

PRAS BRAIN HEART INFUSION SUPPLEMENTED BROTH

INTENDED USE

Remel PRAS Brain Heart Infusion Supplemented Broth is a liquid medium recommended for use in qualitative procedures for the cultivation of anaerobes.

SUMMARY AND EXPLANATION

R.E. Hungate pioneered the development of Pre-Reduced Anaerobically Sterilized (PRAS) media, which are processed in a reduced condition and remain reduced up to and after inoculation¹. Rosenow developed brain heart infusion broth which was prepared by adding pieces of brain tissue to a dextrose broth.² The use of a steam sterilized emulsion of brain tissue in water to support the growth of anaerobic bacilli was first described by von Hibler.³ PRAS Brain Heart Infusion Supplemented Broth is recommended for antimicrobial susceptibility testing methods, such as broth microdilution and modified broth-disk, and as a general purpose medium for the cultivation of anaerobic bacteria.⁴⁻⁷

PRINCIPLE

PRAS Brain Heart Infusion Supplemented Broth contains infusions of brain and heart tissue and peptones to supply protein and other necessary nutrients for the growth of microorganisms. Sodium chloride is added to maintain osmotic equilibrium and dipotassium phosphate acts as a buffer. Dextrose is an energy source. Cysteine, a reducing agent, contains sulfhydryl groups which bind hydrogen ions and produce anaerobic conditions. Resazurin, an Eh indicator, turns from colorless to pink upon exposure to oxygen indicating loss of anaerobic conditions. Vitamin K and hemin are added to satisfy the growth requirements for various anaerobic organisms. PRAS media are prepared and processed in an atmosphere of nitrogen and hydrogen.

REAGENTS (CLASSICAL FORMULA)*

Casein Peptone.....	15.0 g	Dextrose	2.0 g
Brain Heart Infusion/Meat Peptone	12.0 g	L-Cysteine Hydrochloride	0.5 g
Sodium Chloride.....	5.5 g	Hemin	5.0 mg
Yeast Extract.....	5.0 g	Resazurin	1.0 mg
Dipotassium Phosphate	2.5 g	Vitamin K	0.5 mg
		Demineralized Water.....	1000.0 ml

pH 7.4 +/- 0.2 @ 25°C

*Adjusted as required to meet performance standards.

PROCEDURE

Note: Inoculation of test isolate should be performed under anaerobic conditions using an anaerobic chamber, a specialized apparatus which supplies continuous gas flow into the tube during manipulation, or through a rubber stopper (Hungate cap) using a needle and syringe. Consult appropriate references for further guidelines.^{1,6,7}

1. Consult the appropriate references for the recommended procedure for inoculation and cultivation of test isolate.⁴⁻⁶
2. If using the closed or Hungate method, use a syringe and needle to inoculate through the rubber stopper. The diaphragm of the Hungate cap should be decontaminated with alcohol and allowed to dry prior to injection.
3. If using the open method, remove the cap and insert a cannula that has oxygen-free gas flowing from the tip. While the tube is open, inoculate using a Pasteur pipette. If inoculation occurs in an anaerobic chamber, the use of a cannula and oxygen-free gas is not required.
4. Incubate tube(s) aerobically at 35-37°C.
5. Examine for growth after 18-24 hours or longer, as required.

QUALITY CONTROL

All lot numbers of PRAS Brain Heart Infusion Supplemented Broth have been tested using the following quality control organisms and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

CONTROL

Bacteroides fragilis ATCC® 25285
Clostridium perfringens ATCC® 13124

INCUBATION

Anaerobic, 48 h @ 35-37°C
Anaerobic, 48 h @ 35-37°C

RESULTS

Good growth
Good growth

BIBLIOGRAPHY

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Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

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