**ALKALINE METHYLENE VIOLET SOLUTION**

- Methylene Violet 3 RAX
- Citric Acid
- 0.1M Glycine buffer (pH 10.6)
- 2 small, clean (100-125ml) glass bottles
- Sterile pipets
- Foil
- Distilled water
- Mechanical pipeter
- Funnel

**Procedure:**

1. Weigh 0.1g methylene violet 3 RAX and place in small glass bottle.

2. Add distilled water up to 100ml mark on bottle.

3. Swirl bottle to dissolve media. (This is your 0.1% methylene violet stock solution.)

4. Place 10ml of the 0.1% methylene violet solution into a new glass bottle.

5. Dilute stock methylene violet solution up to 100ml with the 0.1M glycine buffer. This solution is ready to use for vitality tests.

6. Place 500ul of the “ready to use” alkaline methylene violet into 1.5ml eppendorf tubes for cell & vitality counting.

**Note:** Discard solution after 6 months.