

# **Enrofloxacin (ENR) Rapid Test Kit**

Technical Manual (GICA)



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## 1 Principle and Application |-

The test kit is used for the detection of enrofloxacin (ENR) in samples such as tissues (fish, shrimp, crab, livestock and poultry meat, viscera), as well as honey.

The kit is developed using the principle of competitive colloidal gold Immunochromatographic Assay (GICA). After the sample solution is added to sample hole, if ENR is present, it will bind with gold labeled antibodies, thereby preventing the labeled antibodies from binding to the ENR conjugates on the nitrocellulose membrane.

If the content of ENR in sample solution is less than detection limit, it will make the test ("T") line colored, and the result is negative. If the content is greater than detection limit, no color reaction will be produced, and the result is positive.

#### 2 Technique Data I-

**Kit Sensitivity:** 2ppb (ppb=μg/kg)

**Limit of detection (LOD):** using fluoroquinolones (QNs) as the standard.

Tissues	6ppb
Honey	6ppb
Milk	4ppb
Poultry eggs	6ppb

#### 3 Kit Content |

Package specification	20T/Kit	40T/Kit
Test device (with disposable dropper)	20	40
Sample reconstitution buffer	30mL×1	30mL×1
Instruction	1	1

# 4 Materials Required but Not Supplied 1-

**Equipment:** grinder (for crushing solid samples), vortex mixer (for shake and mix), graduated transfer pipette, and balance with a division value of 0.01 g.

Micropipettes: single-channel (20-200μL and 100-1000μL)

Reagents: Glacial Acetic Acid (analytical grade).

# 5 Sample Pre-treatment |-

#### 5.1 Instructions

Labware must be clean. Use disposable pipette tips to avoid contamination of interference results.

## 5.2 Solution preparation before sample pre-treatment

Solution 1: 0.1% Glacial Acetic Acid Solution

Measure 0.1mL of glacial acetic acid (analytical grade). Add it to deionized water and mix thoroughly. Make up the volume to 100mL with deionized water to prepare a 0.1% glacial acetic acid solution.

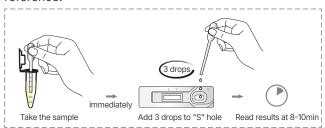
#### 5.3 Sample pretreatment step:

#### 5.3.1 Animal Tissue, Honey, and Egg:

- (1) Take 1±0.05g of defatted homogenized tissue sample and place it in a 15mL centrifuge tube. Add 1mL of 0.1% glacial acetic acid solution (Solution 1) and shake for 5 minutes until the sample forms a uniform paste. Centrifuge at room temperature at 4000r/min for 5 minutes.
- (2)Transfer 0.1mL of the supernatant to another 1.5mL centrifuge tube, add 0.2mL of sample reconstitution buffer, and mix thoroughly. The solution is now ready for analysis.
- 5.3.2 Fresh Milk (It is not recommended to use raw milk that has been frozen and has obvious particles, which can easily lead to test failure):
- (1) Take a fresh milk sample and dilute it with deionized water at a ratio of 1:1. Mix well and set aside for use.

# 6 Test Steps |-

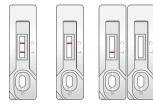
- (1) Tear the foil pouch, take out of the test card, and put it on a flat, clean work surface.
- (2)Pipette the prepared sample solution with the provided dropper, then add 3 drops (approximately  $60\mu L$ ) vertically and slowly (Avoid the generation of bubbles) into the sample hole ("S").
- (3)Read the result at room temperature in 8 to 10 minutes. Results over 10 minutes can only be used as reference.





#### 7 Results Judgement I-

Negative: Test("T") line and control( "C" ) line both appear in the result window. It indicates that the concentration of ENR in the sample is below



Negative Positive Invalid

the detection limit, or absent.

 $\begin{tabular}{ll} \textbf{Positive:} Only control ( "C" ) line appears in the result window. It indicates that the concentration of ENR in the sample is above the detection limit. \\ \end{tabular}$ 

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

#### 8 Notice |-

- 8.1 Don't use the expired or damaged products.
- 8.2 When the test card is taken out of the refrigerator, it should be restored to the room temperature and then opened. The opened test card should be used as soon as possible to avoid failure after being affected by moisture.
- 8.3 Avoid touching the white nitrocellulose membrane in the middle of the detection card.
- 8.4 In order to avoid cross-contamination, the droppers cannot pipet another Solution after pipetting one.
- 8.5 The sample solution to be examined needs to be clear and free of turbid particles. Otherwise, it is prone to lead to blockage, non-obvious color development and other abnormalities, affecting the determination of the experimental results.

# 9 Storage Conditions |

The kit shall be stored at 2°C to 30°C (35.6°F to 86°F)

in dry environment.

Shelf life: 12 months. The date of manufacture is presented in the label of the box.