



TSAF AGAR (6905) (Tryptone Soy Fast Green Agar)

Intended Use

TSAF Agar (Tryptone Soy Fast Green Agar) is used in determining total bacterial counts with the ISO-GRID® and/or NEO-GRID™ Membrane Filtration System in a laboratory setting. TSAF Agar (Tryptone Soy Fast Green Agar) is not intended for use in the diagnosis of disease or other conditions in humans.

Product Summary and Explanation

Tryptone Soy Fast Green Agar is based on Tryptone Soy Agar. The original formula was modified by adding Fast Green FCF dye to enhance visibility of the colonies. TSAF Agar is recommended for the enumeration of total bacteria from all foods using the ISO-GRID and/or NEO-GRID Membrane Filtration Method.^{1,2}

Principles of the Procedure

Enzymatic Digest of Casein and Enzymatic Digest of Soybean Meal are the nitrogen and vitamin sources in TSAF Agar. Sodium Chloride maintains the osmotic environment. Fast Green FCF stains all colonies green or blue-green, enhancing visibility. Agar is the solidifying agent.

Formula / Liter

Enzymatic Digest of Casein	15 g
Enzymatic Digest of Soybean Meal	5 g
Sodium Chloride	5 g
Fast Green (FCF)	0.25 g
Agar	15 g

Final pH: 7.3 ± 0.2 at 25°C

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

Precautions

1. For Laboratory Use Only.
2. Harmful. Possible carcinogen. May cause irritation to eyes, skin and respiratory tract.

Directions

1. Suspend 40.3 g of 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.
4. Cool to 45 - 50°C.
5. Check pH and adjust, if necessary to obtain a final pH of 7.3 ± 0.2 in the solidified medium.
6. Dispense 18 - 20 mL volumes to sterile Petri dishes, cool until plates are solidified.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and green to blue-green-beige.

Prepared Appearance: Prepared medium is clear to slightly hazy and blue-green to green.

Expected Cultural Response: Cultural response on TSAF incubated at 35°C for 48 ± 2 hours using the ISO-GRID and/or NEO-GRID Membrane Filtration System.

Microorganism	Approx. Inoculum (CFU)	Expected Results	
		Growth	Reaction
<i>Escherichia coli</i> ATCC® 25922	10 - 300	Good to excellent	Green colonies
<i>Staphylococcus aureus</i> ATCC® 25923	10 - 300	Good to excellent	Green colonies

Test Procedure

For sample details refer to the ISO-GRID Methods Manual and NEO-GRID Protocols.

1. Prepare a sample homogenate.
2. Filter 1 mL of the homogenate through the prefilter and ISO-GRID membrane filter.
3. Place the membrane filter on the surface of a predried TSAF Agar plate.
4. Incubate the inoculated TSAF Agar in an inverted position in a dry air incubator at $36 \pm 1^\circ\text{C}$ for 45 - 51 hours.
5. Examine the membrane filter for green colonies.

Results

Bacterial colonies stain green.

If positive colonies are present, count the number of squares containing colonies. Convert the number of squares to the corresponding MPN and calculate the confirmed MPN, using the methods described in the ISO-GRID Methods Manual.

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°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 grow poorly or fail to grow on this medium.

Packaging

TSAF AGAR	Code No.	6905A	500 g
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References

1. **Entis, P.** 1986. Hydrophobic grid membrane filter method for aerobic plate count in foods: Collaborative study. J. AOAC **69**:671-676.
2. **Entis, P., and P. Boleszczuk.** 1986. Use of fast green FCF with tryptic soy agar for aerobic plate count by the hydrophobic grid membrane filter. J. Food Prot. **49**:278-279.

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.