THE LANCET
Infectious Diseases

Supplementary appendix 3

This appendix formed part of the original submission and has been peer reviewed. We post it as supplied by the authors.

Supplement to: Zeng G, Wu Q, Pan H, et al. Immunogenicity and safety of a third dose of CoronaVac, and immune persistence of a two-dose schedule, in healthy adults: interim results from two single-centre, double-blind, randomised, placebo-controlled phase 2 clinical trials. *Lancet Infect Dis* 2021; published online Dec 7. https://doi.org/10.1016/S1473-3099(21)00681-2.

同意 HeBCDC IRB 启用日期:2021年2月28日

项目名称：新型冠状病毒灭活疫苗（Vero 细胞）老年人I/II期临床试验

方案名称：评价新型冠状病毒灭活疫苗（Vero 细胞）在 60 岁及以上健康人中的安全性和免疫原性的随机、双盲、安慰剂对照I/II期临床试验

研究产品名称：新型冠状病毒灭活疫苗（Vero 细胞）

申办者：北京科兴中维生物技术有限公司

研究方：河北省疾病预防控制中心

统计方：北京康特瑞科统计科技有限责任公司

方案编号：PRO-nCOV-1002

方案版本日期：2021 年 02 月 08 日

版本号：1.4

方案批准人：高强

批准人签字：高强

批准日期：2021年02月08日

北京科兴中维生物技术有限公司
SINOVAC RESEARCH & DEVELOPMENT CO., LTD.

主要研究者签字

我同意：

- 承担正确指导在本地区进行该项临床研究的职责。
- 确保本研究按照试验方案及临床研究标准操作规程进行。
- 确保参与本项目的人员充分了解研究产品信息和其它本试验方案中规定的研究相关的职责和义务。
- 确保在未经申办者和伦理委员会（IEC）的审查和书面批准的情况下，不对试验方案做任何更改，除非需要消除对受试者的即刻危害或出于遵从注册管理当局的要求（例如：项目的行政管理方面）。
- 我完全熟悉试验方案中所描述的正确使用该疫苗的方法，完全了解申办者所提供的其它信息，包括但不限于以下内容：现行的研究者手册（IB）或等效文件、研究者手册增补件（如果有）。
- 我熟悉并将遵守《药物临床试验质量管理规范》（GCP）、《疫苗临床试验质量管理指导原则（试行）》和所有现行的法规要求。

主要研究者姓名：赵玉良

签名：

日期：2021年2月8日

Project Title: Clinical Study of Phase I/II of SARS-CoV-2 Vaccine (Vero cell), Inactivated in Elderly Population

Protocol Title: A Randomized, Double-blind, Placebo-controlled Clinical Trial, to Evaluate Safety and Immunogenicity of Inactivated SARS-CoV-2 Vaccine (Vero cell), in Healthy Elderly Aged 60 Years and above

Name of Investigational Product: SARS-CoV-2 Vaccine (Vero cell), Inactivated

Sponsor: Sinovac Life Sciences Co., Ltd.

Study Institution: Hebei Provincial Center for Disease Control and Prevention

Statistical Company: Beijing Key Tech Statistics Technology Co., Ltd.

Protocol No.: PRO-nCOV-1002

Protocol Version Date: February 08, 2021

Version No.: 1.4

Protocol Approver: Qiang GAO

Signature of the Approver:

Approval Date:

北京科兴中维生物技术有限公司
SINOVAC RESEARCH & DEVELOPMENT CO., LTD.

Signature of thePrincipal Investigator

I agree to:

- Take the responsibility to correctly guide the conduct of the clinical study in the region.
- Ensure that thisstudy can be conducted in accordance with the trial protocoland the standard operating procedures for clinical studies
- Ensure that the personnel involved in the project fully understand the information of the investigational product and other responsibilities and obligationsrelated to the study specified in the project.
- Ensure that any modificantions of the trial protocol will not be conducted without the without the review and written approval of the sponsor and the Independent Ethics Committee (IEC), unless the immediate hazard for the subjects needs to be eliminated or follow the requirements of the registration regulatory authorities (e.g., administrative management aspect of the project).
- I am completely familiar with the method to correctly use the vaccine described in the trial protocol, have fully understood other information provided by the sponsor, including but not limited to the following contents: current investigator'sbrochure (IB) or equivalent documents and supplements of the IB (if any).
- I am familiar with and will abide by *Good Clinical Practice* (GCP), *Guidances on Vaccine Clinical Trial Quality Management (Trial)* and all current regulations.

Name of the Princeple Investigator: Yuliang ZHAO
Signature:
Date:

Study Team

Sponsor

Sinovac Life Sciences Co., Ltd.
Add.: No. 21, TIANFU Street, Daxing Biomedicine Industrial Base, Zhongguancun Science Park, Daxing District, Beijing, P. R ChinaName of the Contact Person: Qiang GAO
Tel: 13693092396 Fax: 010-62979669 Zip Code: 100085
E-mail：gaoq@sinovac.com

Study Institution

Hebei Provincial Center for Disease Control and Prevention
Add.: No. 97, Huaiandong Road, Shijiazhuang, Hebei Province
Department: Vaccine Clinical Evaluation Institute
Name of the Principle Investigator (PI): Yuliang ZHAO
Tel.: 13315290538 Fax: 0311-86573212 Zip Code: 050021
Email：yuliang_zh@163.com

Clinical Trial Site

Institution: Renqiiu Center for Disease Control and Prevention
Add.: North Side of Taishan Road, Renqiu
Contact Person: Hui JIN
Tel.: 13930778699 Fax: 0317-2267709 Zip Code: 062550
E-mail：1206618652@qq.com

Person in Charge of Monitoring

Name: Yuansheng HU
Sinovac Biotech Co., Ltd.
Add.: No. 39, Shangdi Xi Road, Haidian District, Beijing
Tel.: 13436950182,010-82799318 Fax: 010-82890408 Zip Code: 100085
E-mail：huys@sinovac.com

Institution Responsible for Serum Antibody Testing

Institution: National Institute for Food and Drug Control
Add.: No.31, Huatuo Road, Daxing District, Beijing
Tel.: 010-53851770 Zip Code: 102629

Data Management Institution

Institution: Meta Clinical Technology Co., Ltd.
Add.: Room A, 20 / F, Oriental International Technology building, 58 Xiangcheng Road, Pudong New Area, Shanghai
Contact Person: Hualong SUN
Tel.: 13816706496 Zip Code: 200031 E-mail：hualong.sun@meta-clinical .com

Statistical Analysis Institution

Institition: Beijing Key Tech Statistics Technology Co., Ltd.
Add.: 1018-1119w, Sihui building, Huihe Nan Street, Chaoyang District, Beijing

Contact Person: Zhiwei JIANG

Tel.: 18618483152 Zip Code: 100025 E-mail：zhi.wei.jiang@ktstat.com

Revision Record

No.	Original Version/Date/Revised content	Current Version/Date/Revised content
1	**When Version 1.4 is approved, Version 1.3 is invalid.**	
2	Version 1.3/May 11, 2020/4.1.3 Repeated Dose Toxicity Test in Rats	Version 1.4/February08, 2021/ Updated Repeated Dose Toxicity Test in Rats
3	Version 1.3/May 11, 2021/4.1.5 Reproductive and Development Toxicity Study in Rats	Version 1.4/February 08, 2021/ Updated Reproductive and Development Toxicity Study in Rats
4	Version 1.3/May 11, 2020/4.4 Study of Virus Challenge	Version 1.4/February 08, 2021/ Updated Study of Virus Challenge
5	Version 1.3/May 11, 2020/	Version 1.4/February 08, 2021/ Added "5 Preliminary Clinical Study"
6	Version 1.3/May 11, 2020/ Stability of Vaccine 6.2	Version 1.4/February 08, 2021/ Supplemented the results of stability study
7	Version 1.3/May 11, 2020/6.5 Administration Rout and Schedule	Version 1.4/February 08, 2021/ The immunization program was modified from " at the two doses schedule of day 0,28" to " Two doses of primary immunization were given at 0,28 days, respectively. One dose of booster immunization was given 6 months or 1 year after the second dose (1 year for the phase Ⅰ, 6 months for the phase Ⅱ)".
8	Version 1.3/May 11, 2020/8.2 Study Endpoint	1.4/February 08, 2021/ Added the study endpoint of booster immunization
9	Version 1.3/May 11, 2020/8.3 Study Plan &10.1 Visit Plan	1.4/February 08, 2021/ Added "one dose of booster immunization was given one year after the second dose of immunization in phase Ⅰ", and "one dose of booster immunization was given six months after the second dose of immunization was added in phase Ⅱ".At the same time, corresponding visits and sample collection were added to the visit plan.
10	Version 1.3/May 11, 2020/10.7.2 Defination of Adverse events/Reactions	1.4/February 08, 2021/The definition of adverse events/Reactions were revised according to the Chinese GCP (2020).
11	Version 1.3/May 11, 2020/10.7.6 Reporting of Serious Adverse Events	1.4/February 08, 2021/The reporting process for serious adverse events added "The report object can be adjusted according to existing regulations and the requirements of local regulatory authorities and ethics committees"; 24-hour contact of sponsor has changed.
12	Version 1.3/May 11, 2020/10.11.1 Analysis Set	1.4/February 08, 2021/ According to the revision of this vaccination procedure, the partition of the data analysis set was revised.
13	Version 1.3/May 11, 2020/11.7 Preservation of clinical trial documents	1.4/February 08, 2021/According to the requirements of Chinese GCP (2020), the content fo clinical documents preservation was revised as "The clinical trial documents must be kept according to the requirements of Chineses GCP. The sponsor, study institution and studysite should keep the clinical trial data for at least 5 years after the drug is marketed.
14	**When Version 1.3 is approved, Version 1.2 is invalid.**	
15	Version 1.2/May 4, 2020/5.6 Information of Investigation vaccine	Version 1.3/May 11, 2020/ The batch numbers of medium and high dose trial vaccines are changed, and the corresponding validity period are revised
16	Version 1.2/May 4, 2020/9.1 Visit plan	Version 1.3/May 11, 2020/ The time of Visit 2 from "the 7th day after the first dose" to "the 8th day after the first dose; the time of Visit 4 from "the 7th day after the first dose" to "the

No.	Original Version/Date/Revised content	Current Version/Date/Revised content
		8th day after the second dose
17	**When Version 1.2 is approved, Version 1.1 is invalid.**	
18	Version 1.1/April 29, 2020/1 Introduction	Version 1.2/May 4, 2020/ Updated the results of clinical trials in adults aged 18-58 years
19	Version 1.1/April 29, 2020/7.7 Protocol Violation/Deviation	Version 1.2/May 4, 2020/ Added "protocol deviation / violation report submitted to Ethics Committee for review and approval
20	Version 1.0/April 4, 2020/7.7 Protocol Violation/Deviation: Conditions of protocal violation are listed as follows (including but not limited to): - Subjects do not meet the inclusion criteria or meet exclusion criteria； - Subjects are vaccinated with wrong vaccine; - SAE is not reported within the specified time。 Conditions of protocal deviation are listed as follows (including but not limited to): - Not receiving the investigational vaccine within the protocol-required time window; - Not receiving the blood sampling within the protocol-required time window; - The interval time with other vaccines can not meet the requirement of protocal (Except for rabies or tetanus vaccination in case of emergency).	Version 1.1/April 29, 2020/ Revised as: Refers to any change and non-compliance with the clinical trial protocol design or process. The behavior that does not affect the rights and interests, safety and benefits of the subjects, or the integrity, accuracy and reliability of the data as well as the safety or primary indicatiors belong to the protocol deviation; those that affect the rights and interests, safety and benefits of the subjects, or the integrity, accuracy and reliability of the data and the safety or primary indicatiors belong to serious protocol deviation (i.e. protocol violate). For the protocal deviation / violation during the study, the on-site investigators shall report the fact, process, causes and impact of the incident to the research responsible institution. The principle investigator shall give opinions on the handling of the incident. The investigators should carry out targeted training for relevant staff in the related links of the violation of the protocol to prevent the recurrence of similar incidents, and record the training process.
21	Version 1.0/April 4, 2020/8.2 Exclusion criterion	Version 1.1/April 29, 2020/ Delete the BMI exclusion criteria; Add "History of SARS".
22	Version 1.0/April 4, 2020/9.5 Safety Follow-up Observation	Version 1.1/April 29, 2020/ The safety follow-up observation method was revised to "Subjects will be observed for 30 minutes on site after each dose of vaccination. Diary cards and contact cards are distributed to subjects to record the adverse events within 0~7 days and 8~28 days respectively. The investigators explain the judgment, measurement, recording, precautions and reporting method of adverse events. Systematic observation is carried out within 7 days after vaccination. Subjects are required to closely observe their own symptoms and vital signs and fill in the diary card every day. The investigators verify the adverse events on the 8th days after vaccination through face-to-face interviews on all subjects (those who do not face-to-face interviews are conducted by telephone), collect diary cards and distribute contact cards to record the adverse events within 8~28 days. The investigators verify the adverse events on the 28th days and collect contact cards.
23	Version 1.0/April 4, 2020/9.6 Blood sample collection	Version 1.1/April 29, 2020/ The blood volume is changed from 3ml to 2.5~3.5ml.

No.	Original Version/Date/Revised content	Current Version/Date/Revised content
24	Version 1.0/April 4, 2020/9.7.4 Correlation of Adverse events with Vaccination	Version 1.1/April 29, 2020/The correlation between adverse events and vaccination is modified, as shown in the protocol.
25	Version 1.0/April 4, 2020/2 Participanting Institutions and Responsibilities	Version 1.1/April 29, 2020/ Add the Data Monitoring Committee (DMC) and responsibilities
26	Version 1.0/April 4, 2020/	Version 1.1/April 29, 2020/ Delete the content related to the 0,14 day immunization program
27	Version 1.0/April 4, 2020/3.5 R&D vaccine	Version 1.1/April 29, 2020/ Updated the global vaccine research and development progress
28	Version 1.0/April 4, 2020/7.3.1 Study Plan of Phase I Clinical Trail	Version 1.1/April 29, 2020/ Add “Blood collection 28 days after the first dose”; The sample size is changed from 120 to 72.
29	Version 1.0/April 4, 2020/7.3.2 Study Plan of Phase II Clinical Trail	Version 1.1/April 29, 2020/ The sample size is changed from 600 to 350； Add “the low dosage group”.
30	Version 1.0/April 4, 2020/9.7.5 Treatment of Adverse Events	Version 1.1/April 29, 2020/ During the obervation of the study, subjects with a fever and cough and other respiratory symptoms ,as needed to fixed-point hospital, pharyngeal swab collected/sputum and anal swab, and CT and other imaging examination for analysis to determine whether will be infected by the COVID-19, in the event will be infection, the process for problems according to the SAE, especially analysis the existence of ADE phenomenon.

Protocol Summary

PROTOCOL TITLE	A Randomized, Double-blind, Placebo-controlled Clinical Trial, to Evaluate Safety and Immunogenicity of Inactivated SARS-CoV-2 Vaccine (Vero cell), in Healthy Elderly Aged 60 Years and above
SPONSOR	Sinovac Life Sciences Co., Ltd.
PROJECT PHASE	Phase I/II
OBJECTIVE(S)	To evaluate the safety and immunogenicity of SARS-CoV-2 vaccine in elderly
EXPERIMENTAL DESIGN OF THE TRIAL	A randomized, Double-blinded, Placebo-Controlled, Phase I/II Clinical Trial
PLANNED SAMPLE SIZE	Total of 422 subjects， with 72 in the phase I and 350 in the phase II clinical trial
SUBJECT SELECTION CRITERIA	Healthy older adults aged ≥60 years, with equal percentage of each gender
NAME AND FORMULATION OF DRUG	SARS-CoV-2 Inactivated Vaccine -Inactivated SARS-CoV-2: low dosage: 300SU/0.5ml; medium dosage: 600SU/0.5ml; high dosage: 1200SU/0/5ml -Aluminum hydroxide, disodium hydrogen phosphate, sodium dihydrogen phosphate, sodium chloride, etc.
DOSAGE AND SCHEDULE	Dosage: 0.5ml/dose Primary Immunization: two doses (Day 0,28) Booster immunization: a booster dose 6 months or 1 year after the second dose.
ROUTE OF ADMINISTRATION	Intramuscularly, deltoid region
CHALLENGE SCHEDULE, if applicable	None
BLOOD SAMPLE COLLECTION	Phase I: day 0,28, 56,208, 388, 416 Phase II: day 0,56,208, 215 (or 222), 236, 388, 568
PARAMETERS OF SAFETY	Primary Endpoint – Incidence of adverse reactions within 28 days after each dose of vaccination; Secondary Endpoints – Incidence of adverse reactions within 7 days after each dose vaccination; – Incidence rate of SAEs from the beginning of the vaccination to 6 months after the booster immunization (Phase Ⅰ); – Incidence of SAEs from the beginning of the vaccination to 12 months after booster immunization(Phase Ⅱ).
PARAMETERS OF IMMUNOGENICITY	Primary Endpoint – The seroconversion rate of neutralizing antibodies 28 days after the second dose vaccination. Secondary Endpoints – The seropositive rate, GMT, and GMI of neutralizing antibodies 28 daysafter the second dose vaccination; – The seroconversion rate, seropositive rate, GMT, and GMI 28 days after the first dose vaccination (Phase I). Exploratory Endpoints – The seropositive rate and GMT 6 months after the second dose vaccination – The seropositive rate and GMT 12 months after the second dose vaccination (Phase I); – The seropositive rate, GMT, and GMI 28 days after the booster vaccination (Phase I). – The seropositive rate, GMT, and GMI 7 days (or 14 days) and 28 days after the booster vaccination (Phase Ⅱ). – The seropositive rate and GMT 6 months after the booster vaccination. – The seropositive rate and GMT 12 months after the booster vaccination (Phase Ⅱ).

Glossary

ADE	Antibody Dependent Enhancement
AE	Adverse Event
ALB	Albumin
ALT	Alanine Aminotransferase
AST	Aspartate Aminotransferase
CDC	Center for Disease Control and Prevention
CDE	Center for Drug Evaluation
CFDA	China Food and Drug Administration
CK	Creatine Kinase
COVID-19	Corona Virus Disease 2019
CPK	Creatine Phosphokinase
NMPA	National Medical Products Administration
CFDI	Center for Food and Drug Inspection
CRF	Case Report Form
eCRF	Electronic Case Report Form
ELISA	Enzyme-Linked Immune-sorbent Assay
ELISPOT	Enzyme-Linked Immuno-spot Assay
EDC	Electronic Data Capture
FAS	Full Analysis Set
GCP	Good Clinical Practice
IEC	Independent Ethics Committee
ITT	Intention-to-Treat
LDH	Lactate Dehydrogenase
MedDRA	Medical Dictionary for Regulatory Activities
PI	Principal Investigator
PPS	Per Protocol Set
PT	Preferred Term
SAE	Serious Adverse Event
SARS	Severe Acute Respiratory Syndrome
SARS-CoV-2	Severe Acute Respiratory Syndrome Coronavirus 2
SOC	System Organ Class
SOP	Standard Operation Procedure
SS	Safety Set
SUSAR	Suspected Unexpected Serious Adverse Reaction
TP	Total Protein

Abstract

SARS-CoV-2 Vaccine (Vero Cell), Inactivated developed by Sinovac Life Sciences Co., Ltd. (hereinafter referred as to "Sinovac (R&D)") can induce active immunity and prevent diseases caused by the SARS-CoV-2. According to the preliminary immunogenicity studies, the vaccine can produce good neutralizing antibody responses, and has a good effecitiveness in anmals. At the same time, comprehensive safety evaluations were carried out on animals, showing that the new vaccine is safe. This protocol is drafted on the basis of *Regulation of Drug Registration*[1], *Good Clinical Practice (GCP)* [2], *Guidance on Vaccine Clinical Trial* [3], *Guidance on Vaccine Clinical Trial Quality Management (Trial)*[4] and *Guidance on SARS-CoV-2 Vaccine (Trail)*, etc.

A randomized, double-blind, placebo-controlled design is adopted in Phase I and phase II. In the Phase I clinical trial, 72 healthy older adults aged ≥60 will be selected as study subjects. After the informed consents are signed, subjects who pass the physical examination, meet the inclusion criteria and didn't meet the exclusion criteria will be enrolled into the study. Each enrolled subject will receive two doses of primary immunization according to the immunization schedule of day 0, 28. The dose-escalating manner is used in Phase I, with 36 at medium dosage stage, and 36 at high dosage stage. The sujects enrolled at each dosage stage will be assigned in a 2:1 ratio to receive investigational vaccine or placebo respectively. The medium stage vaccination should be carried out firstly. The high dosage stage vaccination will start only with the condition that safety observation 0~7 days after the first dose of the medium dosage stage vaccination is finished, and the good safety profiles is confirmed. All enrolled subjects will receive one booster dose 1 year after primary immunization.

