Porcine Circovirus type 2 (PCV2) Antibody ELISA Kit

PCV2 Ab Test

Product Number: E30051

Product Unit: 96wells, 192wells and 480wells

Manufacturer Information

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1. Introduction

Porcine circovirus type 2 (PCV2) is the primary causative agent of porcine circovirus-associated disease (PCVAD). The virus preferentially targets the lymphoid tissues, which leads to lymphoid depletion and immunosuppression in pigs. This ELISA kit can be used to detect PCV2 specific antibody in serum or plasma.

2. Description of Test

The microtiter plate was precoated with PCV2 antigen. During testing, samples are added into the microplate wells, in which the precoated antigen will capture the PCV2 antibody in sample and form antigen-antibody complex. Uncombined components are discarded by a washing step. Then HRP conjugated anti-porcine antibody is added into each well, which will combine with the porcine antibody in wells. After another washing step to remove unreacted conjugate, substrate is added and a blue color will be developed if PCV2 antibody is present. The measured intensity is positively proportional to the amount of antibody present in the sample.

3. Precautions

- Store the kit at 2-8℃, Check the lot number and expiration date before use.
- Bring the test kit to room temperature before use. For example, take it out from the cold storage and put at room temperature for at least 30min.
- The stop solution in the kit is acidic, please make sure do not touch it with your hand or skin.
- The component of the kit is noninfectious, but the field sample shall be treated as potentially infectious. Please handle all these materials properly according to your lab regulations.
- After experiment, all lab materials shall be handled properly according to local regulations.

4. Limitations of Test

This ELISA kit is currently designed for veterinary use. We recommend validating in your own lab with different methodologies to confirm the performance. If it is not used for the mentioned purpose, please contact us for help.

5. Reagent Provided

The kit contains the following items.

Item No.	Description	96wells	192wells	480wells
1	Microplate pre-coated with PCV2 antigen	96 wells	2 X 96 wells	5 X 96 wells
2	Positive Control	1 ml	1 ml	2 X 1 ml
3	Negative Control	1 ml	1 ml	2 X 1 ml
4	Enzyme Conjugate	25 ml	25 ml	3 X 25 ml
5	TMB Substrate	25 ml	25 ml	3 X 25 ml
6	Stop Solution	20 ml	20 ml	2 X 20 ml
7	25X Wash Buffer	30 ml	30 ml	3 X 30 ml
8	Sample Buffer	30 ml	30 ml	2 X 30 ml
9	Kit Instruction	1set	1set	1set

6. Instrument Required

- ELISA reader with 450nm
- Micropipette 20-200ul, Micropipette Multi-Channel 50-300ul

7. Reagent Preparation

- Make sure the kit and all test samples are returned to room temperature (18-26°C) before use. Shake each reagent gently before adding into the well.
- The positive control, negative control, enzyme conjugate, sample buffer, and TMB substrate can be used directly. All test samples shall be diluted 1:39 with sample buffer.
- Wash buffer (25X): dilute the 25X wash buffer provided in the kit with deionized water in the volume ratio of 1:24. For example, 10ml 25X wash buffer + 240ml deionized water. The diluted wash buffer can be stored at 2-8°C for 3 days.

8. Assay Procedure

1. Make sure the kit and all test samples are returned to room temperature before use. Shake each reagent gently before adding into the well.
2. Take the microplate from the zip-bag, and mark the location of the sample.
3. Add **100ul positive control** into two wells (100ul per well). Add **100ul negative control** into another two wells (100ul per well).
4. Add **100ul diluted serum sample** (40X, for example: 3ul serum + 117ul sample buffer) into above wells (100ul per well).
5. Cover the plate with plate cover and incubate at **room temperature (22-27 °C) for 60min**.
6. Pour the liquid out from the wells and wash with wash buffer (300ul per well) for 5 times. Tap the residue liquid against absorbent paper to make sure the plate is dry after washing. Note: The condensed wash buffer should be dilute for 25 times with pure water.
7. Add **100ul of enzyme conjugate** into each well. Cover the plate again and then incubate at **room temperature again for 60min**.
8. Repeat the washing step again.
9. Add the **100ul TMB substrate** into each well, 100ul per well.
10. Cover the plate again and then incubate at **room temperature again for 10min**.
11. Add **50ul stop solution** into each well to stop the reaction.
12. Using ELISA reader to read the plate at 450nm.

9. Result Determination

- The experiment is effective when the OD of positive control is greater than 2 times of the OD of negative control.
- **Positive**: OD of sample ≥ (OD of negative control X 2)
- **Negative**: OD of sample < (OD of negative control X 2)

10. Storage and expiration

The kit shall be store at 2-8°C, avoid direct sunlight.

The valid period is 12 months.

11. References

(1) https://en.wikipedia.org/wiki/Porcine_circovirus
(2) https://www.merckvetmanual.com/generalized-conditions/porcine-circovirus-diseases/overview-of-porcine-circovirus-diseases