

**CASEIN HYDROLYSATE BROTH****TM 570**

For production of *Staphylococcus* enterotoxin for use in Cat test and in serological studies

Composition

Ingredients	Gms/Ltr.
Casein acid hydrolysate	20.00
Sodium acetate	7.00
Potassium phosphate	2.00
Magnesium sulphate	0.20
L-Tryptophan	0.075
Ferric citrate	0.025
L-Cystine	0.025
Nicotinic acid	0.0012
Calcium pantothenate	0.0005
Thiamine	0.00004

* Dehydrated powder, store in a dry place, in tightly-sealed containers at 24°C and protect from direct sunlight.

Instructions for Use

Dissolve 29.33gms in 1000ml of distilled water. Gently heat to boiling with gentle swirling and dissolve the medium completely. Sterilize by autoclaving at 15 psi (121°C) for 15 minutes. Cool to 45 - 50°C mix well before pouring into the sterile tubes.

Appearance: Amber colour, slightly opalescent solution with slight precipitate

pH (at 25°C): 7.3 ± 0.2

Principle

CASEIN HYDROLYSATE BROTH is used for production of *Staphylococcus* enterotoxin for use in Cat test and in serological studies. The medium compositions are according to the recommendations and regulations of APHA. *Staphylococcus* is a Gram positive genus of bacteria and is well known food borne pathogens. The organism produces a toxic exotoxin, known as the enterotoxin which targets the intestine and cause the infection. This medium contains Casein acid hydrolysate to provide the carbonaceous and nitrogenous compounds to support the growth of microbes in the medium. The amino acids – L-Tryptophan and L-Cystine along with the vitamin derivatives – Nicotinic acid, Calcium pantothenate and Thiamine can provide the essential nutrients and other growth factors for the organisms. Magnesium sulphate provides cations in the medium required for the metabolism. Sodium acetate can induce the spore germination of the bacteria in the media. The Potassium phosphate can act as a buffering agent to maintain the pH of the medium favourable for the growth of the microorganisms while Ferric citrate can provide the iron cofactors required for the microbial metabolism. After the proper period of incubation, the broth culture will be centrifuged to obtain the supernatant fluid which is then sterilized by Seitz filtration. The filtrate obtained after sterilization is tested for alpha and beta haemolysins and if present, the toxins are denatured by heat or by neutralization with antiserum. After denaturation, filtrates can be injected in the cats to observe whether the vomiting is induced in the cats.

Interpretation

Cultural characteristics observed after inoculating the organisms and subsequent incubation at 35 – 37°C for 18 – 24 hours.

Microorganisms	ATCC	Inoculum (CFU/ml)	Growth
<i>Staphylococcus aureus</i>	25923	10 ³ – 10 ⁵	Luxuriant
<i>Streptococcus pyogenes</i>	19615	10 ³ – 10 ⁵	Luxuriant
<i>Escherichia coli</i>	25922	10 ³ – 10 ⁵	Luxuriant

References

1. American Type Culture Collection, Manassas, Va., USA.
2. Casman. 1958. Public Health Reports, 73:599.
3. Standard Methods for the Examination of Dairy Products. 1960. 11th ed., American Public Health Association, Inc. New York.