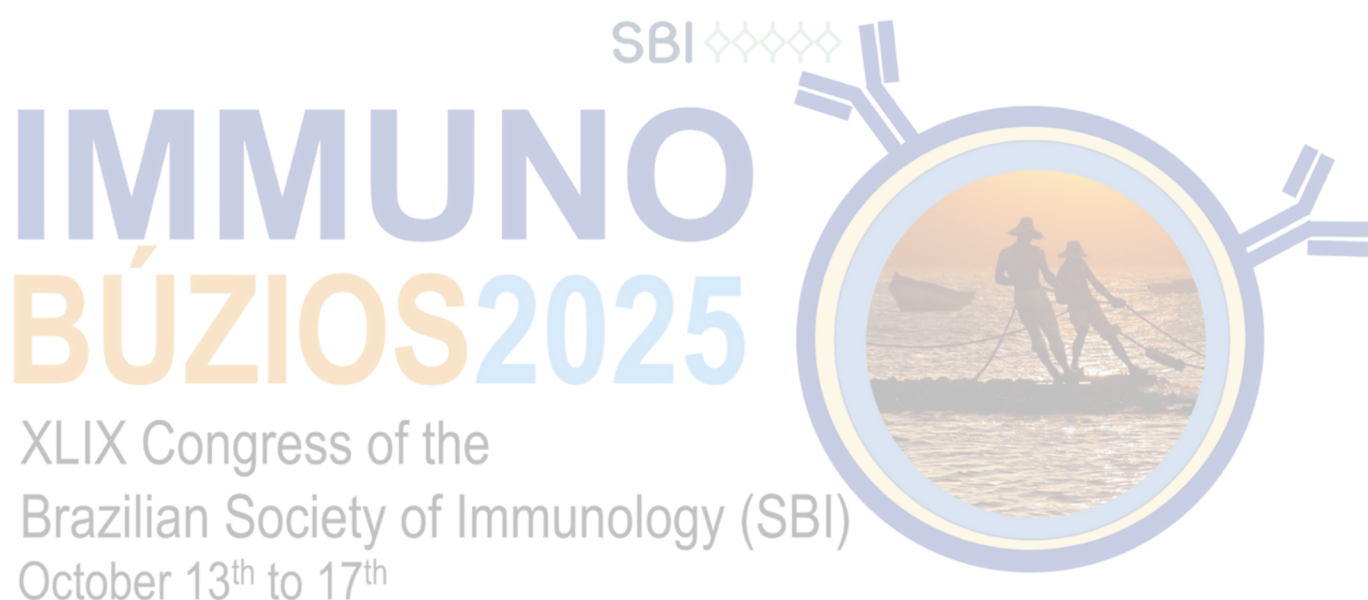


XLIX CONGRESS OF BRAZILIAN SOCIETY OF IMMUNOLOGY

PROGRAM



#ImmunoBúzios2025



2906 - Category: VACCINES (VC)

Enhanced IgG Concentration and Avidity in Bovine Colostrum Following SARS-CoV-2 RBD Immunization with NIBDAF-Based Nanoadjuvants

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Hyperimmune bovine colostrum has emerged as a scalable and safe source of neutralizing antibodies for passive mucosal immunization. The quality and persistence of antibody responses are influenced by the choice of adjuvants. A phagocyte target delivery immunomodulatory nanosystem (NIBDAF; ongoing intellectual property protection) may potentiate both the magnitude and avidity of the antibody response. To compare total IgG concentration and avidity in serum and colostrum of cows immunized with SARS-CoV-2 recombinant RBD protein combined with different adjuvants: Quil-A®, Alúmen®, NIBDAF, and NIBDAF + Alúmen®. Holstein cows in late gestation received three intramuscular doses of recombinant RBD protein formulated with one of the adjuvants. Serum and colostrum samples were collected postpartum. Total IgG levels were measured by indirect ELISA, and avidity was evaluated using a 6 M urea-elution ELISA. Avidity index (AI) was calculated as the ratio of OD values with and without urea. Statistical analysis was performed by two-way ANOVA followed by Tukey and Sidak's multiple comparisons test ($p < 0.05$). All adjuvant formulations elicited detectable IgG responses in serum and colostrum. The NIBDAF + Alúmen® group showed the highest mean IgG concentrations in colostrum (89.5 ± 5.8) and serum (98.6 ± 6.5). Antibodies from all groups exhibited high avidity ($AI > 65\%$). However, colostrum from cows immunized with NIBDAF + Alúmen® had significantly higher AI compared to Alúmen® alone ($p = 0.0425$), and serum AI was significantly higher than in Quil-A® and Alúmen® groups ($p < 0.01$). Immunization with recombinant RBD encapsulated in NIBDAF nanoadjuvant co-administered with Alúmen®, enhances both the quantity and quality of the antibody response in bovine secretions. These findings underscore the potential of NIBDAF-based adjuvants for generating high-avidity antibodies in livestock and support their use in future passive immunization strategies targeting zoonotic respiratory pathogens.

Keywords: Bovine Colostrum ; Nanoadjuvants; SARS-CoV-2.

2974 - Category: VACCINES (VC)

Establishment and Standardization of a Recombinant VSV Platform Pseudotyped with Mpox Virus Envelope Proteins for Vaccine Development and Immunogenicity Studies

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Recombinant vesicular stomatitis virus (rVSV) has attracted considerable interest as a novel platform for virus entry studies and vaccine candidate development. Pseudotyping using heterologous envelope proteins enables this application. However, successful implementation requires the optimization and mastery of key steps, including plasmid amplification and purification, mammalian cell culture, viral rescue, and replication analysis, all of which are critical for the efficient generation of rVSV. The main goal of this study was to develop a robust set of protocols in the production of rVSV while standardizing its characterization. Plasmids containing the green fluorescent protein (GFP), as well as VSV structural genes were amplified in *E. coli* DH5 α and purified using a commercial kit with following the manufacturer's instructions. Quality of the plasmids were assessed by both gel electrophoresis and spectrophotometry. HEK-293T cells were cultured in OptiPRO SFM with either 5% or 10% FBS at 37 °C in 5% CO₂. Lipotransfection was utilized to generate rVSV through the co-delivery of the plasmids. GFP fluorescence and formation of plaques demonstrated viral recovery. Growth curves were reconstructed from plaque assays performed at 0, 24, and 48 hours post-infection. Cell growth looked excellent and there was quite a lot of recovery of virus from the culture with titers doubling from 24 to 48 hours. Additionally, the production of rVSV particles pseudotyped with the Mpox virus envelope proteins is currently underway in an effort to produce functional particles for future entry and immunogenicity studies. This method not only yields high-quality rVSV from a validated system, but also establishes a solid foundation for downstream pseudotyping applications and vaccine vector development involving envelope proteins from a wide range of emerging and re-emerging enveloped viruses.

Keywords: Vaccine vector; Immunogenicity; Mpox virus.