



## Double Sugar Agar, Russell

M057

Double Sugar Agar, Russell is used for the differentiation of gram-negative enteric bacilli on the basis of their ability to ferment dextrose and lactose with or without gas formation.

### Composition\*\*

Ingredients	Gms / Litre
Peptic digest of animal tissue	2.500
Casein enzymic hydrolysate	7.500
Beef extract	3.000
Lactose	10.000
Dextrose	1.000
Sodium chloride	5.000
Phenol red	0.025
Agar	15.000
Final pH ( at 25°C)	7.3±0.2

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

### Directions

Suspend 44.02 grams in 1000 ml distilled water. Heat to boiling to dissolve the medium completely. Dispense in tubes or as desired and sterilize by autoclaving at 118-121°C (correspond to 12-15lbs pressure respectively) for 15 minutes. Allow the tubes to solidify in slanting position to form a generous butt.

### Principle And Interpretation

Gram-negative bacilli belonging to *Enterobacteriaceae* are the most frequently encountered bacterial isolates recovered from clinical specimens. Definitive identification of the members of *Enterobacteriaceae* requires a battery of biochemical tests (1). Double Sugar Agar, Russell is used for the differentiation of gram-negative enteric bacilli on the basis of their ability to ferment dextrose and lactose with or without gas formation. This medium was originally formulated by Russell (2) using litmus indicator. It was later modified by Nichols (3) and Nichols and Wood (4) by replacing the litmus indicator with phenol red. This medium is used for differentiating gram-negative enteric bacilli especially the colon-typhoid-salmonellae-dysentery groups based on the fermentation of the double sugars incorporated namely, dextrose and lactose.

On incubation of inoculated tubed medium, acid production under aerobic condition (on the slant) and under anaerobic condition (in the butt) can be detected by the change in colour of the indicator. Phenol red is the pH indicator in the medium. Gaseous fermentation is indicated by splitting of the agar or by bubble formation in the butt. Organism like *Salmonella* Typhi capable of fermenting dextrose but not lactose will show an initial acid slant in short incubation period. Over a period of time as the dextrose gets consumed the reaction under aerobic condition reverts and becomes alkaline due to the oxidation of acids. Under anaerobic condition (in the butt), the same organism fails to revert the reaction and remains acidic.

Peptic digest of animal tissue, casein enzymic hydrolysate and beef extract serve as sources of carbon, nitrogen, vitamins and other essential nutrients. Lactose and dextrose serve as sources of energy by being the fermentable carbohydrates. Phenol red is the pH indicator in the medium that is pink under alkaline conditions and yellow under acidic conditions. Sodium chloride helps to maintain the osmotic equilibrium of the medium. Pure cultures are used to inoculate the tubed medium (5).

### Quality Control

#### Appearance

Light yellow to pink homogeneous free flowing powder

#### Gelling

Firm, comparable with 1.5% Agar gel

#### Colour and Clarity of prepared medium

Red coloured, clear to slightly opalescent gel forms in tubes as slants

**Reaction**

Reaction of 4.4% w/v aqueous solution at 25°C. pH : 7.3±0.2

**pH**

7.10-7.50

**Cultural Response**

M057: Cultural characteristics observed after an incubation at 35-37°C for 18-40 hours.

Organism	Inoculum (CFU)	Growth	Slant	Butt	Gas
<b>Cultural Response</b> <i>Enterobacter aerogenes</i> ATCC 13048	50-100	luxuriant	acidic reaction, yellowing of the medium	acidic reaction, yellowing of the medium	positive reaction
<i>Escherichia coli</i> ATCC 25922	50-100	luxuriant	acidic reaction, yellowing of the medium	acidic reaction, yellowing of the medium	positive reaction
<i>Proteus vulgaris</i> ATCC 13315	50-100	luxuriant	alkaline reaction, red colour of the medium	acidic reaction, yellowing of the medium	positive reaction
<i>Pseudomonas aeruginosa</i> ATCC 27853	50-100	luxuriant	alkaline reaction, red colour of the medium	alkaline reaction, red colour of the medium	negative reaction
<i>Salmonella Typhimurium</i> ATCC 14028	50-100	luxuriant	alkaline reaction, red colour of the medium	acidic reaction, yellowing of the medium	positive reaction
<i>Shigella dysenteriae</i> ATCC 13313	50-100	luxuriant	alkaline reaction, red colour of the medium	acidic reaction, yellowing of the medium	negative reaction

**Storage and Shelf Life**

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label

**Reference**

1. Koneman E. W., Allen S. D., Janda W. M., Schreckenberger P. C., Winn W. C. Jr., 1992, Colour Atlas and Textbook of Diagnostic Microbiology, 4th Ed., J. B. Lippincott Company
2. Russell F. F., 1911, J. Med. Res., 25:217.
3. Nichols H. J., 1921, J. Infect. Dis., 2982
4. Nichols H. J. and Woods C. B., 1922, J. Infect. Dis., 30, 320
5. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore

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