

TM 1554 - KOHN TWO TUBE MEDIUM NO. 1

INTENDED USE

for identification of *Enterobacteriaceae* on the basis of dextrose and mannitol fermentation and urease production.

PRODUCT SUMMARY AND EXPLANATION

Russell first introduced Double Sugar Medium, a differentiating medium for *Enterobacteriaceae*. Kohn later developed a technique employing two tubes of composite media for study of culture reactions, for the identification of *Enterobacteriaceae*. Gillies further made minor modifications in Kohn's media. Kohn Two Tube Medium No.1 Base is used to study dextrose and mannitol fermentation along with urease production.

COMPOSITION

Ingredients	Gms / Ltr
Peptic digest of animal tissue	15.000
Beef extract	2.000
Yeast extract	2.000
Dextrose	1.000
Mannitol	10.000
Phenol red	0.050
Agar	16.000

PRINCIPLE

Phenol red is the pH indicator in Kohn Two Tube Medium No. 1 and organisms capable of fermenting only dextrose show a yellow butt with or without gas formation and the slant remains unchanged (red). A yellow slant indicates the fermentation of mannitol. A positive urease reaction is shown by a deep cerise (light red) colour of whole medium.

INSTRUCTION FOR USE

- Dissolve 46.05 grams in 975 ml distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 115°C (10 psi pressure) for 15 minutes.
- Cool to 60°C and aseptically add 25 ml of sterile 40% (w/v) Urea solution.
- Mix well and make slants with a generous butt.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder	: Light yellow to light pink homogeneous free flowing powder.
Appearance of prepared medium	: Pink coloured, clear to slightly opalescent gel forms in tubes as slants with a generous butt.
pH (at 25°C)	: 7.2±0.2

INTERPRETATION

Cultural characteristics observed with added sterile 40% w/v Urea Solution after an incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Fermentation of Dextrose	Fermentation of Mannitol	Urease production	Incubation Temperature	Incubation Period
<i>Proteus vulgaris</i>	13315	50-100	Apparent negative reaction, urease activity masks fermentation reaction	Apparent negative reaction, urease activity masks fermentation reaction	Positive reaction, cerise colour	35-37°C	18-24 Hours
<i>Salmonella Typhi</i>	6539	50-100	Acid production, yellow colour	Acid production, yellow colour	Negative reaction, no change	35-37°C	18-24 Hours
<i>Salmonella Enteritidis</i>	13076	50-100	Acid & gas production, yellow colour	Acid production, yellow colour	Negative reaction, no change	35-37°C	18-24 Hours
<i>Shigella flexneri</i>	12022	50-100	Acid production, yellow colour	Acid production, yellow colour	Negative reaction, no change	35-37°C	18-24 Hours
<i>Shigella sonnei</i>	25931	50-100	Acid production, yellow colour	Acid production, yellow colour	Negative reaction, no change	35-37°C	18-24 Hours

PACKAGING:

In pack size of 100 gm and 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. Russell F. F., 1911, J. Med. Res., 25:217.
2. Kohn J., 1954, J. Path. Bacteriol., 67(1): 286.
3. Gillies R. R., 1956, J. Clin. Pathol., 9(4):368.





NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**
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