

ATCC Medium: 0125 Thiobacillus Medium

REAGENTS:

Agar	Broth
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N/A <input type="checkbox"/> Salt Solution			
(NH ₄) ₂ SO ₄	0.2 g	N/A g	0 g
MgSO ₄ x 7H ₂ O	0.5 g	N/A g	0 g
CaCl ₂	0.25 g	N/A g	0 g
KH ₂ PO ₄	3 g	N/A g	0 g
FeSO ₄ x H ₂ O	5 mg	N/A mg	0 mg
Tap water	1000 mL	N/A mL	0 mL

N/A Sulfur

For Flask: place approximately 1.0 g of sulfur powder (precipitated) into a dry flask, per 100ml medium

For Tube: place approximately 0.06 g of sulfur powder (precipitated) into a dry test tube, per 6ml medium

PROCEDURE:

Steps	Check
N/A <input type="checkbox"/> To make Salt Solution:	
Accurately weigh out components for Salt Solution	<input type="checkbox"/>
Dissolve completely into Tap water	<input type="checkbox"/>
Start: _____ End: _____	
Filter sterilize	<input type="checkbox"/>
N/A <input type="checkbox"/> To make MD-0125:	
Weigh out and place sulfur powder (see above for gram needed) into a dry flask and/or tube	<input type="checkbox"/>
Flask only: loosely cover each flask with a screw cap	<input type="checkbox"/>
Tube only: place aluminum foil over the top of the racked tubes	<input type="checkbox"/>
Autoclave sulfur powder @ 100 °C for 30 minutes (P11 cycle @ 100 celcius deg)	<input type="checkbox"/>
Repeat autoclave sterilization for <u>3 consecutive days</u>	<input type="checkbox"/>
Aseptically dispense the filter sterilize salt solution into each of the sterilized Sulfur vessels	<input type="checkbox"/>
***NOTE: make sure to carefully pour the solution down the side of the flask. The sulfur powder should not "wet," it should float on top of the liquid	
Carefully replace cap	<input type="checkbox"/>

NOTE:

Salt solution volume to Sulfur ratio:

1.0 g of precipitated Sulfur powder per 100 mL of medium (flask)

0.1 g of precipitated Sulfur powder per 10 mL of medium (tube)

** Sulfur is insoluble and has a low melting point of 106.8°C. Therefore, it must be processed separately from the salt solution. Amount is determined by vessel size as indicated above.