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**INDUSTRY STANDARD OF THE
PEOPLE'S REPUBLIC OF CHINA**

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Protocol of milk ring test for brucellosis

布氏杆菌病全乳环状试验方法

(English Translation)

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Table of Contents

- Foreword
- 1 Scope
- 2 Reagents and materials
 - 2.1 Reagents
 - 2.2 Milk sample
 - 2.3 Materials
- 3 Method of operation
- 4 Result determination
 - 4.1 Principle of judgment
 - 4.2 Judging criteria

Foreword

This standard is proposed and managed by the National Certification and Accreditation Administration.

This standard was drafted. Tianjin Entry-Exit Inspection and Quarantine Bureau of the People's Republic of China.

1 Scope

This standard specifies the operational technical requirements for the Brucellosis Whole Milk Ring Test.

This standard applies to the monitoring and diagnosis of brucellosis in dairy cattle. The screening test for brucellosis in lactating cows is not applicable to breast cancer.

Inflammation and other breast disease cows' milk, colostrum, skim milk and boiled milk are also not suitable for use in spoilage, sour and frozen milk.

2.1 Reagents

Standard antigen. tetrazolium staining antigen (red), or hematoxylin staining antigen (blue), stored in the dark at 2 °C ~ 8 °C.

2.2 Milk sample

The test milk should be fresh whole milk. When collecting milk, wash the breasts of the dams with warm water, then dry them with clean gauze and squeeze the milk into cleanliness.

In the utensils. If the milk sample is stored at 2 ° C ~ 8 ° C, it can be used within 7 days.

2.3 Materials

2.3.1

Micropipettes and tips.

2.3.2

Agglutination test tube. glass, the specification is 13mm x 100mm, round bottom, clean.

2.3.3

Test tube rack.

2.3.4

Water bath.

2.3.5

Pipette or graduated pipette.

3 Method of operation

3.1

Take 1 mL of milk sample and add it to the sterilization agglutination test tube.

3.2

After fully shaking and mixing the standard antigen, remove 50 μ L of the added milk sample and mix well.

3.3

Set in a 37 ° C ~ 38 ° C water bath for 60 min.

3.4

Take out the test tube, avoid oscillation, and make a judgment immediately.

4.1 Principle of judgment

Determined by the color of the cream and the milk column. When the antigen was stained with tetrazolium, the color was red, and the color of the antigen stained with hematoxylin was blue.

When the milk with less fat or no fat is positive, the cells are agglomerated and sink the bottom of the tube. The judgment is based on the reaction of the milk column.

4.2 Judging criteria

4.2.1

Positive reaction (+++). The upper layer of milk in the milk column forms a red or blue ring with a clear white color and a clear definition.

4.2.2

Positive reaction (++). The annulus of the cream layer is red or blue, but not significant, and the breast is slightly colored.

4.2.3

Weak positive reaction (+). The ring of the cream layer is lighter in color but slightly darker than the color of the milk column.

4.2.4

Suspected reaction ($\pm$). The color of the ring of the cream layer is not obvious, the boundary with the milk column is unclear, and the milk column does not fade.

4.2.5

Negative reaction (-). There is no change in the upper layer of the milk column, and the color of the milk column is uniform.