH-1V administration.
	4. To evaluate the immunogenicity measured by means of total antibody against Receptor Binding Domain of the Spike protein of SARS-CoV-2 quantification, measured by an electrochemiluminescence immunoassay (ECLIA) at Baseline and at Days 14, 91 and 182, and 365 only for Spanish participants, after administration of HIPRA's vaccine (PHH-1V).	4.1 Binding antibodies titer measured for each individual sample and GMT for descriptive statistics analysis at Baseline and Days 14, 91 and 182 for all participants, and 365 only for Spanish participants. 4.2 The geometric mean fold rise (GMFR) in binding antibody titer from baseline to Day 14. 4.3 The percentage of individuals that after PHH-1V vaccine have a ≥ 2-fold and 4-fold change in binding antibodies titer from Baseline to Day 14.
	5. To assess the safety and tolerability of PHH-1V as an additional dose in adult individuals with pre-existing immunosuppressive conditions	5.1. Number, percentage, and characteristics of solicited local and systemic reactions through Day 7 after vaccination. 5.2. Number, percentage, and characteristics of unsolicited local and systemic adverse events (AEs) through Day 28 after vaccination. 5.3. Number and percentage of serious adverse events (SAEs) through the end of the study. 5.4. Number and percentage of adverse event of special interest (AESI) through the end of the study. 5.5. Number and percentage of medically attended adverse events (MAAE) related to study vaccine through the end of the study.

		5.6. Grade 3 and 4 changes from baseline in safety laboratory parameters at Days 14, 91 and 182 for all participants, and 365 only for Spanish participants, after vaccination.
Exploratory Objectives and Endpoints	1. To assess number of participants with SARS-CoV-2 infections ≥ 14 days after PHH-1V administration.	1.1. Number and percentage of participants with SARS-CoV-2 infections ≥ 14 days after PHH-1V administration according to COVID-19 infection criteria throughout the study duration.
	2. To assess number of COVID-19 severe infections ≥ 14 days after PHH-1V administration.	2.1 Number and percentage of COVID-19 severe infections ≥ 14 days after PHH-1V administration and through the end of the study requiring hospitalization. 2.2 Number and percentage of ICU admissions associated with COVID-19 ≥ 14 days after PHH-1V administration and through the end of the study. 2.3 Number and percentage of non-invasive ventilation administration associated with COVID-19 ≥ 14 days after PHH-1V administration and through the end of the study. 2.4 Number and percentage of deaths associated with COVID-19 ≥ 14 days after PHH-1V administration and through the end of the study.
	3. To further evaluate humoral responses by measuring IgG subclasses usage and avidity of humoral immunity at Baseline and Days 14, 91 and 182, and 365 only for Spanish participants.	3.1 Quantification of IgG subclasses (IgG1, IgG2, IgG3 and IgG4) measured by ELISA at Baseline and Days 14, 91 and 182 for all participants, and 365 only for Spanish participants. 3.2 Cross reactivity with Spike glycoproteins from other coronavirus (SARS-CoV, MERS-CoV, HKU1, OC43, NL63, 229E) measured at Baseline and Days 14, 91 and 182 for all participants, and 365 only for Spanish participants.

		3.3 Avidity of the main antibodies elicited against the Spike glycoproteins determined by ELISA at Baseline and Days 14, 91 and 182 for all participants, and 365 only for Spanish participants.
	4. To evaluate T-cell mediated responses, their immune phenotype polyclonally with potential cross-reactivity capacity and gene-expression profiles against the SARS-CoV-2 RBD proteins at Baseline and at Days 14, 91, 182 and 365 in a subset of approximately 50% of participants.	4.1 Frequency of IFNγ positive T-cells measured by enzyme-linked immune absorbent spot (ELISpot) covering the RBD sequence of D614G, RBD Omicron, RBD Beta and RBD Delta and any other relevant Variants of Concern (VOC) in the epidemiologic moment at Baseline and at Days 14, 91, 182 and 365 in a subset of approximately 50% of participants. 4.2 Immune phenotype of SARS-CoV-2 specific CD4^{+}/CD8^{+} T-cell responses measured by intracellular cytokine staining (ICS) flow-cytometry at Baseline and at Day 14 in a subset of 50% of participants. 4.3 T cell transcriptional profiles and TCR usage of RBD-specific T-cells isolated using cell sorting with CD25, CD69 and CD137 activation markers and subjected to single cell transcriptomics analyses (sc-RNAseq) at Baseline and at Day 14, 182 and 365 in a subset of 50 individuals selected based on the humoral, ELISPOT and Flow cytometry data.
Study Population	Phase IIb/III: Approximately 400 adults aged above 18 years old with pre-existing immunosuppressive conditions will receive an additional vaccination with HIPRA's vaccine (PHH-1V). In Spain, the 4th dose of the vaccine has been already approved for this specific group of patients, but there are some people that haven't received this dose yet, due to rejection at that time, a COVID-19 infection, or other non-specified causes. These people, in addition to those that have two doses and an episode of COVID-19 infection, are potential volunteers to be recruited in Spain.	

	In Turkey, at the moment of submission of this amendment, booster doses have already been widely implemented in these populations. Thus, individuals who have received booster doses after a primary vaccination with Coronavac and/or Comirnaty are potential volunteers to be recruited in Turkey.
Study Design	This is a phase IIb/III, open label, single arm, multi-center clinical trial that aims to determine safety, reactogenicity, tolerability and immunogenicity of additional vaccination with a recombinant protein receptor binding domain (RBD) fusion heterodimer candidate against SARS-CoV-2 developed by HIPRA (PHH-1V) in individuals with pre-existing immunosuppressive conditions. In this phase IIb/III study, approximately 400 eligible individuals who have received primary vaccination against COVID-19 with 3 doses of mRNA vaccine regardless of past history of COVID (or 2 doses plus a documented history of COVID) last dose being administered at least 91 days before day 0 will be included. In Spain, the 4th dose of the vaccine has been already approved for this specific group of patients, but there are some people that haven't received this dose yet, due to rejection at that time, a COVID-19 infection, or other non-specified causes. These people, in addition to those that have two doses and an episode of COVID-19 infection, are potential volunteers to be recruited in Spain. Individuals recruited in Turkey which received a primary vaccination regimen using a combination of Coronavac and/or Comirnaty with or without additional boosters of the same vaccines will be allowed to be included provided that last booster dose being administered is at least 91 days before day 0. Individuals with history of COVID-19 disease will be allowed to be included if diagnosed at least 91 days prior (first day of positive test, RAT or PCR), passed 30 days from hospital discharge prior to inclusion and provided they are no suffering from long covid (Post-acute COVID-19 syndrome) Screening and baseline visits will be allowed on the same day. All participants will receive a single dose of PHH-1V on Day 0 and will be observed for 15 minutes, except for those participants reporting previous mild allergy reactions to vaccination will be observed for 30 minutes. All participants will be provided with a paper diary on Day 0 to record solicited local and systemic reactions after vaccination through Day 7. Unsolicited adverse events will be reported through Day 28 except for COVID-19 cases that will be recorded in eCRF throughout the study. SAEs, AESI and MAAE will be reported during all the study.

	On-site follow-up visits will be performed on Days 14, 91, 182 and 365 for Spanish subjects, and Days 14, 91 and 182 for Turkish subjects. All 400 included individuals will be evaluated for high throughput humoral immunogenicity (primary objective). All participants will be evaluated for safety (secondary endpoint). A subset of about 50% of the enrolled individuals will be further characterized in terms of humoral immunity (IgG subclasses and avidity) and T cell responses (exploratory objectives) that requires PBMC isolation which will be performed at some of the Spanish clinical sites. This subset will be selected based on pre-existing conditions. A DSMB may review safety data at any time point during the study as necessary and/or if requested by any of the involved PIs in case of observation of serious adverse events (SAEs). An interim analysis is envisioned after data on humoral response at Day 91 of all enrolled participants in Spain will be available. A final analysis will include exploratory humoral and cellular objectives and the analysis of long-term immunogenicity response of the PHH-1V vaccination.
Study blind	This study is unblinded since it is a single arm clinical trial
Selection criteria	The inclusion and exclusion criteria for enrolling are described below. If there is a question about these criteria, the investigator must consult with the appropriate sponsor representative and resolve any issues before enrolling a participant in the study. Inclusion criteria: Participants must meet all the following criteria to be considered eligible for the study: 1. Male, female or transgender, ≥ 18 years old at Day 0. 2. Participant must provide consent indicating that she or he understands the purpose and potential risks and is willing and able to participate in the study and comply with all the study requirements and procedures (scheduled visits, laboratory tests, complete diaries, etc). 3. Participant who has one of the following SARS-CoV-2 vaccination schemes: For sites in Spain, the following vaccination schemes will be allowed: • Three doses of mRNA vaccines (Comirnaty and/or Spikevax) with or without past history of positive test for COVID-19, provided that last vaccination and diagnose of COVID-19 is at least 91 days before day

	0, and if hospitalization was required, participant was discharged from hospital at least 30 days before day 0. - Two doses of mRNA vaccines (Comirnaty and/or Spikevax) and previous history of positive test for COVID-19, provided that last vaccination and diagnose of COVID-19 is at least 91 days before day 0, and if hospitalization was required, participant was discharged from hospital at least 30 days before day 0. For sites in Turkey where vaccination with Coronavac was started, the following vaccinations schemes will be allowed: - A complete primary scheme of the following vaccines: Coronavac and/or Comirnaty, with or without additional boosters of the same vaccines, being the last vaccination at least 91 days before Day 0. . Participants with or without history of a positive test for COVID-19 are allowed. The infection (first day of positive test, RAT o PCR) must have been at least 91 days before Day 0. 4. Participant who has any of the following underlying immunosuppressive conditions*: - Confirmed HIV infection with persistent CD4 T cell counts <400 within last 6 months prior to Day0 regardless of pVL determination and ARV; - Primary Antibody Deficiency Disorders on immunoglobulin replacement therapy for at least 6 months prior to Day0 (maintenance dose); - Kidney disease on dialysis program for at least 6 months prior to Day0; - Kidney transplant at least >1 year that have never been treated with anti-CD20/anti-CD3 or has been given at least >1 year prior to Day0 AND on maintenance immunosuppressive therapy based on at least 3 drugs: tacrolimus/cyclosporine, glucocorticoids and mycophenolate or everolimus/sirolimus. - Auto-immune disease (AID**) on treatment with rituximab (RTX)/ocrelizumab during within the last 6 months prior to Day0 <i>*approximately 60 participants per condition will be included</i> <i>**AID includes: vasculitis, myositis, SLE/Sjögren, rheumatoid arthritis, multiple sclerosis or other.</i> 5. Contraceptive use should be consistent with local regulation for participants in clinical trials. a. <u>Female</u> participants of childbearing potential [defined as any female who has experienced menarche and until becoming
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	postmenopausal* (defined as having ≥ 12 months amenorrhea prior to screening without an alternative cause) unless is surgically sterile]: • Have a negative pregnancy test on the day of vaccination. • Use of any acceptable contraceptive method that should be started on day 0 and until 8 weeks after vaccination, except hormonal contraception. Acceptable contraceptive methods are: 1. Hormonal contraception (progestogen-only or combined): oral, injectable or transdermal (patch) started at least 28 days before day 0 and until 8 weeks after vaccination 2. Intrauterine device. 3. Vasectomized partner (the vasectomized partner should be the sole partner for that participant). 4. Sexual abstinence **, as a form of contraception, is acceptable if in line with the participant's lifestyle. 5. Condom b. <u>Male</u> participants: • Vasectomized participants. • Refrain from donating sperm for at least 28 days after day 0. • Agree to use a male condom may be considered in women of childbearing potential partners, from screening and for at least 8 weeks after vaccination. • Sexual abstinence**, as a form of contraception, is acceptable if in line with the participant's lifestyle. <i>* A high follicle stimulating hormone (FSH) level in the postmenopausal range may be used to confirm a post-menopausal state in women not using hormonal contraception or hormonal replacement therapy.</i> <i>** Sexual abstinence is considered an effective method only if defined as refraining from heterosexual intercourse from screening until 8 weeks after receiving the vaccine for female participants and from screening until 4 weeks for male participants. Periodic abstinence (e.g., calendar, ovulation) and withdrawal are not acceptable methods of contraception.</i> Exclusion Criteria Participants meeting any of the following criteria will be excluded from the study: 6. History of anaphylaxis to any prior vaccine. 7. Participant received or plans to receive: • Live attenuated vaccines (licensed) within 4 weeks before or after receiving any study vaccine.
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	• Other not live vaccines (licensed) within 14 days before and after receiving any study vaccine • Viral-vectored COVID-19 vaccines, such as Vaxzevria (AstraZenca) or Ad26.CoV2.S from Janssen 8. Pregnancy or breast-feeding at screening or Day 0 (vaccination time-point) or willingness/intention to become pregnant during the entire length of the study for female participants. For male participants, willingness/intention that your partner becomes pregnant during the entire length of the study. <u>Medical conditions</u> 9. A confirmed COVID-19 diagnose (by RT-PCR or RAT, asymptomatic or symptomatic) <90 days prior to Day0. 10. Participant has a clinically significant acute illness (this does not include minor self-limited illness such as mild diarrhea) or fever (temperature $\geq 38^{\circ}$ C (100.4°F) at screening or within 48 hours prior to the planned vaccination (Day 0). 11. Participant had a surgery requiring hospitalization (defined as inpatient stay for > 24 hours) before vaccination and he/she has not received the hospital discharge at day 0; or has a surgery requiring hospitalization planned within 12 weeks after study vaccine administration. Minor surgical procedures not requiring hospitalization are accepted. 12. Participant has ongoing severe and non-stable psychiatric condition likely to affect participation in the study (e.g., ongoing and non-stable severe depression, recent suicidal ideation, severe eating disorder, psychosis). 13. Participant has a problematic or risk use of substances including alcohol (except tobacco) that can compromise the study follow-up. Problematic or risk use of psychoactive substances is understood as the one that causes evident damage, whether it is dependence or any other physical, psychological, or social problem or that carries a high risk of suffering these damages. The negative consequences that consumption causes to third parties could be included. 14. Participant has a bleeding disorder (e.g., factor deficiency, platelet disorder), blood dyscrasia, or continuous use of anticoagulants or has any condition that in the opinion of the investigator contraindicates intramuscular injections or frequent phlebotomy. The use of ≤ 325mg of aspirin or ≤ 75mg of clopidogrel per day as prophylaxis is permitted but not combined. 15. Participant suffering from post-acute COVID-19 syndrome / long-covid
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	<u>Prior/Concomitant Therapy and Clinical Study Experience.</u> 16. Participant received any immunotherapy (monoclonal antibodies, plasma) aimed to prevent or treat COVID-19 within 90 days <u>preceding</u> the planned administration of study vaccine (180 days in case of Evusheld*) 17. Participation in any research involving an investigational product (drug, biologic, device) within 12 weeks prior to vaccination and during the study. 18. Participant has donated ≥ 450ml of blood products within 12 weeks before screening. 19. Participant has any medical condition and/or finding that in the investigator opinion might increase participant risks, interfere with the study or impair interpretation of study data. <i>*treatment with immunotherapies (monoclonal antibodies, plasma) aimed to prevent COVID-19 (PrEP) will be allowed at any timepoint of the study after day 14 following current local guidelines. Those participants will be censored for the clinical endpoints on COVID-19 acquisition and disease outcomes and the immune analysis will be controlled by the treatment received</i>
Investigational Product	Single administration of COVID-19 HIPRA's vaccine (PHH-1V).
Reference product	Not applicable
Route of administration, pharmaceutical form and dosage	The investigational product is administered by the intramuscular route. It is presented in the following form: • COVID-19 HIPRA's vaccine (PHH-1V) multi-dose vial containing 10 doses ready to use. Each dose will consist in a volume of 0.5 ml of PHH-1V (40 µg of protein).
Analysis population	• Enrolled (EP): All participants who have signed the Informed Consent Form (ICF). • Intent-to-treat (ITT): All participants who meet the inclusion/exclusion criteria and received a dose of the study vaccine at Day0. • Modified Intent-to-treat (mITT): All participants in the ITT who meet the inclusion/exclusion criteria, received the study vaccine and did not tested positive for COVID-19 within 14 days of receiving the study vaccine

	• Immunogenicity (IGP): All participants in the immunogenicity subset of mITT who had a valid immunogenicity test result before receiving study vaccine and at least one valid result after dosing. • Safety (SP): All enrolled participants who received the study vaccine. This population will be used for all safety analyses.																								
Statistical Methods and Planned Analyses	Sample size: A sample size of 400 participants is proposed. This is an exploratory study to assess immunogenicity in special populations with some underlying immunosuppressive conditions. No formal sample size calculation is envisioned. As a means to characterize the statistical properties of this study, the table below presents the probability of observing zero, at least one, and two or more safety endpoints among the 400 participants for various “true” event rates: <table><tr><th>Event rate</th><th>P(0 events n=400)</th><th>P(≥1 event n=400)</th><th>P(≥2 events n=400)</th></tr><tr><td>1%</td><td>1.8%</td><td>98.2%</td><td>91%</td></tr><tr><td>5%</td><td>0%</td><td>100%</td><td>100%</td></tr><tr><td>10%</td><td>0%</td><td>100%</td><td>100%</td></tr><tr><td>15%</td><td>0%</td><td>100%</td><td>100%</td></tr><tr><td>25%</td><td>0%</td><td>100%</td><td>100%</td></tr></table> <i>P=Probability</i> Interim analysis: First immunogenicity analysis will be released once data of high throughput humoral immunogenicity at Day 91 of all enrolled participants in Spain. This report will also include data from safety assessments. Final analysis will be released with longitudinal follow-up up to 1 year since vaccination. Other interim analysis will be performed anytime in the study when sponsor deems it necessary. Statistical methods: No formal statistical analysis will be performed on the primary and secondary endpoints. Descriptive analysis will be performed for variables overall by time-point. Categorical variables will be presented by means of number of cases and frequencies (%) and continuous variables will be presented by number of non- missing observations, mean, standard deviation (SD), median, min and max; for the immunogenicity variables the geometric mean titre, geometric mean concentration, GMFR, and standard deviations will be presented, as	Event rate	P(0 events n=400)	P(≥1 event n=400)	P(≥2 events n=400)	1%	1.8%	98.2%	91%	5%	0%	100%	100%	10%	0%	100%	100%	15%	0%	100%	100%	25%	0%	100%	100%
Event rate	P(0 events n=400)	P(≥1 event n=400)	P(≥2 events n=400)																						
1%	1.8%	98.2%	91%																						
5%	0%	100%	100%																						
10%	0%	100%	100%																						
15%	0%	100%	100%																						
25%	0%	100%	100%																						

	appropriate. Dichotomised measures for immunogenicity will be presented as frequencies and percentages. 95% confidence intervals (CI) will be also provided, as appropriate. In general, missing data will not be imputed. For exploratory endpoints related to the immunogenicity endpoints, zero values will be imputed to half of the lower limit of quantification (LLOQ). If other parameters are deemed appropriate for imputation, information will be detailed in the Statistical Analysis Plan (SAP). Geometric mean titer (GMT), geometric mean concentration (GMC), GMFR and standard deviations will be calculated based on the log-transformed titers. Calculation of 95% CI will be based on the t-distribution of the log-transformed titers or the difference in the log-transformed titers for GMT and GMFR, respectively, then back transformed to the original scale. Further details on methodology for summary and statistical analyses of the data collected in this study will be detailed in the study SAP. Immunogenicity analysis The following data will be presented: • The neutralizing antibody titer and the total binding antibodies measured for each individual sample and the GMT with its 95% confidence intervals at baseline and at Days 14, 91, 82 and 365 (only for subjects in Spain) • The geometric mean fold rise (GMFR) in neutralizing antibody titer and in total binding antibodies from baseline to Day 14. • The percentage of participants that after PHH-1V vaccination have a ≥ 2-fold or ≥ 4-fold change in total binding antibodies titer from Baseline to Day 14 Safety analysis Solicited local reactions and systemic events from Day 0 through Day 7 after boost vaccination will be presented by intensity and cumulatively across severity levels. Unsolicited local and systemic reactogenicity adverse events from Day 0 through Day 28 after boost vaccination will be presented by intensity and cumulatively across severity levels.
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	Adverse Events and SAEs will be categorized according to Medical Dictionary for Regulatory Activities (MedDRA) terms and summarized by System Organ Class (SOC) and Preferred Term (PT). Summaries of local and systemic solicited AEs will be presented by SOC and PT for events occurring by Day 7. In addition, AEs will be summarized by maximum intensity and causal relationship to study drug. A separate summary of AESI, including potentially immune-mediated medical conditions (PIMMCs) and MAAE will be reported. Laboratory parameters will be summarized as actual values and change from baseline over time. Shift tables from baseline to worst on-study value and baseline to Days 14, 91 182 and 365 (only for participants in Spain) value will be reported. The safety analyses for the Phase IIb/III will be performed using the SP and for each primary vaccination scheme.
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LIST OF ABBREVIATIONS AND DEFINITION OF TERMS

