Supplementary Information

Purification and Immunogenicity of Nipah Virus-Like Particles from Insect Cells

Urban Bezeljak[1,2]*, Alexander Jerman[1], Tina Kobal[2], Martina Lokar Kosmač[2], Elfi Birsa[2], Marko Kolenc[3], Dániel Déri[4], Bernadett Pályi[4,5], Zoltán Kis[4,5], Matjaž Peterka[2]

[1]Sferogen d.o.o., Ajdovščina, Slovenia

[2]COBIK, Ajdovščina, Slovenia

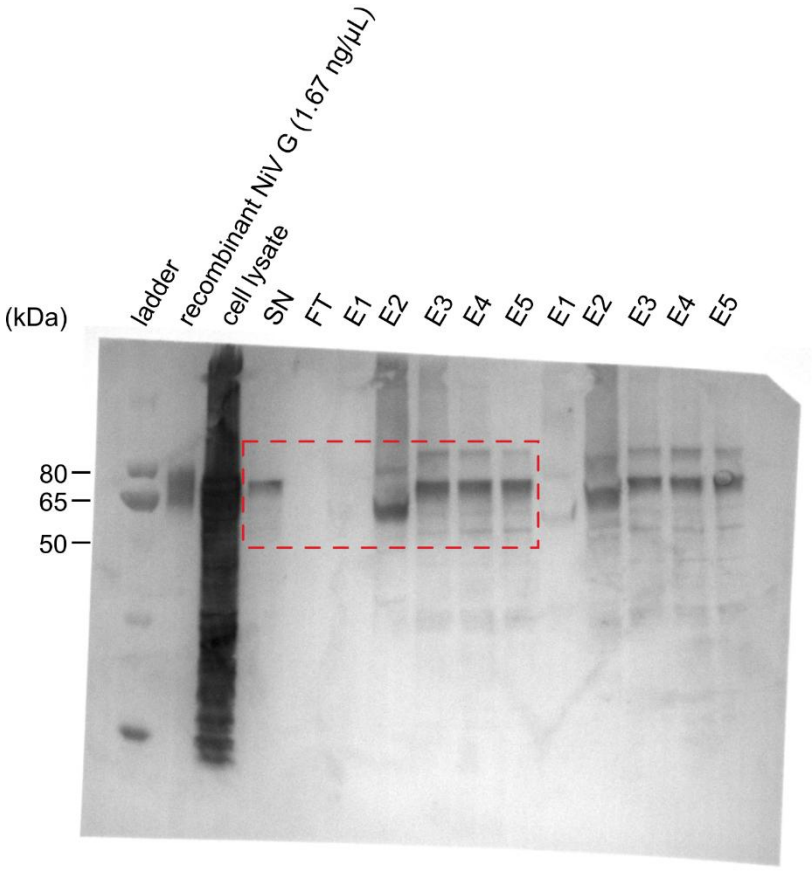
[3]Institute of Microbiology and Immunology, Faculty of Medicine, University of Ljubljana, Ljubljana, Slovenia.

[4]National Biosafety Laboratory, National Center for Public Health and Pharmacy, Budapest, Hungary.

[5]European Research Infrastructure on Highly Pathogenic Agents (ERINHA-AISBL), Brussels, Belgium.

