

**SWINE TRANSMISSIBLE
GASTROENTERITIS(TGE) ANTIBODY ELISA
KIT
MANUAL**

Swine Transmissible Gastroenteritis (TGE) Antibody ELISA Kit

Catalogue Number. IP100183

Product Usage

This kit is used to detect swine Transmissible Gastroenteritis (TGE) antibody in porcine serum.

Notes

- 1) All reagents should return to the room temperature(18~26°C) before using, and store back at 2-8°C after using
- 2) Do not use kit out of expiry date. Do not mix use reagents from kits of different lot numbers.
- 3) The unused micro-wells should seal back to bag and store back at 2-8°C.
- 4) Used materials should be treated innocuously, and be handled in accordance with local, regional and national regulations.
- 5) Avoid Substrate exposure to bright light, avoid contact with oxidants.
- 6) When the quantity of serum sample is big, dilute all serum sample on serum dilute plate firstly, then transfer all diluted serum sample on reaction wells, make the reaction time same.
- 7) Stop solution is corrosive, if splashed on the skin or clothing should immediately rinse with plenty of water.
- 8) Strictly adhere to instruction to get best result. All procedure including pipetting, timing and washing etc. must be accurate to get accurate result.

Specifications: 96 wells × 2.

Components

	Code item	Spec.	
1	TGE-Coated plates		
96 wells	96T X 2		
2	Enzyme Conjugate	11/22 ml	yellow lid
3	20X Concentrated washing buffer	100 ml	white lid
4	Substrate	11/22 ml	Orange lid
5	Sample diluent	100 ml	Transparent lid
6	Stop solution	12 ml	blue lid
7	TGE Negative control	1.5 ml	Green lid
8	TGE Positive control	1.5ml	red lid
9	Adhesive Foil	2pieces	
10	Instruction sheet	1 piece	

Sample preparation

- 1) 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. Negative control and positive control do not need dilute.
- 3) Washing solution preparation: Dilute the 20×concentrated washing buffer with deionized water or distilled water at 20 times. (10ml 20×concentrated washing buffer + 190ml deionized water) It is normal if there is crystallization in the 20×concentrated washing buffer, put at 37°C until completely dissolved.

Operation procedures

- 1 .Adding sample: Take out the required coated plates according to sample quantity (Can be detached) and record the sample position on a worksheet. For single-wave length test, set one blank control well, add nothing; for double-wave length test, do not set blank control well. Set 2 wells for negative control serum and 2 wells for positive control serum, add undiluted negative and positive control serum to its well accordingly, 100 μL/well. Others are wells for samples, add 100μL/well of Sample diluent solution, then add 1 μL serum sample separately (there will be color change after adding sample).
2. Mix gently for 30s, incubate at 37°C for 30 min.
3. Remove adhesive foil. Pour the liquid out of the wells, add Washing solution(dillute the Wash Concentrate at 20 times with deionized water) into each well fully, stand for 1 min. Repeat 5 times, at last time pat to dry on absorbent paper.
4. Add 100 μL enzyme comjugate into each well (except the blank well, also do not add any liquid to blank well).
5. Cover plate with new adhesive foil. Incubate at 37 °C for 30 min.
6. Repeat step 3(washing).
7. Add 50μL substrate A and substrate 50μL B into each well, mix properly, incubate for 15 min at 37 °C in the dark with new adhesive foil.
8. Add 50μL stop solution into each well, mix gently and determine the result within 5-30 min.
9. For single-wavelength test, measure the A value with a photometer at 450 nm, set zero for the blank well, and read A value of each well; For double-wavelength test, measure the A value with a photometer at 450 nm (Reference-wavelength: 630 nm), read A value of each well.

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,

$S/P = \frac{\text{Sample OD}_{450/630} - \text{NCx}(-)}{\text{PCx}(-) - \text{NCx}(-)}$, NCx(—) means Negative control's average OD_{450/630} value(calculate as 0.05 when the value is less than 0.05), PCx(—) means Positive control's average OD_{450/630} value

If $S/P \geq 0.2$, it is positive; less than 0.2, it is negative.

Precautions

All reagents should be adjusted to the room temperature and shake evenly before using, store at 2-8 °C after using

- 2) Do not exchange the reagents from the kits of different lot numbers to use. Avoid reagent pollution when using.
- 3) Substrate and stop solution may have excitant to skin and eyes, pay attention when using.
- 4) Do not expose TMB (Substrate) to light and avoid it contact with antioxidants.
- 5) The wells should avoid damp or touching water after unsealing (Put the un-using microtiter strips back to bag with dehydrator in 2~8 °C soon)
- 6) Deal all waste reasonable before dumping to avoid pollution.
- 7) Strictly adhere to instruction to get best result. All procedure including pipetting, timing and washing etc. must be accurate.
- 8) Serum dilution plate is disposable, do not use for second time; the MAX volume of it is 300µL/well.

storage: store at 2-8°C, dark, sealed, dry place, no frozen..

Expiry date: 12 months; date of production is on box.