The phase II will be initiated only with the condition that the safety observation 0~7 days after the first dose of the high dosage stage vaccination in Phase I is finished, and the good safety profiles is confirmed the Data Monitoring Committee (DMC). A total of 350 healthy older adults aged ≥60 will be selected in the phase II clinical trial. On the premise that informed consents are signed. Subjects who pass the physical examination, meet the inclusion criteria and didn't meet the exclusion criteria will be enrolled. Each enrolled subject will receive two doses of primary immunization according to the immunization schedule of day 0,28. The subjects will be assigned in a ratio of 2:2:2:1 to receive the low dosage, medium dosage, high dosage vaccine, or placebo. All enrolled subjects will receive one booster dose 6 months after primary immunization.

The immediate reactions occur 30 minutes after each dose of vaccination will be observed on site. The local and systemic solicited AEs occurred 0~7 days after each dose vaccination, as well as the unsolicited AEs occurred 0~28 days will be collected. Additionally, the SAEs during the study periodwill be collected to evaluate the safety. Venous blood will be collected from all subjects at different time points before and after vaccination for the neutralizing antibody test, to evaluate primary immune effect, booster immune effect and immune persistence.

The clinical protocol will be independently undertaken by the investigator after being approved by independent ethics committee (IEC). The clinical research associates designated by the sponsor will monitor the whole process of the study to ensure the safety of the trial.

Contents

1 Introduction

Inactivated SARS-CoV-2 Vaccine (Vero cell), developed by Sinovac Life Sciences Co., Ltd. (hereinafter referred to as "Sinovac (R&D)"). can induce the body to produce active immunity and prevent the diseases caused by SARS-CoV-2. Preliminary immunogenicity studies showed the SAR-Cov-2 vaccine can induce good neutralizing antibody responses, and have a good effectiveness in animals. Comprehensive safety evaluations showed the SARS-CoV-2 vaccine was safety in animals.

The Phase I/II clinical trial in healthy adults aged 18~59 years was conducted in April, 2020 in Suining County, Jiangsu Province. A total of 144 subjects in Phase I and 600 subjects in Phase II have been enrolled. In Phase I of medium dosage stage, 14 subjects had reported 16 local and systemic adverse events (AEs) during 0~7 days after the first vaccination. The incidence rate of AEs was 19.44% (14/72), and pain at injection site was the most frequently reported symptoms. The most AEs were grade 1 by severity, and there were only 1 case of grade 2 AE. No grade 3 or above AEs occurred. On the 3rd day after the first dose in medium dosage stage, a total of 4 subjects had abnormal and clinically significant laboratory indicators, and the incidence rate was 5.6% (2/72). In Phase I of high dosage stage, 12 subjects had reported 21 local and systemic AEs during 0~7 days after the first vaccination. The incidence rate of AEs rate was 16.90% (12/71). The most AEs were grade 1 by severity, and there were only 1 case of grade 3 AE. On the 3rd day after the first dose in the high dosage stage, a total of 2 subjects had abnormal and clinically significant results laboratory indicators, and the incidence rate was 2.8% (2/72). Based on the above results, the DMC confirmed that the inactivated SARS-CoV-2 vaccine (vero cell) was safe and meets the requirements for Phase II. Phase II in healthy adults aged 18~59 years is currently in progress. In order to evaluate the safety and immunogenicity of the inactivated SARS-CoV-2 vaccine (vero cell) in older adults aged 60 years and above, this clinical trial protocol is designed.

2 Participanting Institutions and Responsibilities

2.1 Sponsor

2.1.1 Responsibilities

The sponsor is Sinovac (R&D), Ltd..Its responsibilities are:

-Provide the preliminary clinical trial plan, sign and seal to approve the final plan.

-Provide clinical study approval documents, investigator brochure (pre clinical safety information of products), product implementation standards and other field application documents.

- Provide trial vaccine, and issue vaccine qualified report.

- Evaluate and select the institution in charge of the clinical trial and the study site, appoint the CRA to assess the study site and preform the monitor responsibilities according the requirements of GCP; take ultimate responsibility for the quality of the clinical trial.

-Participate in the investigation and treatment of AEs , and provide the medical treatment and related compensation costs for the confirmed vaccine-related AEs according to relevant regulations; disposal of other conditions is specified in the work agreement.

-Provide clinical study funds.

2.1.2 Profile

Sinovac (R&D), formerly known as the R & D center of Sinovac Biotech Co., Ltd., is a biological high-tech enterprise wholly owned by Sinovac (Hong Kong) Co., Ltd. and registered in 2009 with a registered capital of 9.6 million US dollars. The company is Zhongguancun high-tech enterprise and Zhongguancun gold seed enterprise.

Sinovac (R&D) specializes in the research, development and technical services of human vaccines and related products, providing technical support for the prevention and control of major infectious diseases. Relying on the advantages of the group in vaccine R & D and industrialization for many years, the company has gradually formed a R & D mode with the enterprise as the main body and the combination of production, learning and research. It has

constructed a virus separation and identification technology platform, a cell factory platform, a micro carrier fermentation technology platform, a virus pure chemical technology platform, a bacterial fermentation and purification platform, a polysaccharide protein combination technology platform, and a freeze-drying technology platform, animal evaluation platform, quality control platform, diagnostic reagent raw material development platform, each platform's professional and technical advantages complement each other, cross penetrate, and promote the company's research and development to move forward steadily and efficiently.

Sinovac (R&D) has undertaken 2 national major new drug development projects and 1 Beijing Science and technology plan, and has obtained 12 Chinese invention patents. The 23 valent pneumonia polysaccharide vaccine developed by the company has successfully completed clinical studies and industrialization in Sinovac (Beijing). At present, the company is developing Poliomyelitis Hib series combined vaccine, 13 valent pneumococcal combined vaccine, Recombinant hepatitis B vaccine, etc.

2.2 Institution in charge of clinical trial

2.2.1 Responsibilities

The Institution in charge of clinical trials is Hebei Provincial Center for Disease Control and Prevention. Its responsibilities are:

-Participate in making the clinical trial protocol and required forms and cards;

-Participate in the drafting of informed consent form for vaccination, preparation of SOP for clinical trial on-site operation, and application for approval of Ethics Committee; be responsible for organizing and implementing the selection of clinical trial site that meets the requirements of GCP, organizing the evaluation of the trial site, and filing in the "Drug clinical trial organization filing management information platform" of the State Drug Administration;

-Organize the implementation of clinical trials, and control the quality of the implementation process of clinical trials;

-To be responsible for guiding the site to report the serious adverse events (SAEs) occurred during the clinical trial to the provincial drug administration, as well as the sponsor and the ethics committee in a timely manner, and to carry out investigation and disposal of SAEs;

-Participate in database locking and save the locked database backup for inspection;

-Responsible for reporting the progress of clinical trial implementation to relevant administrative departments, and writing the clinical trial summary report.

2.2.2 Profile

Hebei Provincial Center for Disease Control and Prevention (CDC) is a public welfare institution directly under the health and Health Commission of Hebei Province, which was jointly established by the former provincial health and epidemic prevention station, Provincial Institute of endemic disease control, Provincial Institute of occupational disease control, Provincial Institute of radiological health, Provincial Academy of Medical Sciences and Provincial tuberculosis control center in August, 2001. Hebei COC is the provincial technical guidance center for disease prevention, with adding four function fuctions inculding Hebei health testing center, Hebei Academy of Medical Sciences, Hebei Institute of occupational disease prevention and control, and National food safety risk monitoring of Hebei center.

The Hebei CDC currently has 40 institutes (Departments and Offices), including 14 administrative and management offices and 26 departments. The authorized staff member is 558, and there are 472 employees at present, including 144 with master's degree or above, accounting for 30.5% of the total number of employees, 159 with senior titles, accounting for 44.28% of the total number of employees, 22 provincial management experts, provincial young and middle-aged experts, and personnel with special government subsidies, and 4 provincial model workers.

The center has two provincial key medical disciplines, including cardiovascular and cerebrovascular disease prevention and control discipline and food safety risk monitoring laboratory. It is the teaching base of preventive medicine in four universities, including Hebei Medical University, Hebei University, North China University of Science and Technology, and Shanxi Medical University, which undertakes the teaching work of about 30-50 students every year. The Hebei CDC has undertaken more than 250 scientific research projects, including 11 national

science and technology major projects, 30 national cooperation projects and 2 international cooperation projects. It has won more than 110 scientific research awards at department level and above, and 25 of them have won provincial and ministerial level science and technology progress awards.

Since 2008, Hebei CDC began to carry out vaccine clinical trials according to GCP standards. Up to now and it has undertaken 28 clinical trial projects (19 have been completed), and established a relatively complete vaccine clinical trial management and quality control system. Since the implementation of *The Guiding Principles for Quality Management of Vaccine Clinical Trials (Trial)*, the center has adjusted in accordance with relevant requirements. On November 15, 2013, the vaccine clinical trial institution was established, and on November 25, 2013, a specialized management department of vaccine clinical trials, the Vaccine Clinical Research Institute, was established to be specifically responsible for the organization, management and implementation of vaccine clinical trials. There are 8 professionals in Vaccine Clinical Research Institute , including 2 chief doctors and 7 master's degrees. All personnel have received GCP training organized by NMPA system.

2.3 Study Site Institution

2.3.1 Responsibilities

The clinical study site is Renqiu City and the study site institute is Renqiu CDC. Its responsibilities are:
-Cooperate in the evaluation and filing of the test site;
-Organize the personnel with corresponding professional technology and clinical research experience to participate in the work of the research site, and all participants should thoroughly read and understand the contents of the study protocol, and strictly follow the protocol to ensure that there is sufficient time to complete the clinical study within the protocol-specified period;
-Organize on-site implementation, including the organization and selection of the subjects, obtaining the informed consent signed by the subjects, screening and enrollment, vaccination, safety visit, blood sample collection, serum separation, sample freezing and submission, etc;
-Be responsible for data input to ensure that all the collected data are true, accurate, complete and legal;
-Accept the supervision and audit of the CRA or inspector designated by the sponsor and the inspection of drug regulatory anthorities to ensure the quality of clinical research;
-Ensure that the subjects get proper treatment in case of adverse reactions / events during the study period. In case of serious adverse reactions/events, handle and report immediately according to the relevant operation procedures.
-Be responsible for the storage of relevant clinical trial data during the clinical trial.

2.3.2 Profile

Renqiu CDC is located in the North Road of Taishan, adjacent to Zhonghua Road in Renqiu city, covering an area of 10,000 m^2 with a building area of 3800 m^2. Renqiu CDC has 10 departments, 74 staff, 3 deputy chief doctors, 17 with intermediate technical titles, 27 with primary technical titles, 17 doctors, 6 assistant doctors and 9 nurses. It possessed provincial screening laboratory for HIV / AIDS confirmation, P2 laboratory and precise instruments and equipment including biosafety cabinet, autoclave sterilizer, constant temperature incubator and benchtop.

2.4 Sample Testing Institute

2.4.1 Responsibilities

National Institute of Food and Drug Control (NIFDC), its repsonsibities are:
-Be responsible for the detection of serum neutralizing antibody.

2.4.2 Profile

NIFDC is an institution directly under the National Medical Products Administration. It is the legal institution and the highest technical arbitration institution for the quality inspect of pharmaceutical biological products. It is also the designated "World Health Organization (WHO) drug quality assurance cooperation center". According to the regulatory requirements, the NIFDC perform the inspection for registration, import, supervision, safety evaluation, and batch release of biological products of multi-field products including drugs, biological products, medical devices, foods, health foods, cosmetics, laboratory animals, packaging materials, etc. Additionally, the NIFDC is responsible for the national research, distribution and management of the drug, medical device standard substances and the bacterial strains for production verification, and carry out relevant technical research work.

2.5 Monotroing Institution

2.5.1 Responsibilities

The Clinical Research Department of Sinovac Biotech Co., Ltd. is responsible for the supervision of clinical trials.
-Carry out clinical trial supervision according to GCP, protocol and SOP;
-Assist the sponsor to undertake the screening, training of the study institution, and hold the kick-off meeting;
-Check the trial process and progress;
-Check the signing of informed consent;
-Check the qualification of investigators and effectiveness of the implementation equipment;
-Check the transportation, storage, distribution, use, return and treatment of clinical trial vaccine;
-Check the collection, storage and transportation of biological samples;
-Check the handling of adverse events;
-Check the logicality of original records and report documents;
-Complete the monitoring work after the study completion, etc.

2.5.2 Profile

Since its establishment in 2002, Clinical Research Department of Sinovac Biotech Co., Ltd. has independently conducted the organization, implementation, monitoring, data management and statistical analysis of multiple studies such as inactivated hepatitis A vaccine, combined hepatitis A and hepatitis B vaccine, SARS vaccine, influenza A vaccine, H5N1 vaccine, EV71 vaccine, 23 valent pneumococcal vaccine, varicella vaccine, inactivated polio vaccine and quardrivalent influenza vaccine, and has rich experiences in clinical trial organization, implementation and management.

2.6 Data Management

2.6.1 Responsibilities

Meta Clinical Technology Co., Ltd. is responsible for data management of this clinical trial.
-Develop data management plan and data verification plan according to the requirements of the protocol;
-Provide EDC and other related online services;
-Carry out data management in accordance with the *technical guidelines for clinical trial data management* during the trial, and confirm that all data reports and records are correct and complete;
-Clean up the data, raise questions about the research data, and assist the investigators to verify and clarify;
-Write data management report.

2.6.2 Profile

Meta Clinical Technology Co., Ltd. is a contract research organization (CRO) which mainly undertakes data related service outsourcing business in clinical trials of domestic and foreign pharmaceutical enterprises. It was founded in September 2014. Now, there are offices in Shanghai, Beijing, Xi'an and Shenyang etc. The company has a strategic partnership with Colin Likang, which is a CRO providing comprehensive service. Meta Clinical Technology Co., Ltd.has provided data management, statistical analysis and drug pharmacovigilance services for phase I~IV and

bioequivalence clinical trial of innovative drugs and generic drugs of dozens of domestic and foreign pharmaceutical companies. It has:
-Standard operating procedure (SOP) and strict quality management system that meet the requirements of ICH-GCP, FDA 21 CFR Part 11, and other international or domestic requirements of clinical trial;
-Personnel who have experiences in clinical trial design, implementation, data management and statistical analysis in China, United States, European Union, Japan, South Korea, etc., and familiar with relevant drug management regulations and implementation rules;
-A complete education and training system.

2.7 Statistics Analysis

2.7.1 Responsibilities

Beijing Key Tech Statistics Technology Co., Ltd. is responsible for statistical analysis of clinical trials.
- Writing the section of randomization, sample size and statistical analysis section if the clinical trial protocol;
- Prepare statistical analysis plan according to clinical trial protocol;
- Impletation of randomization and blinding;
- Carry out statistical analysis according to the proposed statistical analysis plan and write statistical analysis report.

2.7.2 Profile

Beijing Key Tech Statistics Technology Co., Ltd. (hereinafter referred to as "Key Tech") was registered and established in Beijing in August 2017. It is a domestic funded company specializing in clinical trial data management and statistical analysis services. It takes the biostatistics service of clinical research as the core, mainly for the registration of clinical trials, and provides the statistical strategy consultation, statistical design and statistical analysis throughout the whole clinical trial process. At present, Key Tech has established offices in Beijing, Xi'an and Nanning, with 43 employees, mainly graduated from the Fourth Military Medical University, Peking University, Sichuan University and other domestic first-class universities. Among them, at present, there are 21 statisticians / statistical programmers, 18 data managers, 1 quality control personnel and 3 other non business personnel in the on-the-job employees; according to the education background distribution, there are 3 doctors, 6 masters and 34 undergraduates.

Since its establishment, Key Tech has assisted the applicants to obtain 8 clinical trial approvals, completed 18 new drug applications, including 5 new biological products of class I, and 5 products already approved for marketing, including the first 13 valent pneumonia vaccine, first nasal spray influenza vaccine, the second adamutumab product, the third quadrivalen influenza vaccine and varicella vaccine in China.In 2019, Key Tech signed an agreement with Abbott on statistical consulting services in the Asia Pacific region, and established a long-term partnership with domestic and foreign major innovative pharmaceutical enterprises.

2.8 Data Monitoring Committee

The data monitoring committee is composed of experts in clinical medicine, epidemiology and statistics. Its main responsibilities are:
- Be responsible for reviewing safety data and conducting risk assessment of clinical trials to ensure the safety of the trials.

3 Background and Principle

3.1 Summary

Since December 8, 2019, Hubei Province has reported several cases of unexplained pneumonia, most of whom work or live in the South China seafood market where live animal sales exist. The early stage of pneumonia presents severe symptoms of acute respiratory infection, and some patients develop rapidly into acute respiratory distress syndrome (ARDS). The pneumonia was confirmed to be human to human transmission, and the epidemic escalated rapidly in

early January. There were cases in all provinces of China, Japan, Singapore, the United States and more than 20 countries. A novel coronavirus was detected in the throat swab samples of patients in January 7, 2020 by the China Center for Disease Control and Prevention (CDC). The novel coronavirus pneumonia epidemic was declared as a public health emergency in January 31, 2020 by WHO. In March 12, 2020, WHO declared the epidemic entered the international pandemic stage.

The novel coronavirus gene sequences are most closely related to the two SARS like coronavirus (bat-SL-CoVZC45 and bat-SL-CoVZXC21) [6] derived from bat. The International Committee on Taxonomy of Viruses (ICTV) announced that the official classification name of this noval coronavirus was Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) in February 12, 2020, and the World Health Organization announced on the samen day that the official name of the disease caused by the virus was COVID-19.

3.2 Virology

Coronavirus (COV) is an important pathogen of human and vertebrate. It can infect respiratory tract, gastrointestinal tract, liver and central nervous system of human, livestock, birds, bats, mice and many other wild animals. Since the outbreak of severe acute respiratory syndrome (SARS) in 2003 and the outbreak of Middle East respiratory syndrome (MERS) in 2012, the possibility of CoVs spreading from animals to humans has been proved. CoVs belong to the coronavirinae family of Nidovirales coronavirus family, which includes four genera: α-coronavirus, β- oronavirus, γ-coronavirus and δ-coronavirus [7].

SARS-CoV-2, enveloped, with a diameter of 60-140nm, harbors a linear single-stranded positive sense RNA genome, encoding 4 structural proteins. The genetic characteristics of SARSr-COV and MERSr-COV are significantly different. At present, the homology with bat-SL-COVZC45 is more than 85%. In vitro, the virus could be detected in respiratory epithelial cells in 96 hours, while in Vero E6 and Hun-7 cell lines, it takes about 6 days.

So far, the whole genome sequences of virus are comparable, showing that there is no obvious mutation in the virus. Close monitoring on novel coronavirus also indicated no significant variation existed, from virus isolated from the environment or from early-stage patients or from recent patients [8]. However, novel coronavirus is a positive-strand RNA, hence, mutation and recombination are still possible in the future which would increase or decrease the virulent.

The understanding of the physical and chemical characteristics of coronavirus mostly comes from the literatures of SARS-CoV and MERS-COV. It is sensitive to UV and heat, and it could be inactivated under the condition ether of 56 ℃ for 30 minutes or 75% ethanol, chlorine containing disinfectant, etc. While chlorhexidine cannot effectively inactivate the virus [9].

3.3 Clinical Manifestations

Based on the current epidemiological survey, the latent period of the COVID-19 is from 1 to 14 days, mostly 3 to 7 days. Fever, fatigue and cough are the main manifestations. A small number of patients with nasal obstruction, runny nose, sore throat, myalgia and diarrhoea. Severe patients arise with dyspnoea and/ or hypoxemia one week after the onset of the disease, among which could rapidly progress to acute respiratory distress syndrome, septic shock, metabolic acidosis and coagulation dysfunction. What is noteworthy is that patients with severe or critical condition may be accompany with moderate to low fever, or even without fever.

Atypical symptoms, such as vomiting, diarrhoea, weakness and short breath arise in children and new-borns, moreover, patients with mild manifestation were accompany with symptoms of fever, slight fatigue, without pneumonia. Most patients have a good prognosis, while few of them are in a critical condition. Patient were elderly or with chronic diseases have poor prognosis. The clinical manifestations between pregnant women and non-pregnant were similar[9].

3.4 Epidemiologic Feature

Transmission route and Susceptible population

COVID-19 patients are the main source of infection, and asymptomatic patients may also be contagious. Respiratory secretions through droplets and intimate contact contributed to person-to-person transmission. The virus is transmitted through the droplets produced by patients' coughing, sneezing and talking. The susceptible people are generally susceptible to infection after inhalation. Aerosol transmission is possible if under a relatively closed environment for long time.

The fecal-oral route remains to be determined. Recently, novel coronavirus was detected in faces of patients diagnosed in Wuhan, Shenzhen and even the United States. It indicated that the virus could be duplicated and existed in the digestive tract, suggesting the possibility of fecal-oral route transmission[10] However, the possibility of transmission through intake food contaminated by virus is still undetermined. Others pointed out that aerosol transmission is possible through feces droplets, while further investigation still needed.