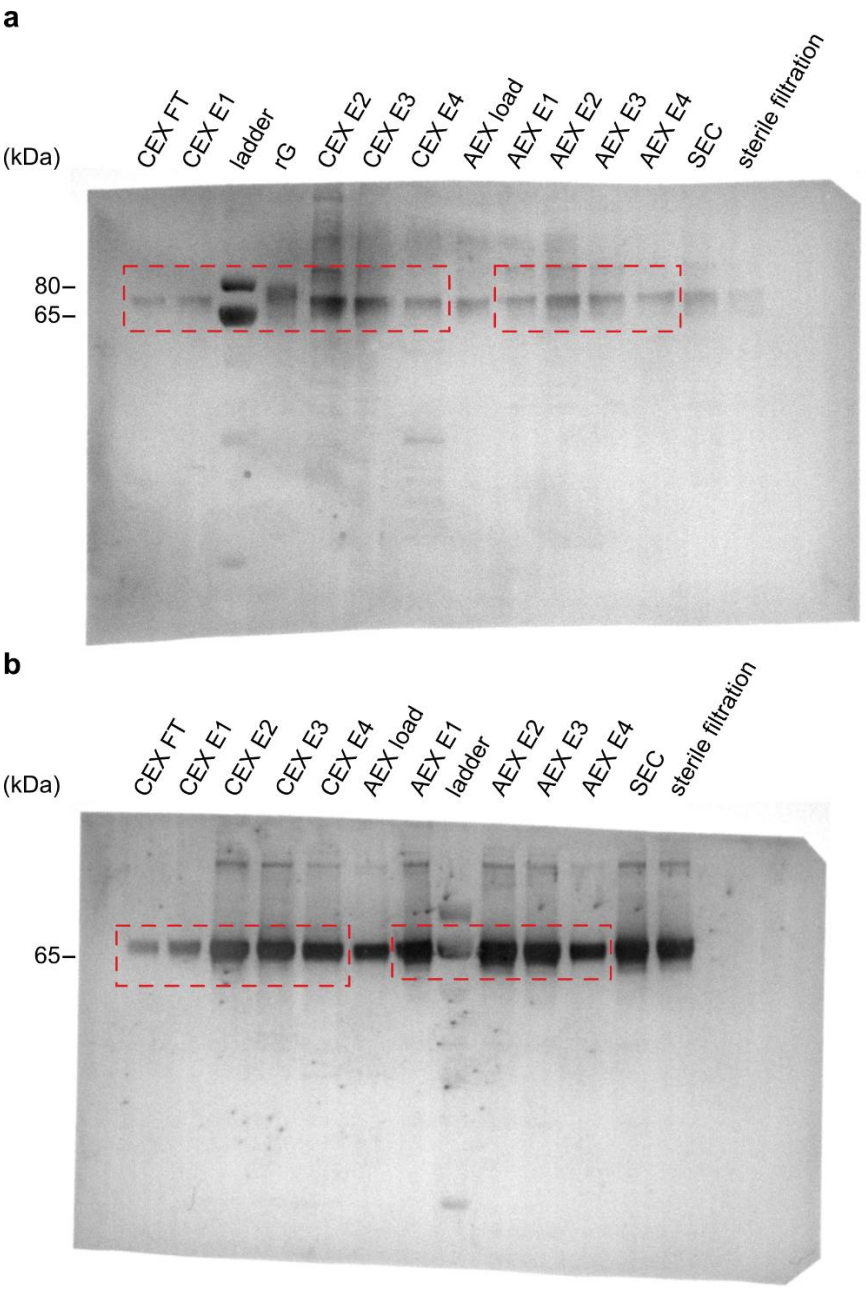
* Corresponding author urban.bezeljak@sferogen.com

Uncropped and unprocessed blot from Figure 1c

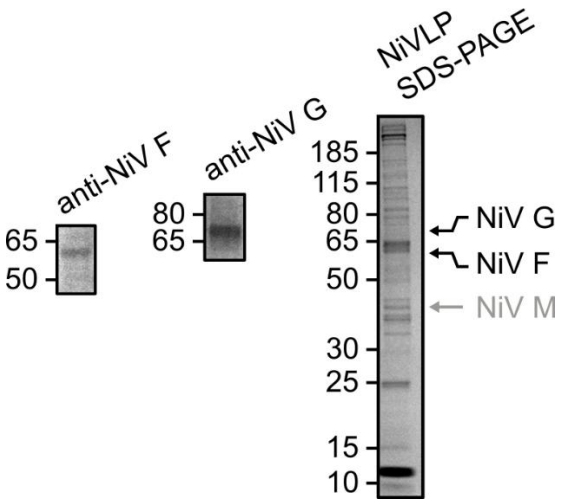


Supplementary Figure 1. Western blot analysis of SXC capture chromatography step. NiV G protein is detected in E2-E5 elution fractions. SN: cell culture supernatant; FT: chromatography flow-through; E1-5: elution fractions. Red rectangle indicates the cropped and processed area used in Fig. 1c.

