

[Cat. No.] **K-2990**

SRS Reverse primer	0.3 µM
SRS Probe (TET)	0.3 µM

Introduction

AccuPower® Salmon Disease Multiplex Master Mix is a real-time PCR product that can simultaneously detect diseases infecting salmon (Infectious Pancreatic Necrosis Virus (IPNV), Infectious Salmon Anaemia Virus (ISAV), Bacterial Kidney Disease (BKD), Salmonid Rickettsial Septicaemia (SRS)). Clinical signs of four diseases include swollen abdomen or eyes or darkening of the skin, pale gills, haemorrhage at the base of the fins, lesions of the skin and death in acute cases. These infectious diseases are causing and increasing huge economic losses in salmon farming industry especially Chile, as well as in Canada, Ireland, Scotland and Norway.

In this product, all elements (RTase, DNA polymerase, primers, dNTPs, reaction buffer) necessary for real-time PCR of 4 pathogens simultaneously or specifically are mixed in a tube, so the user can only add template DNA/RNA, oligo mix and DEPC-D.W. You can easily prepare a PCR reaction solution.

Applications

- Qualitative analysis of multiplex real-time PCR for IPNV, ISAV, BKD, SRS pathogen.

Components

Components	Amount
2X Master Mix	625 µl x 2ea
Oligo Mix	500 µl
Positive control (1*10 ⁸ copies/µl)	50 µl
DEPC-D.W.	1.3 ml

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

Composition

Composition	25 µl reaction
RocketScript™ Reverse transcriptase	0.5 U
2X Master Mix	
Taq DNA Polymerase	3.5 U
dNTPs (dATP, dCTP, dGTP, dTTP)	Each 300 µM
Reaction buffer with 2 mM MgCl ₂	1X
IPNV Forward primer	0.2 µM
IPNV Reverse primer	0.2 µM
IPNV Probe (FAM)	0.2 µM
ISAV Forward primer	0.3 µM
Oligo Mix	
ISAV Reverse primer	0.3 µM
ISAV Probe (Cy5)	0.3 µM
BKD Forward primer	0.2 µM
BKD Reverse primer	0.2 µM
BKD Probe (Texas Red)	0.2 µM
SRS Forward primer	0.3 µM

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® Salmon Disease Multiplex Master Mix, 1.25 ml of 2X Master Mix solution, 100 tests	K-2990




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

Batch Code	Biological Risks	Catalog Number	Caution
Consult Instructions For Use	Contains Sufficient for <n> tests	Do not Re-use	Manufacturer
Research Use Only	Temperature Limitation	Use-by Date	

Experimental Procedures

Steps		Procedure Details																																	
1	 Preparation of PCR material	<p>1. Before use, thaw all components of AccuPower® Salmon Disease Multiplex Master Mix on ice and mix them thoroughly. Then, briefly spin down components.</p>																																	
2	 PCR reaction solution composition	<p>2. Add all components into PCR tubes or a plate 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: center;">Material</th> <th style="text-align: center;">Volume (µl)</th> </tr> </thead> <tbody> <tr> <td>2X Mater Mix</td> <td style="text-align: center;">12.5</td> </tr> <tr> <td>Oligo Mix</td> <td style="text-align: center;">5</td> </tr> <tr> <td>Template DNA/RNA (Positive Control)</td> <td style="text-align: center;">1~5</td> </tr> <tr> <td>DEPC-DW</td> <td style="text-align: center;">Up to 25</td> </tr> <tr> <td>Total reaction volume</td> <td style="text-align: center;">25</td> </tr> </tbody> </table>	Material	Volume (µl)	2X Mater Mix	12.5	Oligo Mix	5	Template DNA/RNA (Positive Control)	1~5	DEPC-DW	Up to 25	Total reaction volume	25																					
Material	Volume (µl)																																		
2X Mater Mix	12.5																																		
Oligo Mix	5																																		
Template DNA/RNA (Positive Control)	1~5																																		
DEPC-DW	Up to 25																																		
Total reaction volume	25																																		
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: center;">Step</th> <th style="text-align: center;">Temperature</th> <th style="text-align: center;">Time</th> <th style="text-align: center;">Cycles</th> </tr> </thead> <tbody> <tr> <td>Reverse Transcription</td> <td style="text-align: center;">50 °C</td> <td style="text-align: center;">15 min</td> <td style="text-align: center;">1 cycle</td> </tr> <tr> <td>Pre-denaturation</td> <td style="text-align: center;">95 °C</td> <td style="text-align: center;">5 min</td> <td style="text-align: center;">1 cycle</td> </tr> <tr> <td>Denaturation</td> <td style="text-align: center;">95 °C</td> <td style="text-align: center;">10 sec</td> <td rowspan="2" style="text-align: center;">45 cycles</td> </tr> <tr> <td>Annealing& Extension</td> <td style="text-align: center;">60 °C</td> <td style="text-align: center;">20 sec</td> </tr> <tr> <td colspan="4">Scan</td> </tr> </tbody> </table> <p>* Note: Users can adjust the protocol according to their instrument and template sequences to get optimal results.</p> <ul style="list-style-type: none"> Perform Real-Time PCR by selecting a total of 4 types of fluorescence. <table border="1" style="width: 100%; border-collapse: collapse;"> <thead> <tr> <th style="text-align: center;">Target</th> <th style="text-align: center;">Flourescence</th> </tr> </thead> <tbody> <tr> <td style="text-align: center;">IPNV</td> <td style="text-align: center;">FAM</td> </tr> <tr> <td style="text-align: center;">ISAV</td> <td style="text-align: center;">Cy5</td> </tr> <tr> <td style="text-align: center;">BKD</td> <td style="text-align: center;">Texas_Red</td> </tr> <tr> <td style="text-align: center;">SRS</td> <td style="text-align: center;">TET</td> </tr> </tbody> </table> <p>5. After the reaction is completed, analyze the results.</p>	Step	Temperature	Time	Cycles	Reverse Transcription	50 °C	15 min	1 cycle	Pre-denaturation	95 °C	5 min	1 cycle	Denaturation	95 °C	10 sec	45 cycles	Annealing& Extension	60 °C	20 sec	Scan				Target	Flourescence	IPNV	FAM	ISAV	Cy5	BKD	Texas_Red	SRS	TET
Step	Temperature	Time	Cycles																																
Reverse Transcription	50 °C	15 min	1 cycle																																
Pre-denaturation	95 °C	5 min	1 cycle																																
Denaturation	95 °C	10 sec	45 cycles																																
Annealing& Extension	60 °C	20 sec																																	
Scan																																			
Target	Flourescence																																		
IPNV	FAM																																		
ISAV	Cy5																																		
BKD	Texas_Red																																		
SRS	TET																																		