VERSION 1.01

# PORCINE TOXOPLASMOSIS IGG ANTIBODY ELISA TEST KIT

MANUAL

ELISAKITS.ONLINE
By Immunomart

**Porcine Toxoplasmosis IgG Antibody ELISA Test Kit**

**Catalogue Number. IP100167**

***Product Usage***

The Porcine Foot And Mouth Disease IgG antibody ELISA Test kit is used for detection of Porcine Foot And Mouth Disease Type O antibody in porcine serum; assessment of immunity conditions against porcine FMD in the pig farm and investigation of the epidemiology of the porcine FMD.

***Principle***

The Porcine Foot And Mouth Disease(FMD) IgG antibody ELISA test kit is made from the antigen coated microtiter plate(coated with FMD antigen) and other reagents. It applies the Solid-phase ELISA principle to FMD-Ab in serum, then add enzyme conjugate to specifically bind with complex of coated antigen+FMD-Ab+enzyme labeled anti-pig-IgG antibody on the microplate. With the TMB substrate, it will generate an amount of color. The depth of color is relative with the content of the FMD-Ab, when the value of color is greater than the cut-off value, the pigs are vaccinated well or natural infected exist

***Technical specifications***

96 wells × 2.

***Components***

| | | |
|---|---|---|
| 1 | FMDAntigen coated microplate | 96T X 2 |
| 2 | FMD-IgG Negative control serum | 1.5 mL/tube |
| 3 | FMD-IgG Positive control serum | 1.5 mL/tube |
| 3 | Enzyme conjugate | 22ML |
| 4 | 20×concentrated washing solution | 50ml |
| 5 | Substrate A | 1 bottle |
| 6 | Stop solution | 12ml |
| 7 | Sample diluent solution | 50 ml |
| 8 | Adhesive Foil | 6 pieces |
| 9 | Instruction | 1 pieces |

***Material required not provided***

1 Microplate Reader (Dual-wave length: 450/630 nm).
2 Microplate Washers.
3 Micropipettes, adjustable (Single-wave length 1-100ul、0.5-10ul、multi-wave length 30-300ul).
4 Constant temperature box o r water bath box.
5 Oscillators.

6 Disposable tips (10ul, 200ul)
7 Deionized water

***Preparation***

1) Bring ELISA reagents to the room temperature (20-25 ℃) for 30 min to get best results. Microplate should return to room temperature and dry before open package.
2) Sample dilute: Dilute sample with the sample diluent at 40 times.(5ul serum + 195ul sample diluent), the diluted sample need to mix evenly to get better results.
3) Washing solution preparation: Dilute the 20×concentrated washing buffer with deionized water at 20 times. (50ml 20×concentrated washing buffer + 950ml deionized water ) It is normal if there is crystallization in the 20×concentrated washing buffer, put at 37℃ until completely dissolved.

***Operation procedures***

1.Take out the coated plates (Can be detached) and record the sample position on a worksheet. Set
2. wells for negative control serum, add undiluted negative control serum, 2 wells for positive control serum, add undiluted positive control serum, 100μL/well (It is OK not to set blank control). Others are sample wells, add the diluted sample, 100μL each.
3. Mix gently, cover and incubate at 37℃ for 30 min.
4. Remove adhesive foil. Pour the liquid out of the wells, add the diluted Washing buffer into each well fully, be static for 10s,pour out. Repeat 3 times, at last time pat to dry on absorbent paper.
5. Add 100μL enzyme conjugate into each well.
6. Cover plate with new adhesive foil. Incubate at 37 ℃ for 30 min.
7.Repeat step 3(washing).
9..Add substrate 100ul into each well, mix properly, incubate for 10 min at 37 ℃ in the dark with new adhesive foil.
10..Add stop solution 50μL into each well, mix gently and determine the result.
11. Measure the OD value of each well with a photometer at dual-wave length 450nm/630nm.

***Results***

For the assay to be valid, the positive control wells' average OD value must be greater than or equal to 0.6, and the negative control wells' average OD value is less than 0.1. Otherwise the test is invalid, need test again.
The result is judged by S/P value,
S/P=(Sample OD450/630- NCx(—))/( PCx(—)- NCx(—)), NCx(—) means Negative control's average OD450/630 value, PCx(—) means Positive control's average OD450/630 value
If S/P≥0.15, it is positive; less than 0.15, it is negative.

***Product performance***

1. Specificity: to test 30 negative control serums, no false positive.
2. Sensitivity: to test 30 positive control serums, no negative.
3. Precision: CV（%）no bigger than 15%（n=10）
4. Stability: Store at 2℃～8℃ for 12 months or store at 37℃ for 3 days, the result can reach the above 3 standards.

***Precautions***

This test kit is for research use only.
2. Do not use reagents expired, do not mix reagents from different lots.
3. Read the Manual carefully before use.
4. Experiment rubbish should be dealt with high pressure steam sterilization at 121 ℃ for 30 minutes, or treated with 5.0g/L sodium hypochlorite disinfectant for 30 minutes, then discard.
5. Micro Well plate removed from the refrigerated environment should be balanced moisture to dry at room temperature, then can be opened. Put back unused Micro Well plate into dry foil bag and sealed at 4 ℃. Unused liquid reagent should cover caps, store at 2-8 ℃ in dark with other group components.
6. Should use Micropipette to add sample and reagents, and often proof its accuracy.
7. When adding washing buffer, should be full but no overflow, avoid appearing free enzyme at mouth of well or cross pollution between wells.
8. Stop solution is corrosive, use large amount of water to wash immediately when touch the skin or clothes.

***Storage:*** store at 2-30℃, dark, sealed, dry place, no frozen.
***Expiry date:*** 24 months; date of production is on box.