

Anaerobe Isolation Agar

Use and description:

This medium has high nutritive value due to its richness in peptones, extracts, haemin and amino acids. A primary isolation medium capable of growing most clinically significant anaerobes. Starch and sodium bicarbonate act as de-toxification agents. It contains for promoting agents *Fusobacterium necrophorum*, *Propionibacterium acnes*, *Bacteroides fragilis*, *Eubacterium* spp. and *Veillonella* spp. Pyruvate helps neutralise hydrogen peroxide.

Kanamycin 75 (MMS22) should be used for the selective isolation of *Clostridium* spp. and other anaerobes. Kanamycin is more inhibitory to anaerobic cocci. Metronidazole (MMS23) and Nalidixic acid (MMS25) are used for the isolation of *Actinomyces* spp. from clinical material. The metronidazole will suppress the growth of most other anaerobes. Nalidixic acid for the isolation of non-sporing anaerobes from clinical material. Vancomycin (MMS26) for the isolation of Gram negative anaerobes from clinical material. When used with other blood agar bases, further enrichment of the medium with haemin and menadione is beneficial. When Neomycin 100 (MMS04) is added to egg yolk medium this supplement will allow the growth of clostridia whilst inhibiting other lecithinase producing organisms. When Neomycin 75 (MMS24) is added to blood agar the resulting medium will allow the growth of clostridia, most *Bacteroides fragilis* strains and some anaerobic cocci.

Composition per liter:

Peptone mixture.....	21.76 g
Sodium chloride.....	5.00 g
Sodium bicarbonate.....	0.50 g
Dextrose.....	1.00 g
Sodium pyruvate.....	0.50 g
Amino acids factors.....	2.00 g
Soluble pyrophosphate.....	0.13 g
Vitamins and growth factors.....	11.00 mg
Agar.....	15.00 g

Final pH of the ready to use medium: 7.2 ± 0.2

Medium preparation:

Add 45.0 grams of dehydrated culture medium to 1 liter of distilled water. Allow to soak for 10 minutes, swirl to mix then sterilise by autoclaving at 121°C for 15 minutes. Cool to 48°C then aseptically add 5-10% of sterile defibrinated horse blood, mix well and pour into petri dishes.

Quality specification:

Dehydrated medium: homogeneous, straw colored fine powder.

Ready to use medium: red due to addition of blood. The blood will darken (reduce) because of the presence of reducing agents.

Microbiological response:

Organism	Result
<i>Bacteroides fragilis</i> ATCC 25285	Growth
<i>Clostridium perfringens</i> ATCC 13124	Growth

Storage:

Dehydrated medium should be stored between 10 to 25°C. Once opened, place the container in a dark, dry place. The dehydrated medium should not be used if there is any lump or if the color has changed from the original.

**For references please turn appendix 1.
For supplement details please turn appendix 2.**