Immunogenicity and Safety of a Three-Dose Regimen of a SARS-CoV-2 Inactivated Vaccine in Adults: A Randomized, Double-blind, Placebo-controlled Phase 2 Trial

Brief summary: The severe acute respiratory syndrome 2 (SARS-CoV-2) inactivated vaccine (KCONVAC) induced robust immune response after two doses but showed limited boosting antibody response after the third dose administered 28 days after the second vaccination.

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Abstract

Background Control of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) pandemic needs effective vaccines.

Methods In a phase 2 randomized, double-blind, placebo-controlled trial, 500 adults aged 18-59 years or ≥60 years were randomized in 2:2:1 ratio to receive 3 doses of 5-μg or 10-μg of a SARS-CoV-2 inactivated vaccine, or placebo separated by 28 days. Adverse events (AEs) were recorded through Day 28 after each dosing. Live virus or pseudovirus neutralizing antibodies, and receptor binding domain (RBD-IgG) antibody were tested after the second and third doses.

Results Two doses of the vaccine elicited geometric mean titers (GMTs) of 102-119, 170-176, and 1449-1617 for the three antibodies in younger adults. Pseudovirus neutralizing and RBD-IgG GMTs were similar between older and younger adults. The third dose slightly (<1.5 folds) increased GMTs. Seroconversion percentages were 94% or more after two doses, which were generally similar after three doses. The predominant AEs were injection-site pain. All the AEs were grade 1 or 2 in intensity. No serious AE was deemed related to study vaccination.

Conclusions Two doses of this vaccine induced robust immune response and had good safety profile. A third dose given 28 days after the second dose elicited limited boosting antibody response.

Key words: SARS-CoV-2 inactivated vaccine; immunogenicity; safety; three-dose regimen

Background

The pandemic of coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has led to more than 250 million confirmed cases and more than 4.3 million deaths worldwide as of 13 August 2021 according to the World Health Organization (WHO) [1]. The significant morbidity and mortality call for an urgent need for effective vaccines and immunization strategies to contain this virus.

In the race to compete with virus spread, vaccine industry has been collaborating with academy, health organizations, and regulatory agencies closely to accelerate vaccine development. Since the first-in-human clinical trial with a recombinant adenovirus type-5 vectored COVID-19 vaccine conducted in China in March 2020 [2], a variety of vaccines have been developed and underwent clinical assessment. As of 13 August, 2021, there are 110 SARS-CoV-2 candidate vaccines in various phases of clinical development, and another 184 in preclinical development according to WHO [3]. Various platforms or technologies are applied for the development of COVID-19 vaccine, including inactivated vaccine, adenovirus vectored vaccine, recombinant protein-based vaccine, RNA vaccine, and DNA vaccine [3].

Given the urgent need for effective vaccines, 22 COVID-19 vaccines have received conditional approval or emergency use authorization (EUA) via accelerated licensure process so far [4]. In China, seven COVID-19 vaccines have received conditional approval or EUA, including five inactivated vaccines, one adenovirus type-5 vectored vaccine, and one recombinant protein-based vaccine. These five authorized inactivated vaccines were manufactured using similar process but different in antigen content and virus seed. The mass vaccination programs with these vaccines have played a critical role for prevention and control of COVID-19.

The safety, immunogenicity, and/or efficacy in human have been demonstrated and continued to be assessed in ongoing clinical trials for many of these vaccines [5-13]. However, studies showed significant trend of decline in antibody titer various months following 2-dose vaccination of inactivated vaccine, adenovirus vectored vaccine, or mRNA vaccine [14-16]. Some studies also

demonstrated that a booster dose (third dose) elicited robust anamnestic response when given 6 months or more following the second dose [14-15].

Here we report the immunogenicity and safety of a 3-dose regimen of an inactivated SARS-CoV-2 vaccine (KCONVAC, manufactured by Shenzhen Kangtai Biological Products Co., Ltd. [China] and Beijing Minhai Biotechnology Co., Ltd. [China]) from an ongoing phase 2 clinical trials conducted in Chinese adults and elders.

Methods

Study design and participants

This 3-dose regimen study was a part of an ongoing phase 2, randomized, double-blind, and placebo-controlled clinical trial which was conducted in Jiangsu Provincial Center for Disease Control and Prevention (JPCDC). The study was done in accordance with the Declaration of Helsinki and Good Clinical Practice. An independent data safety monitoring board was established before the start of the trial to provide oversight of the safety data during the study. The protocol and informed consents were approved by the institutional review board of JPCDC. Written informed consent from all participants was obtained before screening for eligibility.