Abbreviation	Definition
ACE-2	Angiotensin Converting Enzyme 2
ADE	Antibody-Dependent Enhancement
AE	Adverse Event
AESI	Adverse Event of Special Interest
AID	Autoimmune Disease
AZ	Vaxzevria Vaccine abbreviation
BUN	Blood Urea Nitrogen
CBC	Complete Blood Count
CI	Confidence Interval
COPD	Chronic Obstructive Pulmonary Disease
COVID-19	Coronavirus Disease 2019
CRA	Clinical Research Associate
CRO	Clinical Research Organisation
CTCAE	Common Terminology Criteria for Adverse Events
DBL	Data Base Lock
DSMB	Data Safety Monitoring Board
ECMO	Extracorporeal Membrane Oxygenation
eCRF	Electronic Clinical Report Form
ECLIA	Electrochemiluminescence immunoassay
ELISpot	Enzyme-Linked Immune Absorbent Spot
EP	Enrolled Population
ETV	Early Termination Visit
FDA	Food and Drug Administration
FSH	Follicle Stimulating Hormone
GCP	Good Clinical Practice
GMC	Geometric Mean Concentration
GMFR	Geometric Mean Fold Rise
GMT	Geometric Mean Titre
HIV	Human Immunodeficiency Virus
IB	Investigator's Brochure
IC ₅₀	Inhibitory concentration 50
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
ICS	Intracellular Cytokine Staining
ICU	Intensive Care Unit
ID ₅₀	Inhibitory dilution 50
IEC	Independent Ethics Committee
IGP	Immunogenicity Population
IM	Intramuscular
LLOQ	Lower Limit of Quantification
MAAE	Medically Attended Adverse Events
MedDRA	Medical Dictionary for Regulatory Activities

Abbreviation	Definition
Moderna	Spikevax Vaccine Abbreviation
mRNA	Messenger RNA
PBMC	Peripheral Blood Mononuclear Cell
PBNA	Pseudovirion-Based Neutralisation Assay
PCR	Polymerase Chain Reaction
PIMMCs	Potentially Immune-Mediated Medical Conditions
PrEP	Pre-exposure prophylaxis
PT	Preferred Term
RAT	Rapid Antigen Test
RBD	Receptor Binding Domain
RT-PCR	Reverse Transcription Polymerase Chain Reaction
SAE	Serious Adverse Events
SAP	Statistical Analysis Plan
SARS-CoV-2	Severe Acute Respiratory Coronavirus 2
SD	Standard Deviation
SOC	System Organ Class
SP	Safety Population
SpO ₂	Oxygen Saturation
SUSAR	Suspected Unexpected Serious Adverse Reaction
UAR	Unexpected Adverse Reaction
VDE	Vaccine-Dependent Disease Enhancement
VNA	Virus neutralisation assay
VOC	Variants of Concern
WHO	World Health Organization

1. INTRODUCTION

1.1 Disease Overview

At the end of December 2019, the World Health Organization (WHO) was informed about a cluster of cases of viral pneumonia of unknown cause in Wuhan, China. In mid-January 2020, the pathogen causing this atypical pneumonia was identified as a novel coronavirus, severe acute respiratory coronavirus 2 (SARS-CoV-2) and genome sequence data were published¹. Since then, the virus has spread globally. On 30 January 2020, the WHO declared the outbreak a Public Health Emergency of International Concern and on 11 March 2020 the WHO declared a pandemic². The pandemic is ongoing despite unprecedented efforts to control the outbreak.

SARS-CoV-2 is a positive-sense single-stranded RNA (+ssRNA) virus, with a single linear RNA segment. It is enveloped and the virions are 50-200 nanometres in diameter. Like other coronaviruses, SARS-CoV-2 has 4 structural proteins, known as the S (spike), E (envelope), M (membrane), and N (nucleocapsid) proteins.

The spike protein contains a polybasic cleavage site, a characteristic known to increase pathogenicity and transmissibility in other viruses. The spike protein is responsible for allowing the virus to attach to and fuse with the membrane of a host cell. The S1 subunit catalyses attachment to the angiotensin converting enzyme 2 (ACE-2) receptor present on cells of the respiratory tract, while the S2 subunit facilitates fusion with the cell membrane. The spike protein is considered a relevant antigen for vaccine development because it was shown that antibodies directed against it neutralise the virus and elicit an immune response that prevents infection in animals.

Human-to-human transmission of SARS-CoV-2 was confirmed in January 2020. Transmission occurs primarily via respiratory droplets from coughs and sneezes and through aerosols. The median incubation period after infection to the development of symptoms is 4 to 5 days. Most symptomatic individuals experience symptoms within 2 to 7 days after exposure and almost all symptomatic individuals will experience one or more symptoms before Day 12. Common symptoms include fever, cough, fatigue, breathing difficulties, and loss of smell and taste and symptoms may change over time³.

The major complication of severe Coronavirus Disease 2019 (COVID-19) is acute respiratory distress syndrome (ARDS), presenting as dyspnoea and acute respiratory failure that requires oxygen/ventilatory support including mechanical ventilation. In addition to respiratory sequelae, severe COVID-19 has been linked to other sequelae such as myocardial injury, arrhythmias, cardiomyopathy and heart failure, acute kidney injury often requiring renal replacement therapy, neurological complications such as encephalopathy, and acute ischemic stroke⁴.

The management of COVID-19 cases has developed since 2020, and today encompasses multiple treatments, including supportive care, anti-inflammatory agents, targeted immunomodulatory agents, anticoagulants, antivirals, monoclonal antibodies and antibodies administered from convalescent plasma. Guidelines are frequently updated considering emerging data of clinical trials and loss of efficacy against new variants of concern.

These therapies have shown variable and limited impact on the severity and duration of illness, with different efficacies depending on the stage of illness, manifestations of disease and underlying diseases, especially those considered as immunosuppressive conditions.

In addition to treatment, successful development of safe and effective vaccines against COVID-19 has significantly mitigated the impact of this disease, especially on the prevention of severe progression, hospitalization and death.

However, individuals with immunosuppressed / compromised conditions have been considered at risk of severe disease and have received priority access to COVID19 vaccines in most roll-out vaccine programs, even for some diseases, received additional doses in their primary vaccination regimens (<https://www.cdc.gov/coronavirus/2019-ncov/vaccines/recommendations/immuno.html>)

Since immunosuppressive conditions comprises a highly heterogeneous disease/treatment group, vaccine responsiveness as well as severity and fatality rate differ among pathologies and treatments. Pivotal COVID-19 vaccine clinical trials have in general excluded immunocompromised individuals and people receiving immunosuppressive treatments. Thus, vaccine-responsiveness in these individuals is not yet well characterized, especially after booster doses and in the context of current variants of concern which have already shown some degree of antibody viral escape.

Finally, in the current epidemiological context in which COVID19 restrictions are progressively being lifted, there is an urgent need to provide protection of vulnerable groups and mitigating the effects of the pandemic in these populations.

1.2 PHH-1V

Up to date, messenger RNA (mRNA)-based vaccines, recombinant adenovirus vectored vaccines and full spike protein vaccines have established the efficacy of vaccines for SARS-CoV-2 based on full-length Spike (S) protein^{5,6,7}.

Recombinant S protein produced in mammalian or insect cells has also shown immunogenicity and efficacy⁸. For large volume, low-cost production, however, protein-based vaccines incorporating the receptor binding domain (RBD) subunit remain an important alternative to the full S protein. Antibodies to RBD account for most of the neutralising activity elicited in a natural infection.

HIPRA has developed a recombinant protein RBD fusion dimer vaccine (PHH-1V) with an oil-water adjuvant equivalent to the commonly used adjuvant MF59C.1, for the prevention of COVID-19 caused by SARS-CoV-2, prepared as an emulsion for intramuscular administration in adults.

The adjuvant equivalent to MF59C.1 has been shown to be safe and to enhance immunogenicity of the vaccine in pre-clinical studies.

1.2.1 Phase I/IIa study with PHH-1V in adult healthy individuals

A first-in-human, phase I/IIa, randomised, controlled, observer-blinded, dose-escalation, multicentre clinical trial to evaluate safety and immunogenicity of COVID-19 HIPRA vaccine in adult individuals without underlying conditions, was conducted. A total of 30 healthy adults were allocated to one of the following cohorts:

- **Cohort 1:** 5 participants received 2 doses of COVID-19 vaccine HIPRA containing 10 µg of protein and 1 participant received 2 doses of Comirnaty (ARNm COVID-19 vaccine, Pfizer-BioNTech)

- **Cohort 2:** 10 Participants received 2 doses of COVID-19 vaccine HIPRA containing 20 µg of protein and 2 participants received 2 doses of Comirnaty (ARNm COVID-19 vaccine, Pfizer-BioNTech)
- **Cohort 3:** 10 participants received 2 doses of COVID-19 vaccine HIPRA containing 40 µg of protein and 2 participants received 2 doses of Comirnaty (ARNm COVID-19 vaccine, Pfizer-BioNTech)

The participants were allocated to the dose escalation cohort according to the order that they have been preselected. Participants at each cohort were randomised 5:1 to receive COVID-19 vaccine HIPRA or control commercial COVID-19 vaccine.

The safety data evaluated has been the number of participants with at least solicited local and systemic reactogenicity adverse events (AEs) for 7 days following each vaccination and the number of participants with at least one unsolicited AE for 28 days following.

The analysis related to the solicited AEs was performed as follow:

- Number (%) of participants with at least one solicited systemic AE, in general and by kind of systemic AE.
- Number (%) of participants with at least one solicited local AE, in general and by kind of local AE.
- Number (%) of participants with at least one systemic or local AE.

All analyses described above for the solicited AEs for 7 days following each vaccination were also performed by day and grade of intensity (1-mild, 2-moderate, 3-severe or 4 potentially life-threatening). These analyses were also performed for the maximum intensity over 7 days.

After the first vaccination, the major part of the participants in all the cohorts reported solicited AEs, however, the majority of them were local. After the second vaccination, there were fewer cases of reported solicited AEs than with the first vaccination in participants vaccinated with HIPRA vaccine. However, 100% of participants vaccinated with Comirnaty reported solicited AE with the second vaccination. The solicited systemic AE reported with the second vaccination were fewer than with the first vaccination in participants vaccinated with HIPRA vaccines, while there was an increase in % of solicited local and systemic AE reported, in participants vaccinated with Comirnaty (see Table 1 for summary % solicited AEs).

Table 1. Summary of Solicited AEs (% of participants)

Participants with at least one solicited AE during 7 days (%)	Cohort 10 µg n=5	Cohort 20 µg n=10	Cohort 40 µg n=10	Comirnaty n=5
After 1st vaccination				
Solicited AE	60	90	80	80
Solicited AE grade 3 or above	0	0	0	0
Solicited local AE	60	80	80	80
Solicited local AE grade 3 or above	0	0	0	0
Solicited systemic AE	20	60	60	0
Solicited systemic AE grade 3 or above	0	0	0	0
After 2nd vaccination				
Solicited AE	60	60	50	100
Solicited AE grade 3 or above	0	0	0	0
Solicited local AE	60	60	50	100
Solicited local AE grade 3 or above	0	0	0	0
Solicited systemic AE	0	30	20	60
Solicited systemic AE grade 3 or above	0	0	0	0

Abbreviations: AE = adverse event

Only Grade 1-2 solicited local and systemic AE occurred. The most frequent solicited local AE were local sensitivity and pain at the injection site. The most frequent solicited systemic AE were fatigue/tiredness and headache.

In the case of HIPRA vaccines, the most frequent solicited grade 2 local AE was local sensitivity in all the cases (first and second vaccination), while only one case experimented a solicited systemic grade 2 AE, fatigue/tiredness (see Table 2 for summary solicited AE grade 2). There wasn't any systemic grade 2 AE after the second vaccination in participants vaccinated with HIPRA vaccine. Participants vaccinated with Comirnaty suffered fever (50%), headache (66.7%), fatigue/tiredness (50%) and myalgia/muscle pain (50%).

Table 2. Summary of Solicited AE Grade 2 (% of participants)

	Cohort 10 µg n=5	Cohort 20 µg n=10	Cohort 40 µg n=10	Comirnaty n=5
After 1st vaccination				
Local AE				
Local sensitive	100	42.9	12.5	50
Headache	0	0	20	0
Systemic AE				
Fatigue / Tiredness	0	33.3	0	0
After 2nd vaccination				
Local AE				
Pain at the injection site	0	0	0	25
Local sensitivity	33.3	50	25	50
Systemic AE				
Fever	0	0	0	50
Headache	0	0	0	66.7
Fatigue / Tiredness	0	0	0	50
Myalgia / Muscle pain	0	0	0	50

Abbreviations: AE = Adverse Event

No DLTs appeared at any dose (10 µg, 20 µg, 40 µg) after the first or the second immunisations. No pre-specified trial halting rules were met.

No relevant laboratory changes from values at the entry of the study were detected in safety laboratory assessment.

Quantification of neutralising antibodies analysed by sero-neutralisation with pseudovirion (PBNA) and live virus (VNA), at baseline, 3 weeks after the first dose and 2 weeks after the second dose of vaccine was performed.

The results have shown a very good response in neutralising antibodies, for all the variants analysed that are specifically alpha, beta, gamma and delta. This response is already evident after the first dose of vaccine and is magnified after the second dose. In this phase, and even with individuals vaccinated with Pfizer vaccine in the study, statistical comparison has not been performed because the sample size, especially of the Pfizer group, is very low and makes no sense. However, by internal data we have from sample panels of people vaccinated with vaccines already registered, we can confirm that the HIPRA vaccine induces a neutralising antibody response that is quite similar to Pfizer vaccine, and even exceeding the Pfizer vaccine for beta variant. No statistics can be done at this step of clinical studies.

In addition to neutralising antibodies, another secondary objective was the analysis of immunogenicity in terms of total antibodies. To evaluate the immunogenicity by means of total antibody against Receptor Binding Domain of the Spike protein of SARS-CoV-2 quantification, measured by an electrochemiluminescence immunoassay (ECLIA). There is a clear seroconversion both after the first dose and after the second one, at which point the levels increase significantly.

A very important parameter is the fold rise which measures the increase in the level of antibodies between baseline and 3 weeks after the first dose, and between baseline and 2 weeks after the second dose of vaccine. It is accepted that a vaccine induces an antibody response that may be considered relevant, if this value is equal to or greater than 4.

It has been assessed that 2 weeks after the second dose of vaccine 100% of HIPRA vaccinated participants, from all 3 groups, exceed the fold rise value of 4. Moreover, it is observed that 3 weeks after the first dose, 100% of participants in the 20 and 40 microgram per dose of HIPRA vaccine groups, and 80% of participants in the 10 microgram per dose of HIPRA vaccine group, have exceeded the fold rise value of 4.

What concerns cell immunity, in the participants immunized with COVID-19 Vaccine HIPRA, a T cell mediated immunity was observed after vaccination. Specifically, results showed that vaccination with a 40 µg RBD fusion dimer/dose of COVID-19 Vaccine induced a specific T cell response, with a significant IFN-γ production after re-stimulation in vitro with RBD peptide mix from alpha (on D21 and D35), beta (on D21) and delta (on D35) SARS-CoV-2 variants compared with D0 (P<0.05).

In summary, we can confirm that the HIPRA vaccine induces a powerful immune response at both the level of total antibodies and neutralising antibodies. Furthermore, it has been shown that the neutralising antibody response is balanced among all tested variants.

Further details on the investigational vaccine and information on nonclinical studies can be found in the Investigator's Brochure (IB).

1.2.2 Phase IIb double-blind, randomised, active-controlled, multicentre, non-inferiority trial to assess immunogenicity and safety of a booster vaccination with PHH1V in adults fully vaccinated against COVID-19

In the Phase IIb study, 782 eligible participants who have received two doses of the Comirnaty vaccine, and were at least 182 days and less than 365 days after their second dose, were randomly assigned to the following two treatment arms in a PHH-1V: Comirnaty 2:1 ratio:

- Cohort 1: single booster dose of PHH-1V on Day 0
- Cohort 2: single booster dose of Comirnaty on Day 0

765 participants were vaccinated: 513 participants received the booster dose of PHH-1V, and 252 participants received the booster dose of Comirnaty.

Participants were provided with a paper diary to record local and systemic reactions after vaccination from Day 0 through Day 7. Unsolicited adverse events were reported through Day 28. SAEs, AESI and MAAE were reported during all the duration of the study. Participants were contacted through telephone on Day 7 for a safety assessment. Participants returned to the site on Days 14, 28, 182, and 364 (final visit) for blood sample collection and safety follow-up.

Find below the Topline results collected up to the cut-off date of 17-Feb-2022.

Regarding SAFETY:

Table 3. Treatment-emergent adverse events (TEAEs) reported during the study until the data cut off (Safety Population)

	PHH-1V (N=513)		Comirnaty (N=252)		Overall (N=765)	
	Events	Participants (%)	Events	Participants (%)	Events	Participants (%)
Total number of TEAEs	1595	460 (89.7)	1062	238 (94.4)	2657	698 (91.2)
Total number of serious TEAEs	0	0	0	0	0	0
Total number of participants with TEAEs leading to death		0		0		0
TEAE intensity [1]						
Mild	1394	343 (66.9)	886	146 (57.9)	2280	489 (63.9)
Moderate	189	109 (21.2)	165	85 (33.7)	354	194 (25.4)
Severe	12	8 (1.6)	11	7 (2.8)	23	15 (2.0)
Relationship to study treatment [1]						
Not related	151	21 (4.1)	77	7 (2.8)	228	28 (3.7)
Unlikely related	46	2 (0.4)	9	0	55	2 (0.3)
Possibly related	371	88 (17.2)	232	44 (17.5)	603	132 (17.3)
Probably related	291	79 (15.4)	201	35 (13.9)	492	114 (14.9)
Related	736	270 (52.6)	543	152 (60.3)	1279	422 (55.2)
Unrelated [2]	197	23 (4.5)	86	7 (2.8)	283	30 (3.9)
Related [3]	1398	437 (85.2)	976	231 (91.7)	2374	668 (87.3)

Abbreviations: N = the number of participants in the population, TEAE = treatment emergent adverse event.

