

## ATCC Medium: 2764 SP4-Z MEDIUM

### REAGENTS:

#### N/A **Base Medium**

PPLO Broth.....	3.5 g
Tryptone.....	10 g
Peptone.....	5 g
DI Water.....	640 ml
DNA (fish sperm, spectrum DE120 or equivalent).....	0.2 g
Agar (if required)	15 g

#### N/A **Additive Supplement (sterile)**

CMRL-1066 medium (10x concentrated; ATCC 20-2207).....	50 ml
2% Yeastolate Solution. (2%, Autoclaved).....	100 ml
Fetal bovine serum (heat-inactivated).....	85 ml
Swine serum (heat-inactivated).....	85 ml
50% Glucose Solution (50% aqueous solution).....	20 ml
L-cysteine Solution (see below).....	10 ml
NAD Solution (see below).....	10 ml

#### N/A **2% Yeastolate Solution**

TC Yeastolate.....	2 g
DI Water.....	100 ml

#### N/A **50% Glucose Solution**

Glucose.....	10 g
DI Water.....	20 ml

#### N/A **L-cysteine Solution**

L-cysteine.....	0.01 g
DI Water.....	10 ml

#### N/A **NAD Solution**

NAD.....	0.1 g
DI Water.....	10 ml

### PROCEDURE:

#### Steps

#### N/A **To make ATCC Medium 2764:**

Accurately weigh out reagents  
Dissolve completely into DI water

Start:

End:

Adjust pH to 7.4

Autoclave at 121°C.

Aseptically, add sterile additive supplements to the Base Media

#### N/A For agar, placed at 55°C before adding additive supplements

Re-adjust pH to 7.4 after addition of supplements