

[Cat. No.] **K-2905**

Introduction

AccuPower® EAE PCR Kit is a ready-to-use premix for PCR that can be used to detect *E. coli* in livestock by specifically amplifying the *E. coli* attachment effacement (*EAE*) gene.

E. coli enters the organs of animals and causes lesions. It is also a source of many infections such as acute urinary tract infections, peritonitis, hepatitis, sepsis, and meningitis. *E. coli* can be classified into various categories: enteropathogenic *E. coli* (EPEC), enterotoxigenic *E. coli* (ETEC), enteroinvasive *E. coli* (EIEC), enterohemorrhagic *E. coli* (EHEC), etc. Among those, intimin, the product of *EAE* gene, causes EPEC adhesion to intestinal epithelial cells, which may develop into microvilli atrophy, polar actin, etc. Furthermore, intimin protein can also induce pathogenic characteristics to EHEC and EPEC by inducing bacterial adhesion, which develops into A/E lesions.

This product contains vacuum-dried components specific to *EAE* gene including DNA polymerase, primers, dNTPs, and reaction buffer required for PCR. This ready-to-use kit simplifies preparation of PCR mixture as the user only has to add template DNA and nuclease-free water. After the reaction, since tracking dye is included, the samples can be applied directly on agarose gel for analysis without adding extra solution.

Features & Benefits

- **Convenience & Reproducibility:** All reactants necessary for PCR including primers are lyophilized in each PCR tube, providing reproducible results in a convenient way.
- **Sensitivity:** By applying the patented PyroHotStart (Enzyme-mediated HotStart) technology that minimizes non-specific reactions and maximizes reaction efficiency, only the target gene can be effectively amplified even with a trace amount of template DNA.
- **Stability:** Included stabilizer in the PCR reaction mixture provides increased stability compared to solution-type products.

Composition

Composition	20 µl reaction
Top DNA Polymerase	1 U
dNTPs (dATP, dCTP, dGTP, dTTP)	Each 250 µM
Reaction buffer with 1.5 mM MgCl ₂	1X
Stabilizer and tracking dye	0
EAE Forward primer	0.5 µM
EAE Reverse primer	0.5 µM

* **Note:** For research use only. Not for use in diagnostic or therapeutic procedures.

Specifications

Top DNA Polymerase	
5'→3' exonuclease activity	No
3'→5' exonuclease activity	No
3'-A overhang	Yes
Fragment size	790 bp

Storage

Store at -20°C. If stored in the recommended temperature, this product will be stable until the expiration date printed out on the label.

Online Resources



English

Visit our **product page** for additional information and protocols

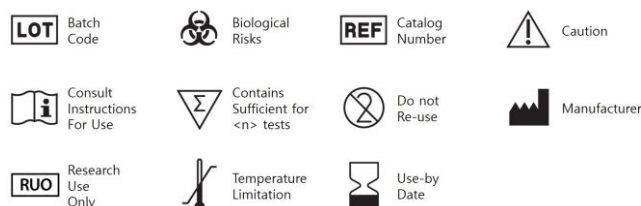
Ordering Information

Description	Cat. No.
AccuPower® EAE PCR Kit, 0.2 ml thin-wall 8-tube strips with attached cap / 96 tubes	K-2905




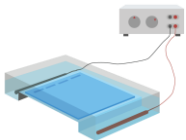
Notice

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Explanation of Symbols



Experimental Procedures

Steps		Procedure Details																								
1	 <p>Add template DNA</p>	<p>1. After preparing the template DNA and nuclease-free water, add the template DNA to the <i>AccuPower®</i> EAE PCR Kit.</p>																								
2	 <p>Preparation of reaction mixture</p>	<p>2. Add nuclease-free water into PCR tubes to make a total volume of 20 µl. (Do not include the volume of the dried premix in the PCR tubes.)</p> <p>3. Completely dissolve the vacuum-dried pellet by vortexing, and briefly spin down.</p>																								
3	 <p>Incubate reactions in a thermal cycler</p>	<p>4. Place PCR tubes on the thermal cycler.</p> <p>5. Perform the reaction under the following conditions.</p> <table border="1"> <thead> <tr> <th>Step</th> <th>Temperature</th> <th>Time</th> <th>Cycles</th> </tr> </thead> <tbody> <tr> <td>Pre-denaturation</td> <td>94°C</td> <td>15 min</td> <td>1 cycle</td> </tr> <tr> <td>Denaturation</td> <td>94°C</td> <td>30 sec</td> <td></td> </tr> <tr> <td>Annealing</td> <td>55°C</td> <td>30 sec</td> <td>40 cycles</td> </tr> <tr> <td>Extension</td> <td>72°C</td> <td>60 sec</td> <td></td> </tr> <tr> <td>Final extension</td> <td>72°C</td> <td>10 min</td> <td>1 cycle</td> </tr> </tbody> </table> <p>* Note: Users can adjust the protocol according to their instrument and template sequences to get optimal results.</p>	Step	Temperature	Time	Cycles	Pre-denaturation	94°C	15 min	1 cycle	Denaturation	94°C	30 sec		Annealing	55°C	30 sec	40 cycles	Extension	72°C	60 sec		Final extension	72°C	10 min	1 cycle
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4	 <p>Analyze with gel electrophoresis</p>	<p>6. After the reaction, maintain the reaction mixture at 4-8°C.</p> <p>7. Load samples on agarose gel without adding a loading-dye mixture, and perform gel electrophoresis for analysis.</p>																								