A TEAE is defined as an adverse event that started on or after the date of administration of study treatment until 28 days thereafter.

[1] If a participant experienced more than one TEAE, the participant is counted once at the most severe or most related event.

[2] Unrelated adverse events are those classified as not related and unlikely related.

[3] Related adverse events are those classified as possibly, probably, and related. If a TEAE has a missing relationship it is assumed to be related to the study treatment for analysis purposes.

Table 4. Summary of Treatment-Emergent Adverse Events by MedDRA System Organ Class and Preferred Term in $\geq 1.0\%$ of Overall Participants (Safety Population)

System organ class Preferred term	PHH-1V (N=513)		Comirnaty (N=252)		Overall (N=765)	
	Events	Participants (%)	Events	Participants (%)	Events	Participants (%)
Total number of TEAEs	1595	460 (89.7)	1062	238 (94.4)	2657	698 (91.2)
General disorders and administration site conditions	1040	428 (83.4)	716	230 (91.3)	1756	658 (86.0)
Injection site pain	754	412 (80.3)	467	225 (89.3)	1221	637 (83.3)
Fatigue	166	141 (27.5)	115	106 (42.1)	281	247 (32.3)
Injection site induration	45	44 (8.6)	44	43 (17.1)	89	87 (11.4)
Injection site erythema	34	34 (6.6)	37	36 (14.3)	71	70 (9.2)
Pyrexia	15	14 (2.7)	24	23 (9.1)	39	37 (4.8)
Asthenia	10	9 (1.8)	3	3 (1.2)	13	12 (1.6)
Injection site swelling	3	3 (0.6)	8	7 (2.8)	11	10 (1.3)
Axillary pain	6	3 (0.6)	6	6 (2.4)	12	9 (1.2)
Chills	1	1 (0.2)	5	5 (2.0)	6	6 (0.8)
Nervous system disorders	207	169 (32.9)	129	106 (42.1)	336	275 (35.9)
Headache	195	161 (31.4)	122	101 (40.1)	317	262 (34.2)
Musculoskeletal and connective tissue disorders	123	112 (21.8)	108	94 (37.3)	231	206 (26.9)
Myalgia	109	101 (19.7)	93	86 (34.1)	202	187 (24.4)
Back pain	5	5 (1.0)	4	4 (1.6)	9	9 (1.2)
Arthralgia	3	3 (0.6)	5	4 (1.6)	8	7 (0.9)
Gastrointestinal disorders	101	69 (13.5)	29	21 (8.3)	130	90 (11.8)
Diarrhoea	41	38 (7.4)	7	6 (2.4)	48	44 (5.8)
Nausea	36	30 (5.8)	15	13 (5.2)	51	43 (5.6)
Vomiting	11	10 (1.9)	3	3 (1.2)	14	13 (1.7)
Infections and infestations	32	32 (6.2)	19	19 (7.5)	51	51 (6.7)
COVID-19*	13	13 (2.5)	8	8 (3.2)	21	21 (2.7)
Nasopharyngitis	6	6 (1.2)	3	3 (1.2)	9	9 (1.2)
Upper respiratory tract infection	5	5 (1.0)	1	1 (0.4)	6	6 (0.8)
Blood and lymphatic system disorders	13	13 (2.5)	17	17 (6.7)	30	30 (3.9)
Lymphadenopathy	9	9 (1.8)	16	16 (6.3)	25	25 (3.3)

System organ class Preferred term	PHH-1V (N=513)		Comirnaty (N=252)		Overall (N=765)	
	Events	Participants (%)	Events	Participants (%)	Events	Participants (%)
Respiratory, thoracic, and mediastinal disorders	19	17 (3.3)	4	4 (1.6)	23	21 (2.7)
Cough	5	5 (1.0)	0	0	5	5 (0.7)
Oropharyngeal pain	5	5 (1.0)	0	0	5	5 (0.7)
Investigations	9	9 (1.8)	5	5 (2.0)	14	14 (1.8)
SARS-CoV-2 test positive*	9	9 (1.8)	4	4 (1.6)	13	13 (1.7)

Abbreviations: N = the number of participants in the population, PT = preferred term, SOC = system organ class, TEAE = treatment emergent adverse event.

* Different trial centres collected cases with different terminology (COVID-19 or SARS-CoV-2) however the meaning is the same.

A TEAE is defined as an adverse event that started on or after the date of administration of study treatment until 28 days thereafter.

If a participant experienced more than one TEAE, the participant is counted once for each SOC and once for each PT.

SOCs are ordered in decreasing frequency of the total number of participants with TEAEs reported in each SOC and PTs are ordered within a SOC in decreasing frequency of the total number of participants with each TEAE.

Adverse events were coded using the MedDRA Dictionary, version 24.1.

One participant experienced one SAE, the event was recovered/resolved and unrelated to the vaccine. There were no deaths in the study.

Vaccination with PHH-1V and Comirnaty vaccines are overall well tolerated. There were no immune-mediated adverse events reported for both, PHH-1V and Comirnaty vaccines.

There were no cases of severe COVID-19 infection in any of the vaccine arms. The cases of COVID-19 infection reported for both vaccine arms were non-severe cases. Such findings may indicate that both vaccines provide protection to moderate, severe, life-threatening, and fatal forms of SARS-CoV-2 infections.

Regarding IMMUNOGENICITY:

- For the Wuhan SARS-CoV-2 variant, the neutralising antibody results do not demonstrate non-inferiority of PHH-1V to the Comirnaty vaccine in immune response at 14 days after the booster. Despite not demonstrating the non-inferiority, the PHH-1V vaccine was able to elicit high levels of neutralising antibodies. Considering that Wuhan SARS-CoV-2 is no longer the dominant strain in the pandemic, delta and mostly omicron are the most concerning variants circulating currently, therefore, the neutralisation against these variants has become much more relevant than that against Wuhan.

- The secondary objective assessed was to determine and compare the changes in immunogenicity against the VOCs (i.e. beta, delta, and omicron) at Baseline and Day 14 in participants who have received two doses of Comirnaty vaccine and PHH-1V as a booster versus participants who have received three vaccinations with the Comirnaty vaccine.
- For the beta SARS-CoV-2 variant, the neutralising antibody titres and the fold rise in neutralising antibody titres demonstrate superiority of the PHH-1V vaccine to the Comirnaty vaccine in immune response at 14 days after the booster vaccination.
- For the delta SARS-CoV-2 variant, results from neutralising antibody titres and the fold rise in neutralising antibodies titres demonstrates non-inferiority of the PHH-1V vaccine to the Comirnaty vaccine in immune response at 14 days after the booster vaccination.
- Results from neutralising antibody titres and the fold rise in neutralising antibodies titres for the omicron SARS-CoV-2 variant demonstrates superiority of the PHH-1V vaccine to the Comirnaty vaccine in immune response at 14 days after the booster vaccination.
- The percentage of participants that after a booster have a ≥ 4 -fold change in binding antibody titres from Baseline and Day 14 are similar between both vaccine arms, and a response in antibody titres was observed in most vaccinated participants.

1.2.3 Phase III, open Label, Single Arm, Multi-centre Trial to Assess the Safety and Immunogenicity of a Booster Vaccination with a Recombinant Protein RBD Fusion Heterodimer Candidate (PHH-1V) Against SARS-CoV-2 in Adults Vaccinated Against COVID-19

In the Phase III study, approximately 3.000 eligible adults aged above 16 years old will be given 1 dose of PHH-1V as booster vaccination. Participants are provided with a paper diary to record local and systemic reactions after vaccination from Day 0 through Day 7. Unsolicited adverse events are reported through Day 28. SAEs, AESI and MAAE are reported during all the duration of the study. Participants will return to the site on Days 14, 28, 182, and 365 (final visit only for the immunogenicity subset) for blood sample collection and safety follow-up.

Up to date (15th March 2022), approximately 2500 participants have been vaccinated with no major safety problems reported.

1.3 Study Design Rationale

Several vaccines against COVID-19 are in development and/or already approved for emergency use of in Europe. As of March 2022, five vaccines have been approved for emergency use in adults and adolescents and other four are under rolling review process. Approved vaccines have been developed using several different platforms, such as adenovirus-vectored vaccines (e.g., Vaxzevria / Covishield from AstraZeneca/Oxford and Ad26.COV2.S from Janssen/Johnson & Johnson), nanoparticles, mRNA vaccines (e.g., Comirnaty from Pfizer and Spikevax from Moderna) and recombinant protein (e.g. Nuvaxovid from Novavax).

Immunogenicity data for most of these vaccines available beyond 6 months after completed vaccination showed waning of immunity and emergence of new variants of the SARS-CoV-2 virus (e.g., Alpha [lineage B.1.1.7], Beta [lineage B.1.351], Gamma [lineage P.1], Delta [lineage B.1.617] and especially latest Omicron [lineage B.1.1.529]) by incorporation of several mutations to the spike protein, enabling antigens' escape. Some of these new variants are associated with high transmissibility and/or moderate or full resistance to the antibody response elicited by the current generation of COVID-19 vaccines and to most of the currently available treatments based on monoclonal antibody, resulting in an increase in the number of COVID-19 cases, more hospitalisations, and potentially more deaths and incurring in a higher demand of healthcare resources.

To address morbidity and mortality secondary to new SARS-CoV-2 variants, protect high-risk or vulnerable populations, and maintain long-term immunogenicity, several approaches are in development. Administration of a booster dose of known COVID-19 vaccines are being recommended to the general population as a third dose and as a fourth dose to the higher risk population, 3 months after the third, in some countries.

Booster vaccinations are being administered either with extra doses of the original prime vaccine (homologous booster), or with new or marketed vaccines different than the prime (heterologous booster). Preliminary clinical data suggests that both the homologous and heterologous booster regimes can be effective and safe ^{9,10,11,12,13}. In Spain, current vaccination strategy (Estrategia de vacunación frente a COVID19 en España, actualización 11 del 8 de febrero 2022) considers individuals at high risk of severe disease ('Grupo 7') individuals with solid organ transplant, recipients of hematopoietic stem cell transplantation, post CAR-T cell treatment, on immunosuppressive treatment, or having graft-versus-host disease regardless of the time since HSCT, as well as individuals with onco-haematological disease and solid organ cancer, all primary immunodeficiencies, excluding IgA deficiency and antibody formation defect, very high risk patients with certain immunosuppressive treatments, HIV infection with low CD4 cell counts, people with renal replacement therapy (haemodialysis and peritoneal dialysis), cystic fibrosis, Down syndrome aged 40 or over. In addition, individuals on chronic immunosuppressive therapies such as high doses of corticosteroids or during long periods of time, and biological and non-biological immunomodulators (i.e. anti-CD20 drugs including rituximab, ocrelizumab, ofatumumab, tositumomab, and ibritumomab among others) have also received priority access to COVID-19 vaccinations.

This group of individuals was recommended to receive a primary vaccination consisting of 3 doses and is now currently recommending a 4th dose 5 months after the last dose. However, its impact on the immune response on immunocompromised people has only been partially clarified and thus, a proper evaluation of the current vaccine regimens and additional boosters in immunocompromised individuals remains an utmost priority. In fact, some people with conditions at high risk of immunosuppression or who have received immunosuppressive treatment with some drugs such biological immunomodulators, it is plausible that insufficient or null response will be generated after booster vaccinations as well, still leaving them unprotected against severe disease after exposure to SARS-CoV-2. Treatments such as passive immunization given as pre-exposure prophylaxis might be an alternative to provide protection.

A Phase I/IIa study with HIPRA COVID-19 vaccine (PHH-1V) was conducted to generate initial data on the safety, reactogenicity, and immunogenicity of two doses, administered 21 days apart,

to adults between 18 and 39 years of age. Also, a Phase IIb is being conducted to compare the immunogenicity and safety of a booster dose of HIPRA's recombinant protein RBD fusion dimer vaccine as a heterologous booster (to individuals who have received the second dose of the Pfizer–BioNTech (Comirnaty) COVID-19 vaccine at least 182 days prior to the booster dose in this study) versus a homologous booster (participants who received the second dose of the Comirnaty COVID-19 vaccine at least 182 days prior to the booster dose in this study will receive a third dose of the Comirnaty vaccine). Finally, a Phase III trial is ongoing to assess the safety and tolerability of a booster dose of HIPRA's recombinant protein RBD fusion dimer vaccine as a heterologous booster to participants who are vaccinated (following local authorities recognized vaccination scheme) at least 91 days before administration of study vaccine. People who had a non-severe or asymptomatic COVID-19 infection has been included. Immunogenicity is being assessed in a subset of participants vaccinated with homologous doses of Vaxzevria or Spikevax vaccines and with heterologous doses.

This Phase IIb/III trial aims to assess the safety and immunogenicity of an additional heterologous dose of HIPRA's recombinant protein RBD fusion dimer vaccine in a subset of immunosuppressive conditions, such as kidney transplant, people under haemodialysis or peritoneal dialysis, primary antibody deficiencies under IgG replacement therapy, chronic HIV infection with low CD4 cell counts and individuals with autoimmune diseases under Rituximab/ocrelizumab treatment. Individuals will be included if having received their last dose of a recognized vaccination scheme at least 91 days before administration of study vaccine. As this population is at higher risk of COVID-19 acquisition and severe disease, people with past history of COVID-19 will be allowed to participate if infection occurred at least 91 days before inclusion provided they have been discharged from hospital at least 30 days before administration of study vaccine.

1.4 Rationale for Phase IIb/III trial to address safety and immunogenicity in special populations

Based on the efficacy to prevent COVID-19 symptomatic infection and severe disease demonstrated in phase IIb/III clinical trials, to date, many COVID-19 vaccines have been granted an emergency use authorization and or have been fully approved. However, the manufacturing challenges posed by the global demand for doses, the need for safe and effective vaccines that are accessible and affordable to diverse populations, the current lack of immunogenicity and efficacy data in certain populations such as individuals with immunosuppressive conditions, and the emergence of more-transmissible viral variants with high level of mutation driving antibody escape highlight the need for approval of a larger number of COVID-19 vaccines to achieve vaccine equity worldwide.

Vaccine-induced immune responses are often multifaceted. Some components (such as antibody responses) may correlate with the level of protection against acquisition and cellular responses, with protection against disease severity. However, antibody responses are easier to measure, standardized and thus more clinically applicable than cellular responses. Many studies connect neutralizing antibody responses to SARS-CoV-2 with vaccine efficacy, which has been considered a correlate of protection for vaccines against COVID-19 acquisition. However, no absolute correlate of protection has been established to date for COVID-19 vaccines due to the continuing research and emerging of new mutants.

Importantly, recent evidence shows that COVID-19 vaccines remain highly effective against COVID-19 related hospitalizations and deaths despite the waning of humoral immunity. However, many of current COVID-19 related hospitalizations and deaths occur in individuals with immunosuppressive conditions that probably remain under protected despite vaccination. Of note, most of pivotal trials with currently approved COVID-19 vaccines have excluded individuals with immunosuppressive conditions.

Waning of immunity increases the risk of acquiring new variants of concern (VOC) despite of the assumption that we are moving from a pandemic to an endemic scenario. For this reason, it is paramount that given the current circulating variants and the current state of the pandemic (even with the establishment of natural immunity in the population and high vaccination rates) that the levels of protection remain high to lessen the burden in healthcare systems and to preserve lives.

Data from phase IIb of the PHH-1V vaccine has shown potent neutralization capabilities against the delta variant 14 post vaccination (With a geometric mean titre ratio of 1.01 and a 95% confidence interval (CI) between 0.86 and 1.19) comparable to Comirnaty and superior to Comirnaty against beta 14 post vaccination (with a geometric mean titre ratio of 0.65 and a 95% confidence interval (CI) between 0.54 and 0.77) and against omicron 14 days post vaccination (with geometric mean titre ratio of 0.60 and a 95% confidence interval (CI) between 0.15 and 0.70, which, are the VOCs that to date, have shown high immune escape properties from the currently approved and or authorized vaccines. A high response in binding antibody titres was obtained in both arms with no difference in fold change between the vaccine arms.

Considering the public health implications of emerging SARS-CoV-2 VOC on the performance of all COVID-19 vaccines, faster development and approval of new vaccines with a better performance against the VOC in individuals with immunosuppressive conditions is urgent.

Immunosuppressive conditions considered at higher risk for severe COVID-19 disease are very heterogeneous and probably imply different degrees of immune suppression with different implications on vaccine responsiveness at either humoral and cellular level. To try to characterize the immune responses after vaccination in this group, the following conditions have been selected based on the immune bases of their underlying condition, potential for being under a stable condition, primary vaccination regimens received (mRNA-based, except in Turkey) and feasibility for recruitment:

Underlying Condition	Main characteristics for the clinical trial	Rationale for selection	Expected sample
HIV infection	CD4 cell counts <400, within last 6 months regardless of their pVL and ART.	Most clinical trials have only included optimally ART suppressed individuals with high CD4 cell counts, showing in this good vaccine responses and low risk of severe disease. Thus, data on those is missing and represent a high proportion of the HIV burden worldwide. Participating sites have already performed immunogenicity studies in these individuals so acceptability to participate in the trial is expected.	65-75 - all with primary vaccination based on mRNA-vaccines
Solid organ transplant - Kidney	>1 year since transplantation to avoid acute period with higher risk of rejection. >1 year since last dose of anti-CD20 and/or anti-CD3 treatment On stable immunosuppressive	After 1 st year from transplantation, in the absence of rejection, most individuals maintain a stable immunosuppressive regimen. Based on tacrolimus levels from last 3 months prior to	65-75 - all with primary vaccination based on mRNA-vaccines

	therapy based on at least 3 drugs (tacrolimus, glucocorticoids and mycophenolate or everolimus/sirolimus)	inclusion, and prior history of opportunistic infections, the degree of immunosuppression can be stratified. Individuals are routinely followed at the hospital so high acceptability to participate in the trial is expected.	30 – with Coronavac/mRNA regimen (Turkey)
Kidney replacement therapy	Individuals on haemodialysis or peritoneal dialysis for at least 6 months.	Underlying kidney conditions, concomitant treatments and history of opportunistic infections will help to address the degree of immunosuppression. Individuals are routinely followed at the hospital so high acceptability to participate in the trial is expected.	65-75 - all with primary vaccination based on mRNA-vaccines 30 – with Coronavac/mRNA regimen (Turkey)
Primary antibody deficiencies	Individuals on chronic IgG replacement therapy	Individuals with disorders characterized by an inability to produce clinically effective immunoglobulin responses, therefore, poorer vaccine responsiveness and are more prone to several type of infections. Participating sites have already performed immunogenicity studies in these individuals so acceptability to participate in the trial is expected.	45-55 - all with primary vaccination based on mRNA-vaccines

Autoimmune diseases (AID) on treatment with Rituximab/ Ocrelizumab	Rituximab / Ocrelizumab treatment for at least 6 months.	Rituximab/ Ocrelizumab is also widely used for onco-hematologic diseases. However, those are expected to be in more unstable conditions for participating in a clinical trial such this one. AID are an heterogeneous group of diseases (vasculitis, myositis, SLE/Sjögren, rheumatoid arthritis and multiple sclerosis) but the immunodeficiency related to COVID-19 seems to be more driven by the immunosuppressive treatment than the underlying condition itself. Rituximab / Ocrelizumab has very long-term effects on lymphocyte populations, therefore, these individuals are at higher risk of poorer vaccine responsiveness and severe disease for long periods of time. Participating sites are all hospitals with rheumatology and neurology units with individuals under rituximab / ocrelizumab therapy	65-75 - all with primary vaccination based on mRNA-vaccines
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1.5 Risk-Benefit Assessment

1.5.1 Known Potential Risks

Nonclinical studies show that PHH-1V at different antigenic dose levels are well-tolerated in relevant animal species without identified safety risks. Regarding the adjuvant, as with every vaccination and based on previous clinical experience with influenza vaccines containing MF59C.1, local reactions (e.g., pain, tenderness, erythema/redness, and induration/swelling at the injection site) and systemic AEs (e.g., fever, headache, fatigue, chills, myalgia, arthralgia, nausea/vomiting, and diarrhoea) are expected side effects that typically resolve within 24 hours after the vaccination with or without treatment.

