



## ***E.coli* Residual DNA Detection Kit**

*E.coli* Residual DNA Detection Kit is designed to detect residual *E.coli* DNA in biological products during production.

Catalog number: ARG83091

Package: 100 tests

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### **INTRODUCTION**

*E.coli* Residual DNA Detection Kit is designed for the quantitative detection of *E.coli* host cell DNA in intermediates, semi-finished products and finished products of various biological products.

*E.coli* Residual DNA Detection Kit adopts the principle of Taqman probe to quantitatively detect *E.coli* residual DNA in samples. The kit is a rapid, specific and reliable device, with the minimum detection limit reaching fg level.

### **PRINCIPLE OF THE ASSAY**

*E.coli* Residual DNA Detection Kit is a test kit that uses quantitative polymerase chain reaction (qPCR) technology to detect residual *E.coli* DNA. *E.coli* Residual DNA Detection Kit includes a set of primers and probes that can amplify and detect specific sequences of *E.coli* DNA. qPCR is a PCR technique that simultaneously amplifies and detects DNA by monitoring the accumulation of product with the use of a fluorescent dye. This kit has high specificity and sensitivity, is easy to use, and suitable in laboratories.

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### **MATERIALS PROVIDED & STORAGE INFORMATION**

Store the unopened kit at -20 °C. Use the kit before expiration date.

| <b>Component</b>                   | <b>Quantity</b> | <b>Storage information</b> |
|------------------------------------|-----------------|----------------------------|
| 10x <i>E.coli</i> DNA standard     | 50 µl           | -20°C                      |
| <i>E.coli</i> Primer and probe mix | 550µl           | -20°C (protect from light) |
| 2x qPCR Reaction Buffer            | 1.1 ml          | -20°C (protect from light) |
| DNA Dilution buffer                | 3 x 1 ml/vails  | -20°C                      |
| ROX (High)                         | 50µl/vails      | -20°C (protect from light) |
| ROX (Low)                          | 50µl/vails      | -20°C (protect from light) |

### **MATERIALS REQUIRED BUT NOT PROVIDED**

- PCR machine
- Pipettes and pipette tips
- DNase/RNase-Free Water
- PCR tube

### **TECHNICAL HINTS AND PRECAUTIONS**

- Wear protective gloves, clothing, eye, and face protection especially while handling blood or body fluid samples.
- Store the kit at -20°C at all times.
- All reagents must be kept on ice during the entire experiment.
- Once the assay has been started, all steps should be completed without interruption.
- It is highly recommended that the standards and samples be assayed in triplicates.
- Change pipette tips between the addition of different reagent or samples.

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### **REAGENT PREPARATION**

- **Standards:** Dilute 10x *E. coli* DNA standard with DNA Dilution buffer to yield a S5 concentration of 300 pg/μl. The DNA Dilution buffer serves as zero standard (0 pg/ml), and the rest of the standard 10-fold serial

Dilute *E. coli* DNA standard as according to the table below:

| <b>Standard</b> | <b>DNA Conc.</b> | <b>μl of DNA Dilution buffer</b> | <b>μl of standard</b> |
|-----------------|------------------|----------------------------------|-----------------------|
| S5              | 300 pg/μl        | 90 μl                            | 10 μl (stock)         |
| S4              | 30 pg/μl         | 90 μl                            | 10 μl (S5)            |
| S3              | 3 pg/μl          | 90 μl                            | 10 μl(S4)             |
| S2              | 0.3 pg/μl        | 90 μl                            | 10 μl(S3)             |
| S1              | 0.03 pg/μl       | 90 μl                            | 10 μl(S2)             |
| S0              | 0 pg/μl          | 0 μl                             | 100μl                 |

- **Sample:** The suggested concentration of the sample is 30 ng/μl.

### **ASSAY PROCEDURE**

- 1 Prepare qPCR mix buffer:

|                                     |                       |
|-------------------------------------|-----------------------|
| 2x qRCR Reaction Buffer             | 10μl                  |
| <i>E. coli</i> Primer and probe mix | 4.6μl                 |
| ROX *                               | 0.4μl                 |
| <b>Total</b>                        | <b>15μl (1 wells)</b> |

- \* Choose the appropriate ROX (High or Low) to match the PCR machine. If the PCR machine does not require ROX, adjust the volume with DNase/RNase-free water to obtain a final volume of 15μl.
- 2 Mix 15μl qPCR mix buffer with 5μl diluent standard / sample / blank in PCR tube. The final volume should be 20 μl.
  - 3 Initial denaturation: 95°C, 10 min
  - 4 PCR cycle:
    - Denaturation: 95°C, 15 sec
    - Elongation: 60°C, 1 min, for **40 cycle** and select fluorescent FAM.