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Virus

PLOS One

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MATERIALS

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Antibiotic-Antimycotic (100X) Thermo Fisher Scientific Catalog #15240062

Phosphate Buffered Saline Contributed by users

FAdV isolate, UPM1137, was obtained from an outbreak of IBH and gizzard erosion in 27 weeks old commercial layer chickens with 2% mortality. Upon necropsy, the liver was pale and friable with erosion in koilin layer of gizzard. Both liver and gizzard samples were positive for FAdV by histological examination and conventional polymerase chain reaction (PCR) [Norfitriah et al., 2018]. Liver from the infected chickens was collected and processed by three times frozen and thawed prior macerated with a sterile mortar and pestle for preparation 1 in 2 (w/v) suspension in sterile phosphate buffered saline (PBS, pH 7.4, 0.1M). For clarification, suspension was centrifuged at 381 x *g* for 30 minutes and the supernatant was collected and purified by filtration through 0.45µm syringe filter. Liver homogenates was treated with commercial antibiotic-antimycotic solution (GIBCO Laboratories, New York, NY, USA) at 1 in 10 (v/v) dilutions and incubated at 4°C for 1 hour prior inoculation [Alemnesh et al., 2012]. The virus inoculum (0.1 mL) was then inoculated in SPF embryonated chicken eggs via chorioallantoic membrane (CAM) route. The eggs were candled daily for mortality. Severe hepatic necrosis was recorded in the dead embryos. The liver was harvested for the next passage (E2) in SPF eggs for the preparation of inoculum used in the present study.

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