

# TECHNICAL DATA SHEET

Article No. 9595

Orange Serum Agar, ready-to-use culture medium

---

## SPECIFICATION

Prepared medium. Solid medium for the culture of aciduric organisms especially those associated with the spoilage of citrus products and their derivatives.

Colour: yellow  
pH: 5.6 ± 0.2 at 25 °C

---

## COMPOSITION IN G/ L

Orange Serum	5.0
Yeast extract	3.0
Tryptone	10.0
Dextrose	4.0
Dipotassium phosphate	3.0
Agar	17.0

---

## PACKAGE DETAILS

### 9595-20x20 ml

Volume	20 ± 0.5 ml
Tube size	17 x 145 mm
Packaging unit	20 tubes

1 box with 20 x 20 ml in 17x145 mm glass tubes. ink labelled, metal-Non injectable cap. - 20 tubes per box .

### 9595-20x200 ml

Volume	200 ± 5 ml
Bottle size	250 ml
Packaging unit	10 bottles

1 box with 10 x 200 ml in 250 ml bottles. Injectable cap: Plastic screw inner cap. The use of syringes needles with a diameter greater than 0.8 mm is not recommended.



## DESCRIPTION/TECHNIQUE

### Description

Orange Serum Agar was developed in the 1950's by Hays and coworkers for the detection, enumeration and isolation of spoilage microorganisms in fruit juices and products derived from citrus. Products with a low pH have microbial growth restricted to that of aciduric microorganisms. In a later study it was shown that Orange Serum Agar pH 5.4 was the most suitable medium for the isolation of lactic acid bacteria, especially (*Lactobacillus* and *Leuconostoc*) and yeasts that produce (buttermilk off-odor) in citrus fruits.

Orange Serum Agar is not a differential Agar but a culture medium in which the orange extract provides a favorable acidic environment in which aciduric microorganisms can be recovered including those damaged by food processing. Tryptone provides the main source of carbon and nitrogen, providing optimal growth conditions. Yeast Extract supplies Group B complex vitamins that stimulate growth and the phosphate provides an osmotic buffer for cell survival. Dextrose is a supplementary source of carbon and the agar is a solidifying agent.

### Technique

The International Fruchtsaft-Union (IFU) recommends the use of Orange serum agar in several standardized methods, using the plate count method:

1. Prepare serial 10-fold dilutions of the sample using a suitable diluent such as Buffered Peptone Water.
2. Distribute aliquots of 1 ml of the diluted sample in sterile Petri dishes.
3. Add 20 ml of molten sterile medium cooled to 45°C, gently swirl the dish to mix the sample and medium properly.
4. Allow it to solidify and incubate at a 30 ± 1°C for 48 hours before enumeration. If there is no growth extend the incubation to 5 days, reading daily before giving a negative result.

Generally the colonies of yeasts and molds are distinguished by their morphology but those of aciduric bacteria need to be Gram stained and examined microscopically to be appropriately categorized.

Note: The solid mediums can be melted in different ways: autoclave, bath and, if the customer considers appropriate, also the microwave. Whenever the microwave option is chosen, it is necessary to take certain safety measures to avoid breaking of the containers, such as loosening the screw cap and putting the bottle or tube in a water bath in the microwave. The fusion temperature and time will depend on the shape of the container, the volume of medium and the heat source. Avoid overheating as both the heating periods.

## MICROBIOLOGICAL CONTROL

Melt Medium - Prepare Plates - Spiral Spreading: Practical range 100 ± 20 CFU. min. 50 CFU (productivity)  
 Microbiological control according to ISO 11133:2014/A1:2018.

Aerobiosis. Incubation at 30 ± 1 °C Reading at 48 h - 5 days

Microorganism	Growth
<i>S. cerevisiae</i> ATCC® 9763, WDCM 00058	Good (≥50 %)
<i>Aspergillus niger</i> ATCC® 16404	Good (≥50 %)
<i>Lactobacillus fermentum</i> ATCC® 9338	Good (≥50 %)

### Sterility control:

Incubation 48 hours at 30-35 °C and 48 hours at 20-25 °C: NO GROWTH.

Check at 7 days after incubation in same conditions.

---

## REFERENCES

- 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.
- HAYS, G.L. & D.W. REISTER (1952) The control of 'off-odour' spoilage in frozen concentrate orange juice. Food Technol. 6:386-389.
- HATCHER, W.S., M.E. PARISH, J.L. WEIHE, D.F. SPLITTSTOESSER & B.B. WOODWARD (2001) Fruit Beverages, en Compendium of Methods for the Microbiological Examination of Foods. 4th ed., F.P. Downes & K. Ito, editors. APHA Inc., Washington D.C., USA.
- IFU Method No. 2 (1996) Total Count of Potential Spoiling Microorganisms of Fruits and Related Products. International Federation of Fruit Juice Producers. Microbiological Methods (2004). Schweizerischer Obstverband. Postfach CH-6302 Zug.
- IFU Method No. 6 (1996) Mesophilic & Thermoduric-Thermophilic Bacteria: Spores Count. D-II Mesophilic Anaerobic Sporeforming Bacteria: Spores Count. International Federation of Fruit Juice Producers. Microbiological Methods (2004). Schweizerischer Obstverband. Postfach CH-6302 Zug.
- IFU Method No. 7 (1998) 'Sterility' Testing of 'Aseptic Filled Products', 'Commercial Sterile Products' and 'Preserved Products'. International Federation of Fruit Juice Producers. Microbiological Methods (2004). Schweizerischer Obstverband. Postfach CH-6302 Zug.
- IFU Method No. 10 (1998) Microbiological Examination of Potential Spoiling Microorganisms of Low Acid and High pH Vegetable Products. International Federation of Fruit Juice Producers. Microbiological Methods (2004). Schweizerischer Obstverband. Postfach CH-6302 Zug.
- ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
- MURDOCK, D.I., J.F. FOLINAZZO & V.S. TROY (1952) Evaluation of plating media for citrus concentrates. Food Technol. 6:181-185.
- MURDOCK, D.I. & C.H. BROKAW. (1958). Sanitary control in processing citrus concentrates. I. Some specific sources of microbial contamination from fruit bins to extractors. Food Technol. 12: 573-576.
- STEVENS, J.W. (1954) Preparation of dehydrated agar media containing orange juice serum. Food Technol. 8:88-91.
- 

---

## STORAGE

8 - 25 °C

---

## SHELF LIFE

12 months unopened from date of manufacture

---

last updated: 25.08.2022