It's reported that new-borns, whose delivered by positive pregnant patients, were diagnosed as positive 30 hours after birth, which indicated possibility of maternal-neonatal transmission [11].

Epidemic situation of COVID-19 in China

As of 10:00 on April 27st, 2020 (CEST), there have been 84,341 cases and 4,643deaths in China[12]. *China-WHO Novel Coronavirus Pneumonia (COVID-19) Joint Investigation Report* [13] pointed out that of 55,924 confirmed patients, the median age is 51 years (2 days to 100 years), and the interquartile spacing is 39 to 63 years old. 77.8% of patients are aged between 30 and 69 years old. Among them, 51.1% are male, 77% are from Hubei Province, 21.6% are farmers or manual workers.

In China, person-to-person transmission of novel coronavirus pneumonia occurs mainly in families according to the cluster case investigation and some family transmission studies in several provinces. A total of 1836 reported cases from Guangdong Province and Sichuan Province, of them1,308 patients were reported in 344 clusters, 78%-85% of which occurred in family members. The research for family internal transmission is in progress, but the preliminary results in Guangdong Province estimate that the second attack rate of family members is about 3%-10%. As the epidemic continues, although familial cluster infection dominates, community cluster infection also increases within hospital[13].

Global Epidemic situation of COVID-19

As of 10:00 on April 27st, 2020 (CEST), there have been 2,878,196 cases and 85,530 deaths were reported globally[12]. The countries with high incidence are the United States (931,698confirmed cases in total), Spain (207,634 confirmed cases in total), Italy (197,675 confirmed cases in total), , Germany (155,193 confirmed cases in total), the United Kingdom (152,844 confirmed cases in total), France (123,279 confirmed cases in total) and Turkey (110,130 confirmed cases in total) etc. The outbreak has influenced 209 countrie all around the world, and has caused global COVID-19 pandemic.

3.5 R&D of Vaccines

At present, there is no approved treatment or vaccine for COVID-19 in the world. According to report of WHO, as of April 26th, 2020, a total of 82 candidate vaccines are under preclinical research stage, and 7 candidate vaccines are under clinical trials, including mRNA-1273 from Modern, INO-4800 from Inovi, mRNA vaccine from Pfizer, inactivated vaccine from Wuhan Institute of Biological Products, ChAdOx1-nCOV from University of Oxford, Ad5-nCOV from CanSino Biological Inc. and inactivated vaccine from Sinovac Research & Development Co., Ltd.

4 Preclinical Study and Laboratory Evaluation of Vaccines

4.1 Safety Study

The single dose toxicity study in rats, active systemic anaphylaxis study in guinea pigs, repeated dose toxicity study on rats, repeated dose toxicity study on cynomolgus monkey and reproductive development toxicity study in rats

were carried out for the experimental vaccine. The results are as follows:

4.1.1 Single Dose Toxicity Study on Rats

Objective: To evaluate the acute toxicity of SARS-CoV-2 Vaccine on Sprague-Dawley (SD) rats within 14 days after a single dose, so as to provide toxic data for acute poisoning.

Design: 20 quarantined SD rats with equal gender and weight, were selected and randomized into vaccine group and control group to receive intramuscularly high dosage vaccine (0.5mL/1200SU [SARS-CoV-2 Unite]/rat) or saline 0.5mL/dose. Acute toxicity was observed for 14 days after injection, then perform anatomical observation.

Results: No death or near-death rats was observed in two groups, and also no clinical abnormal reaction was observed. The body weight in each group showed a normal increasing trend, and compared with the negative control animals of the same gender in the same period, there was no statistical difference in the body weight of the animals in the sample group, and there was no significant effect of the drug on the food intake of the animals. Gross anatomical observation shows no abnormality in the main organs and tissues of animals in each group.

Conclusion: when the SD rats were injected with the vaccine in high dose intended for clinical use, no abnormal changes related to drug administration were observed, and the maximum tolerated dose (MTD) of SD rats was greater than or equal to 1200SU/dose.

4.1.2 Active Systemic Anaphylaxis Test in Guinea Pigs

Objective: Observed the rapid active systemic anaphylaxis of guinea pig by sensitization via intramuscular injection of SARS-CoV-2 vaccine (once every two days for three times) and booster at D19/D26 via intravenous injection, provide animal data for the clinical trials of the tested product.

Design: According to the weight of the animal before administration, choose 36 Hartley guinea pigs of similar weight and randomly divide them into 4 groups, the low dosage group, high dosage group, negative control and positive control group, sensitized by vaccine of 0.5mL/1200SU/dose, saline and human hemoglobin. On D1, D3 and D5, the animals were intramuscularly sensitized, on D19 and D26, booster the animals via intravenous injection. The first three animals in each group received booster vaccination via foot vein, the booster dosage is twice of the sensitization dosage. Perform clinical observation after administration, the design of the study is shown in the following table:

Table 1 Design of Active Systemic Anaphylaxis Study in Guinea Pigs

Group	Sample/ Control	Number of animals	Sensitization (i.m) D1, D3, D5		Booster (i.v) D19, D26	
			Dosage	Volume (mL/GP)	Dosage	Volume (mL/GP)
1	Negative control	9	0	0.5	0	1
2	Positive control	9	20 mg/animal	0.5	40 mg/ animal	1
3	Low dose of test sample	9	0.1dose/ animal	0.05	0.2 dose / animal	0.1
4	High dose of test sample	9	1dose/ animal	0.5	2 dose / animal	1

Results: No abnormal reaction observed in regular clinical observation, the weight of animals was weighted before grouping, before last sensitization and before administration on the day of booster, the increase of weight in each group was normal. The anaphylaxis reaction in low dosage group, high dosage group and negative control group were all negative. The positive control showed positive in anaphylaxis after booster on D19 and D26.

Conclusion: No allergic reaction was observed after the guinea pigs was injected with high dosage vaccine intended for clinical use.

4.1.3 Repeated Dose Toxicity Test in Rats

Objective: To evaluate the possible toxicity reactions of SARS-CoV-2 vaccine after repeatded intramuscular injection in SD rats for 4 weeks. The recovery of the target organs of toxicity and the toxicity reactions were determined, and the safe dose of vaccine was determined, so as to provide basic data for clinical trial and clinical application of the investigational products

Triple administration trial design: According to the animal weight measured before grouping, 150 animals with qual ified quarantine and similar body weight were selected and randomly divided into 7 groups according to the gender section, which were used in the main test group (1-4 groups, low-dose group of test sample, high-dose group of test sample, negative control group and adjuvant control group) and satellite group (5-7 groups, low-dose group of test sample, high-dose group of test sample and negative control group). There were 15 animals of each sex in the main experimental group, 15 animals of each sex in the satellite group and 5 animals of each sex in the satellite group. The low-dose group, high-dose group, negative control group and adjuvant control group were treated with 0.5mL/300SU/dose, 0.5mL/1200SU/dose of test samples, 0.5mL/dose of normal saline, 0.5mL/dose of adjuvant respectively. The safety of the drug was observed by intramuscular injection on the 1st, 8th and 15th day, and the recovery period was 2 week.

Four administration test design: 80 SD rats at 5-6 weeks were selected, half male and half female, and randomly divided into two groups according to body weight:Negative control group (CN group) and SARS-CoV-2 vaccine group (T group) with 40 animals in each group, including 30 animals in the main experimental group and 10 animals in the satellite group, were given intramuscular injection once at week 0, 1, 2 and 3 with a dose of 1200SU/0.5ml per animal per time, and the recovery period was 2 weeks.

Triple administration test results: During the experiment, there were no death or near-death rats, no clinical abnormal reaction, no abnormal change in body weight and no abnormalities in body temperature and ophthalmic examination. No abnormalities in blood coagulation indicators, blood biochemical indicators, urine test indicators, T lymphocyte subsets and cytokines related to drug administration were observed. Compared with the rats in negative control group of same gender in the same period, on Day 4, basophils elevated among female rats in the low dosage group. And 3 days post last dose, neutrophils elevated among male rats and eosinophils elevated among female rats in the low dosage group, and neutrophils elevated among male rats in the high dosage group. Combined with the mode and mechanism of the reaction, it's considered that those changes may be related to the immune responses and/or local irritation induced by the test sample. In addition, compared with the rats in negative control group of same gender in the same period, 3 days (Day 18) post last dose, lymphocytes elevated among male rats in the low dosage group, while decreased in adjuvant groups. At the end of the convalescent period (Day 29), reticulocyte decreased among female rats in adjuvant group, monocytes elevated among female rats in the low dosage group. Since all those changes were not significant and only observed in a single gender, which indicated there was no correlation with the dose, it is not considered to have toxicological significance.

Pathological examination showed that there were no regular changes of toxicological significance in organ weight and organ coefficient of toxicological significance, and no obvious abnormal changes were observed in general, Observation under microscope 3 days after administration (D18), granulomatous inflammation was observed in 17/20, 13/20, and 10/20 animals in the adjuvant control group and the low-dose and high-dose groups, respectively, ranging from mild to moderate. This change was considered to be a local reaction caused by the accumulation of aluminum preparation, which is the expected reaction caused by intramuscular injection of vaccine containing aluminum adjuvant.At the end of the 2-week recovery period (D29), granulomatous inflammation was still observed at the injection site in7/10, 6/10 and 6/10animals in the adjuvant control group and the low-dose and high-dose groups, respectively, suggesting that the local irritation reaction of administration had not been recovered

Four administration test results: During the experiment, there were no death or near-death rats, no clinical abnormal reaction, no abnormal change in body weight and no abnormalities in body temperature and ophthalmic examination. At the end of the last administration and the end of the recovery period, no abnormalities in blood coagulation indicators, blood biochemical indicators, urine test indicators, T lymphocyte subsets and cytokines (IL-2, IL-10 and TNF-α) related to drug administration were observed.

Pathological examination showed that at the end of the last administration and the end of the recovery period, there were no regular changes of toxicological significance in organ weight and organ coefficient of toxicological significance, and no obvious abnormal changes were observed in general.During the whole experiment, there no local reactions visible to the naked eye such as hyperaemia, oedema, induration and necrosisoccurred at the injection site of the rats in the administration group. Microscopic observation showed that at the end of the last administration, there were 12 cases (12/20) with Infiltration of inflammatory cells in the muscle stroma at the injection site, one of which was accompanied by fibroblast proliferation. At the end of the recovery period.

Conclusion: The inactivated SARS-COV-2 vaccine were intramuscularly administered to rats for 3 or 4 doses with one-week interval between doses. During the administration period and at the end of the recovery period, no significant systemic toxic reactions were observed in rats with 300SU/dose or 1200SU/dose. No Observed Adverse Effect Level (NOAEL) was considered to be 1200 SU/rat. Irritation reaction which may be related to the aluminum adjuvant was observed at the injection site, two weeks after drug withdrawal, the degree of irritation was partially reversible, and no immune toxic reaction was observed.

4.1.4 Repeated Dose Toxicity Test on Cynomolgus Monkey

Objective: Evaluate the possible toxicity reaction and target organ after repeat dosing in cynomolgus monkeys for 4 weeks via intramuscular injection of SARS-CoV-2 vaccine and the recovery of the toxicity reaction for 4 weeks after vaccination, providing animal data for clinical trials.

Design: According to the weight of animal before grouping, 40 quarantined animals with similar weight were selected and randomized according to gender into 4 groups, which are low dosage, high dosage, negative control and adjuvant control groups. 10 Macaca fascicularis each, half male and half female. The animals in low dosage group, high dosage group, negative control and adjuvant control groups were administered by 0.5mL/300SU/dose vaccine, 0.5mL/1200SU/dose vaccine, 0.5mL/dose saline and 0.5mL adjuvant solution on D0, D7 and D14 intramuscularly. The safety observation is conducted until 14 days after the last administration. The indicators including: clinical observation including allergenic reaction and local irritation, etc., weight/temperature/food consumption/ophthalmic testing, clinicopathologic indicator (blood cell count, coagulation function, bloodchem, urine analysis), immunological indicators (T-lymphocyte subsets, cell factors, C-reaction protein, alexin, antibodies), pathology testing (gross anatomical observation, histopathological examination).

Results: During the experiment, there were no death or near-death rats, no clinical abnormal reaction. No abnormalities were observed in body weight, body temperature electrocardiogram, blood pressure and ophthalmic examination. No abnormalities in clinicopathologic indicator and immunological indicators were observed. Pathological examination was conducted 3 days post administration (Day 18), local granulomatous inflammation was observed in 5/6, 6/6 and 5/6 of animals in adjuvant group, low dosage group and high dosage group, respectively, with pathological changes ranging from mild to moderate. The change was considered to be local reaction induced by aluminum adjuvant, which belongs to the expected reaction induced by intramuscular injection of aluminum-containing vaccine. At the end of the 2-week recovery period (Day 29), local granulomatous inflammation was observed in 3/4, 4/4 and 4/4 of animals in adjuvant group, low dosage group and high dosage group, respectively, indicating that the local irritation reaction of administration had not yet recovered.

Conclusion: During the administration period and at the end of the two-week recovery period, no significant systemic toxic reactions were observed in Macaca fascicularis using 300 SU and 1200 SU, so the No Observed Adverse Effect Level (NOAEL) was considered to be 1200 SU/ Macaca fascicularis. Irritation reaction which may be related to the aluminum adjuvant was observed at the injection site, and no immune toxic reaction was observed.

4.1.5 Reproductive and Development Toxicity Study in Rats

Objective: To evaluate the effect of the SARS-COV-2 vaccine on the fertility of male and female rats, the

development of pregnant / lactating female rats, embryos and fetuses, to understand the effect of the vaccine on teratogenesis and offspring development of rats, and to investigate the antibody level in the blood of embryo or offspring and to provide reference for safe drug use in special populations.

Design: According to the weight of the animal before administration, the animals were randomized into 4 groups according to gender. 28 male rats and 56 female rats were randomized in low dosage group, high dosage group, negative control group and adjuvant group and administered with 0.5mL/300SU/dose vaccine, 0.5mL/1200SU/dose vaccine, 0.5mL/dose saline and 0.5mL adjuvant solution. The male rats were administered three times before mating on D1, D8, D15 and D29 while females were administered three times before mating on D1, D8 and D15. 1 week after last administration of male rates, the males and females were mated. The female rats are administered on GD6 and PND7. ON GD20, 1/2 pregnant mice in each group were caesarean for inspection of the foetus (appearance, viscera, skeleton), the other 1/2 of the pregnant rats had a normal labor and feed until the end of lactation.

Results: After repeated intramuscular injection of SARS-CoV-2 vaccine at the doses of 300SU/mouse or 1200SU/mouse from pre mating to embryo inplantation and delivery in SD rats, there was no effect on the fertility of parental female and male rats, no obvious adverse reactions in pregnant/lactating female rats, no embryo fetal development toxicity and teratogenicity, and no effect on the growth and development of F1 offspring.

4.2 Immunogenicity Study

In order to evaluate inactivated SARS-CoV-2 Vaccine (Vero cell), mice and rats were immunized intraperitoneally and intramuscularly with vaccine of different dosage, and different adsorption methods at different immunization schedules. Blood samples were collected at different time points for the testing of serum neutralizing antibody titer and IgG antibody titer after immunization, to determine the immunogenicity of the vaccine. The formulation, dosage and immune schedule of the vaccine are determined according to the immunogenicity results.

Study Design:

- **Determination of aluminum adsorption and non-aluminum adsorption processes for vaccine**

Two different processes were employed to prepare aluminium-containing SARS-CoV-2 vaccines of 1200 SU/0.5 ml, 600 SU/0.5 ml, 300 SU/0.5 ml and 150 SU/0.5 ml, and aluminium-free SARS-CoV-2 vaccines of 1200 SU/0.5 ml, 600 SU/0.5 ml and 300 SU/0.5 ml. Mice were intraperitoneally immunized by the above vaccines, 10 mice per group, 0.5 ml per mouse. For the mice immunized with one injection, serum was collected on Day 7, Day 14 and Day 21 after immunization; for the mice immunized with two injections on Day 0, 7 and Day 0, 14, serum was collected on Day 14, Day 21 and Day 28, and serum IgG antibody titer was determined separately. Negative animal control was set. Immunogenicity of vaccines prepared by two different processes was compared via a comparison of the antibody titers, the specific study design is shown in the table below:

Table 2 Study design of the comparison between immunogenicity of aluminium-adsorbed and non-aluminium adsorbed SARS-CoV-2 vaccine

Dosage (SU/0.5 ml)	Inactivated SARS-CoV-2 vaccine				Inactivated non-aluminium adsorbed SARS-CoV-2 vaccine			
	Batch No.	One dose	Two doses (D0, 7)	Two doses (D0, 14)	Batch No.	One dose	Two doses (D0, 7)	Two doses (D0, 14)
1200 SU	20200303-1	10	10	10	20200303-5	10	10	10
600 SU	20200303-2	10	10	10	20200303-6	10	10	10
300 SU	20200303-3	10	10	10	20200303-7	10	10	10
150 SU	20200303-4	10	10	10	/	/	/	/

- **Determination of Immunization Dosage and Immunization Schedule**

Mice group: Fifty mice were randomized into five groups to intraperitoneally receive three kinds of emergency schedule and two kinds of routine schedule using four antigen content vaccine of 300SU/0.5mL, 600SU/0.5mL,

1200SU/0.5mL and 2400SU/0.5mL, respectively.

Rats group: Twenty-five rats were randomized into five groups to intraperitoneally receive three kinds of emergency schedule and two kinds of routine schedule using four antigen content vaccine of 300SU/0.5mL, 600SU/0.5mL, 1200SU/0.5mL and 2400SU/0.5mL, respectively.

Details of immunization and blood sample collection are shown in Table 5.

The proposed dose was determined by the analysis of the immune dose, neutralization antibody titer and enzyme labeled antibody titer. The immune schedule was determined by comparing the immune effects of one, two or three doses.

Table 3 StudyDesign of the Immunization Dosage and Immunization Schedule of SARS-COV-2 vaccine

Immunization Schedule	Immunization Schedule	Date of blood sampling	Amount
Emergency schedule	Day 0	Day 7, 14, 21, 28, 35, 42	10 Mice, 5 Rat
	Day 0, Day7	Day 14, 21, 28, 35, 42	10 Mice,5 Rat
	Day 0, Day 3, Day 7	Day 7, 14, 21, 28, 35, 42	10 Mice,5 Rat
Routine Schedule	Day 0, Day 14	Day 21, 28, 35, 42	10 Mice,5 Rat
	Day 0, Day 14, Day 28	Day 35, 42	10 Mice,5 Rat

Study result:

- **Determination of Aluminum adsorption and non aluminum adsorption**

The vaccines containing aluminum adjuvant and the vaccine free from aluminum in mice are able to produce a certain level of novel coronavirus antibody on the 7th day after initial immunization. The vaccine of 1200SU/0.5mL free from aluminium adjuvant with was the same as that of 300SU/0.5mL with aluminum adjuvant. The immunogenicity of the vaccine containing aluminum is better than that of the vaccine without aluminum.

- **Determination of Immunization dosage and Immunzation schedule**

(1) For the same species of animals immunized by different doses via the same procedure, the neutralizing antibody titer was determined at the same blood sampling time point. The results showed that the immunization doses and produced neutralizing antibody titers showed a good dose-response relationship.

(2) For the same species of animals immunized by the same dose via different procedures (one-injection, two-injection and three-injection), the enzyme-labelled antibody titer was determined at the same blood sampling time point. The results showed that the immunization effect of two-injection and three-injection procedures were both not inferior to that of one-injection procedure in mice, and the immunization effect of two-injection and three-injection procedures were both superior to that of one-injection procedure in rats. Because of the short interval between three injections, the immunization effect of two-injection procedure was comparable to that of the three-injection procedure.

(3) For the two-injection immunization procedure of the same dose at different time points (Day 0, 7 and Day 0, 14), the enzyme-labelled antibody level of Day 0, 14 immunization procedure was an order of magnitude higher than that of Day 0, 7 immunization procedure on Day 21, indicating that the interval between two injections should be more than 14 days in clinical trials.

(4) For two-injection immunization by different doses via the same immunization procedure, the neutralizing antibody titers of 1200 SU and 2400 SU are almost the same.

Conclusion: the formulation containing aluminium adjuvant was selected, the expected doses in clinical trials were determined to be 300 SU/dose, 600 SU/dose and 1200 SU/dose, and the immunization procedure was determined to be two injections.

4.3 Study of Virus Challenge

Objective: Use the SARS-Cov-2 to attack animals that have been immunized with the inactivated SARS-COV-2 vaccine according to different immunization procedures and doses to evaluate the animal protective effect of the inactivated SARS-COV-2 vaccine and to evaluate the existence of antibody-mediated infection enhancement (ADE), so as to provide animal data for clinical research and application.

