IBRV antibody ELISA Kit

IBRV Ab Test

Product Number: E40121
Product Unit: 1 plate-96wells, 2 plates-192wells, 5 plates-480wells,
for 10plates, 2 units of 5plates will be provided

Manufacturer Information
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For veterinary use only

1. Introduction

The infectious bovine rhinotracheitis (IBR) virus belongs to the group of herpes viruses. It causes in cattle a severe disease predominantly in the upper respiratory tract. Morbidity rate is 100 percent, mortality – depending on hygienic and other factors – ranges from 0-15 percent. The IBR virus is serologically indistinguishable from the infect pustulat vulvovaginitis (IPV) virus which causes disorders of the genital tract in both male and female cattle. Possibly the IPV virus is the older of the two.

2. Description of Test

This IBRV ELISA kit is based on blocking ELISA, which is designed to detect IBRV antibody in bovine serum samples. The 96well microtiter plate was precoated with IBRV protein. During testing, samples and enzyme conjugates are added into the microplate wells, if there exist high titer of IBRV antibody in the samples, which will compete with the enzyme conjugate antibody, after washing step, the TMB substrate was added. The measured intensity is negatively proportional to the amount of antibody present in the sample.

This ELISA kit can be used to detect IBRV antibody in **bovine serum**.

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	1 plate	2 plates	5 plates
1	Microplate pre-coated with antigen	1 X 96 wells	2 X 96 wells	5 X 96 wells
2	Positive Control	1 X 1 ml	1 X 1 ml	2 X 1 ml
3	Negative Control	1 X 1 ml	1 X 1 ml	2 X 1 ml
4	Enzyme Conjugate	1 X 12 ml	1 X 12 ml	3 X 12 ml
5	TMB Substrate	1 X 25 ml	1 X 25 ml	3 X 25 ml
6	Stop Solution	1 X 20 ml	1 X 20 ml	2 X 20 ml
7	25X Wash Buffer	1 X 30 ml	1 X 30 ml	3 X 30 ml

8	Sample Buffer	1 X 30 ml	1 X 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 antibody, sample buffer, and TMB substrate can be used directly.
- 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. Take the microplate from the zip-bag, and mark the location of the sample.
2. **Sample preparation**: centrifuge the blood at 4000rpm for 10 min, then take 60ul of supernate into the sample diluent plate, then add equal volume of sample buffer, for example: 60ul serum + 60ul sample buffer.
3. Take out the pre-coated plate, add **50ul of positive control** into two wells, then **add 50ul of negative control** to another two wells, then **add 50ul of diluent sample** into the rest wells, and then add **50ul of enzyme conjugate** into these wells. Cover the plate and incubate at **37°C for 30min**.
4. Pour the liquid out from the wells and wash with wash buffer (300ul per well) for 4-5 times. Tap the residue liquid against absorbent paper to make sure the plate is dry after washing.
5. **Add the TMB substrate** into each well, **100ul per well**. Cover the plate again and then incubate at **room temperature for 10min**. note: avoid direct sunlight.
6. Add **50ul stop solution** into each well to stop the reaction.
7. Using ELISA reader to read the plate at 450nm.

9. Result Determination

Valid criteria: Mean OD of **negative control > 0.5**; Mean **IN% of positive control > 0.5.**

$$\textbf{Block rate [IN]} = \frac{\textbf{(Mean OD of negative control – Mean OD of samples)}}{\textbf{Mean OD of negative control}} \times 100\%$$

Determination of samples:

Negative	Doubt	Positive
IN <= 30	30 < IN < 40	IN >= 40

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://www.oie.int/en/what-we-do/standards/codes-and-manuals/terrestrial-code-online-access/?id=169&L=0&htmfile=chapitre_ibr_ipv.htm
(2) https://pubmed.ncbi.nlm.nih.gov/165129/