

## ORANGE SERUM AGAR (7587)

### Intended Use

**Orange Serum Agar** is used for the cultivation of aciduric microorganisms associated with spoilage of products.

### Product Summary and Explanation

The low pH of fruit juices makes citrus fruit products susceptible to spoilage by yeasts, molds, and the bacteria *Lactobacillus* and *Leuconostoc*.<sup>1</sup> In the 1950's, Hays investigated spoilage in frozen concentrated orange juice. He found that an agar medium containing orange serum (juice) was superior to Lindegren Agar in isolating the microorganisms responsible for spoilage causing a buttermilk off-odor.<sup>2</sup> Murdock, Folinazzo, and Troy<sup>3</sup> found Orange Serum Agar, pH 5.4 to be a suitable medium for growing *Leuconostoc*, *Lactobacillus*, and yeasts.

Orange Serum Agar is recommended for examining fruit beverages.<sup>1</sup>

### Principles of the Procedure

Enzymatic Digest of Casein provides carbon and nitrogen sources for general growth requirements. Orange Serum provides the acid environment favorable to recovering acid-tolerant microorganisms. Yeast Extract supplies B-complex vitamins that stimulate growth. Dextrose is the fermentable carbohydrate. Potassium Phosphate is a buffering agent. Agar is the solidifying agent.

### Formula / Liter

|                                 |        |
|---------------------------------|--------|
| Orange Serum.....               | 200 mL |
| Yeast Extract .....             | 3 g    |
| Enzymatic Digest of Casein..... | 10 g   |
| Dextrose .....                  | 4 g    |
| Potassium Phosphate.....        | 2.5 g  |
| Agar .....                      | 17 g   |

Final pH: 5.5 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

### Precautions

1. For Laboratory Use.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

### Directions

1. Suspend 45.5 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.

### Quality Control Specifications

**Dehydrated Appearance:** Powder is homogeneous, free flowing, and light beige.

**Prepared Appearance:** Prepared medium is slightly hazy and light to medium amber.

**Expected Cultural Response:** Cultural response on Orange Serum Agar at 35°C after 40 - 48 hours incubation.

| Microorganism                              | Response    |
|--|-------------|
| <i>Aspergillus niger</i> ATCC® 16404       | good growth |
| <i>Lactobacillus casei</i> ATCC® 393       | good growth |
| <i>Lactobacillus fermentum</i> ATCC® 9338  | good growth |
| <i>Lactobacillus plantarum</i> ATCC® 8014  | good growth |
| <i>Saccharomyces cerevisiae</i> ATCC® 9763 | good growth |

The organisms listed are the minimum that should be used for quality control testing.

### **Test Procedure**

1. For plate count method, prepare serial 10-fold dilutions of the test material.
2. Add 1 mL of test sample to a petri dish.
3. Add 18 - 20 mL of sterile, molten agar (cooled to 45 - 50°C) and swirl plate gently to mix well.
4. Allow to solidify before incubating at 30°C for 48 hours. Plates can be held up to 5 days.

### **Results**

Record colony morphology for each type of growth.

### **Storage**

Store sealed bottle containing the dehydrated medium at 2 - 8°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

### **Expiration**

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

### **Limitation of the Procedure**

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

### **Packaging**

|                          |                 |              |              |
|--------------------------|-----------------|--------------|--------------|
| <b>Orange Serum Agar</b> | <b>Code No.</b> | <b>7587A</b> | <b>500 g</b> |
|                          |                 | <b>7587B</b> | <b>2 kg</b>  |
|                          |                 | <b>7587C</b> | <b>10 kg</b> |

### **References**

1. **Vanderzant, C., and D. F. Splittstoesser (eds.).** 1992. Compendium of methods for the microbiological examination of foods, 3<sup>rd</sup> ed. American Public Health Association, Washington, D.C
2. **Hays, G. L.** 1951. The isolation, cultivation and identification of organisms which have caused spoilage in frozen concentrated orange juice. Proc. Fla. State Hort. Soc. **54**:135-137.
3. **Murdock, D. I., J. F. Folinazzo, and V. S. Troy.** 1952. Evaluation of plating media for citrus concentrates. Food Technol. **6**:181-185.
4. **MacFaddin, J. F.** 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1. Williams & Wilkins, Baltimore, MD.

### **Technical Information**

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.