

## Title: Evaluation of FCoV1/SARS-CoV2-based pan-CoV vaccine in SPF cats against FCoV1/FIPV1-Black

**Introduction:** The Director (Yamamoto) of Laboratories of Comparative Immunology & Virology for Companion Animals (LCIV-CA, [Laboratories of Comparative Immunology & Virology for Companion Animals \(LCIV-CA\) » Comparative, Diagnostic & Population Medicine » College of Veterinary Medicine » University of Florida](#)) has developed FCoV1/SARS-CoV-2-based pan-coronavirus (pan-CoV) vaccine against feline coronavirus serotype 1 (FCoV1) and its mutant feline infectious peritonitis virus serotype 1 (FIPV1). To date, commercial FCoV1/FIPV1 vaccine is not available, and only attenuated FCoV/FIPV serotype 2 (FCoV2/FIPV2) vaccine is currently available. Unfortunately, FCoV1/FIPV1 and FCoV2/FIPV2 differ in the sequence at receptor binding domain (RBD), which is the main site for the induction of viral neutralizing antibodies (NABs) [1]. Consequently, Feline Veterinary Medical Association (former AAFP) has reported that FCoV2/FIPV2 vaccine will not protect against FCoV1/FIPV1 [2]. Greater than 90% of pet cats seropositive for FCoV/FIPV in U.S. are infected with FCoV1/FIPV1. Similarly, FCoV1/FIPV1 infection is more common worldwide than FCoV2/FIPV2 infection.

**Vaccine Components & Rationale:** LCIV-CA has opted to combine minimized SARS-CoV2 (SCoV2) and FCoV1 spikes as B-cell-based vaccine immunogens together with FCoV/SCoV2-conserved RNA-dependent RNA polymerase (RdRp) peptides as cytotoxic T-lymphocyte (CTL) and T-helper (TH) epitopes. All of the adverse regions (neurotoxins and inflammatory ICAM-1/superantigens) on SCoV2 spike were molecularly removed to produce a third shorter, B-cell spike for Wuhan and also for FCoV1. The serial vaccinations of specific pathogen-free (SPF) cats with FCoV1/SCoV2-based pan-CoV pDNA-LNP vaccines have delayed FIPV2 challenge infection most likely because of the FCoV-conserved CTL/TH epitopes [3]. Furthermore, Wuhan lineage, Gamma variant, Delta variant, Omicron BA.1 variant, and recently Omicron subvariants JN.1 and NB.1 have been shown to infect laboratory and/or pet cats [4,5]. Close to 21% of the sick pet cats admitted to the Fuchong Pet Hospital in Central China were positive for SCoV2 RBD antibodies and neutralizing antibodies (NABs) to JN.1 more than NB.1 NABs from August 2024 to May 2025 [5]. The goal of our FCoV1/SCoV2-based pan-CoV vaccine is to induce sterilizing immunity and to completely prevent FCoV1/FIPV1 and SCoV2 infections in domestic cats and to prevent reverse zoonosis which occurred with Delta variant in Thailand [6]. Our hypothesis is that the B-cell spike epitopes in the pan-CoV vaccine will induce FCoV1/SCoV2 NABs and antibody-dependent cellular cytotoxicity antibodies (ADCC Abs), whereas CD8<sup>+</sup> CTL and CD4<sup>+</sup> TH epitopes will produce CD4<sup>+</sup> TH cells to enhance CD8<sup>+</sup> CTL activity against infected cells. ADCC Abs will target infected cells to mediate early NK cell killing of infected cells, before CD8<sup>+</sup> CTL killing.

**Experimental Design:** LCIV-CA has ten SPF cats for the next pilot study. We plan to vaccinate five SPF cats thrice at 4-week intervals with FCoV1/SCoV2-based pan-CoV pDNA-LNP vaccine, and the other five SPF cats are similarly immunized with LNP alone. Both groups will be challenged IN with 10<sup>5</sup> TCID<sub>50</sub> of FCoV1/FIPV1-Black isolate. Merck Animal Health has requested that we challenge these groups with FCoV1/FIPV1-Black instead of SCoV2. We are currently collecting pre-vaccination sera and performing pre-vaccination CBC and blood chemistry. Vaccinations will be performed in the first week of February, March, and April before IN challenge in early or mid-May 2026. All pre-vaccination sera, post-vaccination sera after each vaccination, and post-challenge sera will be stored at -20°C. The **FVSP student** will perform FCoV1/FIPV1 whole-virus and RBD immunoblot analyses with these collected sera together with the Director. She will also learn how to develop immunoblot strips from LCIV-CA senior staff, who produces these strips for CVM Diagnostics Service. Since the vaccine contains no CoV nucleocapsid (NC) epitopes, the vaccine protection can be determined by the lack of NC Abs in vaccine group but present in the control group. Similarly, vaccine efficacy will be determined by the lack of semi-nested RdRp RT-snPCR and nested NC RT-nPCR reactivities in the feces and nasal swab of the vaccine group but present in the control group. LCIV-CA postdoctoral scientist (BVSc & AH; MS Anatomical Pathology) will perform molecular assays and will train the **student** in these assays if time permits.

- 1) Yamamoto JK, et al., [doi.org/10.3390/v15040914](https://doi.org/10.3390/v15040914) (LCIV-CA publication)
- 2) 2020 AAHA/AAFP Feline Vaccination Guidelines (page 820) DOI: 10.1177/1098612X20941784
- 3) Sinha P, et al., [doi.org/10.3390/vaccines13111172](https://doi.org/10.3390/vaccines13111172) (LCIV-CA publication)
- 4) Park ES, et al., doi: 10.1038/s41598-024-71791-8
- 5) Yuan Y, et al., [Frontiers | Comparable immune escape capacity for NB.1 with that of JN.1 variant and survey of infection with Severe acute respiratory syndrome coronavirus 2 variants among Chinese Felis silvestris catus](#)
- 6) Sila T, et al., doi: 10.3201/eid2807.212605