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Infectious African Swine Fever Virus in Spray-Dried Porcine Plasma Used as Feed Ingredient, South Korea, 2026

Appendix

Appendix Table 1. Information and genetic characterization of ASFV DNA-positive feed samples, South Korea, 2026

Samples	Date of Detection	Numb. of positive samples	Sampling site	Date of Manufacture	Ct values (log ₁₀ copies/g) ^b	B646L genotype	I73R-I329L genotype	E183L genotype	CVR	EP402R serogroup
spray-dried porcine plasma (SDPP) ^a	19 February 2026	2	Feed manufacturer	20-21 November 2025	28.48, 31.18 (6.45, 5.73)	II	I	II	CVR1	VIII
compound feed (CF) ^a	24 February 2026	2	Pig farm	9 February 2026	35.97, 37.71 (4.45, 3.99)	II	I	II	CVR1	VIII

^a All samples were collected during the official epidemiological investigation of the 2026 ASF outbreaks. Because epidemiological links were suspected between specific feeds and some outbreak farms, all available samples of these suspected feeds were collected from the affected farms. As these feeds were manufactured using SDPP from a specific manufacturer as an ingredient, available SDPP samples were also collected from that manufacturer.

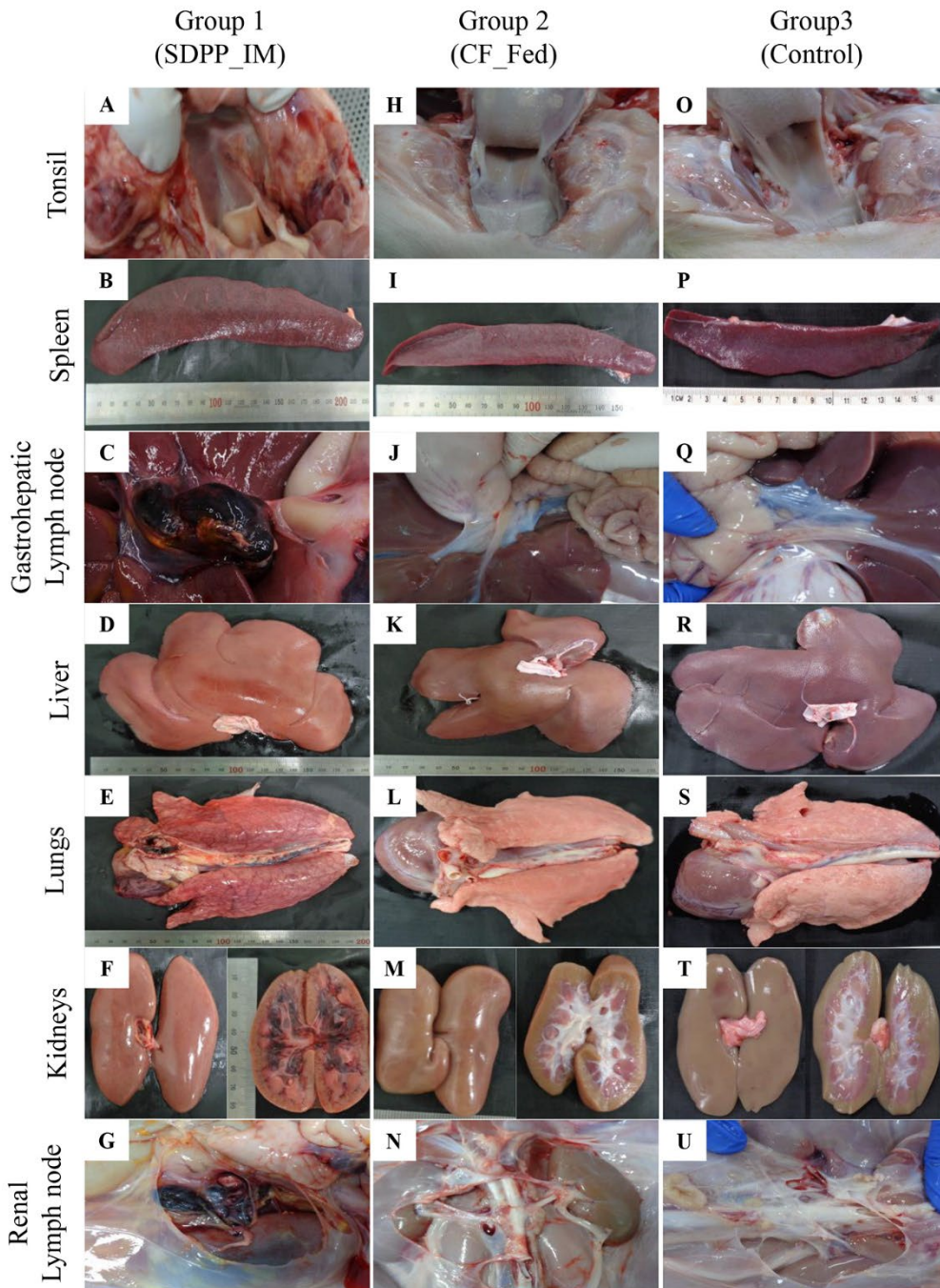
^b Genome copies per gram (copies/g) were calculated from the Ct values using the linear regression equation derived from the standard curve ($R^2 = 0.99$).

Appendix Table 2. Experimental design and administration methods for pigs

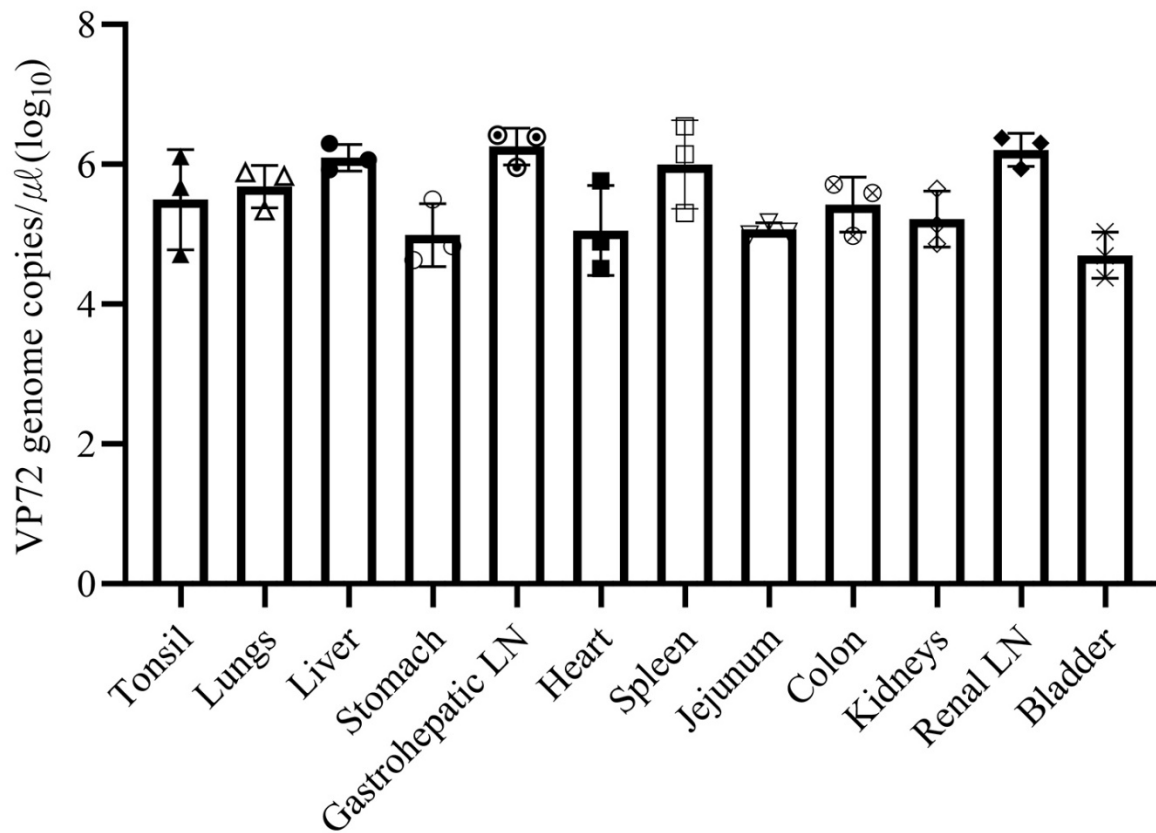
Group	Numb. of Pigs (n)	Inoculum Preparation	Administration	Dietary Feed	Administration
1 SDPP ^a -inoculated	3	50% (w/v) ASFV DNA-positive SDPP suspension in sterile PBS, filtered through a 0.45-μm syringe filter	Intramuscular injection, 1 mL	Clean commercial feed verified free of animal plasma and swine pathogens	Oral feeding, 200g once daily per pig
2 CF ^b -fed	3	-	-	The ASFV DNA-positive CF was manufactured by a feed manufacturer using SDPP produced in the same production batch as the ASFV DNA-positive SDPP.	Oral feeding, 200g once daily per pig
3 Control	3	Sterile PBS	Intramuscular injection, 1 mL	Clean commercial feed verified free of animal plasma and swine pathogens	Oral feeding, 200g once daily per pig

^a SDPP, spray-dried porcine plasma.

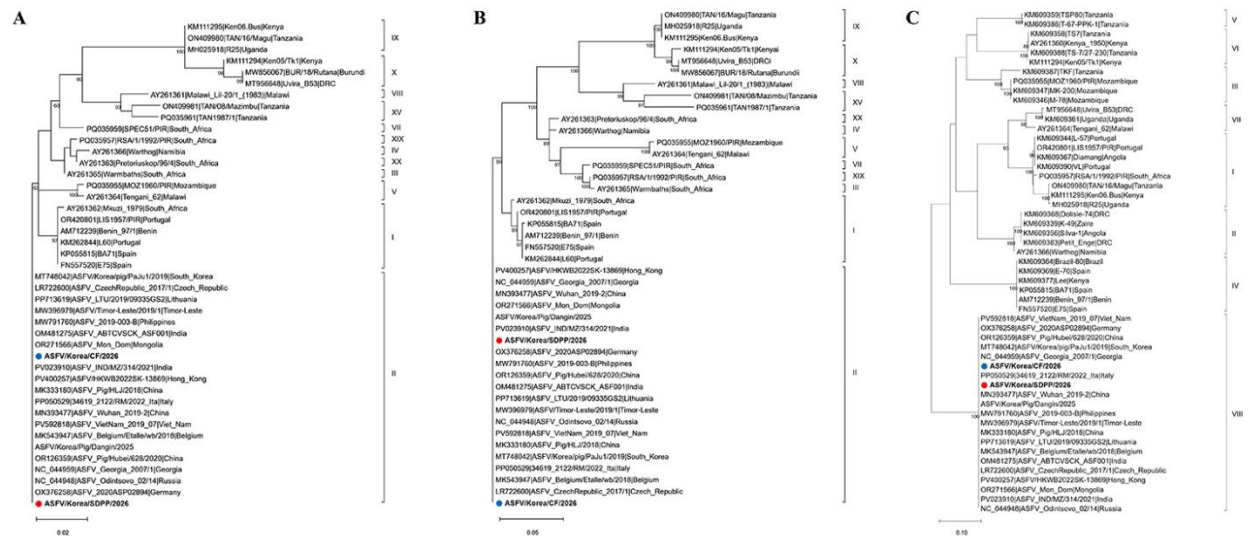
^b CF, compound feed.



