

Bovine Pregnancy-Associated Glycoprotein (PAG) Rapid Test Kit

Technical Manual

(GICA)



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Product Information

Overview

Traditionally, pregnancy in cows has been diagnosed through rectal palpation or ultrasonography (TU). Our Bovine Pregnancy-Associated Glycoprotein (PAG) Rapid Test Kit provides an accurate method for detecting pregnancy-associated glycoproteins (PAG) in whole blood, or serum, starting from 28 days after insemination throughout the entire pregnancy period. This kit provides a crucial tool for detecting pregnant cows, helping to shorten cycles, increase breeding rates, and manage breeding costs.

Principle

Pregnancy-associated glycoproteins (PAG) are specific proteins that appear in the peripheral blood of ruminant animals after pregnancy. They play a crucial role during the pregnancy process and are secreted by the

binucleate cells of the embryo.

The kit uses colloidal gold immunochromatography assay (GICA). After being added to sample hole("S"), the sample will move along the nitrocellulose membrane with the gold markers. If there are PAG in the sample, they will bind with the gold markers as well as antigens on the test ("T") line, resulting in the appearance of a colored test ("T") line. If not, no color reaction will be produced.

Content

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Test device (with disposable dropper)	8
Assay diluent	8
Disposable vacuum blood collection tube (without anticoagulant)	8
Needle	8
Disposable dropper	8
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Storage Conditions

The kit shall be stored at 2°C to 30°C (35.6°F to 86°F) in dry environment. Avoid freezing.

Shelf life: 24 months. The date of manufacture is presented in the label of the box.

Preparation of Sample

This test card is only suitable for testing **whole blood or serum in female cattle from 28 days after mating to parturition** (The test kit is only suitable for pregnancy detection in female cattle after mating and is not intended for testing non-mated cows, bulls, or calves.). Milk and urine from cows cannot be used as samples. (Recommended to use serum testing.)

Blood collection: Blood is collected from root of the cattle tail. The tail of the cow is lifted, and a blood collection needle is inserted into the depression at the midpoint

between the 3rd and 4th coccygeal vertebrae, about 10 cm from the base of the tail. The other end is connected to a blood collection tube, and 1-2mL of blood is drawn.

Whole blood: Blood samples should be collected in a disposable vacuum blood collection tube. (The blood collection tube can be with or without anticoagulant.) Samples collected without anticoagulants should be used immediately. The anticoagulated blood collected can be used within 24 hours after sampling.

Serum: Collect 2-3mL of blood using a collection tube without anticoagulant, let it stand for 30 minutes, and then centrifuge at 4000 rpm for 10 minutes. (Alternatively, the blood can be left undisturbed at 25-40°C, allowing the serum to naturally separate.) Collect the supernatant as the serum sample. Short-term storage can be done at 2-8°C, while long-term storage requires -20°C. Serum should be clear and bright, free from hemolysis and contamination.

Please note that sample should be return to room temperature (15-30°C) before use.

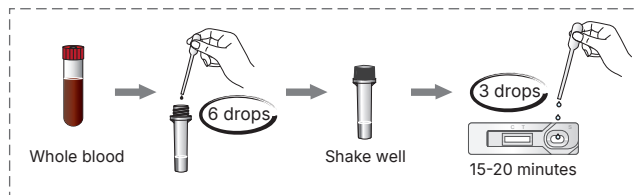
Test Methods

For whole blood sample:

- (1)Use the provided dropper to slowly and vertically add 6 drops of the prepared whole blood into the assay diluent tube, then mix well. What obtained is processed blood sample. (Note: this step is very important; the sample should not be added too much)
- (2)Tear the foil pouch, take out the test card and put it on a flat surface.
- (3)Using another dropper provided inside the bag, slowly add 3 drops of processed blood sample obtained in step 1 (or pipette 70μL using a micropipette)

into the sample well.

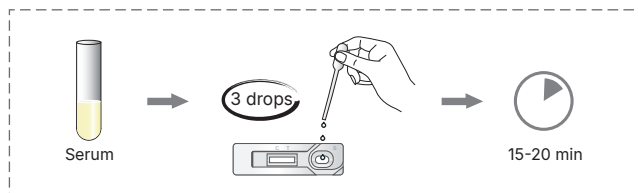
(4) Wait for 15-20 minutes to assess the test results.



For serum sample:

(1) Tear the foil pouch, take out the test card and put it on a flat surface.

(2) Use the provided dropper to aspirate the serum sample, dispense 3 drops (or pipette 70µL using a micropipette) into the sample hole ("S") of the test card, and wait for 15-20 minutes to assess the test results.

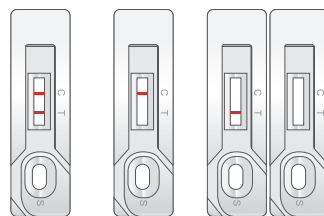


Results Judgement

Negative: Only control ("C") line appears in the result window, **not considered to be pregnant.**

Positive: Both test ("T") line and control ("C") line appear in the result window, **considered to be pregnant.** The higher the PAG content, the darker the color of the test ("T") line.

Invalid: If the control ("C") line does not appear, the result might be considered invalid.



Positive Negative Invalid

Limitation of the Test Method

This test method is only intended for qualitative detection of pregnancy-associated glycoproteins (PAG).

This product cannot differentiate false positives caused by early embryo death. It should not be used as the sole basis for early pregnancy detection in female cattle. Comprehensive consideration should be given to factors such as symptoms and physical signs.

Notice

1) Please read the instructions carefully before testing. And a variety of reagents are only used for this experiment.

2) Like all diagnostic tests, definitive diagnoses should not be based on the results of a single test.

3) Due to the limitations of the testing methodology for the test reagents, operating personnel should pay more attention to negative results of samples. It is recommended to combine other test results for comprehensive judgment. For negative results with doubts, it is advisable to confirm them using other methods.

4) Due to the following reasons, false negative results may occur in the test: unknown components may block the binding reaction between antibodies and antigens; early pregnancy-associated glycoproteins become unstable or degrade with changes in time and temperature, making them unrecognizable by antibodies. These factors may increase the likelihood of false negative results.

5) The kit should be allowed to return to room temperature after being removed from the refrigerator before opening. Once opened, it should be used as quickly as possible to avoid becoming ineffective due to moisture.

6) Avoid using expired or damaged products.

7) Avoid using samples that are contaminated, turbid,

severely hemolytic, and have a large amount of blood lipids.

8) Avoid touching the white nitrocellulose membrane in the middle of the detection card.

9) The waste shall be regarded as pollutants. Please dispose of them properly in accordance with the relevant local regulations.