

## GC AGAR (7104)

### **Intended Use**

**GC Agar** is used with hemoglobin and enrichment for the isolation and cultivation of *Neisseria gonorrhoeae* and other fastidious organisms.

### **Product Summary and Explanation**

In 1945, Johnston described a medium that grew *N. gonorrhoeae* in 24 rather than 48 hours.<sup>1</sup> GC Agar was introduced in 1947 with reduced agar content. While investigating the growth rate of gonococcus, a medium containing growth factors glutamine and cocarboxylase improved recovery.<sup>2</sup> In 1964, Thayer and Martin formulated a medium incorporating the antibiotics Polymyxin B and Rostocetin, with added supplements, into GC Agar.<sup>3</sup> Thayer and Martin improved their medium by replacing the antibiotics with a new solution of Colistin, Vancomycin, and Nystatin (CVN).<sup>4</sup> In 1970, Martin and Lester improved the new Thayer-Martin Medium by increasing the agar and glucose, and adding Trimethoprim Lactate (T), calling it Modified Thayer-Martin (MTM) Medium.<sup>5</sup> Martin and Lewis improved selectivity of MTM by increasing the concentration of Vancomycin and replacing Nystatin with Anisomycin for greater inhibition of yeasts, known as Martin Lewis (ML) Agar.<sup>6</sup> Transgrow Medium is a transport medium incorporating either MTM or ML formulations.<sup>7</sup>

### **Principles of the Procedure**

GC Agar is employed as a basal medium in the preparation of Chocolate Agar, Thayer-Martin Medium, Modified Thayer-Martin Medium, Martin-Lewis Agar, and Transgrow Agar.

Enzymatic Digest of Casein and Enzymatic Digest of Animal Tissue provide nitrogen, carbon, and minerals in GC Agar. Corn Starch absorbs any toxic metabolites produced. The Phosphates are buffering agents. Sodium Chloride maintains osmotic balance of the medium. Agar is the solidifying agent. Chocolate Agar is prepared from GC Agar with the addition of 2% Hemoglobin. Hemoglobin provides Hemin (X factor) required for growth of *Haemophilus* and enhanced growth of *Neisseria* spp. A chemical enrichment composed of cofactors, vitamins, and nicotinamide adenine dinucleotide (NAD) are also required for growth of *Haemophilus* and *Neisseria* spp. If required, antimicrobial supplements are added as inhibitors for improved selectivity of the medium.

### **Formula / Liter**

Enzymatic Digest of Casein .....	7.5 g
Enzymatic Digest of Animal Tissue.....	7.5 g
Corn Starch.....	1 g
Dipotassium Phosphate .....	4 g
Monopotassium Phosphate .....	1 g
Sodium Chloride .....	5 g
Agar .....	10 g

Final pH: 7.2 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

### **Supplements**

Hemoglobin Solution, 2%, 100 mL  
 Growth Enrichment, 2 mL  
 Antimicrobials, if required

### **Precautions**

1. For Laboratory Use.
2. IRRITANT. Irritating to eyes, skin, and respiratory system.

### **Directions, Double Strength**

1. Suspend 7.2 g of the medium in 100 mL of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes. Cool to 45 – 50°C.
4. Prepare 100 mL of a 2% hemoglobin solution and autoclave at 121°C for 15 minutes.
5. Cool to 45 - 50°C and aseptically add to the molten GC Agar.
6. Add 2 mL of growth enrichment. Add antimicrobials, if desired. Mix thoroughly and dispense.

Note: Refer to appropriate references for media formulations of Thayer-Martin Medium, Modified Thayer-Martin Medium, Martin Lewis Agar, and Transgrow Agar.<sup>8,9</sup>

**Quality Control Specifications**

**Dehydrated Appearance:** Powder is homogeneous, free flowing, and light beige.

**Prepared Appearance:** Prepared GC Agar supplemented as Chocolate Agar is opaque and brown.

**Expected Cultural Response:** Cultural response on Chocolate Agar incubated at 35 ± 2°C aerobically or under 7 – 10% CO<sub>2</sub>, as appropriate, and examined for growth after 18 – 24 hours.

Microorganism	Approx. Inoculum (CFU)	Expected Growth
<i>Haemophilus influenza</i> ATCC® 10211	10 - 300	Good to excellent
<i>Neisseria gonorrhoeae</i> ATCC® 43070	10 - 300	Good to excellent
<i>Neisseria meningitidis</i> ATCC® 13090	10 - 300	Good to excellent
<i>Streptococcus agalactiae</i> ATCC® 13813	10 - 300	Good to excellent
<i>Streptococcus pneumoniae</i> ATCC® 6303	10 - 300	Good to excellent

The organisms listed are the minimum that should be used for quality control testing.

**Test Procedure**

For a complete discussion on the isolation and identification of *Neisseria* spp. and *Haemophilus* spp. consult procedures outlined in the references.<sup>8,9</sup>

**Results**

Refer to appropriate references and procedures for results.

**Storage**

Store sealed bottle containing the dehydrated medium at 2 - 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

**Expiration**

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

**Limitation of the Procedure**

Although certain diagnostic tests may be performed directly on GC Agar, biochemical and immunological testing using pure cultures are recommended for complete identification.

**Packaging**

<b>GC Agar</b>	<b>Code No.</b>	<b>7104A</b>	<b>500 g</b>
		<b>7104B</b>	<b>2 kg</b>
		<b>7104C</b>	<b>10 kg</b>

**References**

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3. Thayer, J. D., and J. E. Martin, Jr. 1966. Improved medium selective for cultivation of *N. gonorrhoeae* and *N. meningitidis*. Public Health Rep. **81**:559.
4. Thayer, J. D., and A. Lester. 1971. Transgrow, a medium for transport and growth of *Neisseria gonorrhoeae* and *Neisseria meningitidis*. HSMHA Health Service Rep. **86**:30.
5. Martin, J. E., Jr., and R. L. Jackson. 1975. A biological environmental chamber for the culture of *N. gonorrhoeae* with a new commercial medium. Public Health Rep. **82**:361.

6. **Martin, J. E., Jr., and J. S. Lewis.** 1977. Anisomycin: improved anti-mycotic activity in modified Thayer-Martin Medium. Public Health Rep. **35**:53.
7. **MacFaddin, J. F.** 1985. Media for isolation-cultivation-identification-maintenance of medical bacteria, vol. 1, Williams & Wilkins, Baltimore, MD.
8. **Isenberg, H. D. (ed.)**. 1992. Clinical microbiology procedures handbook. vol. 1. American Society for Microbiology, Washington, D.C.
9. **Murray, P. R., E. J. Baron, M. A. Tenover, and R. H. Tenover (eds.)**. 1995. Manual of clinical microbiology, 6<sup>th</sup> ed. American Society for Microbiology, Washington, D.C.

### **Technical Information**

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.



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