Eligible participants were healthy individuals aged 18 through 59 years, and 60 years or more who were seronegative for SARS-CoV-2 IgM and IgG and negative for SARS-CoV-2 nucleic acid as confirmed by pharyngeal swab reverse transcription polymerase chain reaction (RT-PCR). Individuals with confirmed cases, suspected cases or asymptomatic cases of COVID-19 as referred to the Information System of China Disease Prevention and Control were excluded. Those who had close contact with confirmed or suspected cases, or had travel history to a foreign or domestic epidemic community within 14 days before vaccination were also excluded. To be included, participants should have an axillary temperature of 37.0°C or less; and have general good health as established by medical history, physical examination, and laboratory testing. Pregnant or breastfeeding women were

excluded. People with a previous SARS-CoV infection, mental disease, allergic reaction to any ingredient included in this vaccine or severe allergy to any other vaccines, congenital or acquired immune deficiency, human immunodeficiency virus infection, serious systemic diseases, or other major chronic illnesses were also excluded. A complete list of the inclusion and exclusion criteria is provided in the protocol.

Randomization and masking

The vaccine strain of SARS-CoV-2 virus (19nCoV-CDC-Tan-Strain03, isolated in the Laboratory of National Institute for Viral Disease and Prevention, China Center for Disease Control, from clinical specimens obtained from SARS-CoV-2 positive patient) was cultivated in Vero cells. The harvested virus was inactivated by β-propiolactone, purified, and adsorbed to aluminum hydroxide (adjuvant). Each dose of vaccine contained 5 μg or 10 μg of total protein of inactivated SARS-CoV-2 virus and 0.25 mg of aluminum in a 0.5-mL liquid formulation. The placebo contained the same adjuvant but no viral protein. The experimental vaccines and placebo were blindly labelled with a randomization number on each vial as the only identifier.

The eligible participants were randomized within each age group (18 through 59 years, and 60 years or more) in a ratio of 2:2:1 to receive either 5-μg vaccine, 10-μg vaccine, or placebo. The randomization list was generated by an independent statistician using SAS software (version 9.4, SAS Institute Inc., Cary, NC, USA). A unique randomization number in sequence was allocated to each participant, who then received a vaccine or placebo dose labelled with the same randomization number. The individuals involved in randomization and masking had no involvement in the rest of the trial. Participants, investigators, and staff undertaking lab testing were masked to treatment allocation.

Procedures

Participants were administered 3 doses on Day 0, 28 and 56, and observed for any immediate reaction for 30 minutes following each dosing. Diary cards were provided to participants to record any adverse events (AEs) occurred within 7 days after each dosing. Any AEs occurred from Day 8 through Day 28 after each dosing were also recorded. To verify the AEs, on-site visits were required on Day 7 and 28

after each dosing. Telephone contact were done by investigators on Day 3 and 14 after each dosing. All AEs were graded according to the scale issued by the National Medical Products Administration (NMPA), China in 2019 [17].

Blood samples for antibody assay were taken from all the participants before vaccination, and 28 days post the second and third doses. Binding antibody responses against the receptor binding domain (RBD-IgG) of the SARS-CoV-2 spike glycoprotein were tested by using enzyme-linked immunosorbent assay (ELISA) with a detection limit of 1:20. Serum samples were twofold serially diluted from 1:20 in 96-well plates coated with recombinant RBD, and incubated at 37 ℃ for 60 minutes. Bound RBD-IgG was detected using a horseradish peroxidase-conjugated secondary antibody and substrate tetramethylbenzidine. Optical density was read at 450 nm and 630 nm to calculate the antibody titer. Live virus neutralizing antibody responses were measured by microneutralization assay with a detection limit of 1:4. In brief, heat-inactivated serum samples were serially diluted twofold from 1:4 in 96-well plates, and mixed with same volume of live virus solution (strain: 19nCoV-CDC-Tan-Strain03) of 100 $CCID_{50}$ (50% cell culture infectious dose), and then incubated at 37 ℃ for two hours. Subsequently, Vero cell suspension containing 1.5 to 2.5×10^4 cells was added to each well, and incubated at 37 ℃ for four days. The neutralizing titer was defined as the reciprocal of the highest sample dilution that protected at least 50% of cells from cytopathic effect. Pseudovirus neutralization was tested using a vesicular stomatitis virus pseudovirus system expressing the spike glycoprotein as reported previously [18], with a detection limit of 1:10. Undetectable antibody titre was assigned a value of half the detection limit for calculation.

Outcomes

The primary endpoints for immunogenicity were neutralization antibody seroconversion and titre, and RBD-IgG seroconversion 28 days after the third dose. The secondary endpoints included the proportion of participants experiencing adverse reactions/events within 28 days following each vaccination, occurrence of SAE from the first dose through 12 months post the third dose, titre of

RBD-IgG 28 days after the third dose, and the seroconversion and titre of RBD-IgG and neutralization antibody 28 days, after the second dose.

Seroconversion was defined as antibody titre: 1) < 1:4, < 1:30, or < 1:20 before vaccination and≥1:4, ≥1:30, or≥1:20 post vaccination; or 2) ≥1:4, ≥1:30, or≥1:20 before vaccination and ≥4-fold higher post vaccination for neutralization antibody against live SARS-CoV-2, neutralization antibody against pseudovirus, or RBD-IgG, respectively.

