



Soleris® vial uninoculated (left) and inoculated vial (right).

The Gram Negative (S2-GN) Vial is a screening vial for gram negative organisms in personal care/cosmetic products. The vial has an assay time of 24 hours for most applications. The vial uses a selective media and an antibiotic supplement. As organisms grow in the broth medium, the carbon dioxide (CO<sub>2</sub>) produced diffuses through a membrane layer into a soft agar plug containing a dye indicator. The color change in the dye is read by the BioLumix® instrument. The mem-

brane layer also serves as a barrier, eliminating product interference with the reading frame.

### Materials Required:

1. S2-GN, Gram Negative vial
2. Vancomycin-HCl (Sigma V2002)
3. Sterile water
4. Butterfield's Phosphate Buffer (BPB-99)
5. 10 mL syringes luer lock (Fisher item 03-377-23)
6. Syringe filter (Fisher item 05-713-387)

### Dependent on Sample Tested:

1. Sterile 1 N to 5 N sodium hydroxide (NaOH) and/or hydrochloric acid (HCl)
2. pH meter or pH paper
3. Tryptic Soy broth (BLX-TSB90)
  - a. If required, use a designated neutralization broth, such as D/E Neutralizer, TAT Broth, Modified Lethen Broth, etc.)

### Vial Specifications

1. Vial pH is 6.5 ± 0.2
2. Vial sample capacity: 0.1 ml

### Vial Preparation

1. Remove S2-GN vials from the refrigerator and allow to equilibrate to room temperature.

### Sample Preparation

1. For USP 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.
2. Incubate for 18–24 hours at 35°C.

### Vancomycin Supplement Preparation

1. Weigh 0.1 gram into 10 ml DI water
2. Filter sterilize through 0.45 µm filter membrane.
3. Add 1.0 ml to 9.0 mL sterile DI water.
4. Add 30 µL per vial to deliver 3 mg/L.
5. Prepare fresh daily.

### Inoculation of Vial

1. Add 0.03 mL or 30 µL of vancomycin hydrochloride to vial.
2. Cap the vial and gently invert 3 times to mix sample.
3. Transfer 0.1 mL of the incubated TSB enrichment to the S2-GN vial.
4. Cap the vial and gently invert 3 times to mix sample. Keep cap tight.
5. Insert the vial into the BioLumix instrument set at 35°C and run for the pre-programmed test duration. It is not recommended to adjust the parameters without consulting Neogen Technical Services.

### Algorithm Utilized:

Test	Test Type	Detection Level	Resolution	Ignore	Test Duration	Temp
S2-GN	Yellow	10	1	60	24 Hours	35°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.

Certain product matrices may require parameter adjustments, including increased test duration. For more information contact Neogen Technical Services.