

## APPENDIX A

### CHEMICAL REAGENTS

#### **Minimum essential medium (MEM)**

9.5 g of Minimum essential powder medium is dissolve in 500 ml sterile water. Add 2.2 g of sodium bicarbonate and dilute to 1000 ml with sterile water. Adjust pH to 7.2-7.4 with 10% sodium hydroxide or 10% hydrochloric acid and filter through 0.2 micron membrane filter and keep in sterile bottle.

The complete media is mixture of 400 ml of minimum essential medium (MEM), 40 ml fetal bovine serum and 4 ml penicillin/ streptomycin.

#### **Dulbecco's modified eagle medium (DMEM)**

13.4 g of Dulbecco's modified eagle powder medium is dissolve in 500 ml sterile water. Add 3.7 g of sodium bicarbonate and dilute to 1000 ml with sterile water. Adjust pH to 7.2-7.4 with 10% sodium hydroxide or 10% hydrochloric acid and filter through 0.2 micron membrane filter and keep in sterile bottle.

The complete media is mixture of 400 ml of Dulbecco's modified eagle medium (DMEM), 40 ml fetal bovine serum and 4 ml penicillin/ streptomycin.

#### **RPMI medium 1640**

10.4 g of RPMI medium 1640 powder is dissolve in 500 ml sterile water. Add 2.0 g of sodium bicarbonate and dilute to 1000 ml with sterile water. Adjust pH to 7.2-7.4 with 10% sodium hydroxide or 10% hydrochloric acid and filter through 0.2 micron membrane filter and keep in sterile bottle.

The complete media is mixture of 400 ml of RPMI medium 1640 and 40 ml fetal bovine serum and 4 ml penicillin/ streptomycin.

#### **10% Hydrochloric acid (HCl)**

27 ml of concentrated hydrochloric acid (37%) dissolve and make up to 100 ml with DI water.

**10% Sodium hydroxide (NaOH)**

10 g of sodium hydroxide dissolve and make up to 100 ml with DI water.

**Phosphate buffer saline (PBS)**

1 tablet of PBS dissolve and makes up to 100 ml with DI water, then sterile by autoclave.

**0.4% Sulforhodamine B (SRB)**

0.4 g sulforhodamine B dissolve and make up to 100 ml with 1% acetic acid (protect light)

**40% Trichloroacetic acid (TCA)**

40 g trichloroacetic acid dissolves and makes up to 100 ml DI water

**1% Acetic acid**

100 ml of glacial acetic acid dissolve and make up to 1000 ml with DI water.

**10 mM Tris buffer (pH 10)**

0.121 g Tris Hydroxymethylaminoethane dissolves in 50 ml DI water and adjusts pH to 10 with 1 N hydrochloric acid.