MVA-based GEO-CM04S1 vaccine results in improved cellular immune response in patients with CLL compared with mRNA-based vaccine: initial results of a Phase II randomized study

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ABSTRACT

Patients (pts) with CLL have impaired immune responses to vaccination and are at increased risk for infections, including SARS-CoV-2. A vaccine which could induce a strong cellular immune response warrants investigation in CLL. GEO-CM04S1 is a Modified Vaccinia Ankara-based vaccine which expresses both Spike (S) and Nucleocapsid (N) SARS-CoV-2 antigens.

Aim: To evaluate immune response following administration of GEO-CM04S1 vaccine in CLL pts

We conducted a 1:1 randomized observer-blinded phase 2 trial in pts with CLL (NCT05672355). Pts received two doses of either Pfizer/BioNTech mRNA (with XBB.1.5 S sequence) (Ctrl) or GEO-CM04S1 (Exp; expressing the Wuhan S and N) vaccine on day(d) 1 and d84 of the study. Primary endpoint was T cell immune response at d56 post-injection of either vaccine. Secondary endpoints included safety, the duration/magnitude of T cell response, Th1/Th2 polarization, S- and N-specific IgG levels and neutralizing antibodies levels against SARS-CoV-2 variants up to d365. Simon 2-stage design was used to suspend accrual in an arm after interim analysis if T cell immune response results in that arm was disappointing.

We enrolled 31 pts (16 on Exp and 15 on Ctrl arm) who received a median of 0 prior therapy (range, 0-8); 52% were men. Two pts (1 per arm) withdrew from the study before receipt of first dose. All pts exhibited moderate baseline serum S-specific IgG levels, with no significant change post-vaccine. Pts form both arms mounted a significant increase in RBD-specific IgG response on d28; and on d84 in the Ctrl arm. Predictably, only the Exp arm showed N-specific IgG post-vaccination. Neutralizing antibodies against the ancestral strain Wuhan (D614G) and Omicron variants (BA.1 and XBB.1.5) were present in both arms and reached a statistically significant increase at d28 for Omicron BA.1.

When analyzing the primary endpoint, only the GEO-CM04S1 vaccinated pts generated statistically significant S- and N specific IFN-γ secretion in stimulated T-cells at d56. Neither group had a significant increase in M (membrane)-specific IFN-γ secretion. We observed a significant increase of S-specific IL-4 secretion at d112 in the Ctrl arm and on d28 and d180 in Exp arm. Neither arm had a significant increase in N-specific IL-4 secretion. Expression of activation-induced markers (AIM) by $CD4^+$ T cells (OX40/CD137) was significantly increased only in Exp arm: S-specific on d84 and d180 and N-specific on d84. Meanwhile, AIM on $CD8^+$ T cells (CD69/CD137) were seen only in S-specific $CD8^+$ T cells on d84 and d180 and on d56 and d84 in the Exp and Ctrl arms, respectively.

Both vaccines were well tolerated. No grade ≥3 events occurred. The most common Grade 1/2 adverse events attributed to vaccine were injection site reaction (n=19), headache (n=12), and fatigue (n=11).

CLL pts who received GEO-CM04S1 COVID19 vaccine generated a more robust T cell response compared with mRNA vaccine recipients. With failure of the mRNA vaccine to meet the primary endpoint, enrollment is now restricted to the Exp arm. While the mRNA vaccine stimulated a more pronounced and sustained humoral immune response, GEO-CM04S1 induced a robust cellular immune response, and thus may be a more effective option for preventing COVID-19 in CLL.