As for every vaccine and for general population, the occurrence of allergic/anaphylactic reactions in individuals with immunocompromised conditions cannot be excluded and emergency equipment for the treatment of such reactions must be available at the study site. These events are unexpected and constitute a potential important medical risk. All participants will be closely observed on site for at least 15 minutes after vaccination, and those individuals with known history of allergies to past vaccinations, for at least 30 minutes after vaccination.

Vaccine-dependent disease enhancement (VDE) describes a phenomenon in which pre-existing immunity is not enough to neutralise viral infection and may lead to severe disease progression. The risk of antibody-dependent enhancement (ADE) is defined as when the antibodies generated during an immune response recognise and bind to a pathogen, but they are unable to prevent infection. The risks of developing either VDE or ADE is considered low after the administration of PHH-1V. The factors that are postulated to contribute to VDE (based on observations in animal studies for SARS or MERS) are the use of inactivated virus, recombinant wild-type non stabilised S protein, nucleoprotein vaccines, aluminium, or other adjuvants inducing a Th2 bias; by design these factors are not applicable to PHH-1V.

Furthermore, PHH-1V induces a balanced immune response, as observed in nonclinical studies and documented in the IB in which cellular response was also assessed and a Th1 response was observed.

Participants who experience COVID-19 disease during the study will be followed closely to ensure clinical symptoms and safety data are collected and disease progression is monitored and reported. Results on developmental toxicity studies are not still available for COVID-19 vaccine HIPRA yet. However, the teratogenicity risk is deemed low. Given that human data on pregnancies is not yet available, the teratogenic risk associated with PHH-1V administration cannot be ruled out at this moment. For this reason, inclusion of female participants of childbearing potential age requires use of a highly effective contraceptive measure from the Screening through 8 weeks after vaccination (Day 0) or sexual abstinence beginning at least 21 days before Screening.

In addition, a list of AEs of special interest (AESIs) will be monitored following administration of PHH-1V. If any suspected AESI occurs in a participant who received PHH-1V, a diagnostic workup should be performed by a specialist depending on the type of suspected reaction (e.g., endocrinologist for suspected autoimmune thyroiditis) and this condition will be monitored and documented throughout the study.

PHH-1V has not yet been investigated in individuals with underlying immunosuppressive conditions or treatments but it is not expected to show different safety and tolerability profile. Given the mechanism of action and limited data on primary vaccination responsiveness in this special population, which relies on building up an adequate immune response, it is expected that this group might show reduced induction of a specific immune response against the SARS-CoV-2 RBD of S protein. Thus, it is important to address in depth additional aspects of both humoral and cellular immune response to evaluate potential immune compensation mechanisms. In contrast, drugs that enhance the immune response like certain cytokines (IFN- α , IL-2) may increase the response to the vaccines, which could theoretically result in increased immunogenicity, but also in an increased risk of toxicity.

Risks from phlebotomy for blood sampling are well known and minimal. Venepuncture is a routine procedure that the medical community commonly uses to obtain blood samples. Immediate complications may include slight pain during puncture of the skin and, rarely, dizziness and syncope. Additionally, a hematoma may result from the venepuncture, but this has minimal risk. Monitoring and using aseptic techniques, such as using sterile disposable blood collection apparatuses and adhering to standard medical precautions, reduce any risk to a minimum. The amount of blood to be taken for sampling will be adequate for the analysis envisioned and ensured to not be harmful to the participant's health.

1.5.2 Known Potential Benefits

Participants receiving PHH-1V may or may not mount a similar immunogenicity response to other commercially available vaccine boosters, depending on their primary immunosuppressive condition. Data obtained in Phase I/IIa indicate that PHH-1V is a good vaccine candidate in terms of safety and immunogenicity and also data obtained in Phase IIb indicate that HIPRA's heterologous booster produces similar protection in terms of neutralising antibodies than Comirnaty's homologous booster. In terms of safety, HIPRA's booster demonstrated a good safety profile after Comirnaty's primary vaccination scheme. Interim results from the ongoing Phase IIb trial are confirming safety and immunogenicity data and proceeding with clinical studies in special populations is of public health interest.

1.5.3 Overall Benefit/Risk Conclusion

In the current pandemic scenario, the two objectives of COVID-19 vaccination are the direct protection of the vaccinated individuals against infection and severe disease and the indirect protection of the population (even for those who have not been vaccinated) by reducing overall viral transmission and, thereby, the risk of infection.

The overall benefit/risk assessment for this study is considered acceptable for the following reasons:

1. The selection criteria include provisions to minimise the risk and protect the well-being of participants in the study. Eligibility will be re-assessed before the administration of the vaccine.
2. Safety will be closely monitored during the study.

3. The data gathered in the phase IIb studies has shown that HIPRA's booster immunogenicity in general population is similar to the already marketed Pfizer's vaccine. Therefore, the benefit of an additional dose with HIPRA vaccine in individuals with immunosuppressive conditions needs to be addressed. Associated with its good safety profile, justifies its assessment as an heterologous additional dose in previously vaccinated participants in individuals with immunosuppressive conditions. The safety profile of HIPRA vaccine in Phase IIb and III is indicating that this vaccine has a low reactogenicity profile and is considered well tolerated between participants.

2. STUDY OBJECTIVES

2.1 Study Objectives

2.1.1 Primary Objective

- To determine immunogenicity measured by pseudovirus neutralization against Omicron BA1 at Baseline and at Day 14 after administration of HIPRA's vaccine (PHH-1V).
- To determine immunogenicity measured by live virus neutralization for the HIV cohort against Omicron at Baseline and Day 14 after administration of HIPRA's vaccine (PHH-1V).

2.1.2 Secondary Objectives

- To determine and compare the changes of the immunogenicity measured by pseudovirus neutralization against Omicron, D614G, Beta and any other relevant Variants of Concern (VOC) in the epidemiologic moment at Days Baseline, 14, 91 and 182, and 365 only for Spanish participants, after administration of HIPRA's vaccine (PHH-1V).
- To determine and compare the changes of the immunogenicity measured by live virus neutralization for the HIV cohort against Omicron at days 91, 182 and 365 after administration of HIPRA's vaccine (PHH-1V).
- To determine and compare the changes of the immunogenicity measured by live virus neutralization for the HIV cohort against D614G and any other VOC in the epidemiologic moment at days Baseline, 14, 91, 182 and 365 after administration of PHH-1V.
- To evaluate the immunogenicity measured by means of total antibody against Receptor Binding Domain of the Spike protein of SARS-CoV-2 quantification, measured by an electrochemiluminescence immunoassay (ECLIA) at Baseline and at Days 14, 91 and 182, and 365 only for Spanish participants, after administration of HIPRA's vaccine (PHH-1V).
- To assess the safety and tolerability of PHH-1V as an additional dose in adult individuals with pre-existing immunosuppressive conditions.

2.1.3 Exploratory Objectives

- To assess number of participants with SARS-CoV-2 infections ≥ 14 days after PHH-1V administration.
- To assess number of COVID-19 severe infections ≥ 14 days after PHH-1V administration.
- To further evaluate humoral responses by measuring IgG subclasses usage and avidity of humoral immunity at Baseline and Days 14, 91 and 182, and 365 only for Spanish participants.
- To evaluate T-cell mediated responses, their immune phenotype polyclonally with potential cross-reactivity capacity and gene-expression profiles against the SARS-CoV-2 RBD proteins at Baseline and at Days 14, 91 and 182, and 365 only for Spanish participants in a subset of approximately 50% of participants.

3. STUDY PLAN

3.1 Overall Design

This is a phase IIb/III, open label, single arm, multi-center clinical trial that aims to determine safety, reactogenicity, tolerability and immunogenicity of additional vaccination with a recombinant protein receptor binding domain (RBD) fusion heterodimer candidate against SARS-CoV-2 developed by HIPRA (PHH-1V) in individuals with pre-existing immunosuppressive conditions.

In this phase IIb/III study, approximately 400 eligible individuals who have received primary vaccination against COVID-19 with 3 doses of mRNA (or 2 doses plus a documented history of COVID) last dose being administered at least minimum of 91 days will be included. Individuals recruited in Turkey which received a primary vaccination regimen using a combination of Coronavac and/or Comirnaty with or without additional boosters of the same vaccines will be allowed to be included provided that last booster dose being administered is at least 91 days before day 0. Individuals with history of COVID-19 disease will be allowed to be included if diagnosed at least 91 days and passed 30 days from hospital discharge prior to inclusion and provided they are no suffering from long covid (Post-acute COVID-19 syndrome).

Screening and baseline visits will be allowed on the same day.

All participants will receive a single dose of PHH-1V on Day 0 and will be observed for 15 minutes, except for those participants reporting previous allergy reactions to vaccination that will be observed for 30 minutes.

All participants will be provided with a paper diary on Day 0 to record solicited local and systemic reactions after vaccination through Day 7. Unsolicited adverse events will be reported through Day 28 except for COVID-19 cases that will be recorded in eCRF throughout the study. SAEs, AESI and MAAE will be reported during all the study.

On-site follow-up visits will be performed on Days 14, 91, 182 and 365 for Spanish subjects, and Days 14, 91 and 182 for Turkish subjects.

All 400 included individuals will be evaluated for high throughput humoral immunogenicity (primary objective). All participants will be evaluated for safety (secondary endpoint). A subset of about 50% of the enrolled individuals will be further characterized in terms of humoral immunity (IgG subclasses, cross-reactivity to other coronaviruses and avidity) and T cell responses (exploratory objectives) that requires PBMC isolation which will be performed at some of the Spanish clinical sites. This subset will be selected based on pre-existing conditions.

A DSMB may review safety data at any time point during the study as necessary and/or if requested by any of the involved PIs in case of observation of serious adverse events (SAEs).

An interim analysis is envisioned after data on humoral response at Day 91 of all enrolled participants will be available. A final analysis will include exploratory humoral and cellular objectives and the analysis of long-term immunogenicity response of the PHH-1V vaccination.

3.2 Study Endpoints

3.2.1 Immunogenicity Endpoints

Primary:

- Neutralization titre against Omicron BA1 measured as inhibitory concentration 50 (IC50) by a pseudovirion-based neutralization assay (PBNA) and reported as reciprocal concentration for each individual sample and geometric mean titer (GMT) for descriptive statistics analysis at Baseline and at Day 14.
- The geometric mean fold rise (GMFR) in neutralizing antibody titre from baseline to Day 14.
- Neutralization titre against Omicron measured as ID50 (the reciprocal dilution inhibiting 50% of the infection) by a live virus assay (VNA) to measure cytopathic effect in Vero E6 cells at Baseline and Day 14 after PHH-1V administration.

Secondary:

- Neutralization titer against Omicron, D614G, Beta strains, and any other relevant VOC in the epidemiologic moment, measured as inhibitory concentration 50 (IC50) by a pseudovirion-based neutralization assay (PBNA) and reported as reciprocal concentration for each individual sample and geometric mean titer (GMT) for descriptive statistics analysis at Days Baseline, 14, 91 and 182 for all participants, and 365 only for Spanish participants.
- Neutralization titer against Omicron measured as a ID50 (the reciprocal dilution inhibiting 50% of the infection) by a live virus assay (VNA) to measure cytopathic effect in Vero E6 cells at Days 91 182 and 365 after PHH-1V administration.
- Neutralization titer against D614G and any other VOC in the epidemiologic moment, measured as a ID50 (the reciprocal dilution inhibiting 50% of the infection) by a live virus assay (VNA) to measure cytopathic effect in Vero E6 cells at Days Baseline, 14, 91, 182 and 365 after PHH-1V administration
- Binding antibodies titer measured for each individual sample and GMT for descriptive statistics analysis at Baseline and Days 14, 91 and 182 for all participants, and 365 for Spanish participants.
- The geometric mean fold rise (GMFR) in binding antibody titer from baseline to Day 14.
- The percentage of individuals that after PHH-1V vaccine have a ≥ 2 -fold and 4-fold change in binding antibodies titer from Baseline to Day 14.

3.2.2 Safety Endpoints

Secondary:

- Number, percentage, and characteristics of solicited local reactions and systemic events through Day 7 after vaccination.
- Number, percentage, and characteristics of unsolicited local and systemic adverse events (AEs) through Day 28 after vaccination.

- Number and percentage of serious adverse events (SAEs) through the end of the study.
- Number and percentage of adverse event of special interest (AESI) through the end of the study.
- Number and percentage of medically attended adverse events (MAAE) related to study vaccine through the end of the study.
- Grade 3 and 4 change from baseline in safety laboratory parameters at Days 14, 91 and 182 for all participants, and 365 for Spanish participants, after vaccination.

3.2.3 Exploratory Endpoints

Safety

- Number and percentage of participants with SARS-CoV-2 infections ≥ 14 days after PHH-1V administration according to COVID-19 infection criteria and throughout the study duration.
- Number and percentage of COVID-19 severe infections ≥ 14 days after PHH-1V administration and through the end of the study requiring hospitalization.
- Number and percentage of intensive care unit (ICU) admissions associated with COVID-19 ≥ 14 days after PHH-1V administration and through the end of the study.
- Number and percentage of non-invasive ventilation administration associated with COVID-19 ≥ 14 days after PHH-1V administration and through the end of the study.
- Number and percentage of deaths associated with COVID-19 ≥ 14 days after PHH-1V administration and through the end of the study.

Immunogenicity

- Quantification of IgG subclasses (IgG1, IgG2, IgG3 and IgG4) measured by ELISA at Baseline and Days 14, 91 and 182 for all participants, and 365 for Spanish participants.
- Cross reactivity with Spike glycoproteins from other coronavirus (SARS-CoV, MERS-CoV, HKU1, OC43, NL63, 229E) measured at Baseline and Days 14, 91 and 182 for all participants, and 365 only for Spanish participants.
- Avidity of the main antibodies elicited against the Spike glycoproteins determined by ELISA at Baseline and Days 14, 91 and 182 for all participants, and 365 only for Spanish participants.
- Frequency of IFN γ positive T-cells measured by enzyme-linked immune absorbent spot (ELISpot) covering the RBD sequence of D614G, RBD Omicron, RBD Beta and RBDDelta and any other relevant Variants of Concern (VOC) in the epidemiologic moment at Baseline and at Days 14, 91, 182 and 365 in a subset of approximately 50% of participants.

- Immune phenotype of SARS-CoV-2 specific CD4+/CD8+ T-cell responses measured by intracellular cytokine staining (ICS) flow-cytometry at Baseline and at Day 14 in a subset of 50% of participants.
- T cell transcriptional profiles and TCR usage of RBD-specific T-cells isolated using cell sorting with CD25, CD69 and CD137 activation markers and subjected to single cell transcriptomics analyses (sc-RNAseq) at Baseline and at Day 14, 182 and 365 in a subset of 50 individuals selected based on the humoral, ELISPOT and Flow cytometry data.

3.3 Study Termination

This study may be prematurely terminated if there is sufficient reasonable cause, including, but not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Suboptimal recruitment
- Insufficient compliance to protocol requirements
- Data that are not sufficiently complete and/or not evaluable
- Regulatory authorities request

If the study is prematurely terminated, the Investigator will promptly inform study participants and the authorities as applicable. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule. The Investigator will assure appropriate follow-up for the individuals, as necessary.

The Sponsor will notify regulatory authorities as applicable.

3.4 End of Study

The end of the study is defined as the date of the last visit of the last individual enrolled in the study.

4. STUDY POPULATION

4.1 Number of Participants

In this Phase IIb/III approximately 400 individuals with underlying immunosuppressive conditions will be administered with an additional dose COVID-19 vaccine HIPRA's.

In Spain, the 4th dose of the vaccine has been already approved for this specific group of patients, but there are some people that haven't received this dose yet, due to rejection at that time, a COVID-19 infection, or other non-specified causes. These people, in addition to those that have two doses and an episode of COVID-19 infection, are potential volunteers to be recruited in Spain.

In Turkey, at the moment of submission of this amendment, booster doses have already been widely implemented in these populations. Thus, individuals who have received booster doses after a primary vaccination with Coronavac and/or Comirnaty are potential volunteers to be recruited in Turkey.

4.2 Inclusion Criteria

The inclusion and exclusion criteria for enrolling are described below. If there is a question about these criteria, the investigator must consult with the appropriate sponsor representative and resolve any issues before enrolling a participant in the study.

Participants must meet all the following criteria to be considered eligible for the study:

1. Male, female or transgender, ≥ 18 years old at Day 0.
2. Participant must provide consent indicating that she or he understands the purpose and potential risks and is willing and able to participate in the study and comply with all the study requirements and procedures (scheduled visits, laboratory tests, complete diaries, etc).
3. Participant who has one of the following SARS-CoV-2 vaccination schemes:

For sites in Spain, the following vaccination schemes will be allowed:

- Three doses of mRNA vaccines (Comirnaty and/or Spikevax) with or without past history of positive test for COVID-19, provided that last vaccination and diagnose of COVID-19 is at least 91 days before day 0, and if hospitalization was required, participant was discharged from hospital at least 30 days before day 0.
- Two doses of mRNA vaccines (Comirnaty and/or Spikevax) and previous history of positive test for COVID-19, provided that last vaccination and diagnose of COVID-19 is at least 91 days before day 0, and if hospitalization was required, participant was discharged from hospital at least 30 days before day 0.

For sites in Turkey where vaccination with Coronavac was started, the following vaccinations schemes will be allowed:

- A complete primary scheme of the following vaccines: Coronavac and/or Comirnaty, with or without additional boosters of the same vaccines, being the last vaccination at least 91 days before Day 0. Participants with or without history of a positive test for COVID-19 are allowed. The infection (first day of positive test, RAT or PCR) must have been at least 91 days before Day 0.

