

U.S. English

Product Number:
9734

Reveal[®] 2.0

for E. coli O157:H7



Reveal® 2.0 for *E. coli* O157:H7

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Intended Use

The Reveal® 2.0 for *E. coli* O157:H7 test system provides for the rapid recovery of *Escherichia coli* O157:NM and *Escherichia coli* O157:H7 organisms in various foods, facilitating detection and presumptive identification of the test organism in as little as 12 hours.

In an AOAC Research Institute Performance Tested Method™ study, the Reveal 2.0 for *E. coli* method was found to be an effective procedure for detection of *E. coli* O157:H7 in raw beef trim and raw ground beef. Results indicate the method is capable of detecting *E. coli* O157:H7 in these sample types at initial levels as low as 1 CFU per 375 g.

Assay Principles

The system utilizes specified enrichment media to provide *E. coli* O157:H7 with readily available nutrients and other factors required for its survival and rapid growth. Used with Reveal media, the system can provide results in 12–20 hours.

A portion (200 µL) of the enriched culture is placed into the sample cup followed by a chemical treatment. The test device is placed into the sample cup during the chemical treatment and is incubated at 42°C for 15 minutes. The sample is wicked through the reagent zone that contains specific anti-*E. coli* O157:H7 antibodies conjugated to colloidal gold particles.

If antigens are present in the sample, they will bind to the gold-conjugated antibodies. This antigen-antibody complex then leaves the reagent zone and travels across the membrane, which contains a zone of anti-*E. coli* O157:H7 antibodies. The immune complex with gold conjugate is captured and aggregates in this zone, thus displaying a visible line. The remainder of the sample continues to migrate to the end of the membrane where it is deposited into a waste reservoir.

The reagent zone also contains a gold conjugate of a second antigen, which is eluted by the sample regardless of the presence of *E. coli* O157:H7 antigen to form a line in the control zone, ensuring the test is working properly.

Intended User

This test system is designed for use by personnel familiar with the appropriate aseptic techniques for the isolation and identification of *E. coli* O157:H7. Training, which is available through Neogen®, is recommended for those without a basic knowledge of microbiology.

Materials Provided

Reveal Test Kit

1. 20 Reveal 2.0 for *E. coli* O157:H7 test devices
2. 20 transfer pipettes
3. 20 sample cups
4. 1 promoter reagent

Test Systems Required but Not Provided

Note: The Reveal 2.0 for *E. coli* O157:H7 device requires the use of certain enrichment media and supplies. These can be purchased as part of a test system or individually.

Reveal 2.0 for *E. coli* O157:H7 complete system (contains devices and media for 20, 65 g samples) (Neogen item 9714)

Media Required but Not Provided

Note: Media can be purchased individually or as part of a test system.

1. Reveal 2.0 for *E. coli* O157:H7 media
 - a. Unitized (Neogen item 9727)
 - b. Bulk 500 g (Neogen item 9728)
 - c. 2 kg (Neogen item 9728B)

Materials Required but Not Provided

1. Stomacher-type bags (Neogen item 9736)
2. Scale capable of weighing a minimum of 65 g (Neogen item 9427)
3. Timer (Neogen item 9416)
4. Sample cup rack (Neogen item 9475)
5. Sterile deionized or distilled water
6. Incubator capable of maintaining $42 \pm 1^\circ\text{C}$ (Neogen item 9735)

Optional Materials

Stomacher 400 machine or equivalent

Testing Different Commodities

Please contact a Neogen representative for an updated list of the commodities for which the test has been validated, or for validation studies for specific commodities.

Precautions

1. Do not use expired media.
2. Warm sample to ambient temperature prior to enrichment.
3. Use of incubation times other than those specified may cause erroneous results.
4. Use rehydrated media within 6 hours of preparation.
5. Sample bags must be closed loosely to allow air exchange during incubation, which is vital for sufficient growth of *E. coli* O157:H7.
6. Sterile water should be warmed to $42 \pm 1^\circ\text{C}$ prior to preparing media.
7. Good microbiological laboratory practices should be used.

