VERSION 1.01

SWINE TRICHINELLA ANTIBODY ELISA KIT

MANUAL

ELISAKITS.ONLINE

By Immunomart

Swine Trichinella Antibody ELISA Test Kit

Catalogue Number. IP00181

Brief

The Swine Trichinella antibody ELISA test kit is based on an indirect enzymatic immunoassay (Indirect ELISA).The antigen is coated on plates. When a sample serum contains specific antibodies against virus, they will bind to the antigen on plates. Wash the unbound antibodies and other components. Then add a specific enzyme conjugate. After incubation and washing, add the TMB substrate. A colorimetric reaction will appear, measured by a spectrophotometer 450nm（630nm as a reference）

Principle

This kit use indirect ELISA method, MHP antigen is pre-coated on enzyme micro-well strips. When testing, add diluted serum sample, after incubation, if there is MHP specific antibody, it will combine with the pre-coated antigen, discard the uncombined antibody and other components with washing; then add enzyme labled anti-MHP antibody, combine with the antigen-antibody complex on the plate; discard the uncombined enzyme conjugate with washing; Add TMB substrate in micro-wells, blue product formed enzymatically, after adding stop solution to stop the reaction, use ELISA reader at 450nm wavelength to measure the absorbance A value in reaction wells .

Specifications: 96 wells × 2.

Components

	Code item	**Spec.**
1	Pre-coated Microplate	1 pieces(96 wells)
2	Enzyme Conjugate	11/22 ml
3	10X Concentrated washing buffer	100 ml
4	SubstrateA	15ml
5	BubstrateB	15 ml
6	Sample diluent	100 ml
7	Stop solution	15 ml
8	Negative control	1 ml
9	Positive control	1 ml
11	Adhesive Foil	2/4 pieces
12	Instruction sheet	1 piece

Material required not provided

1. ELISA Reader
2. 450nm and 630nm wavelength
3. 37℃ Incubator
4. measuring pipettes.
5. microplate washing machine

Sample requirement

To obtain best laboratory result, avoid using sample with NaN3、high cholesterol、bacteria and severe hemolytic.
Store serum sample at 2~8℃ for a short term(7 days); store at -20℃ or below -20℃ for a long time, avoid repeated freezing and thawing.

Operation Procedure

1.Washing buffer preparation**:** dilute concentrated washing solution (No.2 solution) with distilled water or deionized water at 1:10(such as 50ml concentrated washing solution+450ml distilled water or deionized water), mix thoroughly to obtain the washing working solution.. Sample dilution: Dilute serum sample with sample diluent (No.5 solution) at 1:100 (such as 5ul serum sample + 495ul sample diluent), mix thoroughly . Negative control and positive control do not need dilute, add directly.
2.Adding sample: for each test, set 2 wells for negative control, add negative control 100ul /well, 1 well for positive control, add positive control 100ul /well, 1 blank control well, add 100ul sample diluent(No.5 solution). For other wells, add 100ul diluted serum sample. Incubate at 37 ℃ in dark for 30 min, pour the liquid out of the wells. Use the diluted washing solution to wash for 3 times, each time stand for 1 min, at last time pat to dry on absorbent paper.
3. Adding enzyme conjugate: Except the blank well, add 1 drop of enzyme conjugate (No.1 solution) to each well. Incubate at 37 ℃ in dark for 30 min, pour the liquid out of the wells. Wash as last step, pat to dry on absorbent paper.
4. Coloration: Add 1 drop of Substrate A (No.3 solution) and Substrate B (No.4 solution) accordingly, mix evenly, Incubate at 37 ℃ in dark for 10 min.
5. Add 1 drop of Stop solution (No.6 solution) to stop reaction (blue will change into yellow after adding stop solution) and determine the result.

Results

Set zero for the blank well, measure with microplate reader at 450nm (Reference-wavelength: 630 nm).
1) Valid determination:
Negative control is colorless or light yellow, Positive control is yellow, it means the test is valid
2) Result judgment
When Average OD value of Negative control is less than 0.08, calculate as 0.08,
S/N= OD Value of Samples/ Average OD value of Negative control
S/N > 2.1, swine Trichinella is positive;
S/N ≤ 2.1, swine Trichinella is negative.
4) Limitation of the test method: This test kit is only applied to swine serum.

Note

1) Return to room temperature before use. Seal the unused micro-well strip and store at 2~8 ℃.

2) Each reagent must be shaken evenly before use.
3) Seal the plate with adhesive film when incubate, do not repeat use the adhesive film.
4) Never mix up reagents from test kits in different batch number. Tighten all caps after use, don't mix caps.
5) The wasted kits and sample should be treated as infection agent. The stop solution is corrosive, be careful to use.
6) When incubate at 37℃, suggested to use water bath. If there is no water bath, put wet box with adhesive foil in 37℃ incubator.

storage: store at 2-8℃, dark, sealed, dry place, no frozen..
Expiry date: 12 months; date of production is on box.