Design: The inactivated SARS-COV-2 vaccine was used to immunize rhesus monkeys according to different immunization procedures and doses, try the virus seed to attack animals 21 to 42 days after the first immunization of the inactivated SARS-COV-2 vaccine,the protective effect of the vaccine was evaluated according to clinical symptoms observation, serum antibody detection and hiopathological examination results of rhesus monkeys, and the presence of ADE under different antibody levels was observed. The study design was shown in the table below:

Table 4 Study Design of Protection Effect of Virus Challenge

Number	Group	Schedule (day)	Dose	Days after the first dose	Pleasant days (After virus challenge)	Number of animals
1	3 doses - vaccine	0,7,14	High dosage (1200SU/0.5ml)	23	7	4
			Miedum dosage (600SU/0.5ml)	22	7	4
2	3 doses - adjuvant	0,7,14	/	21	7	2
3	Model group	/	/	21	7	2
4	2 doses - vaccine	0,14	High dosage (1200SU/0.5ml)	23	7	4
			Miedum dosage (600SU/0.5ml)	22	7	4

Results of two-dose schedule:

In the model group, there was no significant increase in temperature. 2 out of 4 macaques in medium dosage group had high fever over 40℃, and there was no abnormal body temperature in the high dosage group. High viral load was detected in throat swab, anal swab and lung tissues in the model group. Compared with the model group, all of the macaques in medium dosage group was negative in throat swab virus test on Day 3, 5, 7 post virus challenge, all of the macaques were negative in lung tissue virus test on Day 7 post virus challenge, and all of 4 macaques showed mild interstitial pneumonia, suggesting that the medium dosage vaccine had significant protective effect. Compared with the model group, 3 out of 4 macaques in high dosage group was negative in throat swab virus test were negative on Day 3, 5, 7 post virus challenge, all of the macaques were negative in lung tissue virus test on Day 7 post virus challenge and all of 4 macaques showed mild interstitial pneumonia, suggesting that the high dosage vaccine had significant protective effect. The changes of antibody levels in rhesus monkeys in each group were shown in Table 5. According to the results of immune protection of medium dosage group and high dosage group and the level of neutralizing antibody before challenge, it is suggested that the neutralizing antibody titer after 2 doses is greater than or equal to 1:48 had significant protective effect based on the results of immune protection of

Table 5 Changes of antibody levels in rhesus monkeys in each group after administration with the SARS-COV-2 vaccine

	Animal No.	Day 0	Day 7	Day 14	Day 21	Day 3 post virus challenge	Day 5 post virus challenge	Day 7 post virus challenge
Medium dosage group	K21	<8	<8	4	64	64	48	256
	K22	<8	<8	4	128	48	64	128
	K23	<8	<8	6	48	32	96	1024
	K24	<8	<8	32	64	256	128	1024
GMT	/	/	/	7.4	70.8	70.8	78.4	430.5
High dosage group	K17	<8	<8	16	128	1024	512	512
	K18	<8	<8	16	256	256	512	512
	K19	<8	<8	4	96	512	1024	512
	K20	<8	<8	<4	64	192	256	1024
GMT	/	/	/	6.7	119.1	400.7	512.0	608.9

Adjuvant	K9	<8	<8	<4	<4	<4	4	8
group	K10	<8	<8	<4	<4	<4	<4	<8
GMT	/	/	/	/	/	/	/	/
Model	K15	<8	<8	<4	<4	<4	6	12
group	K16	<8	<8	<4	<4	<4	8	8
GMT	/	/	/	/	/	/	6.9	9.8

Results of three-dose schedule:

After the virus challenge, the body temperature of the animals in the model group did not increase significantly, and there was no abnormality in the body temperature of the adjuvant group, the medium-dosage group and the high-dosage group. The WBC count decreased and the percentage of lymphocytes increased after the animals in each group were infected, and there was no statistically significant difference between the medium-dosage group or high-dosage group and the model group. The blood biochemical index values of animals in each group were within the normal range on day 0 and day 14 after immunization, and when the animals were sacrificed. High levels of viral load were detected in throat swabs, anal swads and lung tissues in the model group. Compared with the model group, the average viral load of throat swabs and anal swabs decreased in the medium-dosage group 7 days after challenge. In the high-dosage group, 7 days after challenge, throat swabs and anal swabs tested negative for virus; 3 out of 4 in the middle-dosage group tested negative for virus in the lung tissue 7 days after the challenge, and 4 in the high-dosage group tested negative for virus in the lung tissue 7 days after the challenge.

The neutralizing antibody in the model group and the adjuvant group were both negative and the neutralizing antibody GMT was 61.3 in the medium-dosage group and 50.1 in the high-dosage group 21 days after immunization. 7 days after the challenge, the GMT was 400.7 in the medium-dosage group and 145 in the high-dosage group.

Table 6 Changes of antibody levels in rhesus monkeys in each group after administration with the SARS-COV-2 vaccine

	Animal No.	**Day 0**	**Day 7**	**Day 14**	**Day 21**	**Day 3 post virus challenge**	**Day 5 post virus challenge**	**Day 7 post virus challenge**
Medium dosage group	K5	<8	<8	6	64	32	384	1024
	K6	<8	<8	4	24	32	64	512
	K7	<8	<8	48	384	128	512	768
	K8	<8	<8	6	24	32	64	64
GMT		/	/	9.1	61.3	45.3	168.5	400.7
High dosage group	K1	<8	<8	12	48	24	96	256
	K2	<8	<8	16	64	96	512	384
	K3	<8	<8	6	32	24	48	96
	K4	<8	<8	6	64	16	48	48
GMT		/	/	9.1	50.1	30.7	103.2	145.9
Adjuvant group	K9	<8	<8	<4	<4	<4	4	8
	K10	<8	<8	<4	<4	<4	<4	<8
GMT		/	/	/	/	/	/	/
Model group	K15	<8	<8	<4	<4	<4	6	12
	K16	<8	<8	<4	<4	<4	8	8
GMT		/	/	/	/	/	6.9	9.8

The pathological results of some animals are shown in Figure 1-6.

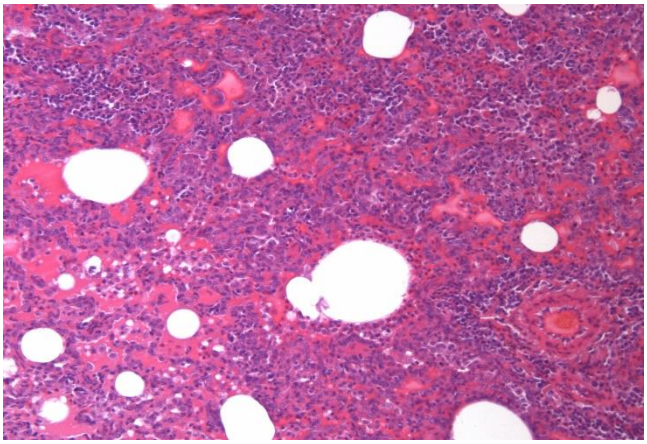

Figure 1 Model group K15 Deputy right lung lobe Severe interstitial pneumonia H.E.×100

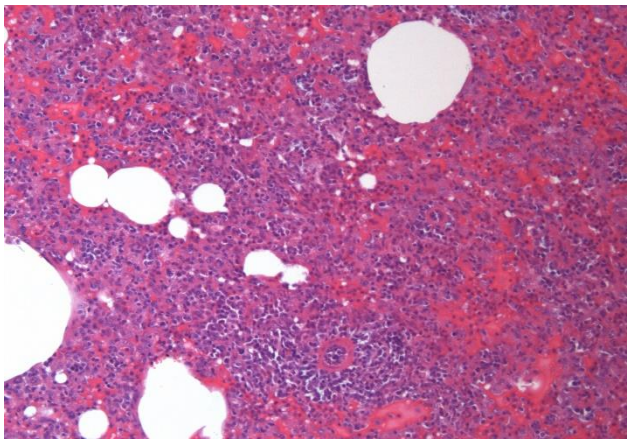

Figure 2 Adjuvant group K10 middle lobe of right lung Severe interstitial pneumonia H.E.×100

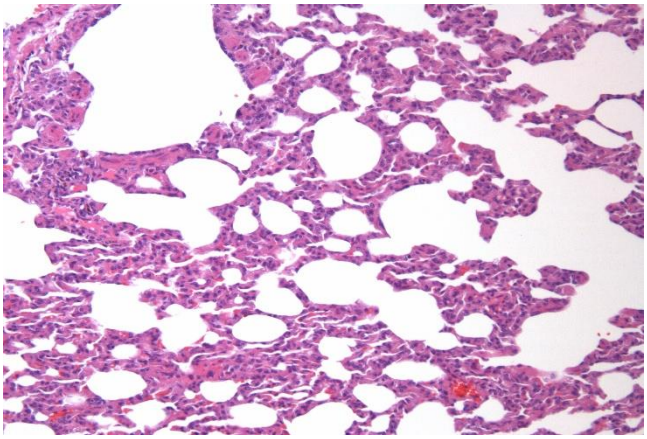

Figure 3 Miedum dosage group K21 superior lobe of right lung mild interstitial pneumoniaH.E.×100

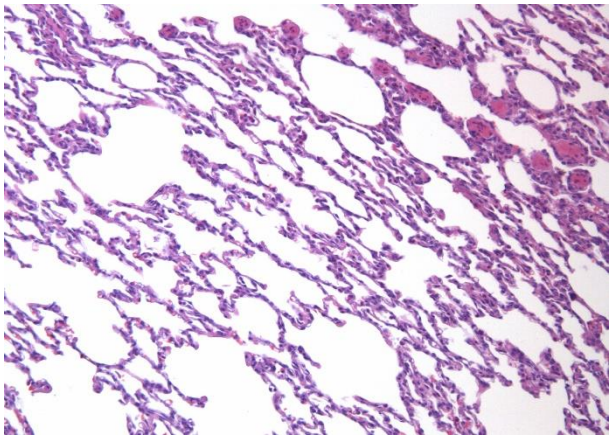

Figure 4 Miedum dosage group K2 superior lobe of left lung nothing abnormal detected H.E.×100

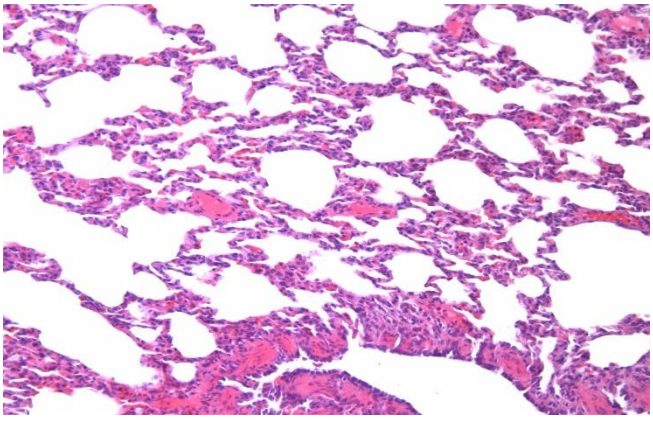

Figure 5 High dosage group K17 inferior lobe of right lung mild interstitial pneumonia H.E.×100

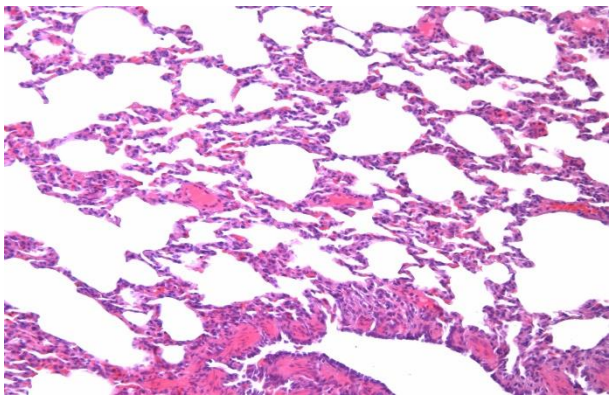

Figure 6 High dosage group K19 inferior lobe of left lung nothing abnormal detected H.E.×100

Conclusion:

The SARS-COV-2 vaccine had significant protective effect in rhesus monkeys. And ADE phenomenon was not observed.

4.4 Study of Cross Neutralization

Objective: To evaluate the cross-neutralizing effect of SARS-CoV-2 immunized serum against different virus strains.

Design: SARS-CoV-2 strain of different time and place of isolation was selected for cross-neutralizing test with SARS-CoV-2 immunized serum from different sources (including serum from convalescent patients and animal serum immunized by inactivated CZ02 strain SARS-CoV-2). The selected SARS-CoV-2 strains are shown in the table below:

Table 7 Basic information of the virus strains

Virus strain	Batch No.	Source
CW01	CW01-W-202002-01	Wuhan (China)
WXY-C3	WXY-202002-01	Zhejiang (China)
CZ01	CZ01-W-202002-01	Zhejiang (China)
CB01	CB01-P5-202002-001	Virus institute (China)
V34	V34-P4（D3）	Military academy of science (China)
CZ12	CZF-202003-01	Zhejiang (China)
CZ30	WGF-202003-01	Zhejiang (China)
HAC	HAC-202003-01	Imported from Italy
HJL	HJL-202003-01	Imported from Italy
QHF	QHF-202003-01	Imported from Spain
SSH	SSH-202003-01	Imported from Switzerland
ZYF	ZYF-202003-01	Imported from Italy

Results: The convalescent serum from patients infected with different sources of novel coronavirus could produce cross-neutralizing reaction to different domestic and foreign coronavirus strains, and the results were basically consistent. The immune serum of novel coronavirus inactivated vaccine prepared with CZ02 novel coronavirus as the candidate strain showed good cross-reaction to different domestic and foreign isolates, which was consistent with the trend of serum reaction in clinically confirmed patients infected with novel coronavirus.

Conclusion: The immune serum of novel coronavirus from different sources can produce cross-neutralizing reaction to novel coronavirus in different situations, and the results of cross-neutralizing reaction is basically consistent.

5 Preliminary Clinical Study

5.1 Phase I/ II clinical trial in adults aged 18-59 years

5.1.1 Safety Assessment

- **Adverse Reactions**

Within 28 days after two doses of the emergency immunization schedule (0,14 days) in 372 subjects of phase Ⅰ/II clinical trial of inactivated SARS-COV-2 vaccine in adults aged 18-59 years, the incidence rates of adverse reactions in the medium dosage group , high dosage group and placebo group were 27.08%,31.94% and15.48% ,respectively, and there was significant statistical difference among groups (P=0.0136), with the high dosage group> medium dosage group> placebo group. Most adverse reactions were mild (grade 1) in intensity, and only one grade 3 adverse reaction occurred in the high-dose group. All adverse reactions occurred within 7 days .The incidence of adverse reactions after the second dose was significantly lower than that after the first dose.The incidence of adverse events was 16.67%, 16.67% and 15.66% in the medium dosage, high dosage and placebo groups, respectively, within 28 days after two doses (0,28 days) of the routine immunization schedule in 371 subjects of Phase Ⅰ/II clinical trial and there was no significant statistical difference in the incidence rates among groups (P=0.9546).Adverse reactions were mainly mild, and there was no grade 3 adverse reactions. Adverse reactions occurred within 7 days after vaccination. The incidence of adverse reactions after the second dose was significantly lower than that after the first dose.

In the three-dose emergency schedule (Day 0,14 and 42) of phase Ⅱ, the incidence rates of adverse reactions were

30.00%, 31.67% and 13.30% in the medium dosage group, high dosage group and placebo group, respectively from the first vaccination to 28 days after the third dose in 150 subjects. There was no significant statistical difference in the incidence rates among groups (P=0.2520). In the three-dose routine schedule (Day 0,28 and 56) of phase II, the incidence rates of adverse reactions were 18.33%, 18.33% and 23.33% in the medium dosage group, high dosage group and placebo group, respectively from the first vaccination to 28 days after the third dose in 150 subjects. There was no significant statistical difference in the incidence rates among groups (P=0.6210). Most adverse reactions were grade 1, and there was no grade 3 adverse reactions occurred. All adverse reactions occurred within 7 days after vaccination. The incidence of adverse reactions after the third dose was significantly lower than that after the first dose and the second dose.

The main symptom of adverse reactions was injection site pain, followed by fatigue. Up to 6 months after the two doses of immunization, a total of 13 serious adverse events occurred in 9 subjects in the phase Ⅰ/II clinical trial, including 3 serious adverse events in 2 subjects in phase Ⅰ clinical trial and 10 serious adverse events in 7 cases in phase Ⅱ clinical trial, respectively, and all were not related to vaccination.

- **Laboratory test index**

The blood routine test, blood biochemical test and urine routine test were carried out before and 3 days after vaccination among all subjects aged 18-59 years in Phase I.

The results showed that the incidence of abnormal laboratory indexes (mainly grade 1) of clinical significance was low. In the emergency immunization schedule, the incidence of clinically significant abnormal laboratory index on the 3th day after each dose of vaccination was 8.33%, 8.33% and 4.17% in the medium dose group, high dose group and placebo group, separately. In the routine immunization schedule, they are 8.33%, 8.33% and 4.35%, respectively. There was no significant difference in the incidence rate among the groups with different immune procedures.

- **Inflammatory factors**

The inflammatory factors of all subjects aged 18-59 years in Phase I were detected before and after immunization. The changes of IL-6, IL-2 and TNF - α in emergency and routine immunization schedules were small, and no significant increase of serum inflammatory factors was found, indicating that the risk of immune pathological reaction induced by vaccine was low.

5.1.2 Immunogenicity Evaluation

5.1.2.1 Immunogenicity results of two-dose schedule

In phase Ⅰ of adults aged 18-59 years, the seroconversion rates of neutralizing antibody to SARS-CoV-2 were 45.83%, 50.00% and 0%, respectively,in the medium dosage group , high dosage group and placebo group at 14 days after two doses accoding to the emergency immunization schedule (Day 0,14), and the GMT (1:1) were 5.6, 7.7 and 2.0, respectively. The seroconversion rates were 83.33%, 79.17% and 4.35% in the medium dosage group, high dosage group and placebo group, respectively, at 28 days after two doses accoding to the routine immunization schedule (Day 0,28), and the GMT were 19.0, 29.6 and 2.2, respectively.

In phase Ⅱ of adults aged 18-59 years, the seroconversion rate of neutralizing antibodies were 92.37%, 98.32% and 3.33%, respectively, in the medium dosage group , high dosage group and placebo group at 14 days days after two doses accoding to the emergency immunization schedule (Day 0,14) (Subject No.: C001-C300), and the GMT were 27.6, 34.5 and 2.3, respectively. The seroconversion rate were 97.44%, 100% and 0% in the medium dosage group, high dosage group and placebo group, respectively at 28 days after two doses according to routine immunization schedule (Day 0,28) (Subjects No: D001-D300), and the GMT of neutralizing antibodies were 44.1, 65.4 and 2.0, respectively. The results showed that the inactivated SARS-COV-2 vaccine had good immunogenicity according to the emergency or routine immunization schedule. The seroconversion rate (98.32%) in high dosage group was slightly higher than that of medium dosage group (92.37%, P=0.0296) 14 days after second dose vaccination of the

emergency schedule. 28 days after the second dose vaccination of the routine schedule, the GMT of neutralizing antibody in high dosage group (65.4) was higher than that of medium dosage group (44.1, P=0.0006). The immunogenicity in medium and high dosage group were comparable given the GMT difference was less than 1.5 fold and the seroconversion rates were all higher than 90%.

The immunogenicity in the phase II was significantly better than that in phase I, showed better immune effect from improved new technology. Analysis the protein composition of purified virus particles showed the S-protein content in vaccine produced with new technology was about twice of it in vaccine produced with old technology. The reason was, compared to cell factory, the bioreactor provides a highly automatic cultivation environment, with dissolved oxygen, pH and CO_2/O_2 under strict control. Based on that, it was assumed that the cultivation environment provided by bioreactor enhanced the S-protein content and improved immunogenicity.

5.1.2.2 Immune persistence of two-dose schedule

In phase Ⅱ, the seroconversion rates of neutralizing antibodies were 16.95%, 24.14% and 0% in the medium dosage group , high dosage group and placebo group at 6 months after the second dose and the GMTs were 4.1, 4.8 and 2.0, respectively, according to two-dose immunization schedule. The seroconversion rates of neutralizing antibodies were 35.19%, 46.43% and 0% in the medium dosage group, high dosage group and placebo group at 6 months after immunization and the GMTs were 6.7, 7.1 and 2.0, respectively. The results showed that neutralizing antibodies dropped to low levels according to different immunization schedule.

5.1.2.3 Immunogenicity results of three-dose schedule

In phase II, the seroconversion rates of neutralizing antibody in the medium dosage group were 94.83%, 93.22% and 98.15% at 14 days after the second dose, 28 days after the second dose and 28 days after the third dose, respectively, and the GMTs were 27.0, 22.2 and 45.8, respectively, according to the three-dose emergency schedule (Day 0, 14, 42); the seroconversion rates in the high dosage group were 98.33%, 98.33% and 98.28%, respectively, at 14 days after the second dose, 28 days after the second dose and 28 days after the third dose and the GMTs were 40.8, 29.1 and 74.2, respectively. The seroconversion rates of neutralizing antibody on the 28th day after the third dose in the medium and high dosage groups were similar, but GMT in the high dosage group was higher than that in the medium dosage group (P=0.0052).

