

# RAINBOW<sup>®</sup> AGAR SALMONELLA

## TECHNICAL INFORMATION

# BIOLOG

**INTENDED USE** For laboratory use as a selective, chromogenic culture medium to aid in the detection and isolation of H<sub>2</sub>S-producing *Salmonella* species. Not for human *in vitro* diagnostic use.

**SUMMARY OF TEST** The United States Department of Agriculture-Food Safety Inspection Service (USDA-FSIS) has instituted *Salmonella* pathogen reduction performance standards for the meat processing and packaging industry<sup>1</sup>. *Salmonella* was targeted “because it is the primary cause of foodborne illness among enteric pathogens”<sup>1</sup>. Rainbow Agar Salmonella utilizes an enhanced detection chemistry to determine H<sub>2</sub>S production among *Salmonella spp.*. Black colonies are formed by even weak H<sub>2</sub>S-producing strains. In addition, novel selective agents increase the recovery rate of *Salmonella* while inhibiting the growth of other enteric bacteria and inhibiting H<sub>2</sub>S production by *Citrobacter* and other H<sub>2</sub>S positive enteric species.

**CONTENTS** This packet contains dry powder to make Rainbow Agar Salmonella. 47 grams of powder will make 1 liter or approximately 50 plates.

**PREPARATION** To prepare 1 liter of Rainbow Agar Salmonella mix one 47 gram packet into 990 ml of purified water, add 10 ml of 35% glycerol. Boil until agar and other components are completely dissolved or autoclave for 5 minutes at 15 lbs. pressure and 121°C. DO NOT exceed 5 minutes at this heat and pressure. After autoclaving, the medium will be green and turbid. Cool agar to 45-50°C before pouring. Stir and dispense approximately 20 ml of media into each petri plate. The medium is ready to use as soon as it has cooled, gelled and the surface has dried. The final medium should be transparent and light yellow in color.

No pH adjustment is needed. The final pH should be pH 7.2-7.6. For samples that have a high background count of enteric bacteria, 100 mg/L sodium selenite can be added to make the medium more selective.

**STORAGE** Rainbow Agar Salmonella powdered medium is extremely hygroscopic and should be stored at 2°C to 25°C in a dry environment. Rainbow Agar Salmonella plates are stable for 12 weeks when stored under refrigeration and packaged in cellophane to inhibit moisture loss.

**USER QUALITY CONTROL** The following organisms are recommended if quality control is desired or required. Inoculate Rainbow Agar Salmonella plates with the following strains by streaking for isolation, then incubate for 24 hours at 35°C without elevated CO<sub>2</sub>. Colony color should be read from isolated colonies.

Organism	ATCC <sup>®</sup> number	Color on Rainbow Agar Salmonella
<i>Salmonella subspecies 1</i>	14028	Black
<i>Escherichia coli</i>	11775	White

**PROCEDURE** Inoculate the medium by streaking or spreading a sample suspected of containing *Salmonella* on the surface of the medium. Incubate the plates for 20 to 24 hours at 35°C without elevated CO<sub>2</sub>. Some strains may need longer incubation for color

development. (See the expected results below). Incubation beyond 36 hours may result in the detection of other H<sub>2</sub>S positive bacteria (e.g., *Citrobacter*). *Salmonella* colonies are detected by their distinctive black coloration. H<sub>2</sub>S production may be concentrated at the edges of heavy growth with central areas gray or white. Isolated colonies will be black. If 100 mg/L sodium selenite is added to make the medium more selective, the background microbial growth may appear orange due to selenite reduction.

### EXPECTED RESULTS<sup>a</sup>

Organism	Black colonies
<i>Salmonella subspecies 1:</i>	
<i>enteritidis</i>	+++
<i>choleraesuis</i>	++
<i>paratyphi A</i>	-
<i>paratyphi C</i>	+
<i>typhi</i>	+/-
<i>gallinarum</i>	-
<i>pullorum</i>	-
<i>Salmonella subspecies 2</i>	+++
<i>Salmonella subspecies 3</i>	+++
<i>Salmonella subspecies 4</i>	+++
<i>Salmonella subspecies 5</i>	+++
<i>Salmonella subspecies 6</i>	+++

<sup>a</sup>Colony coloration is indicative of isolated colonies.

+++	Intensely black colonies in ≤16 hours.
++	Black colonies in 16 to 24 hours.
+	Black colonies in 24 to 36 hours.
+/-	Black or gray colonies in 24 to 48 hours. Some strains are white.
-	Black colonies do not form.

**LIMITATIONS** This medium should not be used as the sole basis for identification of microorganisms. Any colony suspected of being *Salmonella* should be tested further to verify its identity using an approved confirmatory protocol.

Some *Citrobacter* strains will turn black if the plates are allowed to incubate 36 hours or longer. The pyrrolidonylamidase (PYR) spot test may be used to differentiate *Salmonella spp.* (PYR negative) from *Citrobacter spp.* (PYR positive).

### REFERENCES

1. United States Department of Agriculture-Food Safety Inspection Service. 1996. Improving the Safety of Meat and Poultry.

### BIOLOG ORDERING INFORMATION

#### Rainbow Agar Salmonella

Catalog Number	Description
80202	Powder—47 g

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