soleris®

Enterobacteriaceae Vial

Product Number: S2-EBAC9

Pictured: S2-EBAC9 vial uninoculated (left) and inoculated vial (right).

Introduction

The *Enterobacteriaceae* Vial, 9 mL (S2-EBAC9) is a screening vial specific for organisms in the family *Enterobacteriaceae*. The vial has broad inclusivity and an assay time of 14–18 hours for most applications. The vial contains a peptone yeast extract base with glucose as a carbon source. The selective agents include bile salts, sodium lauryl sulfate, and other gram-positive inhibitors. Acidification of the medium due to the glucose utilization changes the pH. As *Enterobacteriaceae* metabolize, the pH indicator changes from a purple to a yellow color. The color change is read by optical sensors in the instrument.

In an AOAC Research Institute Performance Tested Method℠ study, Soleris® for *Enterobacteriaceae* was found to be an effective method for detection of *Enterobacteriaceae* in the following sample types: pasteurized milk, yogurt, mozzarella cheese, ice cream, dried milk, pasteurized liquid egg, frozen cooked chicken, deli ham, lettuce, and dry dog food.

Materials Required

1. S2-EBAC9, *Enterobacteriaceae* (Product No. S2-EBAC9) vial.
   a. Vials are stored at room temperature and expire 6 months after date of manufacture.
2. Soleris 32 instrument (Product No. BSX32), Soleris 128 instrument (Product No. BSX128), or Soleris Next Generation (SNG-INS32). Containing one or four temperature-controlled (15–50 ± 0.5°C) incubator drawers, respectively, with 32 test locations per drawer. Each test location contains a light-emitting diode (LED)-based optical sensor for measurement of changes in absorbance over time.
3. Soleris computer system (Product No. BSC01) — Includes vial rack.
4. Soleris computer only (Product No. SNG-Computer or equivalent).
5. Soleris vial rack (Product No. VR-300 or equivalent) — Holds 32 vials.

Dependent on Sample Tested

1. Sterile 1N to 5N sodium hydroxide (NaOH) and/or hydrochloric acid (HCl).
2. pH Meter or pH paper.
3. Micropipettor and tips — 20–200 µL.
4. Micropipettor and tips — 100–1,000 µL.
5. Butterfield's Phosphate Buffer, 99 mL (BPB-99) or Buffered peptonewater (Product No. NCM0015 or equivalent).

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6. For USP Testing: Tryptic Soy broth, 90 mL (TS-124) or Butterfield's Phosphate Buffer, 90 mL (6654).
   a. If required, use a designated neutralization broth, such as D/E Neutralizer, TAT Broth, Modified Letheen Broth, etc.
7. Stomacher or equivalent.
8. Stomacher-type bags with mesh filter (Product No. 6827).
9. Balance — For weighing samples, minimum 100 g ± 0.1 g capacity.
10. Violet red bile glucose agar (Product No. NCM0041A or equivalent).

Vial Specifications

1. Vial pH is 6.7 ± 0.2.
2. Vial sample capacity up to 1.0 mL.

Sample Preparation

1. For non-USP/non-AOAC testing, add the sample directly or prepare a 1:10 dilution by adding 11 g of sample to 99 mL of sterile Butterfield's Phosphate Buffer Vial sample capacity up to 1 mL.
2. For USP & AOAC testing, perform 1:10 dilution by adding 10 g of sample in 90 mL of Tryptic Soy Broth (See NEOGEN® Rapid Microbiology System Validation Book, Introduction, p.5) or designated neutralization broth.
   a. Check pH and adjust, if necessary, to 7.0 ± 1.0.
3. If using the dilute-to-specification method, complete the dilution required.

Inoculation of Vial

1. Inoculate the vial with no more than 1.0 mL and no less than 0.10 mL of the sample to be tested. If using dilute-to-specification method, add the volume of the appropriate dilution required.
   a. For AOAC testing, use 1.0 mL.
2. Cap the vial and gently invert three times to mix sample. Keep cap tight.
3. Insert the vial into the Soleris instrument set at 35°C or as indicated by trainer. Click start to begin test. The detection curve will be generated in real time. The test will run for preprogrammed time, but positive results may be reported before test is completed. The incubation temperature and test duration can be optimized if required. It is not recommended to adjust parameters without consulting NEOGEN Technical Services.

Incubation Temperatures

Soleris: 35°C ± 2°C or as indicated by trainer.

Algorithm Utilized

For Soleris AOAC Approved Testing

This method was validated by AOAC for detection of *Enterobacteriaceae* in select foods at a positive/negative threshold of ≥ 10 CFU/g of product.

Test	Threshold	Skip	Shuteye	Test Duration	Temperature
S2-EBAC9	10	1	25	18 hours	36°C ± 1°C

For non-AOAC Soleris Testing

Test	Threshold	Skip	Shuteye	Test Duration	Temperature
S2-EBAC9	10	1	25	14–18 hours	35°C ± 2°C

Disclaimers

Information provided is based on validation procedures that NEOGEN performed in NEOGEN laboratories. Deviation from procedures is possible, but should be discussed with NEOGEN Technical Services.

Samples may need to be pH adjusted for all vials.

Appearance of the vials should be inspected prior to use.

If shuteye detections are observed, the threshold may need to be adjusted based on the product matrix. Certain product matrices may require parameter adjustments, including increased test duration. For more information contact NEOGEN Technical Services.

The following organisms produced negative results in inclusivity testing: *Kluyvera ascorbate, Pantoea agglomerans, Proteus penneri, Serratia proteamaculans,* and *Buttiazuxella warmboldiae*. In some cases (e.g., *Pantoea agglomerans*), these reactions have proven to be strain specific, while in other cases the organisms have been described as being temperature sensitive.

Reference the Soleris Operating Manual for troubleshooting, use of instrument, and interpretation of results.

Safety Precautions

Use of this test should be restricted to individuals with appropriate laboratory training in microbiology as some *Enterobacteriaceae* are potentially infectious. Reagents are for laboratory use only. All pipetting transfers must be made using either a disposable pipet and pipetting aid or micropipettor with disposable tips. Culture media contains antimicrobial selective agents and dyes. Wear appropriate PPE and avoid contact with skin and mucous membranes. Refer to the Safety Data Sheet available from NEOGEN for more information. Used Soleris vials should be handled and disposed of as potentially infectious material. The preferred method for disposal of contaminated materials, including used vials, sample homogenates, pipettes, etc., is autoclaving. Items that cannot be autoclaved may be decontaminated by using a disinfectant solution, e.g., 10% household bleach, followed by rinsing with water. Consult with your facility safety director for specific instructions.

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