

## **ATCC Medium: 2883 Modified 337a Agar/Broth Medium**

### **Broth Medium**

KH<sub>2</sub>PO<sub>4</sub>.....1.3 g  
Na<sub>2</sub>HPO<sub>4</sub>.....1.13 g  
(NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>.....0.5 g  
MgSO<sub>4</sub> x 7H<sub>2</sub>O.....0.2 g  
Trace element solution (see below).....5 ml  
DI Water.....1000 ml

Adjust pH to 7.5-8. Autoclave at 121C. Allow to cool down and aseptically add:

Methanol (filter sterilized).....3 ml  
Vitamin B12 (0.1 mg/ml, filter sterilized).....1 ml

Dispense as required.

### **Agar Medium**

Prepare a 2X stock of medium:

KH<sub>2</sub>PO<sub>4</sub>.....1.3 g  
Na<sub>2</sub>HPO<sub>4</sub>.....1.13 g  
(NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>.....0.5 g  
MgSO<sub>4</sub> x 7H<sub>2</sub>O.....0.2 g  
Trace element solution (see below).....5 ml  
DI Water.....500 ml

Adjust pH to 7.5 (not higher because medium will precipitate).

Prepare 2X agar solution:

Agar.....15 g  
DI Water.....500 ml

Autoclave both solutions at 121C. Allow to cool down, mix both solutions and aseptically add:

Methanol (filter sterilized).....3 ml  
Vitamin B12 (0.1 mg/ml, filter sterilized).....1 ml

Dispense as required.

\*\*\*\*Autoclaving of the 2X stock culture medium might cause the formation of a precipitate, but this won't interfere with the growth of the strain\*\*\*\*

**Trace Element Solution**

CaCl <sub>2</sub> x 2H <sub>2</sub> O.....	30.9 mg
FeSO <sub>4</sub> x 7H <sub>2</sub> O.....	20 mg
Na <sub>2</sub> MoO <sub>4</sub> x 2H <sub>2</sub> O.....	10 mg
MnSO <sub>4</sub> x 4H <sub>2</sub> O.....	8.8 mg
DI Water.....	50 ml

\*\*\*Trace elements solution can be autoclaved and stored at 4C. Autoclaving causes the formation of a precipitate, but this won't interfere with the growth of the strain (shake well before use).\*\*\*\*\*