GN Broth • GN Broth, Hajna

Intended Use

GN Broth is used for the selective enrichment of *Salmonella* and *Shigella*.

Summary and Explanation

GN (Gram Negative) Broth was developed by Hajna as an enrichment medium for the recovery of *Salmonella* and *Shigella* from clinical and nonclinical specimens.[1,2] Croft and Miller succeeded in isolating more *Shigella* strains by use of this medium, rather than by direct streaking.[3] Taylor and Schelhart reported that GN Broth enhanced the isolation of enteric pathogens, producing a 53% increase in *Shigella* and a 36% increase in *Salmonella* as compared to direct streaking.[4] In another study, Taylor and Schelhart showed that GN Broth was superior to selenite enrichment media for the isolation of *Shigella*.[5]

GN Broth currently is recommended for use in the microbiological examination of foods.[6]

Principles of the Procedure

Peptones provide amino acids and other nitrogenous substances to support bacterial growth. Mannitol and dextrose are sources of energy. Mannitol is provided in a higher concentration than dextrose to enhance the growth of mannitol-fermenting species, such as *Salmonella* and *Shigella*, and limit the growth of *Proteus* and other dextrose-fermenting bacteria. Phosphate buffers are incorporated to maintain the pH of the medium. Sodium citrate and sodium desoxycholate are added to inhibit gram-positive and some gram-negative bacteria.

Proteus, *Pseudomonas* and coliforms do not overgrow *Salmonella* and *Shigella* in GN Broth during the first 6 hours of incubation.

User Quality Control

NOTE: Differences in the Identity Specifications and Cultural Response testing for media offered as both **Difco**™ and **BBL**™ brands may reflect differences in the development and testing of media for industrial and clinical applications, per the referenced publications.

Identity Specifications

Difco™ GN Broth, Hajna

Dehydrated Appearance:	Off-white to light tan, free-flowing, homogeneous.
Solution:	3.9% solution, soluble in purified water. Solution is light amber, clear to slightly opalescent.
Prepared Appearance:	Light amber, clear to slightly opalescent.
Reaction of 3.9% Solution at 25°C:	pH 7.0 ± 0.2

Cultural Response

Difco™ GN Broth, Hajna

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C for 18-24 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
Enterococcus faecalis	19433	10^3-2×10^3	None to poor
Escherichia coli	25922	10^2-10^3	Good
Salmonella enterica subsp. *enterica* serotype Typhimurium	14028	10^2-10^3	Good
Shigella flexneri	12022	10^2-10^3	Good

Identity Specifications

BBL™ GN Broth

Dehydrated Appearance:	Fine, dry, homogeneous, free of extraneous material.
Solution:	3.9% solution, soluble in purified water. Solution is pale to medium, tan to yellow, clear to slightly hazy.
Prepared Appearance:	Pale to medium, tan to yellow, clear to slightly hazy.
Reaction of 3.9% Solution at 25°C:	pH 7.0 ± 0.2

Cultural Response

BBL™ GN Broth

Prepare the medium per label directions. Inoculate and incubate at 35 ± 2°C; subculture to MacConkey II Agar after 6 hours and again after 18-24 hours of incubation. Incubate subculture plates at 35 ± 2°C for 18-24 hours.

ORGANISM	ATCC™	INOCULUM CFU	RECOVERY
Escherichia coli	25922	10^2-10^3	Good
Salmonella enterica subsp. *enterica* serotype Typhimurium	14028	10^2-10^3	Good
Shigella sonnei	9290	10^2-10^3	Good

Formulae

Difco™ GN Broth, Hajna

Approximate Formula* Per Liter		
Pancreatic Digest of Casein	12.0	g
Proteose Peptone No. 3	8.0	g
Dextrose	1.0	g
D-Mannitol	2.0	g
Sodium Citrate	5.0	g
Sodium Desoxycholate	0.5	g
Dipotassium Phosphate	4.0	g
Monopotassium Phosphate	1.5	g
Sodium Chloride	5.0	g

BBL™ GN Broth

Approximate Formula* Per Liter		
Pancreatic Digest of Casein	10.0	g
Peptic Digest of Animal Tissue	10.0	g
Dextrose	1.0	g
D-Mannitol	2.0	g
Sodium Citrate	5.0	g
Sodium Desoxycholate	0.5	g
Dipotassium Phosphate	4.0	g
Monopotassium Phosphate	1.5	g
Sodium Chloride	5.0	g

**Adjusted and/or supplemented as required to meet performance criteria.*

Directions for Preparation from Dehydrated Product

1. Suspend 39 g of the powder in 1 L of purified water. Mix thoroughly.
2. Dispense and autoclave at 121°C for 15 minutes.
3. Alternatively, the broth may be steamed for 30 minutes at 100°C.
4. Test samples of the finished product for performance using stable, typical control cultures.

Procedure

Inoculate the broth as soon as possible after the specimen arrives at the laboratory. Swab specimens may be inserted directly into the broth. For stool specimens, use 1 g of feces or 1 mL of liquid stool per tube. Consult appropriate references for information about the processing and inoculation of other clinical specimens or food samples.[6-9]

Incubate the tubes with loosened caps at 35 ± 2°C and subculture onto selective and differential media after 6-8 hours of incubation and again after 18-24 hours of incubation.[10]

Expected Results

Growth in broth media is indicated by turbidity compared to an uninoculated control. Subculture onto appropriate selective and differential media to isolate pathogens for identification.

Limitation of the Procedure

Enrichment broths should not be used as the sole isolation medium. They are to be used in conjunction with selective and nonselective plating media to increase the probability of isolating pathogens, especially when they may be present in small numbers. Consult references for detailed information and recommended procedures.[6-9]

References

1. Hajna. 1955. Public Health Lab. *13*:59.
2. Hajna. 1955. Public Health Lab. *13*:83.
3. Croft and Miller. 1956. Am. J. Clin. Pathol. *26*:411.
4. Taylor and Schelhart. 1967. Am. J. Clin. Pathol. *48*:356.
5. Taylor and Schelhart. 1968. Appl. Microbiol. *16*:1383.
6. Downes and Ito (ed.). 2001. Compendium of methods for the microbiological examination of foods, 4th ed. American Public Health Association, Washington, D.C.
7. Murray, Baron, Jorgensen, Landry and Pfaller (ed.). 2007. Manual of clinical microbiology, 9th ed. American Society for Microbiology, Washington, D.C.
8. Forbes, Sahm and Weissfeld. 2007. Bailey & Scott's diagnostic microbiology, 12th ed. Mosby, Inc., St. Louis, Mo.
9. Ewing. 1986. Edwards and Ewing's identification of *Enterobacteriaceae*, 4th ed. Elsevier Science Publishing Co., Inc., New York, N.Y.
10. MacFaddin. 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1. Williams & Wilkins, Baltimore, Md.

Availability

Difco™ GN Broth, Hajna

BS12 CCAM CMPH2 COMPF MCM9

Cat. No.	248610	Dehydrated – 500 g

BBL™ GN Broth

BS12 CCAM CMPH2 COMPF MCM9

Cat. No.	211279	Dehydrated – 500 g
	221729	Prepared Tubes, 8 mL (K Tubes) – Pkg. of 10*
	221730	Prepared Tubes, 8 mL (K Tubes) – Ctn. of 100*

**Store at 2-8°C.*