ATCC Medium 1490: Modified Chopped Meat

Ground Beef (antibiotic & hormone free, low fat)*	500 g (roughly 1 pack)
DI Water	2000 mL
NaOH (1N)	25 mL

Wrap ground beef loosely with cheese cloth. Add all ingredients into large pot. Bring to boil for 20 minutes, simmer for 3 hours. Allow broth to cool, then remove ground beef. Filter through cheese cloth and store remaining broth in a large flask. Store overnight in walk-in refrigerator, allowing fat to solidify**.

The next day, filter the broth again through cheese cloth to remove fat. Filter remaining filtrate through 1L Corning filter using Whatman Glass Microfibre Filters as prefilters. Repeat 3 times. Broth should now be very clear and have a dark brown color. Store unused portion in -20 °C freezer.

Note: When preparing a large batch of chopped meat broth for MD-1490 or if using broth from MD-2922 it is required to have the broth filtered 6 times using Whatman's Filter Papers followed by an easy flow using the sterile filter. The broth must be clear and free of any fat particles unless specifically requested by the biologist. Successful results were identified when autoclaving media first then dispensing without agitating any remaining particles that may result after the autoclave process. Updated 19Jul2017EB

For 1L of complete media, use:

Ground Beef Broth	500.0 mL
Trypticase	30.0 g
Yeast Extract	5.0 g
K ₂ HPO ₄	5.0 g
Resazurin (1mg/ml solution)	1.0 mL
Agar (if necessary)	20.0 g
Hemin Solution (see next page)	10.0 mL
Vitamin K ₁ Solution (see next page) ().2 mL
DI Water	500.0 mL

Boil and cool medium. Add:

L-Cysteine HCl x H₂O..... 0.5 g

Adjust pH to 7.0. Autoclave at 121°C. Dispense medium anaerobically under N₂:H₂:CO₂ (80:10:10). Dispense 7ml of broth per test tube.

Hemin Solution

Hemin	50.0 mg
NaOH (1N)	1.0 mL
DI Water	100.0 mL

Filter sterilize, refrigerate. Discard after 30 days.

Vitamin K₁ Solution

Vitamin K ₁	0.15 mL
Ethanol	30.0 mL

Filter sterilize and store in dark bottle, refrigerate. Discard after 30 days.



Final Product Description

Agar Broth

- Dark brown/red in color
- Broth: translucent; Agar: slightly opaque
- > There may be very slight precipitate
- Recommended Storage: 2-8°C