Statistical analysis

The sample size was determined based on the assumption that the seroconversion percentages for live neutralization antibody in vaccine group and placebo group were 80% and 30%, respectively. A sample size of 100 in vaccine group versus 50 in placebo group would have sufficient power (more than 99%) to demonstrate a real difference in the seroconversion percentages for live neutralization antibody between groups when tested with a two-sided alpha value of 0.05.

All participants who received at least one dose were included in safety analysis. The number and proportion of participants experiencing adverse reactions or AEs in each group are presented. The immunogenicity analysis was done in per-protocol set consisting of participants who did not deviate from the eligibility criterion, received two doses, provided blood samples as scheduled, and had evaluable immunogenic data. Immunogenicity is expressed by seroconversion percentage, geometric mean titre (GMT), and the associated 95% confidence interval (CI). Antibody titres of individuals were log-transformed to calculate GMT per group. GMTs were compared with t test. The $\chi 2$ test or Fisher's exact test was used to compare difference between groups for categorical data. Analysis of variance was used to test the difference between groups for log-transformed antibody titres. All analyses were two-tailed, and $P < 0.05$ was considered statistically significant. The study was registered with ChiCTR.org.cn, number ChiCTR2000039462.

Results

The trial profile is shown in Figure 1. A total of 500 participants were enrolled, of whom 250 were 18-59 years of age and 250 were 60 years of age or more. In each age group, the eligible participants were randomized to receive 3 doses of 5-μg vaccine (N=100), 10-μg vaccine (N=100), or placebo (N=50). All participants received at least one dose of vaccine or placebo and were included in safety analysis. In age group of 18-59 years, four and six participants were not included in the population for immunogenicity analysis of the second and third doses, respectively, due to no immunogenicity data available. In age group of 60 years or more, the corresponding number of participants were 17 and 24. Six participants aged 18-59 years and 24 participants aged 60 years or more discontinued the study. The average age across the three treatment groups were 38.8 to 42.8 years old in age group of 18-59 years and 64.6 to 64.9 years old in age group of 60 years or more. Baseline characteristics were generally similar between the three treatment groups in each age group (Table 1). This study is ongoing to continuously follow up the safety and antibody persistence as planned. The preliminary analysis reported here presents the data through the cutoff of 28 days post the third vaccination.

In age group of 18-59 years, 23 (23%), 23 (23%), and 10 (20%) participants reported at least one AE, of whom 14 (14%), 16 (16%), and 6 (12%) participants reported at least one vaccination-related AE after receiving 5-μg vaccine, 10-μg vaccine, or placebo. Less frequency of AE was reported in age group of 60 years or more, where 4 (4%), 10 (10%) and 2 (4%) participants reported at least one AE, of whom 1 (1%), 10 (10%), and 2 (4%) participants reported at least one vaccination-related AE in the three treatment groups. All the AEs were grade 1 or 2 in intensity. No AE of grade 3 or higher was reported. The most common solicited injection-site AE across the treatment groups and age groups were pain. Solicited systemic AE was less frequent. A total of 5, 4, and 4 participants reported headache, fever, and fatigue, respectively (Table 2). Two SAEs (protrusion of intervertebral disc, and inguinal hernia) were reported in 5-μg and 10-μg vaccine groups aged 60 years or more, respectively. Both were deemed by the investigators not related to study vaccination. No participant discontinued the study due to AE.

The baseline serostatus is summarized in Table 1. Before vaccination, all participants in the age group of 18-59 years were seronegative (under the detection limit) for live virus neutralizing antibody, pseudovirus neutralizing antibody, and RBD-IgG. Only one and six participants in age group of 60 years or more are seropositive for pseudovirus neutralizing antibody and RBD-IgG, respectively. The vaccine induced significant antibody response (Table 3 and Figure 2). After vaccinated with two doses of the investigational vaccine, the antibody titers were significantly elevated from the baseline with GMTs of 102 to 119, 170 to 176, and 1449 to 1617 for live virus neutralizing antibody, pseudovirus neutralizing antibody, and RBD-IgG, respectively, in the age group of 18-59 years. The GMTs for pseudovirus neutralizing antibody and RBD-IgG in the age group of 60 years or more were similar to those observed in the age group of 18-59 years, but GMTs for live virus neutralizing antibody in the age group of 60 years or more were approximately one fourth of that in the age group of 18-59 years. Vaccination with the third dose of vaccine only slightly (less than 1.5 folds in GMTs) increased antibody titers across the treatment groups. 28 days post the second dose of vaccine, 95% (92/97) to 100% (99/99) of participants aged 18 to 59 years, and 94% (86/92) to 100% (94/94) of participants aged 60 years or more underwent seroconversion for the three antibodies. Seroconversion percentages 28 days post the third dose of vaccines were generally similar to those post the second dose. As expected, placebo did not elicit obvious immune response in both age groups with only 0, 2, and 2 participants after the second dose, and 0, 1, and 0 participants after the third dose underwent seroconversion for the three antibodies. We also included live virus neutralizing antibody results from human convalescent serum (HCS) as a reference (Figure 2). The live virus neutralizing antibody titer induced by this investigational vaccine was higher in participants aged 18-59 years than that in HCS, but lower in participants aged 60 years or more than that in HCS.