In phase II, the seroconversion rates of neutralizing antibody in the medium dosage group were 94.92% and 98.11% at 28 days after the second dose and 28 days after the third dose, respectively, and the GMTs were 39.6 and 49.7, respectively, according to the three-dose routine schedule (Day 0,28,56). The seroconversion rates in the high dosage group were 100% and 100% t 28 days after the second dose and 28 days after the third dose, respectively, and the GMTs were 58.4 and 51.9, respectively. The seroconversion rates and GMT at 28 days after the third dose in the medium and high dosage groups were similar.

5.1.3 Conclusion

The inactivated SARS-CoV-2 vaccine (CoronaVac) manufactured by Sinovac has good safety and immunogenicity, and can produce antibodies rapidly after two doses according to 0,14 days and 0,28 days.According to CDE's post-marketing requirements for conditional approval of the inactivated SARS-CoV-2 vaccine:"If the results of subsequent clinical trials suggest that the existing immunization schedule and dose are not optimal, further studies should be carried out to optimize the immunization schedule and dose." Given that the neutralizing antibodies at 6 months after the two doses of primary immunization have decreased to a low level,at present, one dose of booster immunization in 6 months after primary immunization has been carried out in population aged 18 to 59 years in phase || clinical trial with different immunization schedule, so as to further explore the immune effect of booster immunization

schedule and provide a basis for the formulation of the optimal immunization strategy.At present, the results of booster immunization in 6 months after primary immunization are not available.

5.2 Phase I/ II clinical trial in elderly aged 60 years and older

5.2.1 Safety Assessment

A total of 421 subjects received at least one dose of vaccine or placebo in the phase I/II clinical trial. In the safety populations from the Phase I and Phase II, the incidence rates of adverse reactions from the beginning of the first dose to 28 days after the second dose in low dosage group, medium dosage group, high dosage group, and placebo group were 20.00%, 20.00%, 21.95% and 20.55%, respectively. There was no significant difference in the overall adverse reactions among the four groups. All adverse reactions were mild and moderate. No grade 3 adverse reactions occurred. Adverse reactions mainly occurred within 7 days after vaccination. The incidence of adverse reactions after the first and second doses of the low-dose and high-dose groups was similar, and the incidence of adverse reactions after the first dose of the medium-dose group and the placebo group were slightly higher than the second dose, there is no obvious trend of increasing or decreasing adverse reactions with the increase of doses. Pain at injection site was the most frequently reported symptoms, with the incidence rates of 11.00%, 11.20%, 8.94% and 4.11%, respectively. The headache and mucocutaneous eruption in high dosage group were slightly higher than that in other three groups. The incidence of hypoesthesia in the placebo group was slightly higher than that of the other three groups, and the differences in other symptoms were not statistically significant.

A total of 7 subjects reported 8 serious adverse events from the beginning of the first dose to 28 days after the second dose , the incidence rates of serious adverse events were 4.00%, 0.80%, 1.63%, and 0.00%, respectively, in the low-dosage, medium dosage, and high dosage and placebo groups ,and all of the serious adverse events were unrelated to vaccination.

5.2.2 Immunogenicity Evaluation

In phase I, the seroconversion rates (≥1:8) in medium dosage group, high dosage group and placebo group were 100.00%, 95.65% and 0.00% respectively 28 days after the second dose vaccination. GMTs (1:) were 54.9, 64.4 and 2.0, and GMTs were 27.5, 32.2 and 1.0, respectively. In phase II, the seroconversion rates (≥1:8) in low dosage group, medium dosage group, high dosage group and placebo group were 90.72%, 97.96%, 98.98% and 0.00% respectively 28 days after the second vaccination. GMTs (1:) were 23.4, 42.2, 49.9 and 2.1, and GMIs were 11.7, 20.9, 24.2 and 1.0, respectively. The results of the study showed that in both phase I and phase II clinical trials, the seroconversion rate reached more than 95% 28 days after the second dose immunization of the medium-dose group and the high-dose group of the experimental vaccine. The medium and high dose groups did not show a dose-response relationship and the results of phase I and phase II were similar. Phase II clinical trials increased the low dosage group. The results of the study showed that the immunogenicity results of the medium and high-dose groups were significantly better than those of the low dosage group, showing a significant dose-effect relationship between those groups.

5.2.3 Conclusion

The inactivated SARS-CoV-2 vaccine (CoronaVac) manufactured by Sinovac has good safety in people aged 60 years and older according to the 0,28 day immunization schedule. It had better immunogenicity in the elderly in the medium dosage and high dosage groups and the results were similar and significantly better than that in low dosage group. The results were similar to that of the phase II clinical trial of the inactivated SARS-CoV-2 vaccine in adults. Based on the research results in the population aged from18 to 59 and the requirements for drug registration certificate (certificate number: 2021S00156), the subjects of this study will receive one dose of booster immunization in 6 months or 1 year after primary immunization, so as to further explore the immune effect of booster immunization

schedules, to provide evidence for the optimal immune strategy.

6 Product Features

6.1 Preparation Process and Formula of Vaccine

Inactivated SARS-CoV-2 Vaccine (Vero Cell), is prepared from novel coronavirus (CZ02 Strain), which is inoculated on African green monkey kidney cells (Vero Cells), then cultured, harvested, inactivated, concentrated, purified and finally aluminium absorbed. The finished vaccine is a milky white suspension liquid, which can be layered due to precipitation and easily dispersed. The main component of the vaccine is the inactivated noval coronavirus (SARS-COV-2), with the excipients of aluminum hydroxide, disodium hydrogen phosphate, sodium dihydrogen phosphate, sodium chloride, etc., and the vaccine is preservative-free. The vaccine is packaged with prefilled syringes or vials, 0.5ml for each container. The vaccine can induce the immunity against the SARS-COV-2, which can prevent the disease caused by the SARS-COV-2 infection.

The investigational vaccine is manufactuted by Sinovac Research & Development Co., Ltd. and tested eligible by National Institute for Food and Drug Control according to *Manufacturing and Quality Control Requirements of Inactivated SARS-CoV-2 Vaccine（Vero Cell) (Draft Version)* . The vaccine is injectable with the specificantion of 0.5mL/container. The antigen content of low, medium and high dosage vaccine is 300SU, 600SU, and 1200SU/0.5mL respectively.

6.2 Stability

Six lots of vaccine products produced in the bioreactor have completed the thermal accelerated stability studies at 25±1℃ for 56 days and 37±1℃ for 42 days. When stored at 25±1℃, the antigen content at each time point including 42 days meets the quality standard after disintegration,the antigen content is lower than the quality standard after 56 days of disintegration,, and the study will be terminated.When stored at 37±1℃, the antigen content at each time point including 28 days meets the quality standard after disintegration. The antigen content of some lots is less than 50% of the labeled amount after disintegration at the monitoring point on 42 days, and the test will be terminated.
3 lots of products at 2-8℃ in the cell factory has been completed the long-term stability observation for 6 months, and the antigen content of each lots products of the inactivated SARS-CoV-2 vaccine did not significantly decrease after disintegration.Among the 9 lots products under the bioreactor process, 3 lots have completed the long-term stability observation for 6 months with no significant changes in the data of each test item have been found.The other 6 lots have completed the long-term stability observation for 3 months, and the antigen content of each lot products of the inactivated SARS-CoV-2 vaccine has not decreased significantly after disintegration.
The validity of the vaccine was tentatively set at 2-8℃ for storage for 2 years according to the results of accelerated stability.

6.3 Control Vaccine

In this study, placebo produced by Sinovac Research & Development Co., Ltd. was adpopted as the control. The placebo is aluminum hydroxide diluent with trace of milkly white precipitation. The appearance is consistent with the investigational vaccine.

It is tested eligible by National Institute for Food and Drug Control according to the *Manufacturing and Quality Control Requirements of Inactivated SARS-CoV-2 Vaccine（Vero Cell）(Draft Version)*. The vaccine is injectable with the specification of 0.5mL/container. It contains no SARS-CoV-2 antigen.

6.4 Storage and Transportation

Vaccines should be stored and transported at 2~8℃, preventing from light.

6.5 Administration Rout and Schedule

Eligible subjects are intramuscularly injected at the lateral deltoid muscle of the upper arm, with a single dose of 0.5ml investigational vaccine or control vaccine,twodoses of primary immunization schedule will be given at 0,28 days, respectively, one dose of booster immunization will be given 6 months or 1 year after the second dose (1 year for the phase Ⅰ clinical tria, 6 months for the phase II clinical trial 0.5ml per dose ,the vaccine should be shaken well before inoculation.

6.6 Information of Investigational Vaccine

The information of Investigational vaccine is as below:

Table 8 Information of Investigational Vaccine

Group	Name	Pacakge	Antigen content	Manufacturer	Batch number	Expiration date
Low dosage vaccine	Inactivated SARS-CoV-2 Vaccine (Vero Cell)	Pre-filled syringe	300SU/ 0.5ml	Sinovac (R&D)	20200307	2023.03.20
Medium dosage vaccine	Inactivated SARS-CoV-2 Vaccine (Vero Cell)	Pre-filled syringe	600SU/ 0.5ml	Sinovac (R&D)	20200412	2023.04.08
High dosage vaccine	Inactivated SARS-CoV-2 Vaccine (Vero Cell)	Pre-filled syringe	1200SU/ 0.5ml	Sinovac (R&D)	20200411	2023.04.07
Placebo	Aluminum hydroxide diluent	Pre-filled syringe	0SU/ 0.5ml	Sinovac (R&D)	2020022801	2023.02.27

6.7 Package

The vaccine will be packed in a box with a label. The label of the vaccine is shown below. See "8.4 randomization and blinding" for the vaccine numbering rules on the label.

Phase I/II of Clinical Trial of SARS-CoV-2 Vaccine (Vero Cell), Inactivated
PRO-nCOV-1002
E001
Only for clinical study, stored at 2-8℃
Expiration date:

The box diagram is as below:

Phase I/II of Clinical Trial of SARS-CoV-2 Vaccine (Vero Cell), Inactivated
PRO-nCOV-1002
Serial No.:
Only for clinical study, stored at 2-8℃
Expiration date:

7 Objective

To evaluate the safety and immunogenicity of inactivated SARS-CoV-2 Vaccine (Vero cell) in older adults aged ≥60 years old.

7.1 Phase I Clinical Trial

To evaluate the safety, tolerance and preliminary immunogenicity of different dosage vaccine in older adults aged ≥60 years old.

7.2 Phase II Clinical trial

To evaluate the safety and preliminary immunogenicity of different dosage vaccine in older adults aged ≥60 years old so as to determine the appropriate dosage for further clinical evalution.

8 Design

8.1.1 Overall Design

Randomized, double blind and placebo control clincal trial.

8.1.2 Sample Size Considerations

Phase I Clinical trial: according to the requirements of the *Good Clincial Practic* and *Provisions for Drug Registration*, phase I clinical trial is a small-scale study with a sample size of 20-30 people to preliminarily evaluate the safety. The total sample size of phase I are 72 subjects, with 48 sujects receiving medium or high dosage investigational vaccine. The sample size meets the requirements of phase I clinical trial.

Phase Ⅱ Clincal trial: according to the requirments of the *Good Clincial Practic* and *Provisions for Drug Registration*, the phase II clincal trial mainly evaluates the immunogenicity and safety of different dosage vaccine in the targeted population and the sample size is more than 300.
The total number of sujects in phase II is 350, with subjects receiving low, medium, high dosage vaccine and placebo as 100, 100, 100 and 50, respectively. A total of 300 subjects receive the low, medium and high dosage vaccines, which meets the requirement of phase II clinical trial.

8.2 Study Endpoint

8.2.1 Endpoint of Phase I

8.2.1.1 Primary Endpoint

- Incidence of adverse reactions within 28 days after each dose of vaccination.

8.2.1.2 Secondary Endpoint

- Incidence of adverse reactions within 7 days after each dose of vaccination；
- Incidence of SAEs within 6 months after t booster immunization;
- Seroconversion rate, seropositive rate, GMT and GMI of neutralizing antibody on the 28th day after primary immunization；
- Seroconversion rate, seropositive rate, GMT and GMI of neutralizing antibody on the 28th day after the first dose vaccination.

8.2.1.3 Exploratory Endpoint

- Seropositive rate and GMT 6 months and 12 months after primary immunization；
- Seropositive rate, GMT and GMI on 28 days after booster immunization;
- Seropositive rate and GMT 6 months after booster immunization.

8.2.2 Endpoint of Phase II

8.2.2.1 Primary Endpoint

- Seroconversion rate of neutralizing antibody 28 days after two doses of vaccination;

8.2.2.2 Secondary Endpoint

- Seropositive rate, GMT, and GMI of the neutralizing antibody 28 days after two doses of vaccination;

- Incidence of adverse reactions within 28 days after each dose vaccination;
- Incidence of adverse reactions within 7 days after each dose vaccination;
- Incidence of SAEs from the beginning of the vaccination to 12 months after booster immunization.

8.2.2.3 Exploratory Endpoint

- Seropositive rate and GMT of neutralizing antibody 6 months after two doses of vaccination;
- Seropositive rate, GMT, and GMI of the neutralizing antibod at 7 days (or 14 days) and 28 days after booster immunization;
- Seropositive rate and GMT 6 months and 12 months after booster immunization.

8.3 Study Plan

8.3.1 Study Plan of Phase I Clinical Trial

Single centered, randimized, double blinded and placebo controlled clinical trial design is adopted in Phase I. A total of 72 healthy older adults aged≥60 years old are selected as subjects. After informed consent, subjects who pass the physical examination, meet the inclusion criteria and did not meet the exclusion criteria will be enrolled into the study. Enrolled subjects receive two doses of injection according to primary immunization schedule (day 0,28). Subjects are enrolled with a dose-escalation manner, with 36 at medium dosage stage which will run-in first, following by 36 at high dosage stage. The subjects enrolled in each dosage stage will be randomly assigned in a 2:1 ratio to receive vaccine or placebo. The high dosage stage vaccination will start only with the condition that safety observation 0~7 days after the first dose of the medium dosage stage vaccination is finished, and the good safety profiles is confirmed. All enrolled subjects received 1 dose of booster immunization 1 year after primary immunization.

To evaluate the safety, the immediate reactions occur within 30 minutes after each dose of vaccination will be observed on site; the local and systemic solicited adverse events (AEs) occur withi 0~7 days after each dose vaccination, as well as the unsolicited AEs from the beginning of the vaccination to 28 days after the whole schedule vaccination will be collected; additionally, the SAEs from the beginning of the vaccination until 6months after booster immunization will be collected. Venous blood will be collected from all subjects at different time points before and after vaccination for serum neutralizing antibody test, to evaluate the primary immune effect , booster immune effect and immune persistence of the vaccine.

The detailed study plan of phase I clinical trial is shown in the table 6

Table 9 Study Plan of Phase I Clinical Trial

Primary immunization (day)	Booster immunization (day)	Phase	Sample size				Blood sampling time (day)
			Miedum dosage	High dosage	Placebo	Total	
0,28	388	Medium dosage stage	24	-	12	36	0,28,56,208,388,416,568
		High dosage stage	-	24	12	36	
		Total	**24**	**24**	**24**	**72**	

Note: Please refer to "10.1 Visit Plan" for the time window.

8.3.2 Study Plan of Phase II Clinical Trial

Single centered, randomized, double blinded and placebo controlled clinical trial design is adopted. The phase II clinical trial will start only with the condition that safety observation 0~7 days after the first dose of the high dosage stage vaccination in phase I is finished, and the good safety profiles is confirmed by the DMC. A total of 350 healthy older adults aged ≥60 years old are selected as subjects. After informed consent, subjects who pass the physical examination, meet the inclusion criteria and didn't meet the exclusion criteria will be enrolled into the study. Subjects

will receive two doses of injection at the primary immunization schedule of day 0,28. The subjects will be randomly assigned in a 2:2:2:1 ratio to receive the low dosage, medium dosage, high dosage vaccine or placebo. All enrolled subjects received 1 dose of booster immunization 6 months after primary immunization.

The immediate reactions occur within 30 minutes after each dose of vaccination will be observed on site. The local and systemic solicited adverse events (AEs) occur withi 0~7 days after each dose vaccination, as well as the unsolicited AEs from the beginning of the vaccination to 28 days after the whole schedule vaccination will be collected. Additionally, the SAEs from the beginning of the vaccination until 12 months after booster immunization will be collected. Venous blood is collected at different time points before and after the vaccination for serum neutralizing antibody test, to evaluate the primary immune effect, booster immune effect and immune persistence.

The detailed study plan of phase II clinical trial is shown in the table 7.

Table 10Study Plan of of Phase II Clinical Trail

Primary immunization (day)	Booster immunization (day)	Sample Size					Blood sampling time (day)
		Low dosage	Medium dosage	High dosage	Placebo	Total	
0,28	208	100	100	100	50	350	0,56,208,215(or 222)*,236,388,568

* Subjects with study numbers E101 to E275 were tested on the 7th day (Day 215) after booster immunization; Subjects with study numbers E276 to E450 were tested on the 14th day (Day 222) after booster immunization.

Note: Please refer to "10.1 Visit Plan" for the time window.

8.4 Randomization and Blinding

8.4.1 Randomization

In phase I and phase II clinical trial, the blinding code should be generated separately by the randomization statistitian by the method of block randomization using SAS software (version 9.4). The blinding code refers to the list of the correspondence between the random number and the trial products (i.e., vaccine or placebo), which is prepared in duplicate and should be sealed after the completion of the blind coding. The original copy should be kept by the investigator for unblinding of the trial, and the duplicate copy should be kept by the sponsor. In the phase I clinical trial, the vaccine (or placebo) numbers are E001-E072. In the phase II clinical trial, the vaccine (or placebo) number numbers are E101-E450.

The blinding code of the backup vaccine (or placebo) is also generated by the randomization statistian using SAS software (version 9.4). In the phase I clinical trial, the backup vaccine (or placebo) is prepared in a 1:1:1 ratio of medium dosage, high dosage vaccine, and placebo, and the backup vaccine (or placebo) numbers are X001-X012. In phase II clinical trial, the backup vaccine (or placebo) is prepared in a 2:2:2:1 ratio of low dosage, medium dosage, high dosage vaccine, and placebo, and the backup vaccine (or placebo) numbers are Y001-Y056.

In case of the circumstances such as color change and damage of the trial products, the inoculation personnel should report to the person in charge of the site and principle investigator, the initiation procedure of the backup vaccine should be started up, a backup vaccine (or placebo) number should be obtained through the online backup vaccine acquisition system, and the corresponding backup vaccine should be used instead of the problem vaccine.

All trial vaccines and placebos will be pasted with blind labels. See "6.7 vaccine packaging" for label style. The subjects should be inoculated with the vaccine labelled with the number which is in accordance with their study number assigned at the enrolment.

8.4.2 Blinding

In this study, a blind design is adopted, in which the randomization statistician and other personnel who do not participate in the trial will engaged in vaccine (or placebo) blinding, i.e. pasting the printed number label to the

specified location of the vaccine (or placebo), according to the generated blinding code. The whole process of vaccine (or placebo) blinding will be supervised by the randomization statistician. The blinding code should be sealed after the completion of the blind coding. The whole process of blinding must be recorded in writting. Personnel who conduct blinding are forbidden to participate in other relevant work of this clinical trial, and should not disclose the blinding code to any person participating in this clinical trial.

8.4.3 Emergency Unblinding

Except for the blinding, the statistician should prepare emergency envelopes reserved for the potential emergency unblinding. In each envelope, there is a random password which can correspond to any study number, and the actual group of this study number can be disclosed through the online unblinding system. Each random password represents a chance of unblinding, that is to say, only one study number can be unblinded using a certain password, and then it will be invalid, and it is also invalid for the already unblended study number. In this study, three emergency envelopes are prepared in the phase I clinical trial, and 10 emergency envelopes are prepared for the phase II clinical trial. All the emergency envelopes are kept by the personnel in charge of the study site. Sealed status of the emergency envelopes should be checked during the blind audit process.

During the study, if the principle investigator and the sponsor jointly decide it is necessary to unblind in an emergency, the person in charge of the site shall open the emergency letter, log in to the online emergency unblinding system with the random code of unblinding in the envelope and conduct the emergency unblinding following the operation prompts, and make relevant records. Subjects with this study number will discontinue the trial and be treated as dropout, and the principle investigator will record the reason for discontinuation in the case report form (CRF). The opened emergency envelope should be properly kept and returned to the sponsor after the study is completed.