4. Participant who has any of the following underlying immunosuppressive conditions*:

- Confirmed HIV infection with CD4 T cell counts <400 within last 6 months prior to Day0 regardless of pVL determination and ARV;
- Primary Antibody Deficiency Disorder on immunoglobulin replacement therapy for at least 6 months prior to Day0 (maintenance dose);
- Kidney disease on dialysis program for at least 6 months prior to Day0;
- Kidney transplant at least >1 year that have never been treated with anti-CD20/anti-CD3 or has been given at least >1 year prior to Day0 AND on maintenance immunosuppressive therapy based on at least 3 drugs: tacrolimus/cyclosporine, glucocorticoids and mycophenolate or everolimus/sirolimus.
- Auto-immune disease (AID**) on treatment with rituximab (RTX) / ocrelizumab during at least 14 days within the last 6 months prior to Day0

**approximately 60 participants per condition will be included*

*** AID includes: vasculitis, myositis, SLE/Sjögren, rheumatoid arthritis, multiple sclerosis, or other.*

5. Contraceptive use should be consistent with local regulation for participants in clinical trials.

a. Female participants of childbearing potential [defined as any female who has experienced menarche and until becoming postmenopausal* (defined as having ≥ 12 months amenorrhea prior to screening without an alternative cause) unless is surgically sterile]:

- Have a negative pregnancy test on the day of vaccination.
- Use of any acceptable contraceptive method that should be started on day 0 and until 8 weeks after vaccination, except hormonal contraception. Acceptable contraceptive methods are:
 - Hormonal contraception (progestogen-only or combined): oral, injectable or transdermal (patch) started at least 28 days before day 0 and until 8 weeks after vaccination
 - Intrauterine device.
 - Vasectomized partner (the vasectomized partner should be the sole partner for that participant).
 - Sexual abstinence **, as a form of contraception, is acceptable if in line with the participant's lifestyle.
 - Condom

b. Male participants:

- Vasectomized participants.
- Refrain from donating sperm for at least 28 days after day 0.
- Agree to use a male condom may be considered in women of childbearing potential partners, from screening and for at least 8 weeks after vaccination.
- Sexual abstinence**, as a form of contraception, is acceptable if in line with the participant's lifestyle.

** A high follicle stimulating hormone (FSH) level in the postmenopausal range may be used to confirm a post-menopausal state in women not using hormonal contraception or hormonal replacement therapy.*

*** Sexual abstinence is considered an effective method only if defined as refraining from heterosexual intercourse from screening until 8 weeks after receiving the vaccine for female participants and from screening until 4 weeks for male participants. Periodic abstinence (e.g., calendar, ovulation) and withdrawal are not acceptable methods of contraception.*

4.3 Exclusion Criteria

The exclusion criteria for enrolling are described below. If there is a question about these criteria, the investigator must consult with the appropriate sponsor representative and resolve any issues before enrolling a participant in the study.

Participants meeting any of the following criteria will be excluded from the study:

6. History of anaphylaxis to any prior vaccine.

7. Participant received or plans to receive:

- Live attenuated vaccines (licensed) within 4 weeks before or after receiving any study vaccine.
- Other not live vaccines (licensed) within 14 days before and after receiving any study vaccine
- Viral-vectored COVID-19 vaccines, such as Vaxzevria (AstraZenca) or Ad26.CoV2.S from Janssen

8. Pregnancy or breast-feeding at screening or Day 0 (vaccination time-point) or willingness/intention to become pregnant during the entire length of the study for female participants. For male participants, willingness/intention that your partner becomes pregnant during the entire length of the study.

Medical conditions

9. A confirmed COVID-19 diagnose (by RT-PCR or RAT, asymptomatic or symptomatic) <90 days prior to Day0

10. Participant has a clinically significant acute illness (this does not include minor self-limited illness such as mild diarrhea) or fever (temperature $\geq 38^{\circ}\text{C}$ (100.4°F) at screening or within 48 hours prior to the planned vaccination (Day 0).

11. Participant had a surgery requiring hospitalization (defined as inpatient stay for > 24 hours) before vaccination and he/she has not received the hospital discharge at day 0; or has a surgery requiring hospitalization planned within 12 weeks after study vaccine administration. Minor surgical procedures not requiring hospitalization are accepted.

12. Participant has ongoing severe and non-stable psychiatric condition likely to affect participation in the study (e.g., ongoing and non-stable severe depression, recent suicidal ideation, severe eating disorder, psychosis)

13. Participant has a problematic or risk use of substances including alcohol (except tobacco) that can compromise the study follow-up. Problematic or risk use of psychoactive substances is understood as the one that causes evident damage, whether it is dependence or any other physical, psychological, or social problem or that carries a high risk of suffering these damages. The negative consequences that consumption causes to third parties could be included.

14. Participant has a bleeding disorder (e.g., factor deficiency, platelet disorder), blood dyscrasia, or continuous use of anticoagulants or has any condition that in the opinion of the investigator contraindicates intramuscular injections or frequent phlebotomy. The use of ≤ 325 mg of aspirin or ≤ 75 mg of clopidogrel per day as prophylaxis is permitted but not combined.

15. Participant suffering from post-acute COVID-19 syndrome / long-covid.

Prior/Concomitant Therapy and Clinical Study Experience.

16. Participant received any immunotherapy (monoclonal antibodies, plasma) aimed to prevent or treat COVID-19 within 90 days preceding the planned administration of study vaccine (180 days in case of Evusheld)*.

17. Participation in any research involving an investigational product (drug, biologic, device) within 12 weeks prior to vaccination and during the study.

18. Participant has donated ≥ 450 ml of blood products within 12 weeks before screening.

19. Participant has any medical condition and/or finding that in the investigator opinion might increase participant risks, interfere with the study or impair interpretation of study data.

**treatment with immunotherapies (monoclonal antibodies, plasma) aimed to prevent COVID-19 (PrEP) will be allowed at any timepoint of the study after day 14 following current local guidelines. Those participants will be censored for the clinical endpoints on COVID-19 acquisition and disease outcomes and the immune analysis will be controlled by the treatment received*

4.4 Participant Identification and Registration

Individuals who are candidates for enrolment into the study will be evaluated for eligibility by the Investigator to ensure that the inclusion and exclusion criteria (see [Sections 4.2](#) and [4.3](#)) have been satisfied and that the individual is eligible for participation in this clinical study.

Once an individual is determined to be eligible by the clinical study site, the individual will be registered and assigned a sequential and unique participant number. Once a participant number has been assigned, it cannot be reused.

No individual may be enrolled or begin treatment prior to registration and assignment of a participant number.

4.5 Withdrawal and Replacement of Participants

An individual may discontinue study participation for any of the following reasons:

- If he/she is unwilling or unable to meet the protocol requirements.
- If the participant or the Investigator considers it best to end his/her participation in the study.
- Lost to follow-up.
- Withdrawal of consent from the individual or legal tutors.

All participants have the right to withdraw their consent at any time during the study without prejudice to them.

If possible, the participant withdrawing consent or discontinuing the study should complete an early termination visit.

The date and reason for discontinuation or consent withdrawal will be documented in the eCRF.

Participants who withdraw or discontinue from the study will not be replaced.

A participant will be considered lost to follow-up if he or she repeatedly fails to return for scheduled visits and is unable to be contacted by the study site. Reasonable efforts will be made, and documented, by site personnel to contact the participant to continue with their follow-up before determination that the participant is lost to follow-up.

5. STUDY TREATMENTS

5.1 Study Drug Supply and Storage

HIPRA SCIENTIFIC S.L.U. is responsible for manufacturing PHH-1V in accordance with Good Manufacturing Practice.

PHH-1V will be supplied in a vial containing 10 doses of 0.5 ml (40 µg), ready to use. There is no need to dilute or reconstitute. PHH-1V will be shipped to clinical sites and must be stored in a refrigerator (between 2°C and 8°C). Vials must not be frozen.

5.2 Study Drug Dose and Administration

Participants will receive a single dose of PHH-1V (40 µg) on Day 0, administered via IM injection into the deltoid muscle, preferably into the nondominant arm. Refer to the IMP Manual for details of PHH-1V administration.

5.3 Method of Assigning Participants to Treatment

All participants will be administered with a PHH-1V dose.

Participants will be assigned a unique Participant ID and will be identified only by the Participant ID number to protect confidentiality. The site will keep its own participant identification log to match the Participant ID number with the individual's personal data, in accordance with applicable regulatory data protection requirements.

5.4 Blinding, Packaging, and Labelling

5.4.1 Packaging and Labelling

PHH-1V will be packaged, labelled and supplied by the Sponsor in accordance with European regulation (as set in Annex VI of the Regulation EU No 536/2014).

5.5 Duration of Individual Participation

The study consists of a Pre-screening visit (remote by phone or on-site) maximum at -28 days before the Screening/Vaccination Visit and a Follow-up Period of 365 days or 182 days (for Spanish and Turkish participants, respectively). Therefore, an individual may participate in this study for up to 13 or 7 months.

5.6 Assessment of Treatment Compliance

All doses of PHH-1V will be administered at the study site by site personnel. The time and date of dose administration will be recorded in the eCRF. No other assessments of compliance will be conducted.

5.7 Study Drug Accountability

During the study, investigational product accountability will be monitored by the pharmacy receipt, dispensing log, and accountability log (that includes all the information related to the investigational product dispensed and returned).

The site personnel who administer the injection will be responsible for ensuring that the return of the used vials is recorded in the accountability log at the end of the immunisation.

For the Phase IIb/III, study drug accountability will be monitored by the CRA.

At the completion of the study, used and unused vials of the investigational product will be destroyed onsite by pharmacy staff or returned to the supplier according to local pharmacy guidelines and applicable regulations. Authorisation must be granted by the Sponsor before destruction takes place. Final disposition will be registered in the disposition records.

5.8 Prior and Concomitant Treatment

All prior and concomitant medications will be recorded on the eCRF and will include the name of the drug, dose, route, and start and stop dates as well as the reason or indication for administration

Prior immunosuppressive medications are of especial interest in this study to define type and degree of immunosuppression and will be defined as those taken within last 6 months before Day0.

Immunosuppressive therapies of especial interest will include Methotrexate, leflunomide, hydroxychloroquine, Salazopyrin, Mycophenolate, Tacrolimus, Everolimus, Sirolimus, Azathioprine, Rituximab, Ocrelizumab and corticosteroids, among others.

Regimen and dose of Rituximab / Ocrelizumab received within last 6 months before Day0 will be registered.

Cumulative doses of corticosteroids within last 6 months before Day0 will be recorded.

Regimen and dose for last Immunoglobulin replacement therapy before Day0 will be registered.

Prior administration with anti-CD20 and/or anti-CD3 for prevention or treatment of graft rejection given at least > 1year before Day 0 will be recorded.

Other prior medications, including antiretrovirals for the HIV population will be considered those taken within 30 days before Day0.

Prior prohibited medications include any immunotherapy (monoclonal antibodies, plasma) aimed to prevent or treat COVID-19 within 90 days preceding the planned administration of study vaccine (180 days in case of Evusheld).

Concomitant medications will be defined as those started on or after Day 0.

Concomitant medications will include prescription drugs, over-the-counter drugs, herbal medicines, vitamins, and supplements.

Medications prohibited during the study include:

- Any other experimental product against COVID-19 (drug, biologic, device) within 90 days before Day 0 and throughout the study.
- Treatment or immunotherapy (monoclonal antibodies, plasma) aimed to prevent COVID-19 (PrEP) will be prohibited from Day0 to Day14.

After that period participants will be allowed to receive any prevention treatment following current guidelines, based or not on their post-vaccine titers of antibodies measured by local laboratories. Investigators will guarantee that they are aware of any changes on the prevention guidelines for those vulnerable populations and ensure all participants receive the most updated standard of care. Participants receiving those therapies will be invited to remain in the study but will be censored for the clinical endpoints regarding COVID-19 acquisition and disease outcomes. The immune analysis will be controlled for the treatment received. Further information will be detailed on the SAP.

In case that the administration of additional booster doses is recommended for some of the risk groups included in the trial, participants will be offered to continue in the study. New vaccines will be recorded in the medical history and eCRF as concomitant medication. Safety, Immunogenicity and efficacy analysis will be controlled by those administrations.

6. STUDY ASSESSMENTS

6.1 Schedule of Events

Table 3. Schedule of Events (Spanish participants)

Visit number	1	2	3	4	5	6/ETV	USC visit
Visit description	Pre-Screening (on-site or remote)	Screening and Baseline Vaccine	Immunogenicity and safety assessment	Immunogenicity and safety assessment	Immunogenicity and safety assessment	Immunogenicity and safety assessment	As required
Day (+/-days)	-28 to -1	0	14 (-3/+3)	91 (±15)	182 (±15)	365 (±15)	-
Informed consent ^a		x					
Eligibility criteria	x	x					
Medical history and Prior medication		x					
Demographics ^b		x					
Physical examination		x	x	x	x	x	
Vital signs ^c		x	x	x	x	x	
Concomitant medication ^d		x	x	x	x	x	
Vaccination		x					
Participant Diary provided/ collected ^e		x	x				
Adverse events		x	x	x	x	x	
Pregnancy test ^f		x					
Haematology ^g		x (5 mL)	x (5 mL)	x (5 mL)	x (5 mL)	x (5 mL)	

Visit number	1	2	3	4	5	6/ETV	USC visit
Visit description	Pre-Screening (on-site or remote)	Screening and Baseline Vaccine	Immunogenicity and safety assessment	Immunogenicity and safety assessment	Immunogenicity and safety assessment	Immunogenicity and safety assessment	As required
Day (+/-days)	-28 to -1	0	14 (-3/+3)	91 (±15)	182 (±15)	365 (±15)	-
Biochemistry ^h		x (5 mL)	x (5 mL)	x (5 mL)	x (5 mL)	x (5 mL)	
Lymphocyte subpopulations		x (4 mL)					
Quantitative serum Immunoglobulin tests		x (5 mL)					
Quantitative COVID-19 serology per local laboratories		x (5 mL)	x (5 mL)	x (5 mL)	x (5 mL)	x (5 mL)	
Serum for humoral immunogenicity assays ⁱ : COVID-19 serology (N-specific) Binding antibody assays Neutralisation (PBNA/VNA) IgG subclasses quantification Cross-Reactivity to other COV Avidity		x (26 mL)	x (26 mL)	x (26 mL)	x (26 mL)	x (26 mL)	
PBMCs for cellular immunology assays (Cellular Immunogenicity subset - 50% of participants) ^j ELISPOT ICS (Flow cytometry) Sc-RNA seq		x (80mL)	x (80 mL)	x (80 mL)	x (80 mL)	x (80 mL)	
Total		50 / 130 mL	41/ 121 mL	41/ 121 mL	41/ 121 mL	41/ 121 mL	Total 214 / 614mL

Abbreviations: BUN = blood urea nitrogen; WBC = white blood count; ETV = early termination visit; PBMC = peripheral blood mononuclear cells; PCR = polymerase chain reaction; USC = unscheduled visit.

- a Informed consent will be provided via email to the participant after the Pre-Screening and once the study is approved by the ethics committee and competent authorities. ICF will be signed and dated at Baseline Vaccine visit.
- b Demographics will include gender, race, ethnicity, and age. For the different groups, information detailed in section 6.3.1 of the protocol will be recorded for each of the underlying disease.
- c Vital signs will include pulse, blood pressure, temperature, and oxygen saturation.
- d Current concomitant medication, except for immunosuppressive therapy for the last 6 months. See details on section 5.8 of the protocol for immunosuppressive therapy history.
- e A paper Participant Diary will be provided to all the participants to collect solicited local and systemic reactions after vaccination. The Participant Diary will be collected at the Day 14 visit.
- f Urine pregnancy test to be performed at the study site, only for women of childbearing potential on Day 0. A negative urine pregnancy test should be verified before vaccination.
- g Haematology will include WBC, haemoglobin, and platelets.
- h Biochemistry will include alanine aminotransferase, aspartate aminotransferase, gamma-glutamyl transferase, alkaline phosphatase, total bilirubin, BUN or urea (urea can be measured), creatinine, glucose, potassium, sodium, and total protein. 10 mL of blood will be required for haematology and biochemistry.
- i Three SST tubes (8,5ml each) will be obtained to isolate at least 6ml of serum for all humoral immunogenicity assays. Additional serum will be aliquoted for storage. The COVID-19 serology against N protein will be performed if deemed necessary for the interpretation of the results at any timepoint during the study. Serology will be done at HIPRA central laboratory to assess if the participant has ever had a COVID-19 infection.
- j Eight EDTA tubes (10ml each) will be extracted to obtain PBMC for the study of T cell responses in the immunogenicity subset (50% of participants of each group)

Table 4. Schedule of Events (Turkish participants)

Visit number	1	2	3	4	5/ETV	USC visit
Visit description	Pre-Screening (on-site or remote)	Screening and Baseline Vaccine	Immunogenicity and safety assessment	Immunogenicity and safety assessment	Immunogenicity and safety assessment	As required
Day (+/-days)	-28 to -1	0	14 (-3/+3)	91 (±15)	182 (±15)	-
Informed consent ^a		x				
Eligibility criteria	x	x				
Medical history and Prior medication		x				
Demographics ^b		x				
Physical examination		x	x	x	x	
Vital signs ^c		x	x	x	x	
Concomitant medication ^d		x	x	x	x	
Vaccination		x				
Participant Diary provided/ collected ^e		x	x			
Adverse events		x	x	x	x	
Pregnancy test ^f		x				
Haematology ^g		x (5 mL)	x (5 mL)	x (5 mL)	x (5 mL)	
Biochemistry ^h		x (5 mL)	x (5 mL)	x (5 mL)	x (5 mL)	
Lymphocyte subpopulations		x (4 mL)				

Visit number	1	2	3	4	5/ETV	USC visit
Visit description	Pre-Screening (on-site or remote)	Screening and Baseline Vaccine	Immunogenicity and safety assessment	Immunogenicity and safety assessment	Immunogenicity and safety assessment	As required
Day (+/-days)	-28 to -1	0	14 (-3/+3)	91 (±15)	182 (±15)	-
Quantitative serum Immunoglobulin tests		x (5 mL)				
Serum for humoral immunogenicity assays ⁱ : COVID-19 serology (N-specific) Binding antibody assays Neutralisation (PBNA) IgG subclasses quantification Cross-Reactivity to other COV Avidity		x (26 mL)	x (26 mL)	x (26 mL)	x (26 mL)	
Total		45 mL	36 mL	36 mL	36 mL	Total 153 mL

Abbreviations: BUN = blood urea nitrogen; WBC = white blood count; ETV = early termination visit; PBMC = peripheral blood mononuclear cells; PCR = polymerase chain reaction; USC = unscheduled visit.

a Informed consent will be provided via email to the participant after the Pre-Screening and once the study is approved by the ethics committee and competent authorities. ICF will be signed and dated at Baseline Vaccine visit.

b Demographics will include gender, race, ethnicity, and age. For the different groups, information detailed in section 6.3.1 of the protocol will be recorded for each of the underlying disease.

c Vital signs will include pulse, blood pressure, temperature, and oxygen saturation.

d Current concomitant medication, except for immunosuppressive therapy for the last 6 months. See details on section 5.8 of the protocol for immunosuppressive therapy history.

e A paper Participant Diary will be provided to all the participants to collect solicited local and systemic reactions after vaccination. The Participant Diary will be collected at the Day 14 visit.

f Urine pregnancy test to be performed at the study site, only for women of childbearing potential on Day 0. A negative urine pregnancy test should be verified before vaccination.

g Haematology will include WBC, haemoglobin, and platelets.

^h Biochemistry will include alanine aminotransferase, aspartate aminotransferase, gamma-glutamyl transferase, alkaline phosphatase, total bilirubin, BUN or urea (urea can be measured), creatinine, glucose, potassium, sodium, and total protein. 10 mL of blood will be required for haematology and biochemistry.

ⁱ Three SST tubes (8,5ml each) will be obtained to isolate at least 6ml of serum for all humoral immunogenicity assays. Additional serum will be aliquoted for storage. The COVID-19 serology against N protein will be performed if deemed necessary for the interpretation of the results at any timepoint during the study. Serology will be done at HIPRA central laboratory to assess if the participant has ever had a COVID-19 infection.

