Advanced Biomaterials and Oxygen Join Forces to Reinforce COVID-19 Vaccines

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Statement of Purpose: Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has led to an unprecedented global health crisis, resulting in a critical need for effective vaccines that generate protective antibodies. Protein subunit vaccines represent a promising approach but often lack the immunogenicity required for strong immune stimulation.[1] To overcome this challenge, we have engineered advanced biomaterials to leverage and boost the effectiveness of SARS-CoV-2 protein subunit vaccines. Additionally, we investigated the use of oxygen as an immunological co-adjuvant to potentiate vaccine potency further.[2] Our objectives were to demonstrate that oxygen-generating COVID-19 cryogel-based vaccine (O_2-Cryogel$_{vax}$) can (1) induce a balanced Th1 and Th2 immune response and (2) sustain the production of high antibody titers with strong neutralizing activity against the SARS-CoV-2 in a murine pre-clinical model (Figure 1A).

Methods: O_2-Cryogel$_{vax}$ were fabricated by cryogelation.[3] The receptor-binding domain (RBD) of the spike (S) protein and nucleocapsid (N) protein were encapsulated within the scaffolds as antigens alongside the immunomodulatory molecules murine granulocyte-macrophage colony-stimulating factor (mGM-CSF) and CpG oligodeoxynucleotide (ODN) 1826. Controls included Cryogel$_{vax}$ (non-oxygen generating cryogel-based vaccine) and Bolus$_{vax}$ (protein-based vaccines) formulated with the same antigen and immunomodulatory molecule composition. Mice were immunized by subcutaneous injections (one on each flank) at day 0 and 21. Anti-RBD binding IgG titer was determined by ELISA at day 42 and 56. Anti-RBD neutralizing antibody activity was determined at day 21 and 56 using a virus neutralization assay. Th1 cytokine levels in blood were determined using at day 24 using LegendPlex assay. Immunoglobulin (IgG) subclasses ratio was determined at day 56 by ELISA.

Results: Fig. 1 shows the strategy used to fabricate O_2-Cryogel$_{vax}$. (1A) and displays O_2-Cryogel$_{vax}$ 56 days after subcutaneous injection and tissue integration (1B). Overall, O_2-Cryogel$_{vax}$ reinforced substantially the production anti-RBD binding antibodies at day 42 and 56 compared to Cryogel$_{vax}$ and Bolus$_{vax}$, as depicted in Fig. 1C. In addition, O_2-Cryogel$_{vax}$ immunization led to high neutralizing titers within 21 days, which increased after the second dose (Fig. 1D). Finally, O_2-Cryogel$_{vax}$ induced a balanced Th1 and Th2 as demonstrated by the increased Th1-biased cytokine production (Fig. 1E) and the Th2 biased IgG ratio (Fig. 1F).

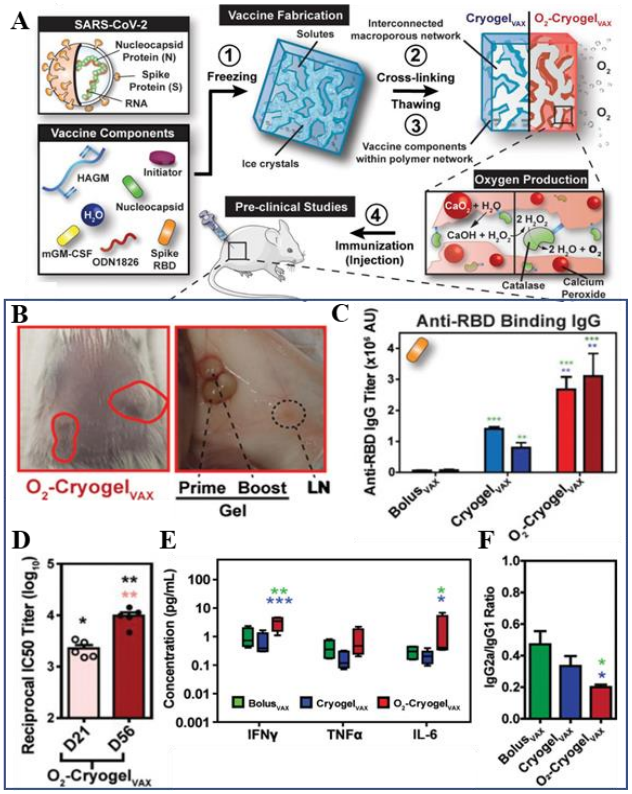

Figure 1: O_2-Cryogel$_{vax}$ induce a strong, long-lasting, and balanced immune response against SARS-CoV-2. A) *schematic depicting O_2-Cryogel$_{vax}$ fabrication.* ***B)*** *Photographs of O_2-Cryogel$_{vax}$ at day 56 post-injection.* ***C)*** *Post-boost endpoint titers of RBD IgG antibody at day 42 and 56.* ***D)*** *Virus neutralization assay after prime (D21) and prime-boost (D56) immunizations with O_2-Cryogel$_{vax}$.* ***E)*** *Th1 cytokine levels in mouse serum at day 24.* ***F)*** *Endpoint titer ratio of the IgG subclasses after 56 days.*

Conclusion: Our study unveils the magnitude of advanced biomaterial-based technology to harness the power of protein subunit vaccines, leading to a rapid and protective anti-SARS-CoV-2 immune response. Additionally, we demonstrated the synergistic effect of vaccines engineered to provide oxygen as a powerful immunological co-adjuvant. Our data suggest that this platform is a promising technology to reinforce vaccine-driven immunostimulation and is applicable to current and emerging infectious diseases.

References: 1. Moyle P. M., ChemMedChem 2013, 8, 360, **2.** Colombani T., Adv. Fun. Mat. 2021, 31, 2170274, **3.** Bencherif S. A., Nat. comm. 2015, 6, 7556.