



## Thermoacidurans Agar

M125

### Intended Use:

Recommended for isolation of *Bacillus thermoacidurans* from food products.

### Composition\*\*

Ingredients	Gms / Litre
Proteose peptone	5.000
Yeast extract	5.000
Dextrose (Glucose)	5.000
Dipotassium hydrogen phosphate	4.000
Agar	20.000
Final pH ( at 25°C)	5.0±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 39 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and pour into sterile Petri plates.

### Principle And Interpretation

*Bacillus coagulans* is commonly found in soil and has been isolated from canned tomato products and dairy products. This organism is responsible for flat-sour spoilage of canned foods (4). *B. coagulans* is also referred to as *B. thermoacidurans* (1). They are of primary importance in spoilage of low-acid foods packed in hermetically sealed containers (1). Spoilage due to bacterial growth is accompanied by a reduction in pH from 0.3 to 0.5 (5) and also the ends of the can remain flat. Thermoacidurans Agar, described by Stern et al (5) is recommended by APHA (4) for cultivation and isolation of *B. coagulans* from canned foods. *B. coagulans* is described as a facultative thermophile, that can grow at 20 to 55°C, and can also grow at pH levels between 5.0 to 7.0. *B. stereothermophilus* can also grow at 55°C but it can not tolerate a pH value of 5.0 and therefore will not grow on Thermoacidurans Agar.

Proteose peptone and yeast extract provide nitrogenous compounds, vitamin B complex and other essential growth nutrients. Dipotassium hydrogen phosphate buffers the medium. Dextrose acts as an energy source.

Extract juice from the canned foods and subject it to heat shock. Transfer 1 ml of the heat shocked sample to 4 sterile Petri plates and to each of 2 plates, add 10-20 ml Dextrose Tryptone Agar (M092) and to the other 2 plates, add 10-20 ml Thermoacidurans Agar. *B. coagulans* will form large, cream to white colonies.

### Type of specimen

Food samples

### Specimen Collection and Handling

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (4).

After use, contaminated materials must be sterilized by autoclaving before discarding.

### Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

### Limitations :

1.This medium is general purpose medium and may not support the growth of fastidious organisms.

## Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

## Quality Control

### Appearance

Cream to yellow homogeneous free flowing powder

### Gelling

Firm, comparable with 2.0% Agar gel.

### Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates

### Reaction

Reaction of 3.9% w/v aqueous solution at 25°C. pH : 5.0±0.2

### pH

4.80-5.20

### Cultural Response

Cultural characteristics observed after an incubation at 55°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery	Sporulation
<i>Bacillus thermoacidurans</i> ATCC 8038	50-100	luxuriant	≥70%	positive

Key : \*Corresponding WDCM numbers.

## Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Use before expiry date on the label.

Product performance is best if used within stated expiry period.

## Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (2,3).

## Reference

1. Becker M. E., Pederson C. S., 1950, J. Bacteriol., 459:717
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2<sup>nd</sup> Edition.
3. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
4. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
5. Stern R. N., Hegarty C. P. and Williams O. B., 1942, Food Research, 7:186.

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### Disclaimer :

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