8.4.4 Unblinding Regulations

The phase I and phase II clinical trials will be unblinded according to the following time points: the unblinding will be conducted after the serum antibody test results of the 28th day after the whole schedule vaccination are obtained. The unblinding will be jointly implemented by the sponsor, the principle investigator and the statistical party, and a record of unblinding should be kept. After unblinding, the investigators responsible for the observation and evaluation of the subjects and the CRAs responsible for the source data verification should be kept blind until the database is finally locked.

8.4.5 Flow Chart

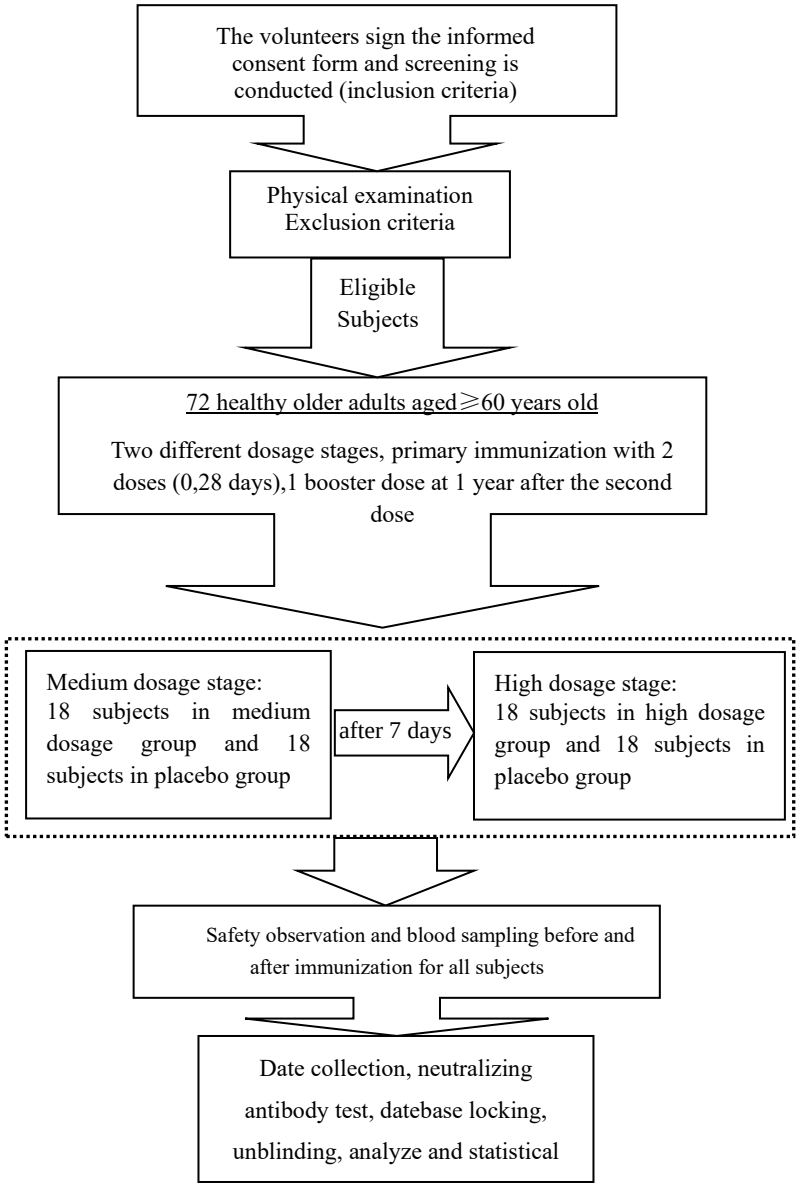

Figure 7 Flow Chart of Phase I Clinical Trial of Inactivated SARS-COV-2 Vaccine (Vero cell)

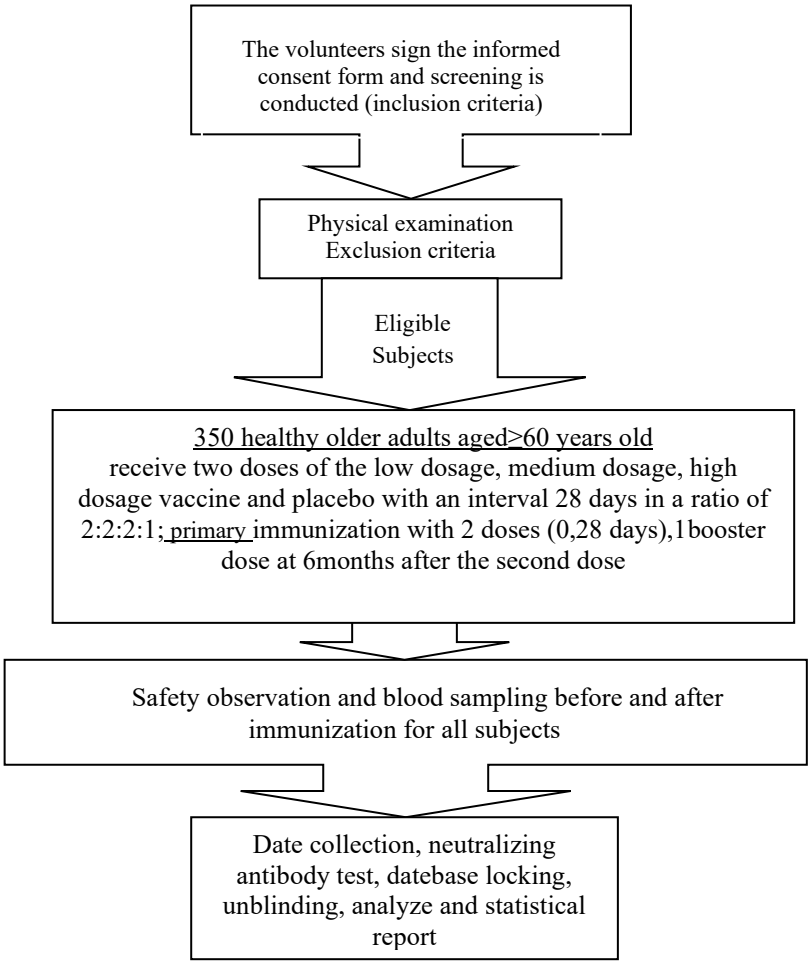

Figure 8 Flow chart of clinical trial phase II of Inactivated SARS-COV-2 vaccine (Vero cell)

8.5 Study Time

8.5.1 Duration of Clinical Trial

Clinical trial is estimated that the study is 22 months

8.5.2 Estimated Time for Subjects to Participate in the Trial

It is estimated that the maximum study duration is 20 months for each subject.

8.6 Trial Suspension and Early Termination

Criteria of trial suspension:

- One or more than one case of the grade 4 adverse events related to vaccination (local, systmic) occur;
- More than 15% of the subjects have grade 3 and above adverse events related to vaccination , including local reaction, systemic reaction and vital signs.

Early Termination Criteria of the Trial:

- After the clinical trial is suspended, the principle investigator and sponsor will jointly discuss and decide whether to early terminate the trial;
- The sponsor requests to fully terminate the trial and has explained reasons;
- The ethics committee requests to fully terminate the trial and has explained reasons;
- The administrative departments require to the fully terminate the trial and has explained reasons.

8.7 Protocal Violation and Deviation

Refers to any change and non-compliance with the clinical trial protocol design or process. The behavior that does not affect the rights and interests, safety and benefits of the subjects, or the integrity, accuracy and reliability of the data as well as the safety or primary indicatiors belong to the protocol deviation; those that affect the rights and interests, safety and benefits of the subjects, or the integrity, accuracy and reliability of the data and the safety or primary indicatiors belong to serious protocol deviation (i.e. protocol violate).

For the protocal deviation / violation during the study, the on-site investigators shall report the fact, process, causes and impact of the incident to the research responsible institution. The principle investigator shall give opinions on the handling of the incident. The protocol deviation / violation report shall be submitted to the Ethics Committee for review and approval.

The investigators should carry out targeted training for relevant staff in the related links of the violation of the protocol to prevent the recurrence of similar incidents, and record the training process.

9 Study Population

9.1 Inclusion Criteria

(1) Healthy subjects aged ≥60 years old;
(2) Be able to understand and sign the informed consent voluntarily;
(3) Provide legal identification.

9.2 Exclusion Criteria

(1) Travel / residence history of Wuhan city and surrounding areas or other communities with case reports within 14 days prior to the entry;
(2) Contact with SARS-CoV-2 infected persons (positive for nucleic acid detection) within 14 days prior to the netry;
(3) Contact patients with fever or respiratory symptoms from Wuhan city and surrounding areas, or from communities with case reports within 14 days prior to the entry;
(4) Two or more cases of fever and / or respiratory symptoms in a small cantact area of subjects, such as family, office, school class or other places within 14 days prior to the entry;

(5) History of SARS;
(6) History of SARS-CoV-2 infection;
(7) History of asthma, allergy to vaccines or vaccine ingredients, and serious adverse reactions to vaccines, such as urticaria, dyspnea, angioneuroedema;
(8) Congenital malformation or developmental disorder, genetic defect, severe malnutrition, etc;
(9) Autoimmune disease or immunodeficiency / immunosuppression;
(10) Serious chronic disease, serious cardiovascular disease, hypertension and diabetes that cannot be controlled by drugs, hepatorenal disease, malignant tumor, etc;
(11) Serious nervous system disease (epilepsy, convulsion or convulsion) or psychosis;
(12) Thyroid disease or history of thyroidectomy, spleenlessness, functional spleenlessness, spleenlessness or splenectomy resulting from any condition;
(13) Diagnosed abnormal blood coagulation function (eg, lack of blood coagulation factors, blood coagulopathy, abnormal platelets) or obvious bruising or blood coagulation;
(14) Immunosuppressive therapy, cytotoxic therapy, inhaled corticosteroids (excluding allergic rhinitis corticosteroid spray therapy, acute non-complicated dermatitis superficial corticosteroid therapy) in the past 6 months;

(15) Long history of alcohol or drug abuse;
(16) Receipt of blood products in the past 3 months;
(17) Receipt of other investigational drugs in the past 30 days;
(18) Receipt of attenuated live vaccines in the past 14 days;
(19) Receipt of inactivated or subunit vaccines in the past 7 days;
(20) Acute diseases or acute exacerbation of chronic diseases in the past 7 days;
(21) Axillary temperature >37.0°C;
(22) According to the investigator's judgment, the subject has any other factors that are not suitable for the clinical trial.

9.3 Exclusion Criteria for the Second Dose and the Tird Dose

The subjects who experience any of events in the following (1) to (4) are forbidden to continue vaccination, but they can continue other study steps according to the investigator' judgement. For the subjects who experience any of the events in the following (5) to (6), the investigator will judge whether vaccination will be continued. For the subjects who experience any of the events in the following (7) to (10), the vaccination can be delayed within the protocol-permitted time window.
(1) Similar vaccines other than the investigational vaccines were used durng the study;
(2) Any serious adverse reactions which have a causal relationship with the vaccination;
(3) Severe anaphylaxis or hypersensitivity after vaccination (including urticaria/rash appears within 30 minutes after vaccination)；
(4) Any confirmed or suspected autoimmune disease or immunodeficiency disease, including human immunodeficiency virus (HIV) infection；
(5) Acute or newly onset chronic disease after vaccination；
(6) Other reactions (including severe pain, severe swelling, severe limitation of movement, persistent high fever, severe headache or other systemic or local reactions) judged by the investigators;
(7) Acute diseases occur during vaccination (acute disease means moderate or severe disease with or without fever);
(8) Axillary temperature >37.2°C during vaccination;
(9) Have vaccinated with subunit vaccine or inactivated vaccine within 7 days, immuned with live attenuated vaccine within 14 days;
(10) According to the investigator's judgment, the subject has any other factors that affect vaccination.

9.4 Subject Withdraw and Suspending Criteria

(1) Subjects request to withdraw；

(2) Intolerable adverse events, whether or not related to the investigational product;

(3) Subjects are not allowed to participate in this trial due to their health status;

(4) In case of any abnormal clinical manifestations of the subjects, the researcher should determine whether it is related to the vaccine, and judge whether the subjects suspend the clinical trial;

(5) Any other reasons considered by the investigator.

If the trial vaccine has been inoculated to the subject before suspending, the clinical trial data of the subject will be used for safety analysis. Subjects could not be replaced in the trail. After the subjects who have been vaccinated in the clinical trial withdraw or suspend the trial, the researcher should provide necessary guidance for any clinical situation related to the trial, and follow up until the a definitive diagnosis is obtained, or the health condition stabilizes or recovers.

10 Method and Procedure

10.1 Visit Plan

(1) Phase Ⅰ clinical trial visit Plan

Table 11 Follow-up Visits Schedule in Phase Ⅰ

Visit No.		1	2	3	4	5	6	7	8	9	10
Date of Visit	D-14 ~D0	D0	D8[e]	D28[e]	D36[e]	D56[e]	D208[e]	D388[e]	D395[e]	D416[e]	D568[e]
Preliminary notification, subject enrolment	X										
Informed consent		X						X			
Demographic information		X									
Regular examination		X									
Inclusion/exclusion criteria screening[a]		X		X				X			
Neutralizing antibody test				X[f]		X	X	X		X	X
Vaccination[b]		X		X				X			
Subject self-recording of the safety observation on diary cards[c]		X	X	X	X	X			X	X	
Adverse reaction/event monitoring (including level 3 or higher, SAE) [cd]		X	X	X	X	X	X	X	X	X	X
Records of concomitant use of drug/vaccine [cd]		X	X	X	X	X	X	X	X	X	X

a) Before each dose vaccination, inclusion/exclusion criteria screening is required.

b) Subjects will be observed for 30 minutes on site to determine the situation of adverse events, especially acute allergic reactions, and then followed by regular follow-ups as required.

c) Safety observation includes assessment of adverse reactions/events and temperature measurement. Body temperature should be measured every day within 0~7 days after each dose vaccination and whenever fever is suspected. Safety observation data are required to be recorded in the diary cards after each dose administration. The investigator regularly interviews the subjects to verify and record adverse events and concomitant use of drugs / vaccines.

d) During D56-D388 and D416-D568, only SAE and drug use associated with SAE are collected.

e) See “Visit Plan” for the time window.

Visit Plan：

Visit1—Day 0—eligible subjects enrolled, collect blood, and the first dose administration.

Visit 2—Day 8 after the first dose - verify the safety observations and concomitant use of drug and other vaccine.

Visit 3—Day 28 day (+10 day) after the first dose- verify safety observations and concomitant use of drug and other

vaccine, collect blood and the second dose administration.
Visit 4—Day8 after the second dose- verify the safety observations and concomitant use of drug and other vaccine.
Visit 5—Day28 (+10 day) after the second dose - verify the safety observations and concomitant use of drug and other vaccine, and collect blood.
Visit 5~Visit 6—verify SAE observation, drug use associated with SAE, and other special circumstances.
Visit 6—Day 180 (+30 day) after the second dose-verify SAE observation, drug use associated with SAE, and other special circumstances, and collect blood.
Visit 6~Visit 7—verify SAE observation, drug use associated with SAE, and other special circumstances.
Visit 7—Day 360 (+30 day) after the second dose-verify SAE observation, drug use associated with SAE, and other special circumstances, collect blood, and booster immunization.
Visit 8—Day 8 after booster immunization - verify the safety observations and concomitant use of drug and other vaccine.
Visit 9—Day 28 (+10 day) after booster immunization - verify the safety observations and concomitant use of drug and other vaccine, and collect blood.
Visit 9~Visit10— verify SAE observation, drug use associated with SAE, and other special circumstances.
Visit 10—Day180 (+30 day) after booster immunization - verify SAE observation, drug use associated with SAE, and other special circumstances, and collect blood.

（2）Phase Ⅱ clinical trial visit Plan

Table 12 Follow-up Visits Schedule ofPhase Ⅱ

	Primary immunization							Booster immunization					
Visit No.		1	2	3	4	5	6	7	8	9	10	11	12
Date of Visit	D-14～D0	D0	D8[e]	D28[e]	D36[e]	D56[e]	D208[e]	D208[e]	D215[e]	D222[e,g]	D236[e]	D388[e]	D568[e]
Preliminary notification, subject enrolment	X												
Informed consent		X							X				
Demographic information		X											
Regular examination		X											
Inclusion/exclusion criteria screening[a]		X		X				X					
Neutralizing antibody test		X				X	X		X[f]	X	X	X	X
Vaccination[b]		X		X				X					
Subject self-recording of the safety observation on diary cards[c]		X	X	X	X	X		X	X	X	X		
Adverse reaction/event monitoring (including level 3 or higher, SAE) [cd]		X	X	X	X	X	X	X	X	X	X	X	X
Records of concomitant use of drug/vaccine [cd]		X	X	X	X	X	X	X	X	X	X	X	X

a) Before each dose vaccination, inclusion/exclusion criteria screening is required.
b) Subjects will be observed for 30 minutes on site to determine the situation of adverse events, especially acute allergic reactions, and then followed by regular follow-ups as required.
c)Safety observation includes assessment of adverse reactions/events and temperature measurement. Body temperature should be measured every day within 0~7 days after each dose vaccination and whenever fever is

suspected. Safety observation data are required to be recorded in the diary cards after each dose administration .
The investigator regularly interviews the subjects to verify and record adverse events and concomitant use of drugs / vaccines.
d) During D56-D208 and D236-D568, only SAE and drug use associated with SAE are collected.
e) See "Visit Plan" for the time window.
f) Only applicable to subjects with study numbers E101 to E275.
g) Only subjects with study numbers E276 to E450 were visited.

Visit Plan：
Visit1—Day 0—eligible subjects enrolled, collect blood, and the first dose administration.
Visit 2—Day 8 after the first dose - verify the safety observations and concomitant use of drug and other vaccine.
Visit 3—Day 28 day (+10 day) after the first dose- verify safety observations and concomitant use of drug and other vaccine, collect blood and the second dose administration.
Visit 4—Day 8 day after the second dose- verify the safety observations and concomitant use of drug and other vaccine.
Visit 5—Day 28 (+10 day) after the second dose - verify the safety observations and concomitant use of drug and other vaccine, and collect blood.
Visit 5~Visit 6—verify SAE observation, drug use associated with SAE, and other special circumstances.
Visit 6—Day 180 (+30 day) after the second dose-verify SAE observation, drug use associated with SAE, and other special circumstances, and collect blood.
Visit 7—Day 180 (+90 day) after the second dose-verify SAE observation, drug use associated with SAE, and other special circumstances, collect blood, and booster immunization.
Visit 8—day 7 (+3 day) after booster immunization - verify the safety observations and concomitant use of drug and other vaccine, in addition, collect blood (only for subjects E101 to E275).
Visit 9—Day 14 (+3 day) after booster immunization - verify the safety observations and concomitant use of drug and other vaccine, and collect blood (Only for subjects E276-E450).
Visit10—Day 28 (+10 day) after booster immunization - verify the safety observations and concomitant use of drug and other vaccine, and collect blood.
Visit 10~Visit11 -verify SAE observation, drug use associated with SAE, and other special circumstances.
Visit 11—Day 180(+30 day) after booster immunization - verify SAE observation, drug use associated with SAE, and other special circumstances and collect blood.
Visit 11~Visit12 -verify SAE observation, drug use associated with SAE, and other special circumstances.
Visit12—Day 360 (+30 day) after booster immunization- verify SAE observation, drug use associated with SAE, and other special circumstances and collect blood.

10.2Recruitment and Informed Consent

Recruitment notices will be issued to volunteers who meet the recruitment criteria. The informed consent should be explained to the volunteers in detail. In the premise of voluntary participation, the volunteers and the investigator jointly sign the informed consent in duplicate, and the volunteers keep the copies.

10.3Screening and Random Enrollment

The subjects who meet the inclusion criteria and don't meet the exlusion criteria are eligible to be enrolled into the study. The screening number is ES+ screening sequence number, such as "ES0001". The enrolled subject will be assigned a study number in the order of enrollment. In the phase I clinical trial and the phase II clinical trial, study numbers of subjects are E001-E072 and E101-E450 respectively.

10.4Vaccination

According to the study number of the subject, the vaccinator takes out the corresponding vaccine labelled with the same number and opens the package box, checks the information of label on the syringe, label in the package box,

and label on the outer surface of package box, the vaccination should be carried outs with the condition that information on the three labels are confirmed consistency. After vaccination, the label in the backage box should be removed and pasted on the specific location of the original logbook, simultaneously, the vaccination information should be recorded in the original logbook.
See "8.3 Study plan" for immunization schedules.

