



PRODUCT SPECIFICATION SHEET

Tributyryn Agar Base w/o Tributyrin (DM270)

Intended Use

Tributyryn Agar Base w/o Tributyrin (DM270) is recommended for detection of lipolytic microorganisms.

Product Summary and Explanation

Lipolytic microorganisms are organisms that are able to decompose vegetable and animal fats with the release of a considerable amount of energy. Many foods contain significant amount of fats that may be susceptible to hydrolysis. The free fatty acids (FFA) liberated by hydrolysis of fat can be responsible for unpleasant flavour or they may oxidize to compounds with undesirable flavour notes. Several problems of fat breakdowns in foods are non-microbial in origin, but many bacteria, yeasts and moulds produce lipolytic enzymes that are capable of causing both hydrolytic and oxidative deterioration of fats when present in food samples.⁽¹⁾

Among the lipolytic microorganisms are the aerobic and the anaerobic bacteria of the genera *Pseudomonas*, *Clostridium*, and molds fungi (*Penicillium*, *Cladosporium* and *Aspergillus*). Anderson⁽²⁾ originally formulated Tributyrin Agar which consists of a stable homogenate of nutrient agar and tributyrin (glyceryl tributyrate) is used for the detection and enumeration of lipolytic microorganisms such as *clostridia*,⁽³⁾ *staphylococci*⁽⁴⁾ marine *Flavobacteria* and *Pseudomonas*⁽⁵⁾ and moulds in foodstuffs and other materials. Tributyrin is the simplest triglyceride occurring in natural fats and oils. It is hydrolyzed by some microorganisms that do not hydrolyze other triglycerides or fats containing longer chain fatty acids. However, it is the substrate of choice for screening purposes, to enumerate lipolytic microorganisms of potential importance in foods.^(6,7)

Principles of the Procedure

Tributyryn Agar Base w/o Tributyrin (DM270) contains peptic digest of animal tissue and yeast extract which provide nitrogen, vitamins and minerals required by the organisms. Tributyrin (glyceryl tributyrate) is used for the detection and enumeration of lipolytic (fat-splitting) bacteria and moulds. Tributyrin degradation by the microorganisms is indicated by clear zones surrounding the lipolytic colonies in the otherwise turbid culture medium. Lipolytic organisms render the medium transparent by converting the fat to water soluble butyric acid.⁽⁸⁾ For the effectiveness of the assay the medium should have a uniform turbid emulsion.⁽⁹⁾

Formula / Liter

Ingredients	Gms / Liter
Peptic digest of animal tissue	5.00
Yeast extract	3.00
Agar	15.00
Final pH: 7.5 ± 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. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions

1. Suspend 23 grams of the medium in 990 ml of distilled water. Add 10 ml of Tributyrin (MS046).





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2. Mix and heat to boiling to dissolve the medium completely.
3. Autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
4. Shake the flask and individual plate so as to maintain uniform turbidity.

Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Light yellow coloured opalescent gel forms with oil droplets in Petri plates
Reaction of 2.3% Solution containing 1% Tributyrin	pH : 7.5 ± 0.2 at 25°C
Gel Strength	Firm, comparable with 1.5% Agar gel

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 24-48 hours with added Tributyrin (MSO46) (under appropriate conditions).

Sr. No.	Organisms	Results to be achieved		
		Inoculum (CFU)	Growth	Lipase Activity
1.	<i>Clostridium perfringens</i> ATCC 12924	50-100	good-luxuriant	negative, absence of clear zone around colony
2.	<i>Clostridium sporogenes</i> ATCC 11437	50-100	good-luxuriant	positive, clear zone around colony
3.	<i>Bacillus subtilis</i> ATCC 6633	50-100	good-luxuriant	positive, clear zone around colony
4.	<i>Escherichia coli</i> ATCC 25922	50-100	good-luxuriant	negative, absence of zone around colony
5.	<i>Staphylococcus aureus</i> ATCC 25923	50-100	good-luxuriant	positive, clear zone around colony

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

Test Procedure

Refer appropriate references for specific test procedures.

Results

Refer appropriate references and test procedures for interpretation of results.

Storage

Store the sealed bottle containing the dehydrated medium at 10 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the 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.





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Limitations of the Procedure

1. For identification, organisms must be in pure culture. Morphological, biochemical and/or serological tests should be performed for final identification.
2. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Tributyrin Agar Base w/o Tributyrin

Product Code : DM270

Available Pack sizes : 100gm / 500gm

References

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9. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore

Further Information

For further information please contact your local MICROMASTER Representative.



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