Discussion

With the intensive efforts exerted collaboratively by industry, academy, health organizations, and regulatory agencies, more and more vaccines against SARS-CoV-2 were developed, underwent clinical assessment, and received conditional approval or EUA. Mass vaccination programs are deploying around the world that significantly contributed to the prevention of excessive morbidity and

mortality. However, with emerging more transmissible and infectious variants of SARS-CoV-2 and time elapsing, the waning antibody titer might not be sufficient to prevent from COVID-19 caused by the emerging variants, warranting a booster dose or third dose to provide adequate protection.

In this ongoing phase 2 trial, a 3-dose regimen of the inactivated vaccine was evaluated. The third dose of the 3-dose series cannot be deemed as a booster dose due to the short interval between the second and third dose, otherwise it is a part of the primary series. This study on 3-dose regimen may provide us with a more insightful understanding on the antibody dynamics following vaccination with inactivated vaccines, and then help develop an appropriate timing for the vaccination of a third dose.

Administration with 3 doses of this investigational vaccine showed good safety profile at both 5-μg and 10-μg dosages in both younger and older adults, consistent with previous reports on 2 doses of inactivated SARS-CoV-2 vaccines [6,7,13]. The predominant AEs were injection-site pain. All the AEs were grade 1 or 2 in intensity. No AE of grade 3 or higher was reported. No SAE was deemed related to study vaccination. Less frequency of AE was reported in older adults of 60 years or more, which is consistent with the experience from many other vaccines. As more safety data were available for various vaccines from clinical trials and post-authorization studies, it is more evident that inactivated SARS-CoV-2 vaccine is better tolerated in term of short-term safety profile as compared with mRNA vaccine or recombinant adenovirus-vectored vaccine [19-21].

Two doses of the investigational vaccine were able to induce significant antibody response for the three antibodies. Most participants in the both age groups underwent seroconversion for the three antibodies. The GMTs and seroconversion percentages observed in the participants aged 18 to 59 years reported here are similar to those reported previously in the same age group vaccinated with the same dosage of the investigational vaccine [13]. Vaccination with the third dose only slightly increase the antibody titers (less than 1.5 folds in GMTs versus 28 days post the second dose/immediately before the third dose). The same 3-dose regimen (Day 0, 28, and 56) with an inactivated vaccine was also reported recently, which showed slight increase in neutralizing antibody titer (GMTs increased from 39.6 post the second dose to 49.7 post the third dose) [14]. It seems that a third dose with a

relatively short dosing interval such as 28 days may not be helpful for induction of higher antibody titer. This is consistent with the interval between the first and second doses. Our previous report showed that a longer dosing interval between the first dose and the second dose (given at Day 0 and 28 versus Day 0 and 14) induced 1.6 to 3.5 times higher antibody titers 28 days post the second dose [13]. A pooled analysis of four randomised trials with a ChAdOx1 nCoV-19 (AZD1222) vaccine also indicated that an interval of 12 or more weeks compared with an interval of less than 6 weeks between the first and second doses induced more than two-fold higher binding antibody responses [22]. Previous experience with inactivated vaccines or recombinant protein vaccines further support that a longer dosing interval may be helpful to elicit higher antibody response, as what was observed with human papillomavirus vaccine, inactivated polio vaccine, and inactivated hepatitis A vaccine [23-26]. All these evidences support a longer interval between the second dose and the third dose for this inactivated vaccine to induce a robust anamnestic response.

Currently accumulating data show that the antibody elicited by inactivated SARS-CoV-2 vaccine was not durable and persisted for 3 to 6 months [14, 27]. However, a booster dose (third dose) given 6 to 8 months later induced significantly elevated antibody titer, indicating that inactivated SARS-CoV-2 vaccine can elicit immune memory [14]. We are also studying the booster response to this investigational inactivated vaccine in a separate study cohort and will publish the results elsewhere.

When comparing the two age groups (18 to 59 years, and 60 years or more), the investigational vaccine elicited similar antibody response in the two age groups with respect to pseudovirus neutralizing antibody and RBD-IgG, but GMTs for live virus neutralizing antibody were significantly lower in the age group of 60 years or more than in the age group of 18-59 years. This phenomenon was found for both post the second dose and post the third dose.

Consistent with our previous finding [13], no significant dosage-dependent antibody response was observed. Both 5-μg and 10-μg dosages elicited similar antibody response in both the age groups. This has supported the EUA for 5-μg dosage instead of 10-μg dosage.

Numerically, the GMT of RBD-IgG was higher than that of either live virus or pseudovirus neutralizing antibodies, and the GMT of pseudovirus neutralizing antibody was higher than that of live virus neutralizing antibody in both the age groups when the vaccines were administered using the same dosage and the antibody titers were assayed at the same timepoints, which is consistent with observations from previous reports [5,13].

In the context that various platforms or technologies are applied by the currently available COVID-19 vaccines, it is interesting to compare the immune response induced by these vaccines. Although head-to-head comparison studies are not available, the accumulating reports indicate that inactivated vaccine is less immunogenic than mRNA vaccine or recombinant adenovirus-vectored vaccine [5-9, 11, 13, 28]. When comparing with human convalescent serum, this investigational vaccine induced higher antibody titer in participants aged 18-59 years but lower in participants aged 60 years or more. However, it should be noted that the human convalescent serum was not assayed concurrently with the blood samples from the study participants. Variation between lot-to-lot assays might exist.

With continued high global incidence, emerging more transmissible and infectious variants, and expected waning antibody titer along with the time, the vaccine industry and academy are increasingly interested in antibody persistence and effect of booster dose. Unfortunately, we have not yet completed our study on these topics, which are the objectives of our ongoing study and will be reported in the future.

In conclusion, the results reported here indicate that vaccination with two doses of this investigational inactivated vaccine induced robust immune response and had good safety profile. A third dose of this inactivated vaccine given 28 days after the second dose was limitedly helpful to further boost the antibody response. A third dose administered with a longer dosing interval may act as a booster dose, which requires further studies.

(**Footnote page**

Potential Conflict of interest

Jiankai Liu is an employee of Shenzhen Kangtai Biological Products Co., Ltd. Guifan Li, Xianyun Chang, and Yafei Liu are employees of Beijing Minhai Biotechnology Co., Ltd. All other authors declare that no conflict of interests.

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29. **Figure 1 Trial profile**

30.

31. **Figure 2 Antibody response post the second and third vaccination.** A, Neutralizing Antibody to Live SARS-CoV-2. B, Neutralizing Antibody to Pseudovirus. C, RBD-IgG. Participants were vaccinated with the indicated dose levels of KCONVAC or placebo on Day 0, 28 and 56. Blood samples were obtain 28 days post the second and third vaccination (i.e., Day 56 and 84). Horizontal bars show GMTs; Error bars indicate 95% CIs; and dots indicate individual antibody titers.

32. Abbreviations: CI, Confidence interval; GMT, Geometric mean titer; SARS-CoV-2, Severe acute respiratory syndrome coronavirus 2; RBD-IgG, Binding antibody responses against the receptor binding domain; HCS, Human convalescent serum.

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Table 1. Baseline characteristics of the study participants

	18-59 years			60 years or more		
	5 µg group (N=100)	10 µg group (N=100)	Placebo group (N=50)	5 µg group (N=100)	10 µg group (N=100)	Placebo group (N=50)
Age (years), mean (SD)	40.0 (9.9)	38.8 (10.8)	42.8 (9.0)	64.9 (3.1)	64.6 (2.4)	64.8 (3.3)
Sex, n (%)						
Male	42 (42)	38 (38)	24 (48)	54 (54)	54 (54)	29 (58)
Female	58 (58)	62 (62)	26 (52)	46 (46)	46 (46)	21 (42)
Completed, n (%)	96 (96)	98 (98)	50 (100)	90 (90)	89 (89)	47 (94)
Discontinued, n (%)	4 (4)	2 (2)	0	10 (10)	11 (11)	3 (6)
Neutralising antibody to live SARS-CoV-2						
Seropositive	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-7.1)	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-7.1)
GMT	2.0 (2.0-2.0)	2.0 (2.0-2.0)	2.0 (2.0-2.0)	2.0 (2.0-2.0)	2.0 (2.0-2.0)	2.0 (2.0-2.0)
Neutralising antibody to pseudovirus						
Seropositive	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-7.1)	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-3.6)	1 (2.0, 0.1-10.7)
GMT	5.9 (5.4-6.4)	5.5 (5.2-5.8)	5.6 (5.1-6.1)	5.9 (5.5-6.4)	6.4 (5.8-7.0)	6.5 (5.6-7.6)
RBD-IgG						
Seropositive	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-3.6)	0 (0.0, 0.0-7.1)	1 (1.0, 0.0-5.5)	3 (3.0, 0.6-8.5)	2 (4.0, 0.5-13.7)
GMT	10.0 (10.0-10.0)	10.0 (10.0-10.0)	10.0 (10.0-10.0)	10.1 (9.9-10.2)	10.3 (10.0-10.6)	10.9 (9.5-12.5)

Data are GMT (95% CI), number of participants (%, 95% CI) for seropositive (antibody titre ≥ detection limit).

Abbreviations: N, number of participants randomized in each treatment group; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; GMT, geometric mean titre; RBD-IgG, antibody to receptor binding domain.

Table 2. Adverse events within 28 days following any vaccination

	18-59 years			60 years or more		
	5 μg group (N=100)	10 μg group (N=100)	Placebo group (N=50)	5 μg group (N=100)	10 μg group (N=100)	Placebo group (N=50)
Any AE	23 (23)	23 (23)	10 (20)	4 (4)	10 (10)	2 (4)
Grade 1	17 (17)	15 (15)	5 (10)	1 (1)	10 (10)	2 (4)
Grade 2	10 (10)	12 (12)	5 (10)	3 (3)	0 (0)	0 (0)
Grade 3 or more	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Vaccination-related AE	14 (14)	16 (16)	6 (12)	1 (1)	10 (10)	2 (4)
Solicited Injection-site AE	11 (11)	13 (13)	3 (6)	0 (0)	9 (9)	1 (2)
Induration	1 (1)	3 (3)	0 (0)	0 (0)	1 (1)	0 (0)
Swelling	1 (1)	2 (2)	0 (0)	0 (0)	0 (0)	0 (0)
Erythema	2 (2)	3 (3)	1 (2)	0 (0)	0 (0)	0 (0)
Pain	10 (10)	10 (10)	3 (6)	0 (0)	8 (8)	1 (2)
Pruritus	2 (2)	2 (2)	0 (0)	0 (0)	1 (1)	0 (0)
Solicited systemic AE	2 (2)	11 (11)	3 (6)	1 (1)	1 (1)	1 (2)
Fever	0 (0)	2 (2)	0 (0)	0 (0)	1 (1)	1 (2)
Diarrhea	0 (0)	1 (1)	0 (0)	0 (0)	0 (0)	0 (0)
Inappetence	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Vomiting	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Nausea	0 (0)	1 (1)	0 (0)	1 (1)	0 (0)	0 (0)
Myalgia	0 (0)	2 (2)	0 (0)	0 (0)	0 (0)	0 (0)
Headache	1 (1)	3 (3)	1 (2)	0 (0)	0 (0)	0 (0)
Cough	0 (0)	1 (1)	1 (2)	0 (0)	0 (0)	0 (0)
Dyspnea	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Skin or mucosa abnormality	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Fatigue	1 (1)	2 (2)	1 (2)	0 (0)	0 (0)	0 (0)
Unsolicited AE	2 (2)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)

Data are n (%) of participants experiencing the relevant adverse events. The participants received three doses on Day 0, 28, and 56.

Abbreviations: N, number of participants included in each treatment group for the safety analysis.

Table 3. Antibody response post the second and third vaccination

	18-59 years			60 years or more		
	5 µg group (N=100)	10 µg group (N=100)	Placebo group (N=50)	5 µg group (N=100)	10 µg group (N=100)	Placebo group (N=50)
28 days post Dose 2,						
m	97	99	50	92	94	47
Neutralising antibody to live SARS-CoV-2						
Seroconversion	96 (99.0, 94.4-100.0)	99 (100.0, 96.3-100.0)	0 (0.0, 0.0-7.1)	89 (96.7, 90.8-99.3)	92 (97.9, 92.5-99.7)	0 (0.0, 0.0-7.6)
GMT	101.8 (83.7-123.7)	119.0 (102.3-138.6)	2.0 (2.0-2.0)	27.6 (22.5-33.9)	26.1 (21.5-31.6)	2.0 (2.0-2.0)
GMI	50.9 (41.9-61.8)	59.5 (51.1-69.3)	1.0 (1.0-1.0)	13.8 (11.3-17.0)	13.0 (10.8-15.8)	1.0 (1.0-1.0)
Neutralising antibody to pseudovirus						
Seroconversion	92 (94.9, 88.4-98.3)	97 (98.0, 92.9-99.8)	1 (2.0, 0.1-10.7)	86 (93.5, 86.3-97.6)	90 (95.7, 89.5-98.8)	1 (2.1, 0.1-11.3)
GMT	176.3 (145.7-213.4)	169.6 (147.1-195.5)	7.1 (6.0-8.3)	117.3 (95.9-143.5)	134.0 (113.5-158.2)	6.9 (5.8-8.1)
GMI	30.4 (24.8-37.3)	31.0 (26.6-36.1)	1.3 (1.1-1.5)	20.0 (16.1-24.8)	21.0 (17.5-25.2)	1.1 (0.9-1.3)
RBD-IgG						
Seroconversion	96 (99.0, 94.4-100.0)	99 (100.0, 96.3-100.0)	2 (4.0, 0.5-13.7)	91 (98.9, 94.1-100.0)	94 (100.0, 96.2-100.0)	0 (0.0, 0.0-7.6)
GMT	1449.1 (1201.8-1747.2)	1616.7 (1414.5-1847.7)	10.8 (9.7-12.2)	1375.9 (1104.2-1714.4)	1473.1 (1251.3-1734.1)	10.9 (9.5-12.6)
GMI	144.9 (120.2-174.7)	161.7 (141.5-184.8)	1.1 (1.0-1.2)	137.6 (110.4-171.4)	143.2 (121.7-168.4)	1.0 (1.0-1.0)
28 days post Dose 3,						
m	96	98	50	90	89	47
Neutralising antibody to live SARS-CoV-2						
Seroconversion	95 (99.0, 94.3-100.0)	98 (100.0, 96.3-100.0)	0 (0.0, 0.0-7.1)	87 (96.7, 90.6-99.3)	89 (100.0, 95.9-100.0)	0 (0.0, 0.0-7.6)
GMT	143.5 (118.5-	159.9 (137.0-	2.0 (2.0-	35.1 (28.2-43.7)	35.5 (29.9-42.1)	2.0 (2.0-

