Viral Transport Media Protocol as Per Centers for Disease Control and Prevention (CDC)

A guided tutorial

July 2020

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Materials Required

- **Ingredients list for Viral or Universal Transport Medium:**
    - Hanks Balanced Salt Solution (HBSS) 1 X with calcium and magnesium ions, no phenol red, 500 mL bottle, sterile and filtered
    - Sterile, heat-inactivated fetal bovine serum (FBS)
    - Gentamicin sulfate (50 mg/mL), sterile
    - Amphotericin B (250 µg/mL), sterile

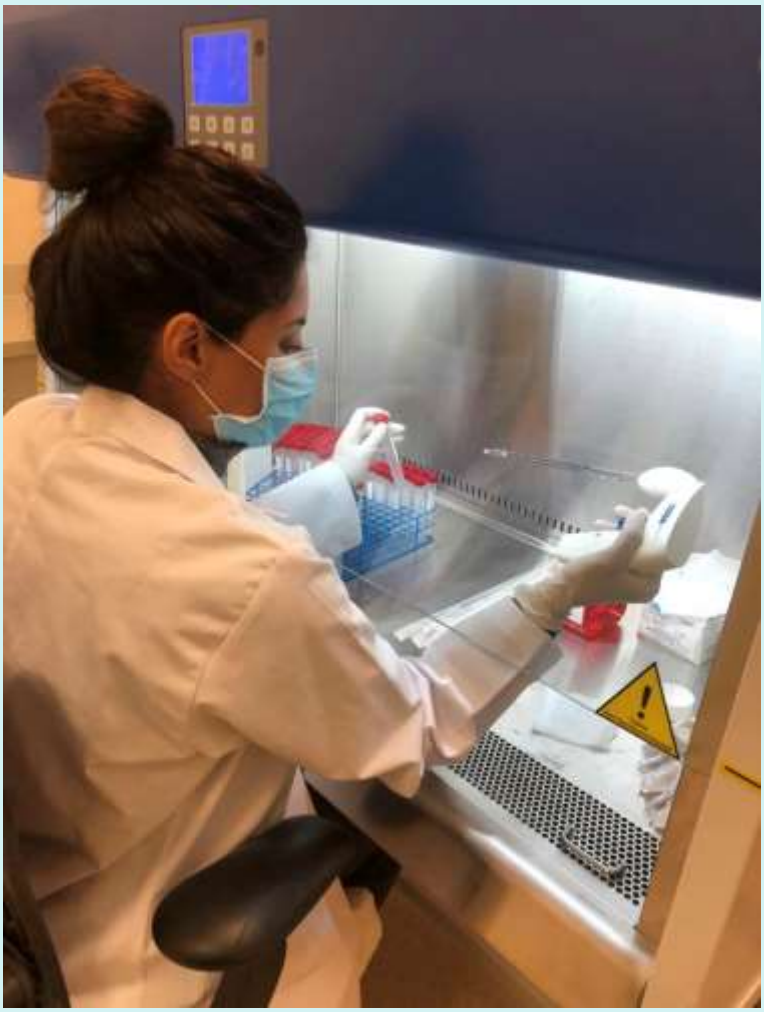
- **Laboratory Materials:**
    - All materials that should be utilized should be disinfected using 70% ethanol
    - Sterile 15 ml conical tubes should be used to transport the viral transport media
    - Individually wrapped sterile pipettes should be used to aliquot the CDC recommended amount of reagent in the conical tubes

Laboratory Personnel and Operating Conditions:

- All personnel should have prior laboratory experience and will be trained in aseptic techniques
- All standard clinical laboratory practices should be followed
- All personnel should be provided with appropriate laboratory attire
- All the reagents required to make the viral transport medium should be pipetted under a laminar flow hood, to ensure sterility.

Figure 1 - Our Set Up

A laminar flow hood, to ensure sterility, in Jaber Al Ahmad Al Sabah Hospital, a facility that is approved as a laboratory facility by the Ministry of Health of Kuwait.



Viral Transport Medium Production

The Viral Transport Medium (VTM) was prepared according to the formulations published by the Centers of Disease Control and Prevention (CDC) (4). Hank's Balanced Salt Solution (HBSS) (Gibco™ Waltham, USA) devoid of the pH-indicator phenol red was implemented as the medium's isotonic solution. The medium was supplemented with sterile heat-inactivated fetal bovine serum (FBS) (Gibco™ Waltham, USA), gentamicin (Gibco™ Waltham, USA), and amphotericin B (Lonza BioWhittaker™ Walkersville, USA), at final concentrations of 2%, 100µg/ml, and 0.5µg/ml, respectively. Using sterile pipettes, 3 mL of the prepared UTM was dispensed into 15 mL sterile conical centrifuge tubes (Labcon™ Petaluma, USA) and stored at 2-8 °C for one year from the date of production. All VTM production was performed at the Jaber Al-Ahmad Al-Sabah Hospital laboratory, Kuwait.

To maintain the sterility of the reagents and prevent contamination of the VTM, the complete procedure was conducted in class II biosafety cabinets. The CDC protocol for sterility testing was performed to determine the sterility of the concocted VTM (2). Depending on the number of tubes prepared per batch, three tubes were randomly selected from the beginning, middle, and end of each batch made to confirm proper sterility assessment of the entire batch. Using a sterile micropipette, 100µL of the prepared VTM was inoculated onto BAP. The plates were invertedly incubated at 35°C ± 2°C for 48 hours.

Figure 2 – Hard at Work

Our dedicated lab team preparing a batch of VTM

For any questions or concerns, please do not hesitate to contact us at info@projectjaber.com