

[Cat. No.] **K-2986**

Introduction

AccuPower® Shewanella putrefaciens Real-Time PCR Kit is a product that can detect *Shewanella putrefaciens* (*S. putrefaciens*), which causes disease in fish, by real-time PCR.

S. putrefaciens is a gram-negative bacterium that is generally pathogenic to fish and rarely to humans. Infected fish seems symptoms such as skin ulcers, darkening of the body, exophthalmos, and abdominal distension, in addition, *S. putrefaciens* causes keratitis in humans. In case of farming loach, carp, particular attention should be paid to *S. putrefaciens* infections.

This product contains all Real-time PCR components specific to *S. putrefaciens*, including DNA polymerase, dNTPs, and reaction buffer. The users can easily prepare reaction mixture simply by adding template DNA, Oligo Mix and DEPC-D.W.

Features & Benefits

- Convenience: All necessary reactants for real-time PCR are included in a tube (i.e., Master Mix Type), allowing the users to perform reaction simply by adding template DNA, Oligo Mix, and DEPC-D.W.
- Sensitivity: By using BIONEER's HotStart Taq DNA Polymerase 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.

Components

Components	Amount
2X Master Mix	625 µl x 2
Oligo Mix	500 µl
DEPC-D.W.	1.8 ml
Positive Control (1x10 ⁸ copies/µl)	50 µl

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

Composition

Composition	25 µl reaction
2X Master Mix	
Taq DNA Polymerase	2 U
dNTPs (dATP, dCTP, dGTP, dTTP)	Each 300 µM
Reaction buffer with 2 mM MgCl ₂	1X
Oligo Mix	
<i>S. putrefaciens</i> Forward primer	0.32 µM
<i>S. putrefaciens</i> Reverse primer	0.32 µM
<i>S. putrefaciens</i> Probe (FAM)	0.32 µM
ROX dye	1X

Specifications

Taq DNA Polymerase	
5'→3' exonuclease activity	Yes
3'→5' exonuclease activity	No
3'-A overhang	Yes

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



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

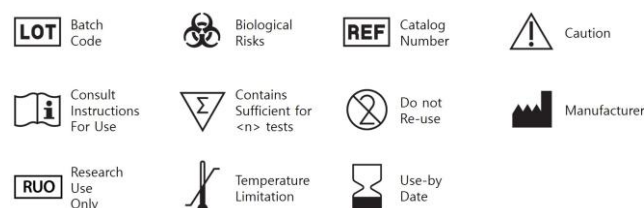
Ordering Information

Description	Cat. No.
AccuPower® Shewanella putrefaciens Real-Time PCR Kit, 1.25 ml of 2X Master Mix solution, 100 tests	K-2986




Notice

BIONEER corporation reserves the right to make corrections, modifications, improvements and other changes to its products, services, specifications or product descriptions at any time without notice.

Explanation of Symbols



Experimental Procedures

Steps		Procedure Details															
1	 Preparation of reaction mixture	<p>1. Thaw all components of <i>AccuPower</i>® <i>Shewanella putrefaciens</i> Real-Time PCR Kit on ice and mix thoroughly before use. Then, briefly spin down all components.</p>															
2	 Composition of reaction mixture	<p>2. Add all components into PCR tubes (not provided) or a plate (not provided) referring to the following list of components (Based on 1 test).</p> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: left;">Components</th> <th style="text-align: left;">Amount</th> </tr> </thead> <tbody> <tr> <td>2X Master Mix</td> <td>12.5 µl</td> </tr> <tr> <td>Oligo Mix</td> <td>5 µl</td> </tr> <tr> <td>Template DNA</td> <td>1-5 µl</td> </tr> <tr> <td>DEPC-D.W.</td> <td>Variable</td> </tr> <tr> <td>Total volume</td> <td>25 µl</td> </tr> </tbody> </table>	Components	Amount	2X Master Mix	12.5 µl	Oligo Mix	5 µl	Template DNA	1-5 µl	DEPC-D.W.	Variable	Total volume	25 µl			
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3	 Real-time PCR	<p>3. Place PCR tubes or plate on the Real-Time Quantitative thermal cycler.</p> <p>4. Perform the reaction under the following conditions.</p> <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: left;">Step</th> <th style="text-align: left;">Temperature</th> <th style="text-align: left;">Time</th> <th style="text-align: left;">Cycles</th> </tr> </thead> <tbody> <tr> <td>Pre-denaturation</td> <td>95°C</td> <td>5 min</td> <td>1 cycle</td> </tr> <tr> <td>Denaturation</td> <td>95°C</td> <td>10 sec</td> <td rowspan="2">45 cycles</td> </tr> <tr> <td>Annealing & Extension</td> <td>55°C</td> <td>20 sec</td> </tr> </tbody> </table> <p>* Note: Users can adjust the protocol according to their instrument and template sequences to get optimal results.</p> <p>5. After the reaction is completed, analyze the results.</p>	Step	Temperature	Time	Cycles	Pre-denaturation	95°C	5 min	1 cycle	Denaturation	95°C	10 sec	45 cycles	Annealing & Extension	55°C	20 sec
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