6.2 Immunogenicity Measurements

Handling of samples for safety and immunogenicity measurements will be detailed in the Laboratory Manual.

6.2.1 Neutralising Antibody Immunity

6.2.1.1 Pseudovirus - PBNA

Neutralising antibodies will be assessed for the non-HIV participants using HIV reporter pseudoviruses expressing SARS-CoV-2 S protein and Luciferase will be generated. pNL4-3.Luc.R-E- will be obtained from the NIH AIDs repository¹⁵.

SARS-CoV-2 SctΔ19 will be generated (Geneart) from the full protein sequence of SARS-CoV-2 spike with a deletion of the last 19 amino acids in C-terminal¹⁶, human-codon optimised and inserted into pcDNA3.1-TOPO.

Expi293F cells will be transfected using Expifectamine Reagent (Thermo Fisher Scientific, Waltham, MA, USA) with pNL4-3.Luc.R-E- and SARS-CoV-2.SctΔ19 at a 8:1 ratio, respectively. Control pseudoviruses will be obtained by replacing the S protein expression plasmid by a VSV-G protein expression plasmid.

Supernatants will be harvested 48 hours after transfection, filtered at 0.45 µm, frozen and titrated on HEK293T cells overexpressing WT human ACE-2 (Integral Molecular, USA). For neutralisation assay, 200 TCID₅₀ of pseudovirus supernatant will be preincubated with serial dilutions of the heat-inactivated serum samples for 1 hour at 37°C and then added onto ACE2 overexpressing HEK293T cells. After 48 hours, cells will be lysed with Britelite Plus Luciferase reagent (Perkin Elmer, Waltham, MA, USA). Luminescence will be measured for 0.2 s with an EnSight Multimode Plate Reader (Perkin Elmer).

Neutralisation capacity of the serum samples will be calculated by comparing the experimental RLU calculated from infected cells treated with each serum to the max RLUs (maximal infectivity calculated from untreated infected cells) and min RLUs (minimal infectivity calculated from uninfected cells), and expressed as percent neutralisation: %Neutralisation = (RLU_{max} - RLU_{experimental}) / (RLU_{max} - RLU_{min}) * 100. All IC₅₀ values will be expressed as reciprocal concentration.

PBNA assays will be conducted at IrsiCaixa AIDS Research Institute in Badalona and in LABORATORIOS HIPRA S.L.U if deemed necessary. Method is duly validated.

6.2.1.2 VNA

Vero E6 cells (ATCC CRL-1586) are cultured in Dulbecco's modified Eagle medium (Invitrogen) supplemented with 10% fetal bovine serum (FBS; Invitrogen), 100 U/ml penicillin, 100 µg/ml streptomycin, (all from Invitrogen). SARS-CoV-2 variant Omicron BA2 was isolated from a nasopharyngeal swab collected at Hospital Germans Trias i Pujol (Badalona) on PPD from a 65-year-old PPD patient. The genomic sequence was deposited at the GISAID repository with ID number PPD. The calculation of the TCID₅₀ per mL of this viral stock was 10^{5.801}. SARS-CoV-2 D614G used was isolated from a nasopharyngeal swab collected at Hospital

Universitari Germans Trias i Pujol de Badalona on PPD from a 85-year-old PPD patient. The genomic sequence was deposited at the GISAID repository with ID number PPD. The calculation of the TCID₅₀ per mL of this viral stock was 10^{6.468}. All viral stocks were titrated in 10-fold serial dilutions on Vero E6 cells to calculate the TCID₅₀ per mL.

Neutralization assays in HIV infected individuals will be performed using a replication competent virus assay and measuring the cytopathic effect in Vero E6 cells in duplicate. Briefly, virus are used at a MOI to achieve a 50% of viral induced cytopathic effect, and preincubated with three-fold serial dilutions (1/60–1/14,580) of heat-inactivated plasma samples from the indicated individuals for 1 h at 37 °C. Pre-incubated viruses are added to 60.000 Vero E6 cells per well in 96 well plates. 72 h later, viral-induced cytopathic effect is measured using the Cell Titer Glow Luciferase reagent (Promega) and the Luminoskan Plate Reader from Thermofisher. The relative light units (RLU) are normalized, and the ID50 (the reciprocal dilution inhibiting 50% of the infection) was calculated by plotting and fitting the log of plasma dilution vs. response to a 4-parameter equation in Prism 8.4.3.

Neutralisation antibody immunity assays by VNA will be conducted at IrsiCaixa AIDS Research Institute in Badalona.

6.2.2 Antibody Binding Immunity Assays

For the 400 participants immunoassay for the in vitro quantitative determination of antibodies to the SARS-CoV-2 S protein RBD in human serum will be used.

The assay uses a recombinant protein representing the RBD of the S antigen in a double-antigen sandwich assay format, which favours detection of high affinity antibodies against SARS-CoV-2. The test is intended as an aid to assess humoral immune response to the SARS-CoV-2 S protein.

Antibody binding immunity assays will be conducted at LABORATORIOS HIPRA, S.A.

Electrochemiluminescence immunoassay (ECLIA) will be conducted with a commercial kit. Specimen type: serum collected using standard sampling tubes; Li-heparin, K2-EDTA-, K3-EDTA-, and sodium citrate plasma.

6.2.2.1 Antibody Binding Immunity Assay to detect possible asymptomatic infections

Immunoassay for the in vitro quantitative determination of antibodies to the SARS-CoV-2 N protein in human serum will be used to detect possible asymptomatic SARS-CoV-2 infection.

The assay uses a recombinant protein representing the N protein in an indirect ELISA format, which favours detection of high affinity antibodies against SARS-CoV-2. The test is intended as an aid to assess humoral immune response to the SARS-CoV-2 N protein to discern those responses induced by the vaccine to those induced by a possible asymptomatic infection.

Antibody binding immunity assays will be conducted at LABORATORIOS HIPRA, S.A.

ELISA will be conducted with a commercial kit. Specimen type: serum collected using standard sampling tubes; Li-heparin, K2-EDTA-, K3-EDTA-, and sodium citrate plasma.

This assay will be performed in all the immunogenicity subset at baseline and at subsequent timepoints (3 months, 6 months and 12 months (V4, V5 and V6) of the study in those participants with negative results in the prior assessment, to determine whether these participants have past history of SARS-CoV-2 infection and/or they acquired during the study. If the results of the ELISA are positive at any of these timepoints, only the results of the mentioned and future timepoints will not be used in the final data interpretation. For example, if a COVID-19 infection is detected at V5, the results from the V4 will be used for the analysis, but not the ones from V6.

The same analysis will be performed at any timepoint of the study, with samples taken previously and available for the analysis, if deemed necessary for the interpretation of the results

6.2.3 High resolution humoral immunity

The levels of specific IgG-subclasses (IgG1, IgG2, IgG3 and IgG4) will be determined in a subset immunosuppressed individuals (n=200). IgG subclasses will be quantified by ELISA. The following secondary antibodies will be used: anti-huIgG1 (clone HP6069, Thermo Scientific), anti-huIgG2 (clone G18-21, BD Biosciences), anti-humanIgG3 (clone HP6050, SouthernBiotech); and anti-human IgG4 (clone JDC-14, BD Biosciences)]. Cross reactivity with Spike glycoproteins from other coronavirus (SARS-CoV, MERS-CoV, HKU1, OC43, NL63, 229E) will be also evaluated in this subset of samples. The avidity of the main antibodies elicited against the Spike glycoproteins will be also determined by ELISA. The avidity index will be calculated by comparing the signal obtained in the presence or absence of an antigen-antibody dissociating reagents.

IgG subclasses quantification, cross-reactivity and avidity of IgG will be conducted at IrsiCaixa AIDS Research Institute in Badalona.

6.2.4 T-cell Mediated Immunity

T-cell mediated immunity will be assessed by measurement of whole PBMC stimulation by ELISpot at different timepoints in a subset of 200 individuals recruited in the Spanish sites representative of the 5 underlying conditions (aprox 40 participants/condition).

To evaluate the SARS-CoV-2-specific T-cell responses, 5 peptide pools overlapping RBD D614G, RBD beta, RBD delta, RBD Omicron BA.1 and RBD Omicron BA.2 variants, were used.

T-cell responses will be analysed as present or absent and reported as the number and proportion of participants responding to each peptide pool and for each time point. The total ELISpot responses will be described as the sum of SFC/10⁶ PBMC of all positive responses per peptide pool, after subtraction of background. Each participant will be classified as a responder if there is at least 1 positive against any of the SARS-CoV-2 peptides pools at any time, and non-responder if ELISpot responses are all negative.

In addition, intracellular cytokine staining (ICS) based T-cell assay will be determined at different timepoints. ICS assays will include Th1/Th2 pathways (e.g., IL-2, IL-4, TNF α , INF γ) CD4 and CD8 T cell determinations using flow cytometry.

An ICS will be considered positive if the percentages of cytokine-positive cells in the stimulated samples are 3 times more than the values obtained in the unstimulated controls and if the background-subtracted magnitudes are higher than 0.02%. Each participant will be classified as a responder if there are at least 1 positive IFN- γ ICS response against any of the SARS-CoV-2 peptide pools at determined timepoints and as a non-responder if responses at these timepoints are all negative.

T-cell mediated response analysis will be conducted at the IDIBAPS of Hospital Clínic of Barcelona, Spain, and at IrsiCaixa AIDS Research Institute, at Hospital Germans Trias i Pujol, Badalona, Barcelona, Spain.

6.2.5 T-cell transcriptional profiles, TCR usage and variant cross-reactivity

To define the impact that the phenotype and functionality of RBD-specific T cells has on the outcome of SARS-CoV-2 vaccination, RBD-specific T cells will be isolated longitudinally and subjected to single cell transcriptomics (sc-RNAseq) analyses. We will isolate RBD-peptide specific T cells from 50 individuals at baseline, D14, D91 and D365 to relate T cell gene expression profiles to the induction, maturation and long-term maintenance of humoral immunity to RBD. This task will consider the humoral activity seen at baseline and at d14, so that transcriptome analyses can be initiated rapidly and then conducted at the same time points as the humoral immune assessments. For single cell transcriptomics, RBD-specific cells will be isolated by employing an activation-induced marker assay that detects activation markers (including CD25, CD69, CD137) on antigen specific cells which then allows for single cell sorting. Using the sc-RNAseq data generated to assess gene expression profiles in vaccine-induced T cells, the TCR repertoire of the analyzed T cells will be reconstructed and the repertoire of single peptide-specific responses defined.

The induction of cross-reactive T cells that can recognize viral variants effectively will thus be critical for combating the current and future SARS-CoV-2 infections. Synthetic overlapping peptides (15mers with 11 amino acids overlap) covering all the described RBD sequence variability in CoV-2, related human coronaviruses and bat coronavirus isolates will be generated and tested individually in IFN γ ELISpot assays. The number of targeted variants will then be related to TCR repertoire breadth, magnitude of total response, and to baseline and post-vaccine functional, phenotypic and transcriptomics parameters in the 50 individuals for which single-cell transcriptomics and, thereby TCR repertoire information.

This deep T-cell mediated response analysis will be conducted at IrsiCaixa AIDS Research Institute, at Hospital Germans Trias i Pujol, Badalona, Barcelona, Spain.

6.2.6 Biological samples for future studies

All participants will be asked to provide consent for the use of the remaining samples obtained during the safety or immunologic assessment for future studies. If the individual consents, leftover samples will be kept and sent to a sample collection located in HIPRA SCIENTIFIC, S.L.U. (Amer, Girona) for future studies related to COVID-19.

If the individual does not consent the storage of the remaining samples for their use in future studies, samples will be destroyed once they are analysed for the purpose of this study in compliance with the law 14/2007 Biomedical Research.

Samples obtained from screening failures will be also kept and sent to the sample collection if individuals have consented their use for future studies.

Participants will be informed that biological samples and their associated data stored at the central lab in HIPRA SCIENTIFIC, S.L.U. will be used for biomedical research in the field of COVID-19 infection. These investigations must be previously approved by an Ethics Committee.

Privacy and confidentiality of data generated in the future on stored samples will be protected by the same standards applicable to all other clinical data.

6.3 Safety Measurements

6.3.1 Demographics and Medical History

Demographics will include age, gender, race and ethnicity. Prior COVID-19 vaccination schemes (and brand) will be recorded as prior medications (brand of the dose/s + last dose date). Previous confirmed COVID-19 infections will also be recorded indicating the date of the RAT or PCR positive test, severity and if hospitalization was required and date of discharge.

To address the degree of immunosuppression, additional information on their underlying conditions will include:

Condition	Variables
HIV infection	Date of HIV diagnosis Nadir CD4 cell count Last pVL, CD4, CD4/CD8 ratio (within 6 months prior study entry) Current Antiretroviral Treatment Time (months) virologically suppressed before study entry
Kidney transplant	Underlying kidney disease: inherited, glomerular, interstitial, urological, others or unknown. Nr of transplants received, living or deceased donors and dates Last treatment with anti-CD20 or anti-CD3 and date (at least >1 year)

	History of opportunistic infections (diagnose and dates) within the last year History of biopsy-proven graft rejection (dates) within the last year
Hemodialysis/ peritoneal dialysis	Underlying kidney disease: inherited, glomerular, interstitial, urological, others or unknown. History of opportunistic infections (diagnose and dates) within the last year
Primary antibody deficiencies	Classification of disease according to 2019 ESID criteria Date of diagnosis Type of IgG deficiency Latest IgG level before inclusion (within last 3 months) Time on IgG replacement therapy
Autoimmune Diseases	Group of AID: vasculitis, myositis, SLE/Sjögren, rheumatoid arthritis and multiple sclerosis. Date of diagnosis. Latest Rituximab / Ocrelizumab dose before inclusion (within last 6 months)

6.3.2 Physical Examination

A complete physical examination will be done at each on-site study visit.

6.3.3 Vital Signs

Vital signs will be recorded at each study visit before blood samples are collected and will include pulse, blood pressure, body temperature, and oxygen saturation. Abnormal values of vital signs will be recorded as well in eCRF but indicated non clinically significant as per investigator criteria.

Blood pressure and pulse measurements will be assessed while the participant is in a supine position and at rest for 5 minutes. The same method of measurement will be used for participants at all visits.

6.3.4 Clinical Laboratory Tests

Details of sample collection, storage, and handling will be provided in the Laboratory Manual. Blood samples to determine haematology and biochemistry tests will be analysed at the local laboratory of each study site. After analysis the samples will be destroyed locally.

The following parameters will be evaluated:

- Haematology: white blood cell count, haemoglobin, platelet count.
- Biochemistry: alanine aminotransferase, aspartate aminotransferase, gamma-glutamyl transferase, alkaline phosphatase, total bilirubin, blood urea nitrogen (BUN) or urea (as per site common procedures), creatinine, glucose, potassium, sodium, and total protein.
- Urine pregnancy test (only for women with childbearing potential; to be performed locally at the study site on Day 0).
- COVID-19 serology test: a sample will be taken and sent to HIPRA's Central Laboratories, where a serology will be performed to determine whether these participants have had a previous COVID-19 infection.
- Quantitative serum Immunoglobulin tests (quantification of IgG, IgM and IgA).
- Lymphocyte subpopulations.

6.3.5 Participant Diary

All participants will be dispensed with a Participant Diary to register all the local and systemic reactions from the time of vaccination until 7 days post-vaccination. On Day 0 participants will be trained on how to fill the diary and the grading system used to report the intensity of events. The diary will be collected at the Day 14 visit and reviewed by the investigator. The participant's Diary will collect the following reaction information at the injection site:

- Pain
- Tenderness
- Erythema/redness
- Induration/swelling

And the following systemic solicited events:

- Fever
- Chills
- Nausea
- Malaise
- Vomiting
- Diarrhoea
- Headache
- Fatigue
- Muscle pain
- Joint pain
- Others

The diary card also contains a section to record any additional concomitant medication used during the 7 days following vaccination

6.3.6 Remote or on-site pre-screening visits

Participants might be contacted by the site staff via telephone or at the clinical site for a pre-screening visit to assess some of the eligibility criteria and willingness to participate in the study.

6.3.7 COVID-19 Infection

6.3.7.1 COVID-19 Infection

All participants should be tested for COVID-19 following the current standard procedures in the health system if present any symptom suggestive of COVID-19. If positive (asymptomatic or symptomatic), participants should contact the study team to inform them about the test. It will be included in the CRF throughout the study as a general AE or AESI if it meets AESI criteria (severe COVID-19 infection). as a general AE throughout the study (non-severe COVID-19, should still be reported throughout the study).

The investigator will follow up the medical evolution by phone and recommend medical assistance in the hospital if required. All this information will be included in the CRF. Non-severe COVID-19 cases will be considered AEs, but only confirmed COVID-19 cases happening after ≥ 14 days post-administration will be taken into account for the exploratory endpoint's analysis mentioned in section 3.2.3.

Severe COVID-19 Infection

Severe cases of COVID-19 after vaccination will be assessed. Severe cases are considered as any episode of COVID-19 requiring ≥ 24 hrs of hospitalization.

All this information will be included in the eCRF and severe COVID-19 infections will be considered as AESIs, as described in the list of AESIs (Appendix 1). Severe COVID-19 cases which meet seriousness criteria will be reported as SAE as described in section 6.3.8.4.1 below.

Only confirmed COVID-19 cases happening after ≥ 14 days post-administration of the vaccine, regardless of severity, will be taken into account for the exploratory endpoints' analysis mentioned in [section 3.2.3](#).

6.3.8 Adverse Events

Adverse events will be coded by preferred term (PT) and system organ class (SOC) using NCI Common Terminology Criteria for Adverse Events. CTCAE terms are grouped by MedDRA Primary SOC. Within each SOC, AEs are listed and accompanied by descriptions of severity (Grade) (version 5.0 of 27Nov2017) (US Department of Health and Human Services).

6.3.8.1 Definitions, Documentation, and Reporting

Adverse event (AE): an AE is any untoward medical occurrence in a clinical study participant administered a medicinal (investigational or non-investigational) product. An AE does not

necessarily have a causal relationship with the treatment. An AE can therefore be any unfavourable and unintended sign (including an abnormal finding), symptom, or disease temporally associated with the use of a medicinal (investigational or non-investigational) product, whether or not related to that medicinal (investigational or non-investigational) product (Definition per International Conference on Harmonisation [ICH]).

This includes any occurrence that is new in onset or aggravated in severity or frequency from the baseline condition, or abnormal results of diagnostic procedures, including laboratory test abnormalities.

Serious adverse event (SAE): an SAE is any untoward medical occurrence that at any dose: Results in death, is life-threatening, requires inpatient hospitalisation or prolongation of existing hospitalisation, results in persistent or significant disability/incapacity, is a congenital anomaly/birth defect, is a suspected transmission of any infectious agent via a medicinal product, is medically important (according to the treating physician), other important medical events that may not be immediately life-threatening or result in death or hospitalisation but may jeopardise the participant or may require intervention to prevent one of the other outcomes listed in the definition above.

If a serious and unexpected AE occurs for which there is evidence suggesting a causal relationship between the study drug and the event (e.g., death from anaphylaxis), the event must be reported as a SUSAR even if it is a component of the study endpoint (e.g., all-cause mortality).