METHODS

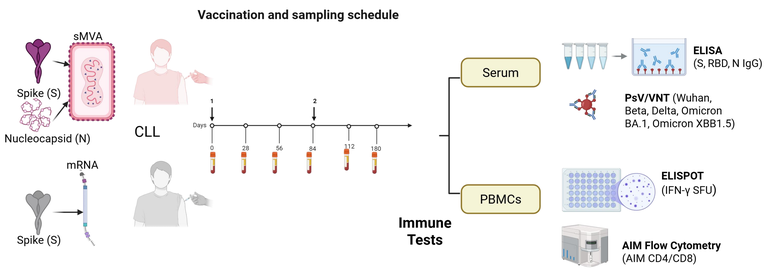


Figure 1: Study Design and Immune Response Assessments Following COH04S1 Vaccination.
Thirty-one CLL patients were randomized to receive 2 doses of either Pfizer/BioNTech mRNA (with XBB.1.5 S sequence) or GEO-CM04S1 (expressing the Wuhan S and N) vaccine (Days 0 and 84). Blood samples were collected for immune monitoring.
Serum samples were analyzed by:
- ELISA to measure IgG binding antibodies against spike (S), receptor-binding domain (RBD), and nucleocapsid (N) proteins.
- Pseudovirus neutralization assays (PsV/VNT) to assess neutralizing titers (NT50) against SARS-CoV-2 ancestral (Wuhan) and variant strains (Beta, Delta, Omicron BA.1, and Omicron XBB.1.5).

PBMCs were evaluated by:
- ELISPOT to quantify IFN-γ-producing T cells in response to S, N, and M peptide stimulation.
- Activation-Induced Marker (AIM) Flow Cytometry to measure antigen-specific AIM+ CD4+ and CD8+ T cell frequencies.

RESULTS

Table 1. Patient and Treatment Characteristics

Characteristic	Pfizer Vaccine n=15[1]	GEO-CM04S1 n=16[1]	Overall N=31[1]
Age at consent (years)	70.0 (54.9 - 85.4)	70.3 (62.6 - 78.8)	70.0 (54.9 - 85.4)
Sex			
Male	10 (67%)	6 (38%)	16 (52%)
Female	5 (33%)	10 (62%)	15 (48%)
Ethnicity, Race			
Non-Hispanic, White	15 (100%)	15 (94%)	30 (97%)
Hispanic, White	0 (0%)	1 (6.2%)	1 (3.2%)
Number of prior therapies	0 (0 - 1)	1 (0 – 8)	0 (0 – 8)
Prior CLL-directed therapy[2]	4 (27%)	8 (50%)	12 (39%)
Protocol vaccine given [3]	14 (93%)	15 (94%)	29 (94%)

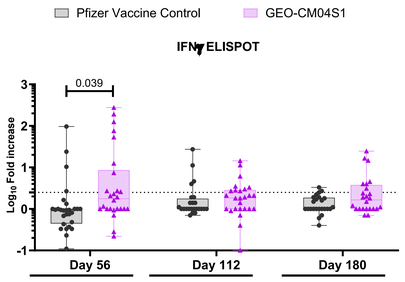
[1] Median (Range); n (%)
[2] No patient had ongoing treatment during study.
[3] Two participants (1 per arm) withdrew from the study before receipt of first dose.

Table 2. Adverse Events Definitely, Probably, or Possibly Attributed to Vaccine

Adverse Event[1]	Treatment Arm				Total (%) (N=28)
	Pfizer Vaccine (n=14)		GEO-CM04S1 (n=14)[2]		
	Grade 1	Grade 2	Grade 1	Grade 2	
Injection site reaction	8	0	10	1	19 (68%)
Headache	4	0	8	0	12 (43%)
Fatigue	6	0	5	0	11 (39%)
Nausea	2	0	1	0	3 (11%)
Fever	1	0	2	0	3 (11%)
Weight loss	1	0	1	0	2 (7%)
Anorexia	1	0	1	0	2 (7%)
Myalgia	1	0	1	0	2 (7%)
Dysgeusia	1	0	1	0	2 (7%)
Chills	0	0	2	0	2 (7%)
Cough	1	0	0	0	1 (4%)
Tenderness at injection site	1	0	0	0	1 (4%)
Hemolysis	0	0	0	1	1 (4%)
Vomiting	0	0	1	0	1 (4%)
Malaise	0	0	1	0	1 (4%)
Bilateral Lower Extremity ache	0	0	1	0	1 (4%)
Pain in extremity	0	0	1	0	1 (4%)

