

# 30787 Deoxyribonuclease Test Agar (DNase Test Agar)

Deoxyribonuclease Test Agar is recommended for the detection of deoxyribonuclease activity of bacteria and fungi, and especially for identification of pathogenic Staphylococci.

## **Composition:**

Ingredients	Grams/Litre
Tryptose	20.0
Deoxyribonucleic acid	2.0
Sodium chloride	5.0
Agar	15.0
Final nH 7 3 +/- 0 2 at 37°C	

Final pH 7.3 +/- 0.2 at 37°C

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

Appearance: Faintly beige colored, homogeneous, free flowing powder.

Gelling: Firm

Color and Clarity: Slightly brownish-yellow colored, clear to slightly opalescent gel forms in petri

plates.

#### **Directions:**

Suspend 42 g in 1 litre of distilled water and heat to the boiling and constant stiring to dissolve completely. Sterilize by autoclaving at 121°C for 15 minutes. Cool to 50°C and pour it into the plates. Bacteria are streaked on to the surface of the agar medium and incubated. After inoculation and 18-24 hours incubation the growth on the surface of the agar is flooded with 1N hydrochloric acid. Polymerised DNA precipitates in the presence of 1N HCl and makes the medium opaque. If the organisms produce DNase enzymes, in sufficient quantity to hydrolyse the DNA, then clear zones are seen around the colonies.

### **Principle and Interpretation:**

Tryptose is a source of nitrogen, vitamins, amino acids and other essential growth nutrients. Deoxyribonucleic acid (DNA) can be hydrolysed by microorganisms producing DNase. If the medium is then flooded with 1 N HCl not hydrolysed DNA precipitates (turbidity) and around DNase-positive colonies clear zones can be observed. Sodium chloride maintains the osmotic balance of the medium and Agar is the solidifying agent.

Instead of flooding the medium with 1N HCl it is also possible to use toluidine blue, crystal violet or methyl green as an indicator [4,5,6]. With the addition of indicator a faster identification of *Serratia marcescens* is achieved. Gram-negative DNase producing bacilli grow on this medium with indicator may accepted as *Serratia* species. May some strains of Staphylococci are inhibited by using indicator. Staphylococci can also be differentiated by ability to ferment mannitol. This can be determined simultaneously by adding 10g/l mannitol and 0.025 g/l of phenol red (a pH indicator turns from red to yellow) to the culture medium.



Cultural characteristics after 24-48 hours at 35±2°C.

Organisms (ATCC)	Growth	DNase
Serratia marcescens (274)	+++ (red colonies)	+
Serratia marcescens (13880)	+++	+
Pseudomonas aeruginosa (27853)	+++	-
Staphylococcus aureus (25923)	+++	+
Staphylococcus aureus (6538)	+++	+
Escherichia coli (25922)	+++	-

### References:

- 1. C.D. Jeffries, D.F. Holtmann, D.G. Guse, Rapid method for determining the activity of microorganisms on nucleic acid, J. Bact., 73, 590-591 (1957)
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- 3. J.W. DiSalvo, Med. Tech. Bull., U.S. Armed Forces Med. J. 9,191 (1958)
- 4. M.M. Streitfield, E.M. Hoffmann, H.M. Janklow, Evaluation of extra-cellular deoxyribonuclease activity in Pseudomonas, J. Bact., 84, 77-80 (1962)
- 5. J.B. Schreier, Modification of Deoxyribonuclease Test Medium for rapid identification of *Serratia marcescens*, Amer. J. Clin. Pathol., 51, 711-716 (1969)
- 6. P.B. Smith, G.A. Hancock, D.L. Rhoden, Improved Medium for Detecting Deoxyribonuclease-Producing Bacteria, Appl. Microbiol., 18, 991-993 (1969)
- 7. International Organization for Standardization (ISO), Meat and meat products Detection and enumeration of *Staphylococcus aureus* (Reference methods), Draft ISO/DIS 5551 (1977).

### **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

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