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## TECHNICAL PRODUCT INFORMATION

### Todd Hewitt w/CNA Broth (LIM Broth)

Catalog No.: T1865

#### INTENDED USE:

Lim Broth (Todd Hewitt Broth w/Colistin and Nalidixic Acid) is a selective enrichment medium which may be used in qualitative procedures to isolate Group B Streptococci from clinical specimens containing mixed bacterial flora. Latex or coagglutination tests may be performed on the broth.

#### HISTORY/SUMMARY:

Group B Streptococci (GBS) are frequently responsible for neonatal sepsis, meningitis and respiratory distress. Infants usually become infected during parturition. Studies have shown that women, heavily colonized with Group B Streptococci, gave birth to infants which were also heavily colonized.<sup>1-4</sup> In 1986 and 1987; Lim et al. reported the usefulness of screening women in the third trimester of pregnancy for GBS using Lim Broth and coagglutination testing. Todd Hewitt Broth with colistin and nalidixic acid is a medium recommended to maximize the likelihood of recovering group B streptococci when sub-culturing to TSA w/Sheep Blood.

Group B streptococci have also been seen in sepsis of nonparturient women and men, in joint infections, osteomyelitis, UTI and wound infections. GBS may be associated with endocarditis, pneumonia and pyelonephritis in immunosuppressed patients.<sup>8</sup>

#### PRINCIPLES:

Lim Broth is prepared from beef heart infusion (nitrogen source), Yeast Enriched Peptone (vitamins and essential minerals), which provide an excellent nutritional base for streptococcal development. Dextrose is used in the medium as a source of carbon and energy and hemolysin production.<sup>8, 9</sup> Disodium phosphate and sodium carbonate provide buffering action to counteract the acidity produced during the fermentation of the carbohydrate, thus protecting the hemolysin from inactivation by the acid. The addition of colistin and nalidixic acid inhibits the growth of gram negative bacteria.

#### FORMULA:

Final pH: 7.8 ± 0.2 @ 25°C

INGREDIENT	AMOUNT PER LITER
Beef Heart Infusion (dehydrated)	3.1 g
Yeast Enriched Peptone	20.0 g
Dextrose	2.0 g
Sodium Chloride	2.0 g
Disodium Phosphate	0.4 g
Sodium Carbonate	2.5 g
Colistin	0.013 g
Nalidixic Acid	0.015 g

#### PRECAUTIONS:

This medium is for IN VITRO DIAGNOSTIC USE. It supports the growth of pathogens and should be handled with caution by adequately trained personnel under the supervision of a microbiologist. Media showing signs of deterioration or contamination must not be used. Media must be brought to room temperature before use.

**STORAGE:**

This media should be stored in the dark at 2-8°C. Do not use media beyond the expiration date.

**SPECIMEN COLLECTION:**

Specimens suitable for culture may be obtained using various techniques. For detailed procedures consult appropriate texts.<sup>5-7</sup> Specimens should be obtained before administration of antimicrobial agents. Specimens should be delivered promptly to the laboratory.

**PROCEDURE:**

1. Inoculate the Lim Broth with swab specimens, recap loosely.
2. Incubate tubes at 35° ± 2°C in an aerobic atmosphere, with or without CO<sub>2</sub>, for 18-24 hours or up to 48 hours if needed.
3. Slide co-agglutination tests may be performed after five (5) hours of incubation.
4. If broth is cloudy after 18-24 hours, subculture to a TSA w/5% Sheep Blood or CNA with blood.
5. Observe for growth suggestive of group B streptococci (beta or non-hemolytic colonies, gram positive and catalase negative).
6. Confirm identification by performing streptococcal grouping sera testing, CAMP test or other procedures.
7. Refer to appropriate texts for further information regarding procedures.<sup>1,6,7</sup>

**GROWTH CHALLENGE PERFORMANCE CHARACTERISTICS:**

NEL approval for each lot of LIM Broth is based on results obtained on a number of tests, among them bacteriological performance with challenged organisms.

Organisms	Results
ATCC# 12386 <i>Streptococcus agalactiae</i>	Growth (moderate to heavy)
ATCC# 29212 <i>Enterococcus faecalis</i>	Growth (moderate to heavy)
ATCC# 25922 <i>Escherichia coli</i>	Inhibition(partial to complete)

**QUALITY CONTROL:**

It is recommended the user confirm the performance characteristics of this medium. Careful selection of organisms must be made to assure maximum testing success. Incubation temperatures and other environmental conditions must also be controlled to further warrant effective test results.

**REFERENCES:**

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- 6) Baron, E.J., and S.M. Finegold, 1990. *Bailey & Scott's Diagnostic Microbiology*, 8<sup>th</sup> edition, The Mosby Co., St. Louis.
- 7) Ballows, A., W.J. Hausier, Jr., K.L. Herrmann, H.D. Isenberg, and H.J. Shadomy (ed.). 1991. *Manual of Clinical Microbiology*, 5<sup>th</sup> edition, American Society for Microbiology, Washington, D.C.
- 8) Difco & BBL Manual, *Manual of Microbiological Culture Media*, 2003, Pages 305-306
- 9) Acumedia Product Information Sheet Todd Hewitt Broth (#7161) PI, Rev 04, Nov. 2010