

COOKED MEAT MEDIA

PRODUCTS:

Tube Media:

Cooked Meat Media	T6300
Cooked Meat Media with Glucose	T6301

PURPOSE:

Cooked Meat Media is used for the cultivation and maintenance of aerobic, microaerophilic, and anaerobic organisms, especially *Clostridium* species. Cooked Meat Media with Glucose is used for the cultivation of organisms for the purpose of studying volatile fatty acids and alcohol produced by the organism, using gas-liquid chromatography.

PRINCIPLE:

In 1890 Theobald Smith⁶ first used animal tissue for culturing anaerobic organisms. Later, von Hible⁷ used brain tissue for cultivating and classifying anaerobic bacilli. This tissue was less susceptible to the toxic metabolic waste products produced by bacteria. Robertson⁵ substituted beef heart for the brain tissue and employed this media for differentiating putrefactive and saccharolytic species.

Cooked Meat Media incorporates beef heart, peptone, and a small amount of dextrose, providing the essential nutritional requirements for most bacteria. The dextrose content is low enough that toxic by-products do not accumulate. Cooked Meat Media has a growth-stimulating influence on bacteria. The by-products of growth do not kill the organism, making it possible to reisolate bacteria which would have otherwise died out or become overgrown by other bacteria. It is thought that the meat particles act as buffers to the extremes of conditions produced by the growth and reproduction of bacteria.² Evidently the meat absorbs undue amounts of acids. The changes of reaction in the media such as color, odor, and proteolysis make it useful in differentiating different species. Proteolysis of meat is particularly useful in the identification of *Clostridium* species. Proteolytic enzymes break down the beef heart to form amino acids, resulting in the breakdown of the solid particles. Proteolytic fermenters decompose and blacken the meat, forming foul-smelling sulfur compounds.

This media is also useful in initiating growth from a very small inoculum. Viability of organisms remains over a long period of time. Slow-growing organisms are able to proliferate in the presence of other bacteria, and therefore may still be recovered. The media is ideal for the maintenance of stock cultures; whereas, the Cooked Meat Media with Glucose, by nature of its increased dextrose concentration, is not.

FORMULAS:

Approximate, per liter deionized filtered water.

(1) Cooked Meat Media:

Beef Heart.....	454.0 g
Peptic Digest of Animal Tissue.....	10.0
Pancreatic Digest of Casein.....	10.0
Dextrose.....	2.0
Sodium Chloride	5.0
Final pH 7.2 ± 0.2 at 25°C	

(2) Cooked Meat Media with Glucose:

Same as (1) above except it also contains:	
Pancreatic Digest of Casein.....	25.0 g
Monopotassium Phosphate	5.0
Yeast Extract	5.0
Glucose.....	4.5
L-Cysteine HCl.....	0.5
Hemin.....	5.0 mg
Vitamin K ₁	0.1
Final pH 7.2 ± 0.2 at 25°C	

PRECAUTIONS:*

For *in vitro* diagnostic use. Observe approved biohazard precautions.

Storage: Upon receipt store at 2-30°C away from direct light. Media should not be used if there are signs of contamination, deterioration (evaporation or discoloration), or if the expiration date has passed.

Limitations: Media may become turbid when incubated at 35°C for 24-48 hours. Formation of gas within the first 24 hours of incubation is evidence of carbohydrate fermentation and not proteolysis. Proteolysis is not considered a contributory factor for production of gas with a saccharolytic organism. Acidity produced during carbohydrate

fermentation is sufficient to completely inhibit proteolysis. When gas production of saccharolytic organisms accumulates, growth is arrested. However, proteolysis produces ammonia and increases alkalinity, thereby arresting the acidity. Therefore, each type of anaerobic organism in the specimen aids and favors growth of the other type.

Blackening may be delayed or absent if the media remains acid. Blackening of meat particles only occurs in the presence of alkali; the reaction is a combination of action on iron and an alkaline environment.

While Cooked Meat Media is an excellent resource for organism maintenance, especially for anaerobes, the reactions that are observed in the media may only be used for characterization, and are not sufficient to speciate the organism. Further biochemical tests must be performed, and purity must be confirmed from this media. Cooked Meat Glucose Media may be used for a holding media only when gas liquid chromatography is performed.

PROCEDURE:*

Specimen Collection: These are not media generally used for primary isolation of organisms from clinical specimens. However, specimens from deep-seated sites where anaerobic infections are suspected may be inoculated directly into the media with the liquid, or macerated tissue. These media are also used in characterizing pure cultures of isolated cultures, or maintaining organisms in a pure or mixed environment. Established isolation techniques and tests for purity should be performed when performing biochemical tests from this media. Information on specimen collection may be found in standard reference texts.^{1,3}

Method of Use: Prior to inoculation, heat tubes in flowing steam or boiling waterbath for 15 minutes to drive off dissolved oxygen. Cool without agitating the tubes. Inoculate the meat particles of Cooked Meat Media with a pure culture of a well-isolated colony or macerated tissue or liquid from a clinical specimen. For clinical specimens, tubes may be held anaerobically for up to 21 days. For maintenance of stock cultures, tubes may be held in the dark at room temperature for 4-6 months.

Interpretation: Several reactions may occur in the Cooked Meat Media such as production of acid and gas, digestion of meat particles, blackening of the meat, and characteristic odor. A detailed account of these reactions and the organisms that produce them may be found in standard reference texts.⁴ Reactions in this media are not sufficient to speciate the organism. Additional physiological, biochemical, and serological reactions are required for definitive identification. The purity of the organism must always be checked by subculture onto a purity plate before additional biochemical tests are performed.

Materials Required but Not Provided: Standard microbiological supplies and equipment commonly found in a microbiology laboratory are not provided.

QUALITY CONTROL:*

Microorganisms Used (ATCC #):

Clostridium perfringens (13124)
Bacteroides fragilis (25285)

Expected Results:

Cooked Meat Media

Growth

Growth

Key: See "Interpretation"

with Glucose

Growth; proteolysis, (+) gas

Growth

User Quality Control: Check for signs of contamination and deterioration. Cooked Meat Media should appear as a clear liquid, amber in color, and with dark granules.

BIBLIOGRAPHY:

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*For more detailed information, consult appropriate references.

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