Any event requiring hospitalisation (or prolongation of hospitalisation) that occurs during the course of a participant's participation in a study must be reported as an SAE, except hospitalisations for the following: hospitalisations not intended to treat an acute illness or AE (e.g., social reasons such as pending placement in long-term care facility), surgery or procedure planned before entry into the study.

Adverse reaction (AR): An AR is an AE suspected to be causally related to a medicinal product.

An AR is a response to a medicinal product which is noxious and unintended. Response in this context means that a causal relationship between a medicinal product and an AE is at least a reasonable possibility (i.e., the relationship cannot be ruled out). Adverse reactions may arise from use of the product within or outside the terms of the marketing authorisation or from occupational exposure. Conditions of use outside the marketing authorisation include overdose, misuse, abuse, and medication errors.

Unexpected Adverse Reaction (UAR): Any adverse reaction, whose nature or severity of which is not consistent with the applicable product information (e.g., IB for an unauthorised investigational product or Summary of Product Characteristics for an authorised product).

Suspected Unexpected Serious Adverse Reaction (SUSAR): An adverse reaction that is both serious and unexpected.

Adverse events of special interest (AESI): An AE for which additional data is required. AESIs in this study will include potential immune-mediated medical conditions happening after vaccination and through the end of the study (see Appendix 1). An AESI is a pre-specified medically-significant event that has the potential to be causally associated with a vaccine product that needs to be carefully monitored and confirmed by further special studies. The purpose is to

identify per-specified specific (rare) events by a set criterion to determine if a COVID-19 vaccine could have caused it.

Medically attended adverse events (MAAEs): A new onset or a worsening of a condition that prompts the participant or participant's parent/legally acceptable representative to seek unplanned medical advice at a physician's office or Emergency Department. Physician contact made over the phone or by email will be considered a physician office visit for the purpose of MAAE collection. This includes medical advice seeking during the study visit or routine medical care. This definition excludes paediatric check-ups, follow-up visits of chronic conditions with an onset prior to entry in the study, and solicited reactions.

6.3.8.2 Assessment of Intensity and Causality

An assessment of intensity will be made using the general categorical descriptors outlined in the toxicity grading scale for healthy adult and adolescent participants enrolled in preventive vaccine clinical studies¹⁷.

For AEs not identified in the grading table, the following guidelines will be followed:

- Grade 1 – Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
- Grade 2 – Moderate; minimal, local or non-invasive intervention indicated; limiting age-appropriate instrumental ADL (Activities of Daily Living) *.
- Grade 3 – Severe or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL**.
- Grade 4 – Life-threatening consequences; urgent intervention indicated.

*Instrumental ADL refer to preparing meals, shopping for groceries or clothes, using the telephone, managing money, etc.

**Self-care ADL refer to bathing, dressing and undressing, feeding self, using the toilet, taking medications, and not bedridden. An assessment of causality will be made based on the following criteria:

- Related: The temporal relationship of the AE with the study drug indicates a possible causal relationship and it cannot be explained by factors such as the patient's clinical condition or therapeutic interventions.
- Unrelated: The temporal relationship of the AE with the study drug indicates an unlikely causal relationship, or other factors (concomitant medication or conditions) or other therapeutic interventions provide a satisfactory explanation for the AE.

6.3.8.3 Procedures for AE and SAE Reporting

Adverse events will be assessed at each visit based on careful clinical observation of the participant, laboratory tests or spontaneous reports by the participant discovered as a result of general questioning by the study staff. All AEs will be recorded in the eCRF.

After the date of approval of this amendment pv5.0, non-severe COVID-19 cases will be collected and recorded in the eCRF within 72 hours from learning the event and notification to Asphalion S.L. will not be needed. Non-severe COVID-19 cases will be recorded in eCRF throughout the study as general AEs.

Open-ended and non-leading verbal questioning of the trial participant is the preferred method to inquire about AE occurrences.

The following will be recorded for each event: indication, intensity (grade 1, 2, 3 and 4), duration (start and stop dates), severity, causal relationship with the intervention vaccine (according to the previously attributability criteria), actions taken, and outcome. The Investigator should report any underlying condition when a surgical or medical procedure is required as the event term, and the procedure as an action taken. For a pre-existing condition that has worsened in terms of severity or frequency, the meaning of the change should be specified (e.g., worsening of hypertension).

If the AE is an overdose, the nature of the overdose must be stated (for example, medication error, accidental overdose, or intentional overdose) and the Investigator shall notify Sponsor or whoever assumes the tasks delegated by the Sponsor, within 24 hours from the time of knowing about the event. The reporting circuit and form will be the same as for the SAE.

For all AEs, the Investigator must pursue and obtain information adequate both to determine the outcome of the AE and to assess whether it meets criteria for classification as an SAE requiring expedited notification (see Section 6.3.8.4.1). Adverse events should be followed until the event resolves or stabilises at a level acceptable to the Investigator.

Serious AEs that have not resolved by the end of the study, or that have not resolved upon discontinuation of the individual's participation in the study, must be followed until any of the following occurs: the event resolves, the event stabilises, the event returns to baseline, if a baseline value/status is available, the event can be attributed to agents other than the study drug or to factors unrelated to study conduct, it becomes unlikely that any additional information can be obtained (participant or health care practitioner refusal to provide additional information, lost to follow-up after demonstration of due diligence with follow-up efforts).

6.3.8.4 Expedited Reporting of SAEs, AESIs, MAAEs, and SUSARS

6.3.8.4.1 Serious Adverse Events

Any SAE due to any cause, whether or not related to the study medication, occurring from the time of signing the informed consent and up to the last visit of the study, must be reported within 24 hours of occurrence or during the next 24 hours after the Investigator becomes aware of the event. It will only be included in the safety database SAEs occurring in patients that are part of the safety analysis group.

The initial report of SAE should be as complete as possible, including details of the current disease and SAE and assessment of the causal relationship between the SAE and the investigational product. Reporting will be made using the SAE/AESI/MAASE Form within 24 hours from first knowledge by the Investigator, completing all information on the form in the following two days.

Serious Adverse Event Report Forms will be sent by email to Asphalion S.L., the contracted CRO for monitoring activities at Spain (FLS-CRO) or Turkey (Monitor CRO), and the Sponsor.

The information missing at the time of the initial report must be reported in the SAE/AESI/MAASE follow-up form. All documentation relating to the event should be included (e.g., additional laboratory tests, hospital records, death certificates). Additional information may be requested by the study coordinator, the Ethics Committee, or regulatory authorities.

In the case of a medication error or if the investigational medicinal product is used outside the provisions of the protocol, while conducting the study, the Investigator shall notify Asphalion S.L. within 24 hours from the time of knowing about the event using the Serious Adverse Event Report Form.

6.3.8.4.2 Adverse Events of Special Interest and Medically Attended Adverse Events

If an AESI occurs, the Investigator will record it in the eCRF and notify Asphalion S.L. within 24 hours from the time of knowing about the event.

All MAAEs will be collected, recorded in the eCRF and notified by the Investigator to Asphalion S.L. within 24 hours from learning the event, from the date of the participant consent until date of approval of protocol amendment 1 v5.0.

After the day of the approval of amendment 1 protocol v5.0, those MAAEs that are considered related, probably related or possibly related to the vaccine by Investigator will be collected in the eCRF and notified to Asphalion S.L. within 24 hours with the correspondent form. Those MAAEs not related to the vaccine by investigator will be collected in the eCRF within 72 hours, and notification to Asphalion S.L. will not be needed.

6.3.8.4.3 Suspected Unexpected Serious Adverse Reaction

Asphalion S.L. will report any event that is serious and unexpected that may be related to the investigational product to the competent authority in Spain (AEMPS), through Eudravigilance and to the Turkish authorities in accordance with local regulations.

Reporting will be made using a specific SUSAR form.

The maximum deadline for reporting will be 15 calendar days from the time the Investigator is aware of the SUSAR. For SUSARs causing death or that are life-threatening for the participant, the maximum reporting time will be 7 calendar days from the time the Investigator is aware of them. This information will be completed, when possible, in the following 8 days.

6.3.8.5 Pregnancy

Pregnancy will not be recorded as an AE. However, the Investigator must notify Asphalion S.L. of any pregnancy that occurs during the study in a female participant or partners of male participants within 24 hours of knowing about the event.

To ensure participant safety, female individuals will be asked for consent to be followed up to determine pregnancy and delivery outcome. If they consent, a follow-up of the pregnancy to document its outcome and the state of health of the new-born will be completed. If the pregnancy outcome meets the SAE criteria, or if the new-born presents an SAE, the procedures for reporting an SAE will be followed.

The report shall be made using the specific form for the notification of pregnancy, which must be sent by fax or email.

7. STATISTICAL PROCEDURES

7.1 Sample Size Estimation

No formal sample size calculation has been performed for this phase IIb/III study.

A sample size of 400 participants is proposed, with 60-80 participants per condition. This is an exploratory study to assess safety and immunogenicity in special populations with some underlying immunosuppressive conditions. No formal sample size calculation is envisioned. As a means to characterize the statistical properties of this study, the table below presents the probability of observing zero, at least one, and two or more safety endpoints among the 400 participants for various “true” event rates:

Event rate	P(0 events n=400)	P(≥ 1 event n=400)	P(≥ 2 events n=400)
1%	1.8%	98.2%	91%
5%	0%	100%	100%
10%	0%	100%	100%
15%	0%	100%	100%
25%	0%	100%	100%

P=Probability

Overall, we consider that the sample size proposed is considered adequate for this specific kind of vaccine.

7.2 Populations for Analysis

The following analyses populations are included in this study:

- **Enrolled (EP):** All individuals who have signed the Informed Consent Form (ICF).
- **Intent-to-treat (ITT):** All participants who are randomised, regardless of the individual’s treatment status in the study.
- **Modified Intent-to-treat (mITT):** All individuals in the ITT who meet the inclusion/exclusion criteria, received a dose of study vaccine and did not tested positive for COVID-19 within 14 days of the receiving study vaccine
- **Immunogenicity (IGP):** All individuals in the mITT who had a valid immunogenicity test result before receiving study vaccine and at least one valid result after dosing. Participants will be analysed according to their underlying disease, history of prior COVID-19 and/or positive N-serology and primary vaccination schemes (mRNA or Coronavac/mRNA)
- **Safety (SP):** All randomised individuals who received the study vaccine. This population will be used for all analyses of safety. Participants will be analysed according to their underlying disease and primary vaccination schemes (mRNA or Coronavac/mRNA)

7.3 Procedures for Handling Missing, Unused, and Spurious Data

In general, missing data will not be imputed. For the continuous variables related to the immunogenicity endpoints and T-cell, zero values will be imputed to half of the lower limit of quantification (LLOQ). If other parameters are deemed appropriate for imputation, information will be detailed in the Statistical Analysis Plan (SAP).

7.4 Interim Analysis

First immunogenicity analysis will be released once data of high throughput humoral immunogenicity at Day 91 of all enrolled participants in Spain. This report will also include data from safety assessments.

Final analysis will be released with longitudinal follow-up up to 1 year since vaccination.

Other interim analysis will be performed anytime in the study when sponsor deems it necessary.

7.5 Statistical Methods

7.5.1 General Methods

No formal statistical analysis will be performed on the primary and secondary endpoints.

Descriptive analysis will be performed for variables overall by time-point. Categorical variables will be presented by means of number of cases and frequencies (%) and continuous variables will be presented by number of non-missing observations, mean, standard deviation (SD), median, min and max; for the immunogenicity variables the geometric mean titre, geometric mean concentration, GMFR, and standard deviations will be presented, as appropriate. Dichotomised measures for immunogenicity will be presented as frequencies and percentages. 95% confidence intervals (CI) will be also provided, as appropriate.

Geometric mean titre (GMT), geometric mean concentration (GMC), GMFR and standard deviations will be calculated based on the log-transformed titres. Calculation of 95% CI will be based on the t-distribution of the log-transformed titres or the difference in the log-transformed titres for GMT and GMFR, respectively, then back transformed to the original scale.

Further details on methodology for summary and statistical analyses of the data collected in this study will be detailed in the study SAP.

7.5.2 Disposition of Individuals

Participant disposition will be tabulated and descriptively summarised for all enrolled individuals. Analysis populations will be summarised. Entry criteria and protocol deviations will be listed.

7.5.3 Baseline Comparisons

Demographic and baseline data will be tabulated using the safety population. No statistical comparisons will be performed for any of the baseline characteristics.

7.5.4 Safety Analysis

Solicited local reactions and systemic events from Day 0 through Day 7 after dosing will be presented by intensity and cumulatively across severity levels.

Unsolicited local and systemic reactogenicity adverse events from Day 0 through Day 28 after boost vaccination will be presented by intensity and cumulatively across severity levels.

Adverse Events and SAEs will be categorised according to NCI Common Terminology Criteria for Adverse Events. CTCAE terms are grouped by MedDRA Primary SOC. Within each SOC, AEs are listed and accompanied by descriptions of severity (Grade).

Summaries of local and systemic solicited AEs will be presented by SOC and PT for events occurring through Day 7. In addition, AEs will be summarised by maximum intensity and causal relationship to study drug. A separate summary of AESIs, including potentially immune-mediated medical conditions (PIMMCs) and MAAEs will be reported.

Laboratory parameters changing to grades 3 or 4, will be summarised as actual values and change from baseline over time. Shift tables from Baseline to worst on-study value and Baseline to Days 14, 91, 182 and 365 (if applicable) value will be reported.

The safety analyses will be performed using the safety population (SP).

7.5.5 Immunogenicity analysis

The following data will be presented:

- The neutralizing antibody titre and the total binding antibodies measured for each individual sample and the GMT with its 95% confidence intervals at baseline and at Days 14, 91, 182 and 365
- The geometric mean fold rise (GMFR) in neutralizing antibody titre and in total binding antibodies from baseline to Day 14.
- The percentage of individuals that after the additional dose have a ≥ 2 -fold and 4-fold change in total binding antibodies titre from Baseline to Day 14

7.5.6 Procedures for Reporting Deviations to Original Statistical Analysis Plan

A SAP will be written to detail further methodology for summary and statistical analyses. Any deviations on the planned analysis from this SAP would be documented on a SAP amendment and in the Clinical Study Report (CSR).

8. ADMINISTRATIVE REQUIREMENTS

8.1 Good Clinical Practice

This study will be conducted in accordance with the protocol and ethical principles stated in the Declaration of Helsinki or the applicable guidelines on Good Clinical Practice (GCP), and all applicable local laws, rules, and regulations.

Requirements for ethical review as set forth in Directive 2001/20/EC of the European Parliament and of the Council of 4 April 2001 on the approximation of the laws, regulations and administrative provisions of the Member States relating to the implementation of GCP in the conduct of clinical studies on medicinal products for human use or other relevant local regulations for institutional review will be followed.

The Sponsor and his delegates, in collaboration with the Investigator, will be responsible for reporting to the Ethics Committee all changes in research activity, including protocol amendments, updates of IBs, annual safety reports, all unanticipated problems involving risks to human participants, and study termination.

8.2 Ethics

In parallel to the submission to the Independent Ethics Committee (IEC), the Sponsor must obtain an authorisation from the appropriate competent authority to conduct the clinical study. Participants must not be entered into the study until the relevant IEC has issued its opinion and the CA has given authorisation to conduct the study.

All substantial amendments must be submitted to the IEC and/or to the Competent Authorities for approval.

Neither the Investigator nor the Sponsor will modify this protocol without a formal amendment by the Sponsor. All protocol amendments must be issued by the Sponsor and signed and dated by the Investigator. Protocol amendments must not be implemented without prior IEC approval, or when the relevant competent authority has raised any grounds for non-acceptance, except when necessary to eliminate immediate hazards to the participants, in which case the amendment must be promptly submitted to the IEC and relevant competent authority.

Documentation of amendment approval by the Investigator and IEC must be provided to the Sponsor. During the study, in situations where a departure from the protocol is unavoidable, the Investigator or other physician in attendance will contact the appropriate Sponsor representative. Except in emergency situations, this contact should be made before implementing any departure from the protocol. In all cases, contact with the Sponsor must be made as soon as possible to discuss the situation and agree on an appropriate course of action. The data recorded in the eCRF, and source documents will reflect any departure from the protocol, and the source documents will describe this departure and the circumstances requiring it.

8.2.1 Independent Ethics Committee

The Protocol, informed consent forms, IB, and other required documents must be approved by the IEC before enrolment of participants in the study. The letter of approval from the IEC, as well as a list of documents reviewed, will be filed in the Investigator Site File (ISF) and a copy will be filed in the study master file (TMF) held by the Sponsor.

8.3 Data and Safety Monitoring Board

The DSMB is a group of experts that monitors the main safety and tolerability outcome measures and the overall conduct of the study with the aim of protecting the safety and interests of the study participants. The membership, frequency, and method of the DSMB and the study aspects to be reviewed would be specified in the DSMB Charter.

After any DSMB meetings, DSMB would make a recommendation to continue the study, to stop vaccination administration new participants in the same part of the study or terminate the study.

The DSMB Chairman will be responsible for providing a written report of findings and recommendations to the Sponsor in a timely manner. Contracted CRO would be responsible for informing the study sites and the Sponsor will be responsible for informing appropriate regulatory authorities of any DSMB recommendation relating to conduct of the study.

8.4 Participant Information and Informed Consent

Each individual must give written consent according to local requirements after the nature of the study has been fully explained. The ICFs must be signed before performance of any study related activity. The ICF(s) that is/are used must be approved by both the Sponsor and by the reviewing IEC and be in a language that the participant can read and understand. The ICF(s) should be in accordance with principles that originated in the Declaration of Helsinki (updated according to its last version, Fortaleza, Brazil, 2013), current ICH and GCP guidelines, applicable regulatory and Sponsor policy. A community advisory panel has been created to ensure that the information delivered to participants involved in the clinical trial is accessible, complete, transparent and comprehensible for what concerns the objective of the project and their personal participation.

Before enrolment in the study, the Investigator or an authorised member of the study site must explain to potential participants the aims, methods, reasonably anticipated benefits, and potential hazards of the study, and any discomfort that the participation in the study may entail.

Individuals will be informed that their participation is voluntary and that they may withdraw consent to participate at any time. Finally, they will be told that the Investigator will maintain a participant identification register and that their records may be accessed by health authorities and authorised Sponsor personnel without violating the confidentiality of the participant, to the extent permitted by the applicable law(s) or regulations.

The individual will be given enough time to read the ICF(s) and the opportunity to ask questions. After this explanation and before entry into the study, consent should be appropriately recorded by means of the participant's personally dated signature. After having obtained the consent, a copy of the ICF(s) must be given to the participant.

8.5 Participant Confidentiality

The collection and processing of personal data from participants enrolled in this study will be limited to those data that are necessary to fulfil the objectives of the study. Individual's data will be codified to ensure confidentiality and compliance with applicable data privacy protection laws and regulations.

Participants will be codified with a Participant ID that prevents their identity from being deduced.

The Investigator and duly authorised collaborators will maintain personal data strictly confidential, according to regulatory requirements. The link between the numeric code and real personal data from participants will be rigorously kept by the Investigator. The ICF(s) obtained from the participant includes explicit consent for the processing of personal data and for the Investigator/institution to allow direct access to his or her original medical records (source data/documents) for study-related monitoring, audit, IEC review, and regulatory inspection.

In the eCRF, the participant will only be identified by the participant Participant ID. The name of participants will not appear in any publication or report of the study results.