10.5Safety Follow-up Observation

Subjects will be observed for 30 minutes on site after each dose of vaccination. Diary cards and contact cards are distributed to subjects to record the adverse events within 0~7 days and 8~28 days respectively. The investigators explain the judgment, measurement, recording, precautions and reporting method of adverse events. Systematic observation is carried out within 7 days after vaccination. Subjects are required to closely observe their own symptoms and vital signs and fill in the diary card every day. The investigators verify the adverse events on the 8th days after vaccination through face-to-face interviews on all subjects (those who do not face-to-face interviews are conducted by telephone), collect diary cards and distribute contact cards to record the adverse events within 8~28 days. The investigators verify the adverse events on the 28th days and collect contact cards.
The subjects are informed to record the adverse events at any time. Acute allergic reactions, serverity level 3 and above adverse events and SAE should be reported to the investigators timely. After the investigators are informed, they should conduct investigation, verification and follow-up until the adverse event is solved, and finally complete the detailed investigation and follow-up records, which should include the following contents:

- Description of adverse events
- Start time and end time of adverse events
- Severity level
- Relevance to vaccination
- Laboratory testing results
- Treatment measures

Timely treatment should be provided with regard to the acute allergic reactions and severity level 3 and above advere events, in order to relieve the sufferings of the subjects as soon as possible; drug treatment and medical treatment during each follow-up should be recorded in detail.

10.6Blood Sample Collection

- **Sample Collection Plan & Numbering Principle**

Blood samples (2.5-3.5ml each time) from subjects are collected before and after immunization according to “10.1 Visit Plan”. The sample numbering rule is “study number + collection serial number”.

- **Sample Management**

All the samples collected on site should be sent to laboratory timely, completing the handover with the laboratory personnel. Serum should be isolated timely and placed in two tubes (i.e. A tube for detection and B tube for backup, the amount of serum in A tube should be no less than 0.5ml). The serum isolation process should be recorded, and the serum should be stored under -20℃ or lower temperature. All the processes of sample handover, serum insolation, and sample preservation should be recorded. Sample submission record should be filled in for all samples, and the temperature control record of the submission process should be kept.

10.7Safety Assessment

10.7.1 Safety Obervation Index

Solicited local adverse events: pain, induration, swelling, redness, rash, pruritus
Solicited systemic adverse events (including vital signs): fever (axillary temperature), acute allergic reaction, skin and mucosa abnormality, diarrhea, anorexia, vomiting, nausea, muscle pain, headache, cough, fatigue.

10.7.2 Definition of Adverse Events/Reactions

The safety of vaccines will be evaluated according to the scope, intensity, and severity of the local adverse events, systemic adverse events as well as abnormality of vital signs, and the correlation of the above events with vaccination. All adverse medical events occurring during the trial (since signing of the informed consent form) should be collected and recorded.

- Adverse events (AE): Adverse medical events that occur after vaccination, but are not necessarily causally related to the trial vaccine.
- Adverse reactions: the adverse events related to the trial vaccination during the vaccination according to the prescribed dose and procedure.
- Serious adverse event (SAE): it refers to the events during the clinical trial that need hospitalization treatment, prolong hospitalization time, disability, affect working ability, endanger life or death, cause congenital malformation, etc.
- Solicitation/non-solicitation adverse events: In this trial, the solicitation period is 0-7 days after each dose of vaccination, and the non-solicitation period is 8-28 days. The solicited adverse events refer to the solicited symptoms occur within the solicitation period, and the unsolicited adverse events refer to the unsolicited symptoms occur within the solicitation period, and any symptoms occur within the non-solicitation period.

10.7.3 Outcome of adverse events

The outcomes of adverse events included: (1) recovered (2) not yet recovered (3) recoverd but has sequela (4) death (5) loss of follow-up/unknown.

10.7.4 Correlation of Adverse Events with Vaccines

The investigators should try their best to explain the adverse events, and assess the possible casual relationship, i.e. the causal relationship between investigational vaccine and alternative causes (e.g. history of underlying disease, concomitant treatment). This applies to all AEs including serious and non-serious ones.

Causality assessment will be determined by the extent to which an event can be reasonably explained in one or more of the following areas:

Reactions with similar nature have been observed for the similar products;

The same event has been reported in the literatures of the drug products of the similar type;

The event appears with vaccination of the investigational vaccine and recurs after re-vaccination of the investigational vaccine.

Causality of AE with vaccination should be assessed by the investigator on the basis of the following questions to determine whether there is a reasonable possibility that the AE is caused by vaccination:

a. Certainly related : there is evidence of vaccination of experimental vaccine; the time sequence of adverse events and vaccination is reasonable; the occurrence of adverse events is more reasonable than other reasons; repeated vaccination is positive; adverse events are consistent with previous knowledge of this or this kind of vaccine..

b. Probably related: there is evidence of vaccination of experimental vaccine; the time sequence of adverse events and vaccination is reasonable. It is more reasonable to explain adverse events by experimental vaccine than by other reasons.

c. Possibly related: there is evidence of vaccination of experimental vaccine; the time sequence of adverse events and vaccination is reasonable. The causes of adverse events can not be excluded from the experimental vaccine, but also may be caused by other reasons.

d. Possible unrelated: there is evidence of vaccination of experimental vaccine; adverse events are more likely to be caused by other reasons; repeated vaccination are negative or uncertain.

e. Definitely unrelated: the subjects do not use the experimental vaccine; or the occurrence of adverse events was illogical with the time sequence of vaccination; or there were other significant reasons that could lead to adverse events

10.7.5 Treatment of Adverse Event

The reactions such as redness, swelling, pain or (and) fever and general discomfort below grade 2 usually can

spontaneously disappear and special treatment is not needed.
The investigators will carry out the investigation and medical follow-up such as disease history, physical examination, necessary laboratory test, and necessary treatment if the subjects experience any adverse events of grade 3 and higher occurred within 28 days after the whole-schedule immunization, until the adverse events are solved. The corresponding investigation records including the symptoms, vital signs, diagnosis, and laboratory test results should be completed.
In case of the serious adverse event, investigator should promptly take the necessary measures and report within 24 hours.
During the study period, subjects with fever and respiratory symptoms such as cough should immediately go to the designated hospital for treatment. The throat swabs and anal swabs should be collected for nucleic acid testing, and CT examination should also be conducted to deterimine whether it is COVID-19. Once a COVID-19 occurs, it will be treated as SAE, especially it should be analysed that whether there is ADE phenomenon.

10.7.6 Reporting of Serious Adverse Events

(1) The study institution should establish the emergency plan for handling of SAE. The investigator should immediately take measures and make records after he/she is informed of SAE.
The investigator should report to the sponsor, the ethics committee and the local provincial drug regulatory authorities within 24 hours after the SAE is informed and submit the subsequent report.
The study institution/ the investigator should timely transfer the latest safety information report related to the clinical trial of the sponsor to the ethics committee. The report object can be adjusted according to existing regulations and the requirements of local regulatory authorities and ethics committees.
The ethics committee should receive the safety information reports such as SAE reports, timely grasp the occurrence and handling situations of SAE of the whole clinical trial, and carry out follow up review of the handling and reporting of SAE in the process of the clinical trial.

(2) When the sponsor receives information on vaccine safety from any source, an analysis and evaluation should be conducted, including the severity, relevance to the investigational vaccine, and whether it is an unexpected event.
During the drug clinical trial, the sponsor should quickly report the suspected unexpected serious adverse reactions (SUSARs) which are considered definitely or suspiciously related to the investigational drug in a maner of case safety report, according to the *Standards and Procedures for Rapid Reporting of Safety Data during Drug Clinical Trials*.

With regard to the fatal or life-threatening SUSARs, the sponsor should report them as soon as possible after beining informed within 7 natural days, and the relevant follow-up information should be reported within the subsequent 8 days (the day on which the sponsor is firstly informed is day 0);
with regard to non-lethal or life-threatening SUSARs, the sponsor should report them as soon as possible within 15 naturak days; For other information indicating serious safety risk, the sponsor should also report them to the national drug evaluation institution, and make a medical and scientific judgment on each situation.

After the initial report, the sponsor should continue to follow up the SAE and submit new information or changes to the previous report in the form of follow-up report within 15 days since the date of obtaining new information. The sponsor should not arbitrarily change the investigator's judgment on the correlation between SAE and vaccine. If the opinions of the sponsor and the investigator are inconsistent, opinions of both parties should be recorded in detail in the repor, and the adverse event should be reported according to higher management requirements.

Under special circumstances, the invesgator and sponsor should promptly provide SAE-related information and safety reports as required by regulatory authorities and ethics committees.

(3) The contact person of sponsor, ethics committee and Hebei Provincial Drug Administration is as follows:

The contact person of Sinovac：Jiayi Wang；Telephone：18518337983；Email:wangjy1755@sinovac.com；Fax:010-82890408.

The contact person of Ethics Committee of Hebei Center for Disease Control and Prevention: Yong doctor；Telephone：0311-86573167；Fax:0311-86573167.

Hebei Provincial Drug Administration: Email:hbfdasae@163.com.

10.7.7 Safety Evaluation Criteria

Solicited local adverse events, systemic adverse events and vital signs: The grading standard of solicited adverse events mainly refer to the *Guiding Principles for Grading Standards of Adverse Events in Clinical Trials of Vaccines for Prevention* (2019) [14] of NMPA. As shown in the table below, solicited adverse events and non-Solicited adverse events with the same symptoms are graded according to the following criteria:

Table 13 Severity Grading Criteria for Local Adverse Events

	Grade 1	Grade 2	Grade 3	Grade 4
Pain	Not affecting or slightly affecting physical activity	Affecting physical activity	Affecting daily life	Loss of basic self-care ability, or hospitalization
Induration*#	Diameter 2.5 to <5 cm or area 6.25 to <25 cm^2 without affecting or slightly affecting daily life	5 to <10 cm in diameter or 25 to <100 cm^2 in area or affecting daily life	Diameter ⩾10 cm or area ⩾100 cm^2 or ulceration or secondary infection or phlebitis or aseptic abscess or wound drainage or seriously affecting daily life	Abscess, exfoliative dermatitis, dermal or deep tissue necrosis
Swelling #	Diameter 2.5 to <5 cm or area 6.25 to <25 cm^2 without affecting or slightly affecting daily life	5 to <10 cm in diameter or 25 to <100 cm^2 in area or affecting daily life	Diameter ⩾10 cm or area ⩾100 cm^2 or ulceration or secondary infection or phlebitis or aseptic abscess or wound drainage or seriously affecting daily life	Abscess, exfoliative dermatitis, dermal or deep tissue necrosis
Redness#	Diameter 2.5 to <5 cm or area 6.25 to <25 cm^2 without affecting or slightly affecting daily life	5 to <10 cm in diameter or 25 to <100 cm^2 in area or affecting daily life	Diameter ⩾10 cm or area ⩾100 cm^2 or ulceration or secondary infection or phlebitis or aseptic abscess or wound drainage or seriously affecting daily life	Abscess, exfoliative dermatitis, dermal or deep tissue necrosis
Rash* #	Diameter 2.5 to <5 cm or area 6.25 to <25 cm^2 without affecting or slightly affecting daily life	5 to <10 cm in diameter or 25 to <100 cm^2 in area or affecting daily life	Diameter ⩾10 cm or area ⩾100 cm^2 or ulceration or secondary infection or phlebitis or aseptic abscess or wound drainage or seriously affecting daily life	Abscess, exfoliative dermatitis, dermal or deep tissue necrosis
Pruritus	Itching at injection site, relieved within 48 hours	Itching at injection site, did not alleviate within 48 h after treatment	Affecting daily life	NA

* In addition to directly measuring the diameter for grading evaluation, sclerosis and rash should also record the progress of measurement results.

The maximum measured diameter or area should be used for induration and swelling, rash and red; evaluation and grading should be based on functional grade and actual measurement results, and higher grading indicators should be selected.

Table 14 Severity Grading Criteria for Systemic Adverse Events

	Grade 1	Grade 2	Grade 3	Grade 4
Acute allergic reaction*	Local urticaria (blisters), no treatment required	Local urticaria need treatment or mild angioedema, no treatment required	Extensive urticaria or angioedema treated or mild bronchospasm	Anaphylactic shock or life-threatening bronchospasm or laryngeal edema
Skin and mucosa abnormality	Erythema/pruritus/color change	Diffuse rash/maculopapular rash/dryness/desquamation	Blister/exudation/desquamation/ulcer	Exfoliative dermatitis involving mucosa, erythema multiforme, or suspected Stevens-Johnsons syndrome
Diarrhea	Mild or transient, 3-4 times/day, abnormal stool, or mild diarrhea lasting less than 1 week	Moderate or persistent, 5-7 times/day, abnormal stool, or diarrhea >1 week	>7 times/day, abnormal stool, or hemorrhagic diarrhea, orthostatic hypotension, electrolyte imbalance, requiring intravenous infusion >2L	Hypotensive shock, hospitalization
Anorexia	Decreased appetite, not affecting food intake	Decreased appetite, reduced food intake, not affecting body weight	Decreased appetite, and significantly reduced body weight	Need intervention (such as gastric tube feeding, parenteral nutrition)
Vomiting	1-2 times/24 hours without affecting activity	3-5 times/24 hours or affecting activity	>6 times within 24 hours or requiring intravenous fluid infusion	Hospitalization or other nutrition routes due to hypotensive shock
Nausea	Transient (<24 hours) or intermittent and basically normal food intake	Persistent nausea leads to reduced food intake (24-48 hours)	Persistent nausea leads to almost no food intake (>48 hours) or requires intravenous fluids	life threatening (e.g., hypotensive shock)
Muscle pain (non-inoculated site)	Does not affect daily activities	Slightly affects daily activities	Severe muscle pain, seriously affects daily activities	Emergency or hospitalization
Headache	Not affecting daily activities, no treatment required	Transient, slightly affecting daily activities, may need treatment or intervention	Seriously affecting daily activities, need treatment or intervention	Intractability, need emergency or hospitalization
Cough	Transient, no treatment required	Persistent cough, effective treatment	Paroxysmal cough, uncontrolled treatment	Emergency or hospitalization
Fatigue	Normal activity is weakened <48 hours, without affecting the activity	Normal activity is weakened by 20%~50%>48 hours, slightly affecting the activity	Normal activity is weakened by >50%, seriously affecting daily activities, unable to work	unable to take care of oneself, emergency or hospitalization
Vital Signs				
Fever, axillary temperature	37.3～＜38.0	38.0～＜38.5	≥38.5	≥39.5, Lasting more than 3 days

* Refers to type I hypersensitivity

For adverse events not covered in the above grading table, the intensity of adverse events was graded according to the following criteria:
Grade 1 (Mild): transient (<48 hours) or mild discomfort; no medical intervention/therapy required.

Grade 2 (Moderate): mild to moderate limitation in activity; some assistance may be needed; no or minimal medical intervention/ therapy required.

Grade 3 (Severe): Marked limitation in activity; some assistance usually required; medical intervention/therapy required, hospitalizations possible.

Grade 4 (Life threatening): Extreme limitation in activity, significant assistance required; significant medical intervention/therapy required, hospitalization or hospice care probable

Grade 5: Death.

10.8Concomitant Drugs and Vaccines

10.8.1 Concomitant Drugs

- In case of adverse events during the trial, necessary drug treatment and medical treatment are allowed.
- In case of serious allergic reactions or life-threatening events, first aid measures should be taken immediately.
- Investigators should record any information of concomitant drugs, including name, dosage form, dosage and administration route, administration time, etc.

10.8.2 Concomitant Vaccines

- Other vaccines should be administered at least 7 days apart after the investigational vaccine administration.
- Subjects can be administered with other vaccine such as rabies vaccinbe and tetanus vaccine against the emergency events during the clinical trial.
- Relevant information of the concomitant vaccine should be recorded in detail, including the vaccine name, administration, vaccination time etc.

10.9Immunogenicity Evaluation

Blood samples collected at different time points were tested for neutralizing antibody, and the seroconversion rate, seropositive rate, GMT and GMI of neutralizing antibody will be calculated.

10.9.1 Evaluation Criteria

The criteria for determining serum antibody positivity are as follows:
- The seropositivity is defined as neutralizing antibody titer is≥1:8

The criteria for determining serum antibody seroconversion are as follows
- The seroconversion is defined as a post-vaccination Nab titer ≥1:8 if seronegative (<1:8) at beseline, or a 4 fold increase of Nab titer if seropositive (≥1:8) at baseline.

10.9.2 Laboratory Test Methods

Microneutralization test method.

10.10 Data Management

10.10.1 Original Materials

The original materials include informed consent form, diary card, original logbook, etc., recording the following basic data:
- Trial name, subject number

- Demographic data
- Inclusion/exclusion criteria
- Vaccination records
- Follow-up date and date of discontinuation of the trial discontinuation date of the subject
- Adverse events/reactions and the corresponding treatment and outcome
- Concomitant medical treatment and other vaccinations

All data should have original records, which should be properly kept by investigaors in a dedicated space. The original data should be archived in the study site, which is the true and complete evidence for the particpantion of the subjects in the clinical trial.

The investigators should carefully, accurately and timely make the original records. All the collected original data should be recoreded on the same day with that of the data collection. Additionally, the raw data should be recorded using the black sign pen, and the mistake record should be crossed out with the correct content being written beside it along with the signature of the modifier, instead of be altered directly.

10.10.2 Case Report Forms (CRF)

"Electronic Data Capture (EDC) System" is adopted to establish the electronic CRF in this trial.

Electronic CRF is used to record the data of clinical trials, which is an important component of clinical trials and study reports. The electronic CRF is required to be inputted according to the system using instructions and CRF filling-in instructions, using the normative language.

All data on the electronic CRF are derived from the original material and are consistent with original data. Any entry, verification, modification, cleaning and quality control processes of electronic CRF data will be recorded in the EDC system. After data cleaning, the principle investigator should confirm the data in each CRF and sign with electronic signature.

Only investigator and approved staff are allowed to access to the EDC system during the trial period.

10.10.3 Data Lock

Final data verification should be carried out after the completion of all the data entry, verification and data cleaning work. The analyzed population, the situation of protocol violation as well as its impact on the analyzed population should be determined according to the assessment indicators, and then the database is locked.

10.10.4 Subject Privacy Protection and Data Utilization Scope

All information concerning the identity of the subject will be kept confidential and the name of the subject will not appear in any publication or report of the study. The study records will be provided to the sponsor representative in the presence of the investigator for the purpose of collecting medical data. In addition, the CRA, auditors, representatives of the Ethics Committee of the Hebei CDC, and representatives of the National Drug Administration (NMPA) can review the original material of subjects related to this study as required, to confirm the accuracy of the data collected in this study. The original data obtained in this study are only for publication of papers or results related to this project.

10.11 Statistical Analysis

10.11.1 Analysis Set

10.11.1.1 Safety Analysis Set (Safety Set, SS)

All randomized subjects who completed at least one vaccination were included in the safety evaluation set. Subjects who are vaccinated with the wrong vaccine will be tansfered to the group of actually administered vaccine according to the ASaT principle (All Subjects as Treated), for the safety evaluation. Safety sets in this study include generl safety set (SS), safety set of each dose. safety set of each dose was carried out according to the actual number of people vaccinated in each dose.

10.11.1.2 Immunogenicity Analysis Set

Phase Ⅰ clinical trials

Full Analysis Set of Primary Immunization (FAS): A population defined according to the principle of intent analysis (ITT), including all the subjects who have been randomized, have received at least one dose of vaccination, have finished blood collection before/after vaccination for at least one time with the corresponding antibody assay results provided. The subjects who are vaccinated with the wrong vaccine will be kept in the originally assigned group according to the ITT principle, for the immunogenicity evaluation.

28 days after 1st dose vaccination per-protocol set（Per-Protocol Set 1，PPS1）: A subset of FAS, including all the subjects who meet the inclusion criteria and don't meet the exclusion criteria, have been randomized, have finished the 1st dose vaccination within the protocol required time window, and have finished the blood collection before and 28 days after 1st dose vaccination with the corresponding antibody assay results provided. Subjects who meet the following conditions are not allowed to enter PPS1:

- Those who violate the study protocol;
- Those who are vaccinated with the wrong vaccine;
- Blood collection on 28 days after 1st dose vaccination beyond the protocol-required time window;
- Use of protocol prohibited vaccines or drugs:
  ① Other investigational or unlicensed products (drugs or vaccines);
  ② Long-term use (lasting more than 14 days) of immunosuppressive or other immunomodulatory drugs (inhaled or topical steroids are allowed);
  ③ Immunoglobulin and/or blood preparations.
- Newly diagnosed autoimmune diseases, including human immunodeficiency virus (HIV) infection;
- Other situations affecting the evaluation of immunogenicity of 28 days after 1st dose vaccination.

28 days after second dose vaccination per-protocol set（Per-Protocol Set 2，PPS2）: A subset of FAS, including all the subjects who meet the inclusion criteria and don't meet the exclusion criteria, have been randomized, have finished the 1st and 2nd dose vaccination within the protocol required time window, and have finished the blood collection before immunization and 28 days after 2nd dose vaccination with the corresponding antibody assay results provided. Subjects who meet the following conditions are not allowed to enter PPS2:

- Those who violate the study protocol;
- Those who are vaccinated with the wrong vaccine;
- 2nd dose vaccination or blood collection on 28 days after 2nd dose vaccination beyond the protocol-required time window;
- Use of protocol prohibited vaccines or drugs:
  ① Other investigational or unlicensed products (drugs or vaccines);
  ② Long-term use (lasting more than 14 days) of immunosuppressive or other immunomodulatory drugs (inhaled or topical steroids are allowed);
  ③ Immunoglobulin and/or blood preparations.
- Newly diagnosed autoimmune diseases, including human immunodeficiency virus (HIV) infection;
- Other situations affecting the evaluation of immunogenicity of 28 days after second dose vaccination.

