SOP-P020

Preparation of Phosphate Buffered Saline (PBS)

Objective: To prepare phosphate buffered saline solution for use as sheath fluid in the Coulter Elite flow cytometer or as buffer for washing, suspending, or diluting cells

Procedure:

1. Weigh out 1864 g BBL medium (FTA hemagglutination buffer; Scientific Products #11248-BT).
2. Place the buffer in a clean autoclaved 20 L Nalgene carboy with spigot.
3. Add 20 L Millipore filtered water.
4. Replace cap on carboy and agitate contents to dissolve. Solution should be clear with no visible solids.
5. Open spigot and fill a Coulter vial with the solution.

6a. Determine the pH of the solution using an Orion Research EA920 Expandable Ion Analyzer (refer to SOP#-P024 for Determination of pH Using an Orion Research EA920 Expandable Ion Analyzer). The analyzer is located in the Hansen building, room B050. The pH should be 7.2 + 0.1

6b. If necessary, adjust the pH of the solution in the carboy to 7.2. Using a 5.25" Pasteur pipette fitted with a rubber pipette bulb, add drop wise 1 N hydrochloric acid (HCl) if the pH is higher than 7.2, or 1 N sodium hydroxide (NaOH) if the pH is lower than 7.2, until the correct pH is obtained.

7. Label the carboy with content information, date, and your initials.
8. Store refrigerated at 0-5°C. Expiration date is approximately one month from date of preparation.

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Verified by:___________________________Date:__________ ______________________________

Print Name Sign Name