BIO-RAD

Which Method Fits Your STEC Testing Needs?

An Illustrative Guide to Understanding the Best Workflow for Your Lab

Shiga toxin-producing *E. coli* (STEC):
E. coli isolates that contain an *stx* gene, an *eae* gene, and genetically identify as one or more of the regulated serogroups

USDA Food Safety Inspection Service

Challenges with STEC Testing

High presumptive positives and low culture-confirmation rates

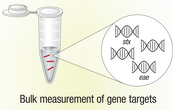
Are *stx*+/*eae*+ results from a single *E. coli* bacterium or multiple bacteria?

Cost-prohibitive and labor-intensive confirmatory method

Technology

Real-Time PCR

Enabled by increased fluorescence, which is proportional to the total amount of amplicon in a single PCR reaction.



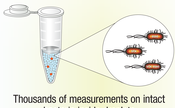
Bulk measurement of gene targets

96

Independent amplification events per run

Droplet Digital™ PCR (ddPCR™)

Enabled by thousands of PCR measurements performed on a single sample partitioned into thousands of nanoliter-sized droplets.



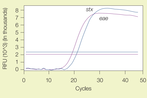
Thousands of measurements on intact bacteria inside droplets

20,000 x 96

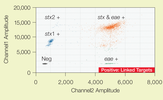
Amplification events per run

Data

Real-time PCR analysis does not distinguish between unlinked and linked *stx*+/*eae*+



ddPCR analysis enables individual identification of *stx* and *eae* presence and verifies their linkage



AOAC-Approved Workflow Methods

Enrichment

Screen with Real-Time PCR
(Bio-Rad or Other)

iQ-Check STEC VirX Kit + and
iQ-Check STEC SerO II Kit +

Confirm

6 hr: dd-Check STEC Kit or 3–5 days: Culture

Screen with ddPCR

dd-Check STEC Kit true positive

Confirm

iQ-Check STEC SerO II Kit

Matrix Compatibility

AOAC RESEARCH INSTITUTE Approved by AOAC for raw beef trim, MicroTally Swab, and fresh spinach

Key ddPCR Advantages

1. Improved Accuracy
2. Decreased Time to Results
3. Reduced Costs and Labor

Applicability

STEC Incidence Rate	Low Confirmation Rate	High Confirmation Rate
Low Presumptive Positive	Real-time PCR screening and ddPCR confirmation	Real-time PCR screening and ddPCR confirmation
High Presumptive Positive	ddPCR screening	Real-time PCR screening

Highlights

Real-Time PCR

- Ideal screening method for STEC
- Fast and high-throughput
- Does not distinguish whether *stx* and *eae* are harbored within a single organism that generated the presumptive-positive results

Droplet Digital PCR (ddPCR)

- Encapsulates intact bacteria to enable measurement of the coexistence of *stx* and *eae* on a single bacterium within a droplet
- Enhances screening accuracy by reducing false positives
- A culture-independent confirmatory method for true STEC-positives

Scan the QR Code to fill out the form for a discussion with a technical specialist on the return on investment (ROI) calculator

Visit **bio-rad.com/ddCheck** to learn more.

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