Immune Persistence Set of Primary Immunization, 6 months after second dose vaccination (IPS-6): includes all the subjects who have received the 1st and 2nd dose vaccination, and blood sample was collected 6 months after the second dose vaccination, and there was an effective antibody titer value.

Immune Persistence Set of Primary Immunization, 12 months after second dose vaccination (IPS-12): includes all the subjects who have received the 1st and 2nd dose vaccination, and blood sample is collected 12 months after the second dose vaccination, and there is an effective antibody titer value.

Full Analysis Set for Booster (bFAS): A population defined according to the principle of intent analysis (ITT), including all the subjects who have been randomized, completed primary immunization, entered the booster immunization process, and have received the booster dose vaccination, and have finished blood collection before booster dose vaccination with the corresponding antibody assay results provided. The subjects who are vaccinated with the wrong vaccine will be kept in the originally assigned group according to the ITT principle, for the immunogenicity evaluation.

28 days after booster dose vaccination per-protocol set（Per-Protocol Set for Booster, bPPS）: A subset of bFAS, including all the subjects who meet the inclusion criteria and don't meet the exclusion criteria, have been randomized, have finished the 1st and 2nd dose vaccination and the booster dose 12 months after the second dose within the protocol required time window, and have finished the blood collection before booster immunization and 28 days after booster dose vaccination with the corresponding antibody assay results provided. Subjects who meet the following conditions are not allowed to enter bPPS:

- Those who violate the study protocol;
- Those who are vaccinated with the wrong vaccine;
- Booster dose vaccination or blood collection on 28 days after booster dose vaccination beyond the protocol-required time window;
- Use of protocol prohibited vaccines or drugs:

  ① Other investigational or unlicensed products (drugs or vaccines);

  ② Long-term use (lasting more than 14 days) of immunosuppressive or other immunomodulatory drugs (inhaled or topical steroids are allowed);

  ③ Immunoglobulin and/or blood preparations.
- Newly diagnosed autoimmune diseases, including human immunodeficiency virus (HIV) infection;
- Other situations affecting the evaluation of immunogenicity of 28 days after booster dose vaccination.

Immune Persistence Set of Booster Immunization, 6 months after booster dose vaccination (bIPS-6): includes all the subjects who have received the 1st and 2nd dose vaccination, completed the booster dose vaccination 1 year after the second dose, and blood sample is collected 6 months after the booster dose vaccination, and there is an effective antibody titer value.

Phase Ⅱ clinical trials

Full Analysis Set of Primary Immunization (FAS): A population defined according to the principle of intent analysis (ITT), including all the subjects who have been randomized, have received at least one dose of vaccination, have finished blood collection before/after vaccination for at least one time with the corresponding antibody assay results provided. The subjects who are vaccinated with the wrong vaccine will be kept in the originally assigned group according to the ITT principle, for the immunogenicity evaluation.

Per-protocol set of Primary Immunization（Per-Protocol Set，PPS）: A subset of FAS, including all the subjects who meet the inclusion criteria and don't meet the exclusion criteria, have been randomized, have finished the 1st and 2nd dose vaccination within the protocol required time window, and have finished the blood collection before immunization and 28 days after 2nd dose vaccination with the corresponding antibody assay results provided. Subjects who meet the following conditions are not allowed to enter PPS:

- Those who violate the study protocol;
- Those who are vaccinated with the wrong vaccine;
- 2nd dose vaccination or blood collection on 28 days after 2nd dose vaccination beyond the protocol-required time window;
- Use of protocol prohibited vaccines or drugs:
  ① Other investigational or unlicensed products (drugs or vaccines);
  ② Long-term use (lasting more than 14 days) of immunosuppressive or other immunomodulatory drugs (inhaled or topical steroids are allowed);
  ③ Immunoglobulin and/or blood preparations.
- Newly diagnosed autoimmune diseases, including human immunodeficiency virus (HIV) infection;
- Other situations affecting the evaluation of immunogenicity of 28 days after second dose vaccination.

Immune Persistence Set of Primary Immunization, 6 months after second dose vaccination (IPS-6): includes all the subjects who have received the 1st and 2nd dose vaccination, and blood sample is collected 6 months after the second dose vaccination, and there is an effective antibody titer value.

Full Analysis Set for Booster (bFAS): A population defined according to the principle of intent analysis (ITT), including all the subjects who have been randomized, completed primary immunization, entered the booster immunization process, and have received the booster dose vaccination, and have finished blood collection before booster dose vaccination with the corresponding antibody assay results provided. The subjects who are vaccinated with the wrong vaccine will be kept in the originally assigned group according to the ITT principle, for the immunogenicity evaluation.

7 days after booster dose vaccination per-protocol set（Per-Protocol Set 1 for Booster, bPPS1）: A subset of bFAS, including all the subjects who meet the inclusion criteria and don't meet the exclusion criteria, have been randomized, have finished the 1st and 2nd dose vaccination and the booster dose 6 months after the second dose within the protocol required time window, and have finished the blood collection before booster immunization and 7 days after booster dose vaccination with the corresponding antibody assay results provided. Subjects who meet the following conditions are not allowed to enter bPPS1:

- Those who violate the study protocol;
- Those who are vaccinated with the wrong vaccine;
- Booster dose vaccination or blood collection on 7 days after booster dose vaccination beyond the protocol-required time window;
- Use of protocol prohibited vaccines or drugs:
  ① Other investigational or unlicensed products (drugs or vaccines);
  ② Long-term use (lasting more than 14 days) of immunosuppressive or other immunomodulatory drugs (inhaled or topical steroids are allowed);
  ③ Immunoglobulin and/or blood preparations.

- Newly diagnosed autoimmune diseases, including human immunodeficiency virus (HIV) infection;
- Other situations affecting the evaluation of immunogenicity of 7 days after booster dose vaccination.

14 days after booster dose vaccination per-protocol set（Per-Protocol Set 2 for Booster, bPPS2）: A subset of bFAS, including all the subjects who meet the inclusion criteria and don't meet the exclusion criteria, have been randomized, have finished the 1st and 2nd dose vaccination and the booster dose 6 months after the second dose within the protocol required time window, and have finished the blood collection before booster immunization and 14 days after booster dose vaccination with the corresponding antibody assay results provided. Subjects who meet the following conditions are not allowed to enter bPPS2:

- Those who violate the study protocol;
- Those who are vaccinated with the wrong vaccine;
- Booster dose vaccination or blood collection on 14 days after booster dose vaccination beyond the protocol-required time window;
- Use of protocol prohibited vaccines or drugs:
  ① Other investigational or unlicensed products (drugs or vaccines);
  ② Long-term use (lasting more than 14 days) of immunosuppressive or other immunomodulatory drugs (inhaled or topical steroids are allowed);
  ③ Immunoglobulin and/or blood preparations.
- Newly diagnosed autoimmune diseases, including human immunodeficiency virus (HIV) infection;
- Other situations affecting the evaluation of immunogenicity of 14 days after booster dose vaccination.

28 days after booster dose vaccination per-protocol set（Per-Protocol Set for Booster, bPPS）: A subset of bFAS, including all the subjects who meet the inclusion criteria and don't meet the exclusion criteria, have been randomized, have finished the 1st and 2nd dose vaccination and the booster dose 6 months after the second dose within the protocol required time window, and have finished the blood collection before booster immunization and 28 days after booster dose vaccination with the corresponding antibody assay results provided. Subjects who meet the following conditions are not allowed to enter bPPS:

- Those who violate the study protocol;
- Those who are vaccinated with the wrong vaccine;
- Booster dose vaccination or blood collection on 28 days after booster dose vaccination beyond the protocol-required time window;
- Use of protocol prohibited vaccines or drugs:
  ① Other investigational or unlicensed products (drugs or vaccines);
  ② Long-term use (lasting more than 14 days) of immunosuppressive or other immunomodulatory drugs (inhaled or topical steroids are allowed);
  ③ Immunoglobulin and/or blood preparations.
- Newly diagnosed autoimmune diseases, including human immunodeficiency virus (HIV) infection;
- Other situations affecting the evaluation of immunogenicity of 28 days after booster dose vaccination.

Immune Persistence Set of Booster Immunization, 6 months after booster dose vaccination (bIPS-6): includes all the subjects who have received the 1st and 2nd dose vaccination, completed the booster dose vaccination 6 months after the second dose, and blood sample is collected 6 months after the booster dose vaccination, and there is an

effective antibody titer value.

Immune Persistence Set of Booster Immunization, 12 months after booster dose vaccination (bIPS-12): includes all the subjects who have received the 1st and 2nd dose vaccination, completed the booster dose vaccination 6 months after the second dose, and blood sample is collected 12 months after the booster dose vaccination, and there is an effective antibody titer value..

10.11.2 Statistical Analysis Methods

10.11.2.1 General Principles

Measuring data are described by means, standard deviations, medians, maximum, and minimum value; counting data or grade data are described by frequencies. All statistical analyses are performed using statistical software SAS 9.4.

10.11.2.2 Characteristics of Subject Population

The number of subjects who were screened, enrolled and completed the trial, and the number of subjects in each statistical analysis data set should be summarized, and the reasons for the dropout should be analyzed. The list of subjects who failed in screening, who dropped out and who did not enter each analysis set were listed separately.

10.11.2.3 Evaluation of Immunogenicity

The seroconversion rate and positive rate of antibodies in the medium dosage group, high dosage group, and placebo group should be calculated respectively, and Clopper-Pearson method will be adopted to calculate the corresponding 95% confidence interval. Chi-square test/Fisher exact probability method will be used to conduct statistical test on difference between groups.
Geometric mean titer (GMT) and geometric mean increase (GMI) of the serum antibody, as well as the corresponding 95%CI in the experimental group and the control group should be calculated, and
and the difference between groups was statistically tested using the ANOVA after log-transfermation of the anytibody titer.

10.11.2.4 Evaluation of Safety

Adverse events were medically coded using MedDRA. This study mainly analyzes the adverse events after vaccination, and the adverse events before vaccination will be listed.
The number of episodes, number of involved subjects, as well as the incidence rate of overall AEs, the vaccine-related AEs, and the vaccine-unrelated AEs in all the groups should be calculated separately, and the differences between groups will be statistically tested using the Fisher exact probability method. The severity, dose distribution and time distribution of general AEs as well as the vaccine-related AEs should be statistically analyzed. The list of the vaccine-related AEs, vaccine-unrelated AEs should be made separately. The AEs after each dose should be statistically analyzed based on the safety set of each dose respectively.
The number of epidoses, number of involved subjects, as well as the incidence rate of overall SAEs, the vaccine-related SAEs, and the vaccine-unrelated SAEs in all groups should be calculated separately, and the differences between groups will be statistically tested using the Fisher exact probability method. The list of the SAEs should be made.

10.11.2.5 Processing of Missing Data

With regard to the statistical analysis of FAS, the missing data of the post-vaccination antibody test result will be filled by the method of Last Observation Carried Forward (LOCF). The missing data of the pre-vaccination antibody result will be filled with the maximum of pre-vaccination antibody results among all the subjects. In terms of the evaluation of the exploratory and safety endpoints,

missing data will not be filled.

11 Monitoring of Clinical Trials

11.1Responsibilities of the Sponsor

The sponsor executes and maintains the quality assurance and quality control system, compiles quality management documents to ensure that the clinical trial is carried out in accordance with regulations, and that data, records and reports meet the requirements of GCP, other regulations and the protocol.

11.2Responsibilities of the Investigator

The principal investigator should manage and clearly divide all personnel involved in the clinical trial. The personal data of the subjects should be kept confidential by the investigators. Documents provided to the sponsor should be identified only by the subject number. The identification list of subjects is kept in the investigator documents. In accordance with GCP principles, the original materials of each subject are allowed to be monitored, audited and verified.

11.3Personnel Training

Before the start of the trial, the staff should be trained. The training contents include GCP principles, clinical trial protocol, SOP, etc. If the sponsor or principal investigator deems it necessary, retraining may be conducted. Each training should have training records.

11.4Subject Compliance Guarantee

According to the clinical trial protocol, a concise, clear and well organized volunteer recruitment form and informed consent form were formulated.

Train the doctor responsible for the informed explaination to communicate with volunteers in plain and understandable language so as to be fully informed.

Screen the subjects strictly according to the inclusion and exclusion criteria.

The Follow-up personnel should have a high sense of responsibility and dedication to improve their communication skills and affinity through training. In the process of safety follow-up, measures should be taken to ensure effective contact between subjects and investigators, and adverse reactions should be disposed timely with the related health consultation provided.

11.5Vaccine Management in Clinical Trials

11.5.1 Definition and Treatment of Cold Chain Failure

Once the refrigerator storing the vaccine has a temperature of <2℃ and >8℃, it is recorded as cold chain failure. Once there is a cold chain failure, the vaccine should be transported to a light-protected environment for storage as soon as possible, with reprting to the sponsor in time. The decision of whether stop or continue using the vaccine should be made according to the written/e-mail responses from sponsor.

11.5.2 Receiving of Vaccine for Trial

When the sponsor delivers the trial vaccine to the study site, the investigator must sign the vaccine receipt form, on which the information (e.g. complete package, and normal cold chain system indication etc.) should be described briefly. When the investigator finds that the vaccine is damaged, spoiled, or has lumps that cannot be shaken in it, such vaccine should be prohibited to be used, and should be returned to the sponsor. In the case of cold chain failure or freeze during the transport or storage process, the vaccine can not be used. The vaccine with the above problems should be marked with '×' on the surface of the outer package and stored separately, managed by a dedicated staff and finally returned to the sponsor.

11.5.3 Management of Trial Vaccines

Trial vaccines should be managed by a dedicated staff and supervised by CRA. The vaccine receipt and transfer record should include the number of received vaccine, the number of vaccinated subjects, the number of remaining vaccine and the number of losses. The investigators will calculate the number of all the trial vaccine.When the field work is completed, the remaining trial vaccines are counted and returned to the sponsor when the study site.

11.6Management of Clinical Trial Sample

Specimens used for neutralizing antibody test should be disposed by the testing institution as medical waste after completion of the testing. The backup serum is temporarily stored by the study site institution, until a verified immunogenicity test report is issued by the testing institution. The backup serum can be stored or processed by the sponsor after the project is completed, and its use needs the approval of the ethics committee and the informed consent of the subjects.

11.7Preservation of Clinical Trial Documents

The clinical trial documents must be kept according to the requirements of Chineses GCP. The sponsor, study institution and studysite should keep the clinical trial data for at least 5 years after the drug is marketed.

11.8Ending Criteria for Clinical Trials

-Samples collected in the clinical trial are sent to the testing institution, and the corresponding testing reports are issued.
-All subjects completed the required visit, and the original data and documents of the clinical trial are transferred to the archivist for archiving and preservation;
-The remaining amount of the trial vaccine is accurate and handed over to the sponsor;
-The statistical analysis report and summary report meet the requirements of the sponsor.

12 Ethical Approval

12.1Review and Approval

This clinical trial protocol should be approved by the local ethics committee. The principle investigator submits the clinical trial protocol and all the necessary additional documents to the ethics committee. After the approval of the ethics committee, the investigator provides the sponsor with a certificate of approval from the ethics committee.

12.2Implementation of On-site Supervision

In the whole process of the trial, the ethics committee should supervise whether there are ethical problems of harming the subjects, whether the subjects get treatment, compensation and corresponding measures when they are harmed by the trial, and evaluate the degree of risk they bear.

12.2.1 Informed Consent Form and Informed Consent

It should be ensured that the method of selection of subjects and relevant information provided to subjects are complete and easy to understand, and the method of obtaining informed consent is appropriate. During the whole process of the trial, the ethics committee should regularly review the progress of the trial and assess the risks and benefits of the subjects.

12.2.2 The Potential Hazards and Hazard Minimization

If the adverse reactions are determined to be related to vaccination (the injection site abscess and rash after vaccination), the subjects will be treated in time according to relevant regulations.

If a life-threatening event occurs, the subject will be escorted to the hospital for treatment immediately and the corresponding report should be made.

Under strict supervision, the trained and experienced medical personnel conduct vaccination and venous blood collection in accordance with the prescribed procedures, so as to minimize the injury and suffering caused by vaccination and blood collection (including pain and local infection at the venipuncture site with little probability).

12.2.3 Protection Measures for Subjects

Clinical trials were conducted in county/city centers for Disease Control and prevention with vaccination qualifications. The sponsor examine the study site strictly according to the requirement of the GCP, before the start of the clinical trial. The environment and facilities of the study site should meet the requirements of *The Guiding Principles for Quality Management of Vaccine Clinical Trials (Trial)*. The emergency plan for the damage and emergency of the subjects should be prepared by the study site. Doctors and nurses with corresponding qualifications and experience should be arranged in the physical examination room and blood collection room in order to strictly grasp the inclusion/exclusion criteria and collect blood smoothly. The first-aid room should be equipped with appropriate first-aid facilities, equipment and drugs, and the first-aid doctors shall have corresponding qualifications and capabilities. When the subjects have adverse events at the study site, they should be treated in the on-site emergency room in time. If they need emergency hospitalization treatment, the ambulance equipped on the study site will send the subjects to the agreement hospital for treatment after the condition is stable though the on-site treatment. The ambulance should also be equipped with necessary first-aid facilities and drugs.

The study site signs a green channel agreement with a local county-level and above general hospital.

During the enrollment of the subjects, the agreement hospital should be notified to prepare for timely treatment. Measures shall be taken to ensure that the emergency adverse events can be dealt with in a timely manner, such as personnel responsibilities, telephone number and rescue route. The effective contact between the subjects and the investigator should be maintained, so that any adverse events can be reported and disposed quickly. When the subjects need to be hospitalized for emergency treatment after serious adverse events, the agreement hospital can provide green channel services such as medical treatment, hospitalization and medical security, to ensure that the subjects can be treated in time. The investigator followed up the progress of the event and completed the investigation record until the end of the serious adverse event.

12.3Confidentiality

It should be ensured that the personal secrets of the subjects are not disclosed under the conditions of the trial enrollment, biological sample collection, report and publication. The recorded information of the test samples only includes the subject number, sample number, sampling time, and testing indicators. Only the main personnel of the study have the anthority to obtain electronica or written copies.

13 Revision of Clinical Trial Protocol

After the sponsor and investigator sign the clinical trial protocol, if there is any modification to the protocol, all the modified protocols shall be re signed and dated by the main investigator and sponsor, and the protocol before modification shall be attached.

All modification plans shall be reported to the ethics committee and approved by the ethics committee before implementation. When modifying the scheme, it is necessary to point out whether it is necessary to modify the informed consent form and electronic CRF form.

14 The Publicity and Publication of Data

After the completion of this clinical trial, if the test results need to be open and/or published, the positive results and the negative results will be open and/or published together.

15 References

1. Former CFDA, *Measures for the Administration of Drug Registration*, 2007.
2. Former CFDA, *Code for Quality Management of Drug Clinical Trials*, 2003.
3. Former CFDA, *Technical guidelines for clinical trials of vaccines*, 2004.
4. Former CFDA, *Guidelines for quality management of vaccine clinical trials (Trial)*, 2013.
5. Drug Evaluation Center. *Guidelines for Novel Coronavirus Preventive Vaccine Research and Development Technology (Trial)* .March, 2020.
6. Lu R, Zhao X, Li J, Niu P, Yang B, Wu H, Wang W, Song H, Huang B, Zhu N, et al. Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. Lancet (London, England). 2020;6736(20):1-10.
7. Chen Y, Liu Q, Guo D. Coronaviruses: genome structure, replication, and pathogenesis. Journal of medical virology. 2020:0-2.
8. Information Office of the State Council. Novel coronavirus infection prevention and control work conference. Beijing, Jan 26, 2020.
9. Diagnosis and treatment of novel coronavirus infection pneumonia (trial version 7) .
10. Holshue ML, DeBolt C, Lindquist S, et al. First Case of 2019 Novel Coronavirus in the United States. N Engl J Med. 2020 Mar 5;382(10):929-936. .
11. Expert group on novel coronavirus pneumonia prevention and control of China Preventive Medicine Association . The latest understanding of novel coronavirus pneumonia epidemiology . Chinese Journal of Epidemiology, 2020-02-14.
12. https://www.who.int/emergencies/diseases/novel-coronavirus-2019/situation-reports.
13. Disease control and Prevention Bureau of national health and Health Commission . China - A novel coronavirus pneumonia in WHO (COVID-19) Joint investigation report .
14. Former SFDA, *Guidelines for the classification of adverse events in clinical trials of preventive vaccines*, 2019.