Uncropped and unprocessed blots from Figures 2c and 2f

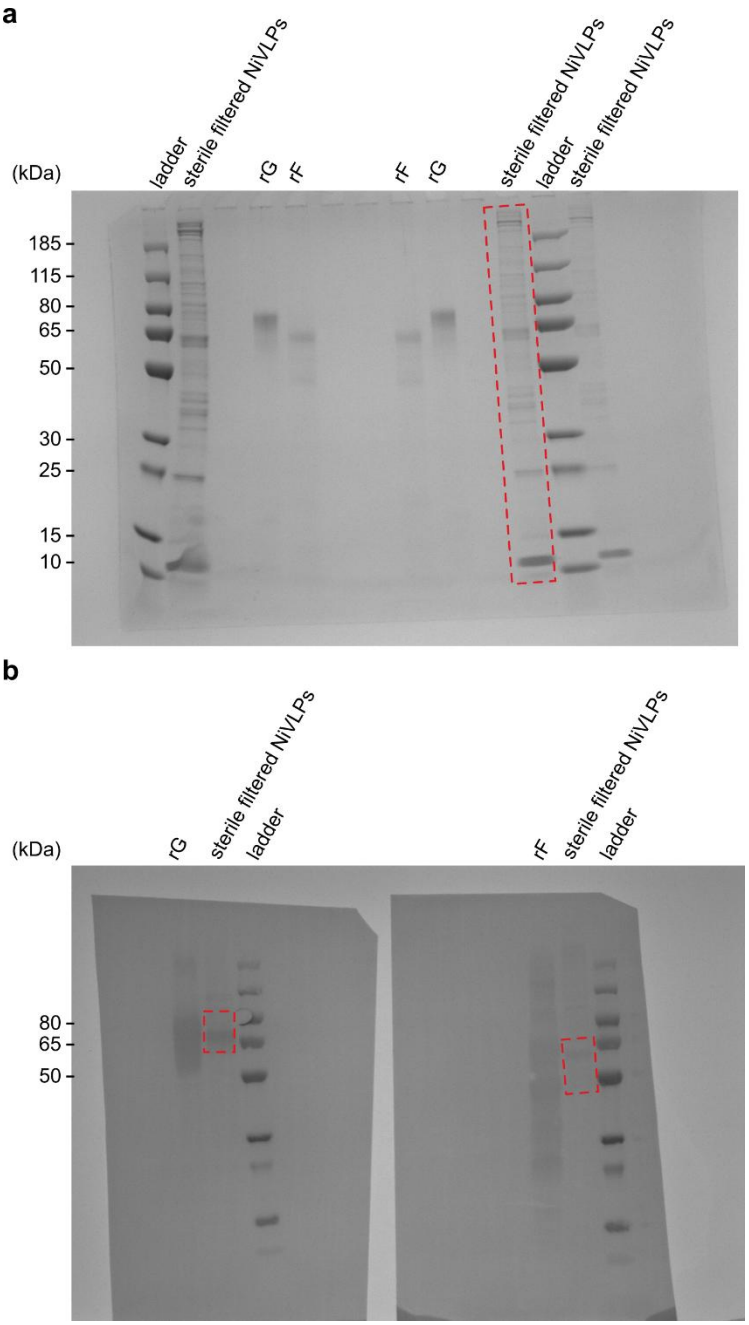


Supplementary Figure 2. NiVLP purification with CEX and AEX chromatography. **(a)** NiV G detection with western blot after CEX, AEX and final sterile filtration. Fraction CEX E2 was loaded on AEX column for final polishing. Finally, fractions AEX E1 and E2 were pooled, buffer-exchanged with SEC and sterile filtered through 0.22 µm filter for immunogenicity study. Red rectangles indicate cropped and processed areas used in Fig. 2c (right) and Fig. 2f (left). **(b)** BV gp64 detection with western blot after CEX, AEX and final sterile filtration. CEX: cation exchange chromatography; AEX: anion exchange chromatography; E1-4: elution fractions; SEC: size-exclusion chromatography. Red rectangles indicate cropped and processed areas used in Fig. 2c (right) and Fig. 2f (left).



Supplementary Figure 3. Characterization of sterile filtered NiVLPs. SDS-PAGE and western blot analysis of sterile filtered NiVLPs. We used polyclonal primary antibodies to detect NiV G and F proteins. Expected molecular weights for NiV G, F and M proteins on SDS-PAGE gel are indicated with arrows. Full-length SDS-PAGE gel and blots are presented in Supplementary Figure 4.

Uncropped and unprocessed SDS-PAGE gel and blots from Supplementary Figure 3



Supplementary Figure 4. Characterization of sterile filtered NiVLPs. **(a)** Separation of sterile filtered NiVLPs with SDS-PAGE. 4-20 % gel was stained with InstantBlue coomassie protein stain. **(b)** NiV G and F detection with western blot after final sterile filtration. rG: recombinant NiV G; rF: recombinant NiV F. Red rectangles indicate cropped and processed areas used in Supplementary Figure 3.