

Fowl Adenovirus Group 1 Antibody ELISA test kit

INSTRUCTION MANUAL

FOR PRODUCT No: LTFP01DV/BZ10



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INTRODUCTION

The Fowl Adenovirus Group 1 (FADV) Antibody Elisa kit is used to detect specific antibody against FADV in serum qualitatively. For monitoring antibody after FADV immune and serological diagnostic of infection in Avian species.

PRINCIPLE

This kit uses indirect ELISA method, purified FADV antigen is pre-coated on micro-well strips. When testing, add diluted serum sample to the microplate, after incubation, if there is FADV specific antibody, it will combine with the pre-coated antigen, discard the uncombined antibody and other components with washing; then add enzyme labeled antibody, discard the uncombined enzyme conjugate with washing; Add TMB substrate in micro-wells, the blue signal by Enzyme catalysis is in direct proportion of antibody content in sample, use ELISA reader at 450nm wavelength to measure the absorbance value in reaction wells after adding stop solution to stop the reaction.

MATERIALS SUPPLIED WITH THIS KIT

1	FADV antigen coated strip-plates	96T×10	7	Substrate	24×5 ml
2	Negative control	1.6×5 ml	8	Stop solution	11×5 ml
3	Positive control	1.6×5 ml	9	Dilution Plate	10 no's.
4	Sample diluent	100×5 ml	10	Adhesive Foil	20 pieces
5	Washing solution 10X	100×5 ml	11	Instruction	1
6	Enzyme conjugate	11×5 ml			

MATERIALS REQUIRED BUT NOT PROVIDED

1. Micropipettor: 0.5µl-10µl, 10µl-100µl, 100µl-1000µl.

2. Disposable pipette suction head.
3. Graduated cylinder: 500ml.
4. Enzyme mark with 96 microplate.
5. Distilled water or deionized water.
6. Bottle washing machine.

SAMPLE PREPARATION

Take animal whole blood, get serum by using regular method, the serum should bright and no hemolysis

WASHING BUFFER PREPARATION

Keep 10X Concentrated washing buffer into room temperature before use, if there are salt crystals, shake to dissolve them, then dilute it at 10 times with distilled water or deionized water. The diluted washing buffer can be stored at 4°C for about 1 week.

PROCEDURE

- 1) Take the antigen coated plate (the plate can be opened and used multiple times according to sample quantity each time), add the diluted serum (at dilution stated in kit) to reaction wells at 100µl/well; meanwhile, set 2 wells for positive control and 1 well for negative control, both positive and negative control do not need to be diluted, take 100µl directly and add into its well, mix gently (do not overflow);
- 2) Cover it with Adhesive plate sealer Incubation at 37 °C for 30 minutes.
- 3) Open the adhesive plate sealer, Discard the liquid of the well, add diluted washing buffer to each well, 250µl/well, then discard the liquid, repeat the above step for 3 times, at last wash to dry with the absorbent paper;

- 4) Add Enzyme Conjugate, 100µl/well, Cover it with Adhesive plate sealer, incubate at 37 °C for 30 minutes;
- 5) Open the adhesive plate sealer, discard the liquid of the well, wash 3 times as step 3, remember at last wash to dry with absorbent paper;
- 6) Add substrate: add Substrate 100µl/well, then cover it with Adhesive plate sealer, incubate at 37 °C in dark for 10 minutes;
- 7) Add stop solution 50µl/well to stop the reaction, measure the result in 10 minutes.

INTERPRETATION OF RESULTS

Read the OD value with ELISA Reader at 450nm (620/630nm as reference).

For the assay to be valid:

Negative control (N) OD value < 0.2, and positive control (P) OD value > 0.4;

Calculate method:

$$\frac{\text{Sample OD value}}{\text{Average positive control OD value}} = \text{S/P value}$$

Results interpretation

S/P value ≥ 0.3: Positive

S/P value < 0.3: Negative

NOTES

- 1) Return all reagents into room temperature before use put the reagents at room temperature for at least 1 hour. Shake it evenly before use, and store back to 2-8°C

after usage.

- 2) Do not mix use reagents from different kits and different lot no., prevent the reagents from cross contamination.
- 3) Substrate and stop solution may have cause irritation to skin and eyes, be careful to use.
- 4) Do not expose Substrate to strong light and avoid contact with the oxidant.
- 5) The coated plates should be sealed and moisture-proof. Put back unused Micro-Well plate into dry foil bag and seal at 2-8 °C.
- 6) All wastes should be treated well to avoid pollution before discarding.
- 7) Strict compliance with the operating instructions can get the best results. Pipetting operation, timing, and washing of the whole process must be precise.
- 8) The Coated plates are disposable, do not re-use used strips.

Specifications: 96 wells × 10.

Expiry date: 12 months.

Storage: at 2-8 °C, don't expose in strong light.

THIS IS A DRAFT MANUAL FOR REFERENCE PURPOSES ONLY.

PLEASE REFER TO MANUAL SUPPLIED WITH

KIT FOR EXACT DETAILS.