## **SOP-P028**

## **Preparation of Stock 1 M Tris**

Objective: To prepare a stock solution of Tris for use in preparation of HBSS buffers

## Procedure:

- 1. Measure out 400 ml Millipore filtered water in a graduated cylinder.
- 2. Pour the water into a clean beaker containing a stir bar.
- While stirring gently using a magnetic stirplate, add the following to the beaker:
  8.0 g Tris base (tris[hydroxymethyl]aminomethane; Sigma #T-1503)
  68.5 g Tris HCl (tris[hydroxymethyl]aminomethane hydrochloride; Sigma #T-3253)
- 4. Stir until dissolved.
- 5a. Continue to stir and 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.
- 5b. Adjust the pH of the solution to 7.3. 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 7.3, or 1 N sodium hydroxide (NaOH) if the pH is lower than 7.3, until the correct pH is obtained.
- 6. Remove the stir bar and pour the solution into a graduated cylinder.
- 7. Add Millipore filtered water to the solution until the volume reaches 500 ml.
- 8. Pour the 1 M Tris into a container such as a glass bottle with cap.
- 8. Label the bottle with content information, date, and your initials.
- 9. Store refrigerated at 0-5°C. Expiration date is six months from date of preparation.

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Date: