**Supplementary Files - Protective efficacy of live-attenuated ASFV vaccine strains against the emerging recombinant ASFV genotype I/II strain in Vietnam.**

|  |  |  |
| --- | --- | --- |
| **Clinical signs** | **Severity level** | **Points** |
| 1: Temperature | <40.0 | 0 |
| 40.0≤ to <40.5 | 1 |
| 40.5≤41 | 2 |
| >41 | 3 |
| 2: Anorexia |  Normal appetite  | 0 |
|  Mild (reduced eating) | 1 |
|  Moderate (picking at feed) | 2 |
| Severe (no interest in feed) | 3 |
| 3: Recumbence | No abnormality | 0 |
| Mild (stillness) | 1 |
| Moderate (get up only when encouraged) | 2 |
| Severe (remain recumbent when encouraged) | 3 |
| 4: Skin Hemorrhage/Cyanosis | No abnormality | 0 |
| Mild | 1 |
| Moderate | 2 |
| Severe | 3 |
| 5: Joint Swelling | No abnormality | 0 |
| Mild (joint swelling) | 1 |
| Moderate (Joint swelling + lameness) | 2 |
| Severe (severe lameness, impaired walking) | 3 |
| 6: Labored breathingand/or coughing | No abnormality | 0 |
| Mild | 1 |
| Moderate | 2 |
| Severe | 3 |
| 7: Ocular discharge | No abnormality | 0 |
| Mild | 1 |
| Moderate | 2 |
| Severe | 3 |
| 8: Digestive findings |  No abnormality | 0 |
|  Mild (diarrhea<24 h) | 1 |
|  Moderate (diarrhea >24 h + vomiting) | 2 |
|  Severe (bloody diarrhea) | 3 |

**Table S1.** In-house developed scoring system used in the study.

|  |  |
| --- | --- |
| **Name**  | **Sequence**  |
| Forward Primer | 5'-ATTGTTGGTGTGGGTCACCT-3' |
| Reverse Primer | 5'-GGATTTGGTGAATGAATTTCCTGG-3' |
| Phan G1 Probe | 5’- Q670/ACGCCAGTCGCGTTTTATACC/BHQ2-3’ |
| Phan G2 Probe | 5’- FAM/ACGTATCAGCGAAAAGCGAACG/ BHQ1-3’ |

 A

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **Virus**  | **Dilution** | **Tignon** | **G1 ACDP** | **Phan G1** | **Phan G2** | **β-actin** |
| **ASFV Matla’78**  | 100 | 23.9 | 22.3 | 25.3 | 0 | 29.4 |
|  | 10-1 | 27.9 | 26.31 | 28.7 | 0 | 33.1 |
|  | 10-2 | 30.9 | 29.64 | 32.0 | 0 | 35.8 |
|  | 0-3 | 35. 0 | 33.08 | 35.4 | 0 | 0 |
|  | 10-4 | 35.9 | 35.25 | 37.3 | 0 | 0 |
|  | 10-5 | 38.1 | 0 | 0 | 0 | 0 |
|  | 10-6 | 0 | 0 | 0 | 0 | 0 |
| **ASFV Georgia 2007/1** | 100 | 23.9 | 0 | 0 | 23.6 | 33.8 |
|  | 10-1 | 26.6 | 0 | 0 | 26.2 | 30.2 |
|  | 10-2 | 33.4 | 0 | 0 | 32.7 | 37.5 |
|  | 10-3 | 35.9 | 0 | 0 | 35.3 | 0 |
|  | 10-4 | 37.7 | 0 | 0 | 0 | 0 |
|  | 10-5 | 0 | 0 | 0 | 0 | 0 |
|  | Ext. Ctrl. | 0 | 0 | 0 | 0 | 0 |
|  | NTC | 0 | 0 | 0 | 0 | 0 |

B

**Table S2:** The primer and probe sequences of the Phan G1 and G2 RT-PCR assays (A), and the performance of the two assays compared to published Tignon and G1 ACDP RT-PCR assays against a dilution series (in PBS) of ASFV Malata’78 (p72 genotype I) and ASFV Georgia 2007/1 (p72 genotype II) amplified in PPL cultures (B). NTC = No template control. Ext. Ctr. = Extraction control.