	173.7)	186.7)	2.0)			2.0)
GMI	71.7 (59.3-86.9)	80.0 (68.5-93.4)	1.0 (1.0-1.0)	17.5 (14.1-21.8)	17.7 (14.9-21.0)	1.0 (1.0-1.0)
Neutralising antibody to pseudovirus						
Seroconversion	93 (95.9, 89.8-98.9)	98 (99.0, 94.5-100.0)	0 (0.0, 0.0-7.1)	86 (95.6, 89.0-98.8)	88 (98.9, 93.9-100.0)	1 (2.1, 0.1-11.3)
GMT	205.1 (171.7-245.0)	213.0 (184.6-245.8)	7.5 (6.4-8.9)	146.8 (122.2-176.5)	155.0 (131.1-183.3)	7.0 (5.8-8.5)
GMI	35.3 (29.2-42.7)	38.9 (33.5-45.3)	1.4 (1.1-1.6)	24.9 (20.5-30.3)	24.0 (19.9-28.8)	1.1 (0.9-1.4)
RBD-IgG						
Seroconversion	95 (99.0, 94.3-100.0)	98 (100.0, 96.3-100.0)	0 (0.0, 0.0-7.1)	89 (98.9, 94.0-100.0)	89 (100.0, 95.9-100.0)	0 (0.0, 0.0-7.6)
GMT	1699.8 (1421.3-2032.7)	1809.2 (1598.6-2047.5)	10.0 (10.0-10.0)	1593.2 (1292.8-1963.6)	1794.8 (1566.0-2057.1)	10.9 (9.5-12.5)
GMI	170.0 (142.1-203.3)	180.9 (159.9-204.8)	1.0 (1.0-1.0)	159.3 (129.3-196.4)	174.2 (151.5-200.3)	1.0 (1.0-1.0)

Data are GMT (95% CI), number of participants (%, 95% CI) for seroconversion. The participants received three doses on Day 0, 28, and 56.

Abbreviations: N, number of participants randomized in each treatment group; m, number of participants included in each treatment group for the per-protocol immunogenicity analysis; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; GMT, geometric mean titre; GMI, geometric mean titre increase; RBD-IgG, antibody to receptor binding domain.