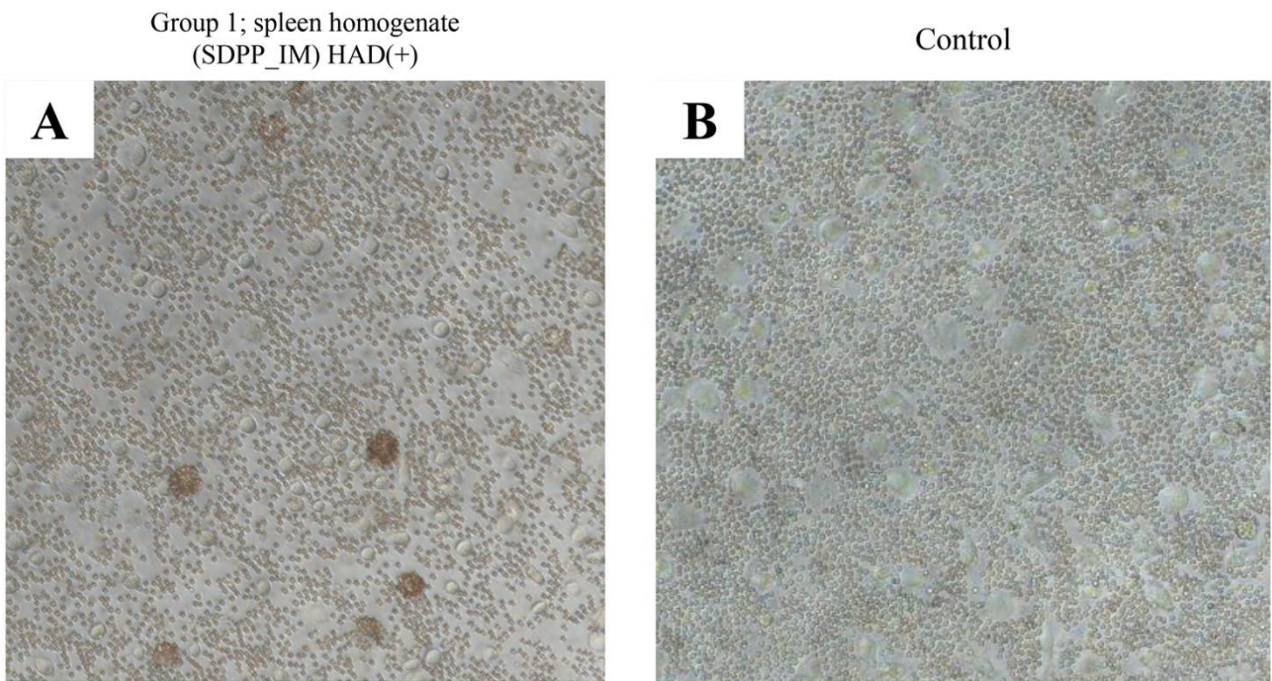
Appendix Figure 1. Major macroscopic lesions in pigs experimentally inoculated with SDPP. (A) tonsil, erythema; (B) spleen, enlargement and hemorrhagic infarction; (C, G) gastrohepatic and renal lymph node, erythema, enlargement and hemorrhagic infarction; (D) liver, petechiae; (E) Lungs, erythema, interlobular edema; (F) Kidneys, petechiae on renal cortex and hemorrhagic renal medulla. (H-N) CF-fed group and (O-U) control group showing no significant lesions.



Appendix Figure 2. Comparison of viral genome loads in different post-mortem tissues of pigs experimentally inoculated with the SDPP (n = 3; LN, lymph nodes).



Appendix Figure 3. Phylogenetic trees of the African swine fever virus (ASFV) strains detected in 2026 based on the (A) *B646L*, (B) *E183L*, (C) *EP402R* genes. Spray-dried porcine plasma (SDPP, red dots) and Compound feed (CF, blue dots) samples clustered with genotype II for *B646L*, genotype II for *E183L*, and serogroup VIII for *EP402R* gene. Phylogeny trees were constructed by maximum likelihood with 1,000 bootstrap replicates. Nodes show bootstraps >80%. Branch lengths indicate substitutions per site.



Appendix Figure 5. Recovery of viable ASFV on porcine alveolar macrophage cells (PAMs) using spleen homogenates from SDPP-inoculated pigs. (A) PAMs exhibiting HAD positivity inoculated with the homogenates and (B) uninfected control cells showing normal morphology.