Duly authorised persons by the Sponsor and the health authorities and the IEC may audit or inspect the study. Personal information will not be publicly available, in compliance with General Data Protection Regulation (GDPR) (EU) 2016/679 of 27th April 2016.

In addition, the Sponsor cannot make decisions as to the significance of any findings resulting from exploratory research. Therefore, exploratory research data will not be returned to participants or Investigators, unless required by law or local regulations. Privacy and confidentiality of data generated in the future on stored samples will be protected by the same standards applicable to all other clinical data.

Only data collected for the study that does not bear any information that could directly identify the participant will be transferred to third parties or other countries. Should this transfer occur, it will be for the same purposes as the study and guarantee confidentiality with at least the level of protection afforded by applicable regulations.

Participants will be informed that their clinical data will be incorporated into an automated study-specific file after, and the results of the clinical studies and different studies conducted with samples can be communicated at scientific meetings, medical conferences or publications. However, the participant's identity or identifiable data will never be disclosed.

8.6 Personal Data Protection

According to the Council of 27 April 2016 on the protection of natural persons as to the processing of personal data and the free circulation of data, and in accordance with the provisions of the Article 9 of the EU Regulation 2016/679 the following personal data protections will be followed in this study.

The data collected for the study will be collected only identified by a code, so that no information will be included to identify the participants. Data will be processed with the only purpose of carry out all activities related to the clinical study in compliance with pharmacovigilance regulations (for the drug safety control). The legal basis for the processing of the data is the participant consent and Article 9.2 of the Regulation. Only the Investigator and his collaborators have the right to

access the source data (clinical history) and will be able to relate the data collected in the study with the participant's medical history.

The identity of the participant will not be available to any other person except for a medical emergency or legal requirement. Health authorities, the Research Ethics Committee and personnel authorised by the Sponsor of the study, may have access to the personal data identified when necessary to verify data and study procedures, but always maintaining confidentiality in accordance with current legislation.

Only encrypted data will be transferred to third parties and to other countries, which in no case will contain information that can identify the participant directly (such as name and surnames, initials, address, social security number, etc.). If this assignment occurred, it would be for the same purpose of the study described and guaranteeing confidentiality.

If a transfer of encrypted data is carried out outside the EU, either in entities related to the hospital where the participant participates, to service providers or to researchers who collaborate with them, the data of the participants will be protected by safeguards such as contracts or other mechanisms established by the data protection authorities.

The Sponsor of the study commits to carry out the data processing according to EU Regulation 2016/679 and, therefore, to keep a record of the processing activities to carry out and to make a risk assessment of the data processing, to establish what measures will be applied and how it will be done.

In addition to the rights already covered by the previous legislation (access, modification, opposition and cancellation of data, deletion in the new Regulation) participants can now limit the processing of data collected for the project that has to be rectified, request a copy, or move to a third party (portability). To exercise these rights, the participant should be directed to the Investigator of the study or the Data Protection Delegate of their site. The participant also has the right to contact the Data Protection Agency if not satisfied.

The data cannot be deleted even if a participant discontinues the study, to guarantee the validity of the investigation and comply with the legal duties and the medication authorisation requirements.

The Investigator and the Sponsor are obliged to keep the data collected for the study at least up to 25 years after its completion. Subsequently, personal information will only be kept by the site for the care of their health and by the Sponsor for other scientific research purposes if the participant has given their consent to do so, and if the law and applicable ethical requirements so permit.

In accordance with the provisions of recital 33 of the regulations and the corresponding provisions of each country involved in the study regulations, the data may be preserved in such a way that the clinical data are kept separate from the identifiers, to be used in future investigations, applying all technical precautions necessary to avoid their re-identification, and in accordance with all ethical and legal requirements.

8.7 Quality Assurance and Quality Control

According to ICH/GCP guidelines, the Sponsor should ensure that the study is adequately monitored.

The purpose of monitoring is to verify that the rights and wellbeing of human participants are protected; that the study is accurate, complete, and verifiable with source data and that the study is conducted in compliance with the protocol, GCP, and the applicable regulatory requirements.

A monitoring plan will be designed by the contracted CRO. The monitoring plan will establish the guideline for conducting all the monitoring activities.

To ensure homogeneity and the same quality standards, monitors in all study centres will be trained with the same procedures. The training sessions will be done before the initiation of the study and training periodic updates if new sites or personnel are included or when overall quality of data collected through the eCRF is not optimal. The training session will be focused on the protocol implementation, target population and recruitment, electronic CRF and site initiation visit (SIV) procedures and monitoring forms to be used along the study conduct.

During the SIV, the monitors will train the local Investigators and collaborators about study protocol, requirements, interventions, visits, follow-up activities and electronic CRF completion. These tasks will ensure the quality of the data and the quality of the conduct of the clinical study.

Source data will be verified during on-site monitoring visits. During the visits, the monitor will compare the data entered in the eCRF with the source documents. The nature and location of all source documents will be identified to ensure that all sources of original data required to complete the eCRF are known to the monitor and study-site personnel and are accessible for verification. During monitoring visits, the relevant study-site personnel should be available, the source documentation accessible, and a suitable environment provided for review of study-related documents. The monitor will meet the Investigator. The number of visits will be described in detail in the monitoring plan.

A close-out visit will be performed before the final closure of each centre. During this visit, medication accountability will be checked and arranged according to the indications of the final version of the protocol. Besides, a general overview and a final revision of pending queries will be implemented, ensuring that all data and corresponding source documents are in place before the site closure.

8.7.1 Direct Access to Source Data

Investigators will ensure access to the source documents of the staff responsible for guaranteeing data quality and data analysis. In addition, access to documentation will be provided, if necessary, to the staff duly authorised by the Sponsor (study monitors), to regulatory authorities, and to the IEC if they request to inspect the study.

Source documents will be stored in the Investigator's File. These documents will be kept for a minimum period of 25 years by the Investigator, after which they will be destroyed.

8.7.2 Electronic Case Report Form Completion

Data generated and/or collected at clinical sites will be registered using an electronic data management system with remote data entry. Completeness and plausibility checks will ensure the collection of high-quality data. An eCRF will be designed, validated and implemented and will provide electronic data capture functionality (EDC) to the Investigators. The electronic data management system consists in two modules: the eCRF design and the SQL database; and so allows the link between case forms and the database in order to make the generated eCRF consistent and valid, as well as project customised. It complies with the standard¹⁸ and specific¹⁹, GCP as well as the higher software^{20,21,22,23} validation requirements, with restricted access based on user level, and with inconsistency filters and traceability of all data until the database closure.

The eCRF and its database are developed and maintained by the Data management group of the Veristat, LLC.

An eCRF for each participant will be completed by authorised personnel.

Each study site will have access to their participants only and access will be secured using unique personal logins. Each data recording or editing event will be logged in the database to allow better monitoring and database coordination.

The Sponsor or its delegates must ensure that data are recorded in the eCRF correctly and completely by authorised personnel. The Investigator must confirm the integrity of the data transferred to the eCRF by signature.

The data entered in the eCRF must be consistent with the project source documents (specific project records, medical history, medical and nursing notes, laboratory reports, etc.) The data must be entered by authorised personnel, after receiving specific training in the use of the eCRF.

Any change in the initial data recorded will be justified and traceable to make the data as clean and accurate for analysis. Data quality will be enhanced by a series of scheduled checks that automatically detect out-of-range or abnormal data.

The data entry should not be delayed regarding the obtaining of the data sources, trying to be as simultaneous as possible, to avoid the loss of data.

The eCRF will be designed to require only the necessary data following a system of locks and dependencies.

Those data not registered in the source data will be considered lost data.

8.7.3 Data Handling

Demographic and biological data will be collected at each clinical site and entered in an eCRF. Defined Investigators at each of the study sites will be in charge of data entry to an eCRF. The collection and processing of personal data from participants enrolled in this study will be limited to those data that are necessary to fulfil the objectives of the study. These data must be collected and processed with adequate precautions to ensure confidentiality and compliance with applicable data protection, European, and National regulations. Appropriate technical and organisational measures to protect the personal data against unauthorised disclosures or access, accidental or unlawful destruction, or accidental loss or alteration will be put in place. The clinical Investigator team at each clinical centre will compromise to keep the identity of participants confidential. For

this aim, participants will be assigned an alphanumeric code (Participant ID). The link between the code and real personal data will be safely stored at the study site, and only the local Investigator will have access to it.

8.8 Record Retention

The Investigator is responsible for maintaining all records which enable the conduct of the clinical study at the site to be fully documented, in compliance with ICH GCP filing standard. Timeliness and completeness of the documentation will be regularly checked by the clinical monitor. The documentation of the clinical study including all the relevant correspondence should be kept by the Investigator for the minimum period required by applicable regulatory.

All completed study related documents (e.g., eCRF, ICF, drug accountability logs, staff signature lists, participant identification log) must be archived for 25 years.

8.9 Financing, Liability, and Insurance

The Sponsor or his delegate will procure insurance for this clinical study to cover study related injuries of the participants according to local regulatory requirements.

Clinical study participants will be provided on request with the conditions of insurance along with the ICF.

8.10 Publication of Study Findings and Use of Information

All data generated as a result of this study will be considered confidential and remain the sole property of the Sponsor.

The Sponsor, in agreement with the Investigators of the study will publish the results of the study in internationally indexed journals. Authorship will consider members of the study management, participating Investigators and persons responsible for coordination, data analysis and article writing.

8.11 Return of Research Results and Management of Incidental Findings

All data generated as a result of this study will be considered confidential and remain the sole property of the Sponsor.

The Sponsor, in agreement with the Investigators of the study, will publish the results of the study in internationally indexed journals. Authorship will consider members of the study management, participating Investigators, and persons responsible for coordination, data analysis, and article writing.

8.12 Post-Study Access

No post-study access to the study drugs will be provided.

9. REFERENCES

1. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, et al; China Novel Coronavirus Investigating and Research Team. A Novel Coronavirus from Patients with Pneumonia in China, 2019. *N Engl J Med*. 2020 Feb 20;382(8):727-733.
2. Ghebreyesus TA. WHO director-general's opening remarks at the media briefing on COVID-19—11 March 2020 [Available from: <https://www.who.int/dg/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-COVID-19-11-march-2020>].
3. Li Q, Guan X, Wu P, Wang X, Zhou L, Tong Y, et al. Early Transmission Dynamics in Wuhan, China, of Novel Coronavirus-Infected Pneumonia. *N Engl J Med*. 2020 Mar 26;382(13):1199-1207.
4. Wiersinga WJ, Rhodes A, Cheng AC, Peacock SJ, Prescott HC. Pathophysiology, transmission, diagnosis, and treatment of coronavirus disease 2019 (COVID-19): a review. *JAMA*. 2020;324(8):782–793.
5. Keech C, Albert G, Cho I, Robertson A, Reed P, Neal S, et al. Phase 1-2 Trial of a SARS-CoV-2 Recombinant Spike Protein Nanoparticle Vaccine. *N Engl J Med*. 2020 Dec 10;383(24):2320-2332.
6. Baden LR, El Sahly HM, Essink B, Kotloff K, Frey S, Novak R, et al; COVE Study Group. Efficacy and Safety of the mRNA-1273 SARS-CoV-2 Vaccine. *N Engl J Med*. 2021 Feb 4;384(5):403-416.
7. Sadoff J, Le Gars M, Shukarev G, Heerwegh D, Truyers C, de Groot AM, et al. Interim Results of a Phase 1-2a Trial of Ad26.COV2.S COVID-19 Vaccine. *N Engl J Med*. 2021 Jan 13;NEJMoa2034201. doi: 10.1056/NEJMoa2034201. Online ahead of print.
8. Dalvie NC, Rodriguez-Aponte SA, Hartwell BL, Tostanoski LH, Biedermann AM, Crowell LE, Kaur K, Kumru OS, Carter L, Yu J, Chang A, McMahan K, Courant T, Lebas C, Lemnios AA, Rodrigues KA, Silva M, Johnston RS, Naranjo CA, Tracey MK, Brady JR, Whittaker CA, Yun D, Brunette N, Wang JY, Walkey C, Fiala B, Kar S, Porto M, Lok M, Andersen H, Lewis MG, Love KR, Camp DL, Silverman JM, Kleanthous H, Joshi SB, Volkin DB, Dubois PM, Collin N, King NP, Barouch DH, Irvine DJ, Love JC. Engineered SARS-CoV-2 receptor binding domain improves manufacturability in yeast and immunogenicity in mice. *Proc Natl Acad Sci U S A*. 2021 Sep 21;118(38):e2106845118. doi: 10.1073/pnas.2106845118. PMID: 34493582.
9. Borobia A, Carcas A, Pérez-Olmeda M, et al. Immunogenicity and reactogenicity of BNT162b2 booster in ChAdOx1-S-primed participants (CombiVacS): a multicentre, open-label, randomised, controlled, phase 2 trial. *Lancet*. 2021 Jun 25;S0140-6736(21)01420-3. doi: 10.1016/S0140-6736(21)01420-3.
10. Groß R, Zanoni M, Seidel A, et al. Heterologous ChAdOx1 nCoV-19 and BNT162b2 prime-boost vaccination elicits potent neutralizing antibody responses and T cell reactivity. *medRxiv* 2021.05.30.21257971; doi: <https://doi.org/10.1101/2021.05.30.21257971>

11. Hillus D, Schwarz T, Tober-Lau P, et al. Safety, reactogenicity, and immunogenicity of homologous and heterologous prime-boost immunisation with ChAdOx1-nCoV19 and BNT162b2: a prospective cohort study. medRxiv 2021.05.19.21257334; doi: <https://doi.org/10.1101/2021.05.19.21257334>
12. Liu X, Shaw R, Stuart A, et al. Safety and immunogenicity report from the Com-COV Study – a single-blind randomised non-inferiority trial comparing heterologous and homologous prime-boost schedules with an adenoviral vectored and mRNA COVID-19 vaccine. https://papers.ssrn.com/sol3/papers.cfm?abstract_id=3874014
13. Wu K, Choi A, Koch M, et al. Preliminary analysis of safety and Immunogenicity of a SARS-CoV-2 variant vaccine booster. medRxiv 2021.05.05.21256716; doi: <https://doi.org/10.1101/2021.05.05.21256716>
14. FDA Guidance for Industry: Clinical Data Needed to Support the Licensure of Seasonal Inactivated Influenza Vaccines, May 2007 (<https://www.fda.gov/regulatory-information/search-fda-guidance-documents/clinical-data-needed-support-licensure-seasonal-inactivated-influenza-vaccines>)
15. Connor, R. I., Chen, B. K., Choe, S. & Landau, N. R. Vpr is required for efficient replication of human immunodeficiency virus Type-1 in mononuclear phagocytes. Virology 206, 935–944 (1995).
16. Ou, X. et al. Characterization of spike glycoprotein of SARS-CoV-2 on virus entry and its immune cross-reactivity with SARS-CoV. Nat. Commun. 11, 1620 (2020).
17. FDA. Guidance for Industry. Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials (September 2007)
18. CPMP/ICH/135/95 Note for Guidance on Good Clinical Practice, URL: http://www.ema.europa.eu/ema/pages/includes/document/open_document.jsp?webContentId=WC500002874
19. Good Clinical Data Management Practice, Version 4, Society for Clinical Data Management (SCDM), October 2005
20. EMEA. Reflection on expectations for electronic source documents used in clinical trials. London, 17 October 2007
21. Directive 9 Guidance for Industry. Part 11, Electronic Records; Electronic Signatures – Scope and Application (August 2003)
22. FDA. Guidance for Industry. Computerized Systems Used in Clinical Investigations (May 2007)
23. FDA. Guidance for Industry. Part 11, Electronic Records; Electronic Signatures – Scope and Application (August 2003)

10.APPENDICES

10.1 Appendix 1. List of Adverse Events of Special Interest

Potential Immune-Mediated Medical Conditions (PIMMCs)

(of New onset, in addition to the already pre-existing underlying autoimmune diseases)

Categories	Diagnoses (as MedDRA Preferred Terms)
Neuroinflammatory Disorders	Acute disseminated encephalomyelitis (including site specific variants: e.g., non-infectious encephalitis, encephalomyelitis, myelitis, myeloradiculomyelitis), cranial nerve disorders including paralyses/paresis (e.g., Bell's palsy), generalised convulsion, Guillain-Barre syndrome (including Miller Fisher syndrome and other variants), immune-mediated peripheral neuropathies and plexopathies (including chronic inflammatory demyelinating polyneuropathy, multifocal motor neuropathy and polyneuropathies associated with monoclonal gammopathy), myasthenia gravis, multiple sclerosis, narcolepsy, optic neuritis, transverse myelitis, uveitis.
Severe COVID-19 cases happening ≥ 14 days post-vaccination	Severe COVID-19 case confirmed through RT-PCR or RAT. Severity explained as >24 hours of hospitalisation.

Categories	Diagnoses (as MedDRA Preferred Terms)
Musculoskeletal and Connective Tissue Disorders	Antisynthetase syndrome, dermatomyositis, juvenile chronic arthritis (including Still's disease), mixed connective tissue disorder, polymyalgia rheumatic, polymyositis, psoriatic arthropathy, relapsing polychondritis, rheumatoid arthritis, scleroderma (including diffuse systemic form and CREST syndrome), spondyloarthritis (including ankylosing spondylitis, reactive arthritis [Reiter's Syndrome] and undifferentiated spondyloarthritis), systemic lupus erythematosus, systemic sclerosis, Sjogren's syndrome
Vasculitis	Large vessels vasculitis (including giant cell arteritis such as Takayasu's arteritis and temporal arteritis), medium sized and/or small vessels vasculitis (including polyarteritis nodosa, Kawasaki's disease, microscopic polyangiitis, Wegener's granulomatosis, Churg-Strauss syndrome [allergic granulomatous angiitis], Buerger's disease [thromboangiitis obliterans], necrotising vasculitis and anti-neutrophil cytoplasmic antibody [ANCA] positive vasculitis [type unspecified], Henoch-Schonlein purpura, Behcet's syndrome, leukocytoclastic vasculitis)
Gastrointestinal Disorders	Crohn's disease, celiac disease, ulcerative colitis, ulcerative proctitis Hepatic Disorders: Autoimmune hepatitis, autoimmune cholangitis, primary sclerosing cholangitis, primary biliary cirrhosis
Renal Disorders	Autoimmune glomerulonephritis (including IgA nephropathy, glomerulonephritis rapidly progressive, membranous glomerulonephritis membranoproliferative glomerulonephritis, and mesangio proliferative glomerulonephritis
Cardiac Disorders	Autoimmune myocarditis/cardiomyopathy

Categories	Diagnoses (as MedDRA Preferred Terms)
Skin Disorders	Alopecia areata, psoriasis, vitiligo, Raynaud's phenomenon, erythema nodosum, autoimmune bullous skin diseases (including pemphigus, pemphigoid and dermatitis herpetiformis), cutaneous lupus erythematosus, morphoea, lichen planus, Stevens- Johnson syndrome, Sweet's syndrome
Hematologic Disorders	Autoimmune haemolytic anaemia, autoimmune thrombocytopenia, antiphospholipid syndrome, thrombocytopenia
Metabolic Disorders	Autoimmune thyroiditis, Grave's or Basedow's disease, Hashimoto thyroiditis ^a , diabetes mellitus type 1, Addison's disease
Other Disorders	Goodpasture syndrome, idiopathic pulmonary fibrosis, pernicious anaemia, sarcoidosis, hypersensitivity.

^a For Hashimoto thyroiditis: new onset only