Media Preparation

65 g Samples

1. Transfer the contents of 1 pouch of unitized Reveal 2.0 for *E. coli* O157:H7 media (Neogen item 9727) or 10.7 g bulk Reveal 2.0 for *E. coli* O157:H7 media (Neogen item 9728) into a Stomacher-type bag.
2. Add 325 mL of $42 \pm 1^\circ\text{C}$ sterilized water. Grasp the bag tightly about 2–3 inches from the top, support bottom, and shake vigorously until medium is dissolved.
3. Place Stomacher-type bag in a rack or other support. The medium can be held at 42°C until used, but no more than 6 hours.

375 g Samples

1. Transfer 61.7 g bulk Reveal 2.0 for *E. coli* O157:H7 media (Neogen item 9728) into a Stomacher-type bag.
2. Add 1875 mL of $42 \pm 1^\circ\text{C}$ sterilized water. Grasp the bag tightly about 2–3 inches from the top, support bottom, and shake vigorously until medium is dissolved.
3. Place Stomacher-type bag in a rack or other support. The medium can be held at 42°C until used, but no more than 6 hours.

Sample Enrichment

1. Combine sample (65 g or 375 g) with the media, grasp the bag tightly at top and knead until mixed. Shake bag vigorously using a side-to-side motion to ensure complete mixing.
Alternative: Place bag in Stomacher machine and mix 30 seconds at normal speed.
2. Close bag loosely. Incubate sample bag at $42 \pm 1^\circ\text{C}$ for 12–20 hours. Do not close bag tightly, as air exchange is required for optimum growth.
3. Remove bag from incubator after designated time. Grab the bag tightly 2–3 inches from the top, support the bottom, and gently mix the contents of the sample bag using a side-to-side motion.

Reveal Test Procedure

1. Remove the required number of Reveal 2.0 for *E. coli* O157:H7 test devices from container.
2. Transfer 200 μL or 8 drops of enriched sample to Reveal sample cup.
3. Add 1 drop of promoter reagent to the sample cup.
 - a. After transferring the enriched sample and promoter reagent to the sample cup, do not allow the sample to sit more than 10 minutes before placing the Reveal device into the sample cup (step 4).
4. Place Reveal device into well mixed sample cup containing reagent and incubate at $42 \pm 1^\circ\text{C}$ for 15 minutes.
5. Remove cups and devices from incubator and record results after 15 minutes. Observe and record the test results immediately (within 60 seconds) after 15 minutes.

Interpretation of Results

Visual Interpretation

1. Line in both control and test zone in 15 minutes is considered positive.
2. Line in control zone only is considered negative.
3. If no line appears in the control zone, the test is considered invalid and the sample should be tested with another device.

Confirmation (Optional)

Neogen recommends that presumptive positive results are verified by plating the enriched sample onto media described in USDA-FSIS procedures.

Disposal

Dispose of (autoclave, bleach, etc.) the Reveal test devices, pipettes, cups, and media in accordance with all applicable local, state, and federal regulations.

Customer Service

Neogen Customer and Technical Services can be contacted through [Neogen.com](https://www.neogen.com) and product training is available by request.

SDS Information Available

Safety data sheets are available for all test kits at [neogen.com](https://www.neogen.com) or by calling 800.234.5333 or 517.372.9200.

Terms and Conditions

Neogen's full terms and conditions are available online.

Warranty

Neogen makes no warranty of any kind, either expressed or implied, except that the materials from which its products are made are of standard quality. If any materials are defective, Neogen will provide a replacement of the product. Buyer assumes all risk and liability resulting from the use of this product. There is no warranty of merchantability of this product, or of the fitness of the product for any purpose. Neogen shall not be liable for any damages, including special or consequential damage, or expense arising directly or indirectly from the use of this product.

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