



***E. coli* O157:H7 Test Kit (20 cassettes)**

Cat. #: K ETLF-020

INTENDED USE

BioAssay Works[®] *E. coli* O157:H7 test is a qualitative rapid test for the detection of *E. coli* O157:H7 in enriched media. This test is for point-of-care use or reference lab settings. This test is designed as an aid in the detection of *E. coli* O157:H7 and should be used as an adjunct to culture.

INTRODUCTION

Most foods harbor microorganisms in differing concentrations. *E. coli* O157:H7 species are one of the major causes of foodborne illness worldwide. There are hundreds of *E. coli* serotypes (species). Many serotypes are capable of causing enteric diseases in humans; many of the hemorrhagic serotypes cause severe intestinal disorders and even kidney failure.

The O157:H7 serotype is associated with severe foodborne illness and has been implicated in most foodborne outbreaks.

Symptoms appear 8-42 hours after ingestion of *E. coli* O157:H7 contaminated food. Symptoms include diarrhea, vomiting, chills, fever, nausea, abdominal cramps, and prostration.

In order to detect low levels of *E. coli* O157:H7 in foods, a series of sequential pre-enrichment culturing steps are needed. The method includes a pre-enrichment on selective or differential media for 16 to 24 hours to increase the levels of *E. coli* O157:H7 organisms.

PRINCIPLE OF THE TEST

BioAssay Works[®] *E. coli* O157:H7 test is designed to detect *E. coli* O157:H7 antigens in contaminated food samples. The test is a rapid qualitative assay that is based on the use of *E. coli* O157:H7-specific antibodies. The reaction between the enriched positive sample and the colored particle-conjugated antibody will form a complex that migrates along the test membrane. An immobilized capture antibody will form a red line at the test line (T) area upon reacting with the sample/gold complex. An internal control line (C) is included with the test cassette to assure that the test has been processed correctly.

TEST PROCEDURE

Materials Provided

- 20 × F ETLF BioAssay Works[®] *E. coli* O157:H7 test cassettes (20 cassettes/kit)
- 1 × F PBS-010 Sample Diluent Buffer (1X PBS Buffer Solution) - 1 bottle (10 mL)

Materials Required but NOT Provided

Enrichment media – Recommend: Selenite Cysteine Broth (GN Broth - Hanja) or (Difco, Cat. # 0486-05 or Accumedia, Cat. # 7218). Alternatively, Selenite Cysteine Broth (Difco, Cat. # 0687-05 or Accumedia Cat. # 7283) or equivalent may be used.

Also required are the necessary ancillary equipment, such as stomacher bag, test tubes, pipettes, and safety equipment used in a microbiology testing laboratory for preparation, storage, and handling of samples.

Sample Collection and Handling

Only food samples should be tested.

Samples that will not be tested within 48 hours should be refrigerated at 2-8°C.

Protocol

Allow the test cassettes, pre-enriched samples, controls, and buffers to reach room temperature, 20-25°C, prior to use.

Sample Preparation/Test Procedure

1. A ratio of 1:10 sample to enrichment media (EM) is recommended. For example, 10 g of ground meat in 90 mL of EM or 25 mL of juice in 225 mL of EM. Incubate the sample-EM broth mixture at 37°C for 16-24 hours.
2. Add 100 µL of Sample Diluent Buffer (F PBS-010) solution (1X PBS) to a test tube.
3. Add 100 µL of the enriched sample (from Step 1) to the test tube. Shake well or vortex to mix.
4. Add 120 µL of the buffer/sample mixture (from Step 3) to the sample well of the test cassette (F ETLF).
5. Read results within 15-20 minutes and no later than 30 minutes. Some positive results may be observed within a few minutes depending on the concentration of the antigen in the sample tested.

QUALITY CONTROL

Each test cassette has a built in procedural control. Correct procedural technique and test device performance is confirmed when a colored line appears in the control (C) area of the test cassette. It is recommended that when a new shipment of product is received, negative and positive controls be tested, and the appropriate results obtained.

INTERPRETATION OF RESULTS

A reddish colored line developing in the control line area (C) of the test cassette indicates proper sample migration, capillary flow, and reagent integrity. If this line does not develop, the test is INVALID and must be repeated.

A reddish colored line in the test line area (T) of the test cassette is indicative of a POSITIVE result. No discernible development of a reddish colored line is indicative of a NEGATIVE result.

Report as follows:

POSITIVE = Sample Positive for detection of *E. coli* O157:H7.

NEGATIVE = Sample absent or below the level of analytical sensitivity for *E. coli* O157:H7.

Depending on the concentration of antigen and consistency of sample, POSITIVE results may be observed in a few minutes; however, to confirm NEGATIVE results, the complete reaction time of 20 minutes is required.

If test results are indeterminate (very faint reddish color on the test line [T]), the test should be repeated or another sample collected. If a faint line repeatedly develops on the test line of the cassette, this should be considered a POSITIVE test result.

A reddish colored line that develops in the test line area (T) of the test cassette after 30 minutes is not indicative of a positive result, is not diagnostic, and should be ignored.

STORAGE

Store all components of the BioAssay Works® *E. coli* O157:H7 Test Kit at room temperature (20-25°C). The test cassettes must remain sealed in the desiccated pouch until ready for use.

REAGENTS

Test Cassettes (20 × Part #: F ETLF):

Immunochromatographic assay cassettes containing dried colloidal gold labeled anti- *E. coli* O157:H7 antibody and immobilized polyclonal anti- *E. coli* O157:H7 antibody

Sample Diluent Buffer (1 × Part #: F PBS-010):

1X Phosphate Buffered Saline (1X PBS), 10 mL

LIMITATIONS OF THE PROCEDURE

This test is designed as an aid in the detection of *E. coli* O157:H7 and should be used as an adjunct to culture. Samples containing large amounts of food matter may migrate slowly up the test cassette membrane. Follow the instructions recommended in the test procedure for transferring these samples to a test tube; and allow a maximum of 30 minutes for migration to occur. On occasion, samples containing very high concentrations of *E. coli* O157:H7 lipopolysaccharide antigens may produce a strong POSITIVE test line (T) and a weak or faint CONTROL line (C) on the test cassette. This is due to large amounts of conjugate being deposited on the TEST line (T), depleting the amount of conjugate available for binding at the CONTROL line (C). If a strong positive sample is suspected, the sample may be serially diluted in distilled water to view the CONTROL line (C) and ensure proper performance of the test procedure.

PRECAUTIONS

The BioAssay Works® *E. coli* O157:H7 Test Kit is intended for Food Testing Only. All samples and test materials should be handled as if capable of transmitting disease. Strict adherence to established microbiological safety precautions should be observed when handling samples, test materials and in the disposal of processed assay materials. Do not use reagents beyond the indicated expiration date.

SHELF LIFE

The *E. coli* O157:H7 Test Kit is guaranteed to perform as designed up to the date recorded on the product label when stored as instructed.

This product is for internal research use only and may not be resold or used for provision of any services to a third party for paid consideration.