Technical Information

GN Broth, Hajna

Product Code: DM 1242

Application: - GN Broth, Hajna, is recommended for selective enrichment of gram-negative enteric organisms.

Composition**

Ingredients	Gms / Litre
Tryptose	20.000
Dextrose	1.000
Mannitol	2.000
Sodium citrate	5.000
Sodium deoxycholate	0.500
Dipotassium phosphate	4.000
Monopotassium phosphate	1.500
Sodium chloride	5.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

Hajna [1] developed Gram Negative (GN) Broth as an enrichment medium for recovery of *Salmonella* and *Shigella* from clinical and non-clinical specimens such as urine, blood clots, throat swabs, swabs from eating and drinking utensils etc [1, 2]. GN Broth, Hajna is also recommended by APHA [3] for the microbiological examination of foods. Croft and Miller isolated more strains of *Shigella* from rectal swabs using this medium [9]. Taylor and Schelhart showed the superiority of GN Broth to selenite enrichment media for isolation of *Shigella* [10]. Hajna [2, 4] also suggested the enrichment of organisms by keeping rectal swabs in this medium for 4-6 hours before plating on solid media. The medium contains tryptose, which provides amino acids and other nitrogenous substances to support bacterial growth. The combination of sodium citrate and sodium deoxycholate inhibit growth of gram-positive and some gram-negative bacteria such as coliforms. Phosphates serve as a buffering system. Sodium chloride maintains osmotic equilibrium. The higher concentration of mannitol over dextrose limits the growth of Proteus and enhances growth of mannitol fermenting *Salmonella* and *Shigella.* During the first 6 hours of incubation *Proteus, Pseudomonas* and coliforms do not overgrow *Salmonella* and *Shigella* in GN Broth. To increase the probability of isolating pathogens [3, 5, 7] this enrichment broth should be used in conjunction with selective and nonselective plating media.

GN Broth, Hajna should be inoculated directly with the specimen. In case of stool specimens, approximately 1 gram should be used for inoculation. Appropriate references for processing of clinical and food samples should be followed [3, 5, 6, 8]. After incubation of 6-8 hours and again after 24 hours, sub culturing on selective agar media should be carried out [7].

Methodology

Suspend 39 grams of powder media in 1000 ml distilled water. Shake well & dispense in test tubes. Sterilize by autoclaving at 115°C for 15 minutes. AVOID EXCESSIVE HEATING.

Quality Control

Physical Appearance
Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium
Light amber coloured, clear to slightly opalescent solution in tubes.

Reaction
Reaction of 3.9% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH range 6.80-7.20

Cultural Response/ characteristices
DM 1242: Cultural characteristics observed after an incubation at 35 - 37°C for 18 - 24 hours.

Organism	Inoculum (CFU)	Growth in GN broth	Growth after 24 hours on MacConkeyAgar	Colour of colony
Escherichia coli ATCC 25922	50-100	good	good	pink-red with bile ppt
Enterococcus faecalis ATCC 19433	50-100	none-poor	none-poor	pale pink-red
Proteus mirabilis ATCC 25933	50-100	good	good	colourless
Pseudomonas aeruginosa ATCC 27853	50-100	good	good	colourless
Salmonella Typhimurium ATCC 14028	50-100	good	good	colourless
Shigella flexneri ATCC 12022	50-100	good	good	colourless

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label.
Prepared Media: 2-8° in sealable plastic bags for 2-5 days.

Further Reading

1. Hajna A. A., 1955, Publ. Health Lab., 13:59.
2. Hajna A. A., 1955, Publ. Health Lab., 13:83.
3. Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.
4. Hajna A. A., 1956, Air. Univ. Sch. Ar. Med., USAF, 56:39
5. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
6. Forbes B. A., Sahm A. S., and Weissfeld D. F., Bailey & Scotts Diagnostic Microbiology, 10th Ed., 1998, Mosby, Inc., St. Louis, Mo.
7. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. 1, Williams and Wilkins, Baltimore.
8. Ewing, 1986, Edwards and Ewings Identification of Enterobacteriaceae, 4th Ed., Elsevier Science Publishing Co., Inc., New York, N.Y.
9. Croft C. C., Miller M. J., 1956, Am. J. Clin. Pathol., 26:411.
10. Taylor W.I., Schelhart D., 1968, Appl. Environ. Microbiol., 16:1383.

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