DESCRIPTION

The STEC qPCR Kit is designed for identifying Siga-toxin-producing E. coli (STEC) strains in a one PCR amplification reaction using real-time quantitative polymerase chain reaction(qPCR) and probe label. The detection of target DNA confirms ingredient authenticity or prevents food fraud, ethical issues, or health concerns.

PRINCIPLE

Authenticating ingredients using real-time PCR is based on the amplification of a specific region of the relevant target genome. The amplified product is detected using target-specific fluorescent probes that bind to the amplified product. As the PCR product accumulates, there is an increased fluorescent signal from the bound probes. Monitoring the fluorescence intensities during the PCR run allows the detection of the accumulating PCR product in real time.

The STET qPCR Kit include internal controls (Positive and Negative), 2x qPCR super mix, Prime-Probe Mix in which the probe is labeled with FAM. These aids in the straightforward interpretation of the results (see the table "Summary of possible PCR outcomes").

APPLICATIONS

Detect STEC-derived DNA in plant, cannabis, grain, food and animal feed.

KIT CONTENTS

Name	Volume
Tribo™ 2x qPCP Super Mix	1.0 mL
Primer-probe Mix (FAM)	1 x 100 µL
Positive Control DNA	50 µL
Negative Control DNA	50 µL

Sufficient reagent for 100 x 20µL

The Probe is labeled with FAM

STORAGE CONDITIONS

The kit is shipped on ice and stored at -20°C for long-term storage. Shelf life of 12 months after receipt.

For research use only.

KEY FEATURES

❖ Highly sensitive and specific for STEC strains.

❖ High efficiency: the optimal buffer condition and specific engineered Taq DNA polymerase have increased the efficiency of PCR amplifications.

❖ Streamlined protocol: Just add DNA Template, and water.

❖ No cross reactivity with other species.

PCR PROTOCOL

1. Set up PCR reaction for each sample in 20 µL

Reaction Component	Volume (µL) Per Sample
Tribo™ 2x qPCP Super Mix	10.0
Primer-probe Mix (FAM)	1.0
DNA sample	1-3
Water	up to 20 µL

Internal control should be included as below:
Positive Control (3 µL DNA /reaction) Negative Control (3 µL DNA/reaction)
Blank Control: no DNA template

2. Suggested PCR conditions

Step	AmpliTaq Activation	PCR	
	HOLD	CYCLE (40 cycles)	
		Denature	Anneal/ Extend
Temperature	95 °C	95 °C	60 °C
Time	3 min	10 sec	30 min

DATA ANALYSIS

Positive Reaction: Sample Ct < or = 35， and Positive, Negative and Blank controls are normal.

Negative Reaction: Sample Ct > 35， and Positive, Negative and Blank controls are normal.

Repeat Reaction: If one of the control reactions is not normal, PCR reaction is failed, and should be repeated.