Figure 1

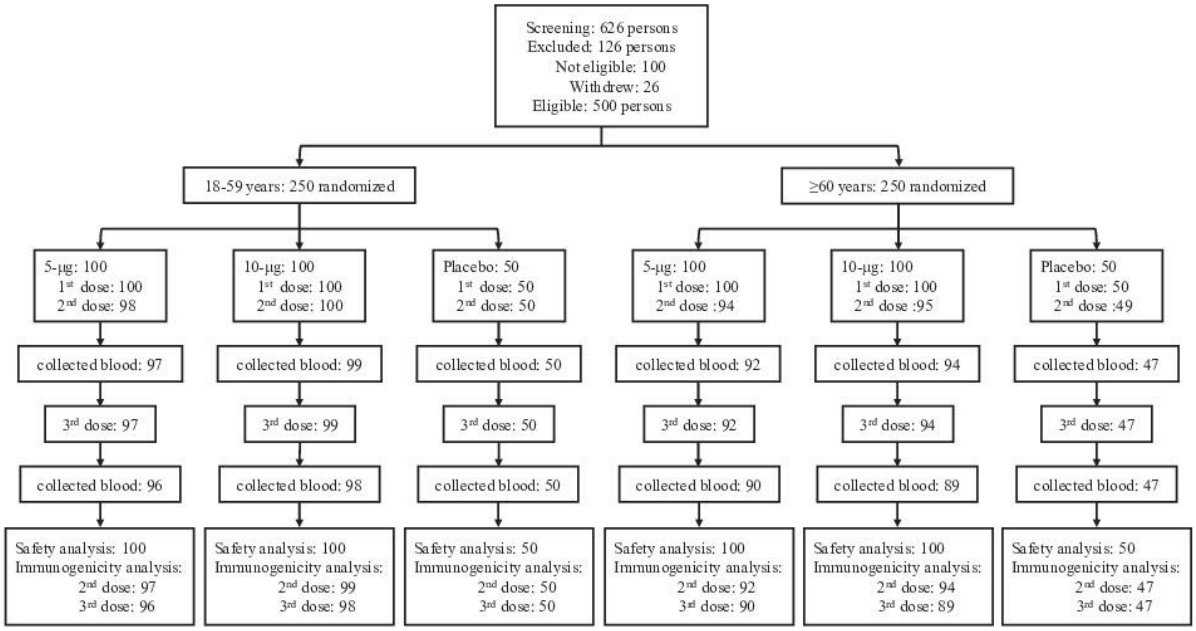
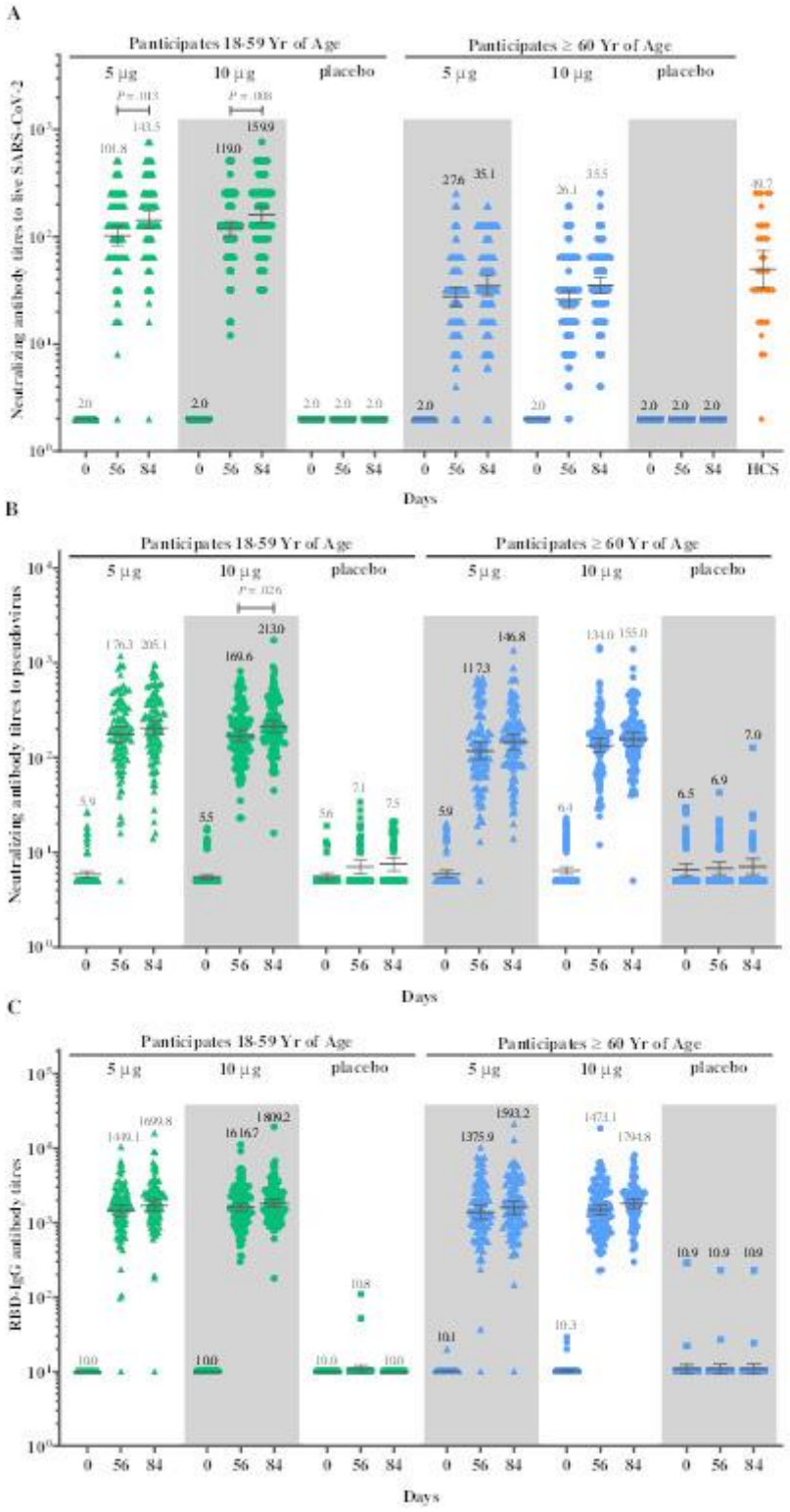


Figure 2


A
Panticipates 18-59 Yr of Age
Panticipates ≥ 60 Yr of Age
5 μg
10 μg
placebo
Neutralizing antibody titres to live SARS-CoV-2
Days
B
Neutralizing antibody titres to pseudovirus
C
RBD-IgG antibody titres
0
56
84
HCS