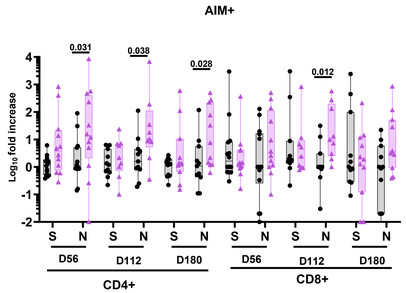
[1]No grade 3 or higher adverse events were observed.
[2]One subject in GEO-CM04S1 arm received the protocol treatment on day 0 and withdrew consent before day 1 follow-up.

Figure 2. GEO-CM04S1 induces IFN-γ cellular response



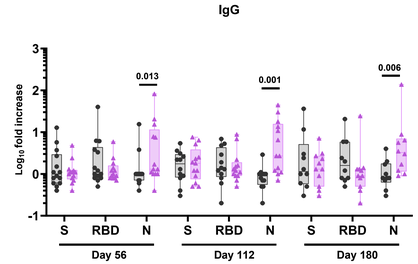
IFNγ T cells were quantified via ELISPOT following stimulation of PBMCs with Spike and Nucleocapsid peptide libraries. Shown is the fold rise response relative to baseline of IFN γ spot forming units (SFU) measured in 10^6 PBMCs. This analysis was conducted using the human IFNγ-IL-4 FluoroSpot FLEX kit (Mabtech), following the manufacturer's instructions. The fold rise results are shown in Figure 2, with the data categorized into two groups: the experimental arm (GEO-CM04S1) and the control arm (Pfizer vaccine).

Figure 3. GEO-CM04S1 induces T-cell activation



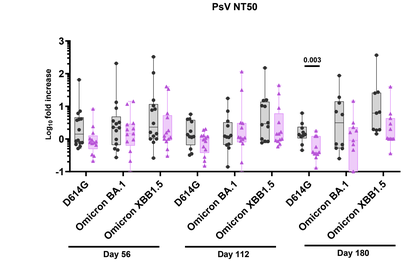
PBMCs were stimulated with S and N peptide libraries and specific S and N T cells were quantified as activation induced markers (AIM) by cytofluorimetry at the indicated timepoints. The number of CD4+AIM+ and CD8+ AIM+ T cells per microliter of blood was calculated. Shown is the fold rise response relative to day 0, with the data categorized into two groups: the experimental arm (GEO-CM04S1) and the control arm (Pfizer vaccine).

Figure 4. GEO-CM04S1 induces N-specific humoral response



The quantification of S-, RBD-, and N-specific binding antibodies was performed using ELISA, calibrated with the WHO international standard serum. IgG results were divided into two groups: the experimental arm (GEO-CM04S1) and the control arm (Pfizer vaccine). Fold increase relative to day 0 is presented

Figure 5. mRNA vaccine elicited stronger D614G pseudovirus neutralizing antibody responses in patient serum



SARS-CoV-2 specific neutralization against ancestral (D614G), Omicron BA.1 and Omicron XBB1.5 variant viruses were measured by microneutralization assay using SARS-CoV-2 pseudovirus (PsV). The NT50 represents the antibody titer required to achieve 50% neutralization of a given sample. The fold-increase response was calculated relative to the baseline.

CONCLUSIONS

- In patients with CLL, those who received the GEO-CM04S1 COVID-19 vaccine generated a more robust T cell response compared to recipients of the mRNA vaccine.
- Due to the failure of the mRNA vaccine to meet the primary endpoint, enrollment is now restricted to the experimental arm.
- GEO-CM04S1 induced a strong cellular immune response, which may make it a more effective option for preventing COVID-19 in CLL patients.

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