SOP-P019

Preparation of 2% Paraformaldehyde (PF)

Objective: To prepare 2% paraformaldehyde solution for use in fixing stained cells

Procedure:

- 1. Measure out 175 ml PBS (refer to SOP#-P020 for Preparation of Phosphate Buffered Saline) in a graduated cylinder.
- 2. Pour the PBS into a clean Erlenmeyer flask containing a stir bar.
- 3. While stirring gently on a magnetic stirplate, add 4.0 g paraformaldehyde.
- 4. Seal the top of the flask with parafilm and cover the flask with aluminum foil to protect the contents from light.
- 5. Leave the flask on the stirplate to stir overnight. Solution should be clear with no visible solids.
- 6. Continue to stir and determine the pH of the solution using a pH meter (refer to SOP#-P048 for Determination of pH Using a VWR model 9100 pH meter, located in Bindley 122).
- Adjust the pH of the solution to 7.4. Using a 5.25" Pasteur pipet fitted with a rubber pipet bulb, add dropwise 1 N hydrochloric acid (HCl) if the pH is higher than 7.4, or 1 N sodium hydroxide (NaOH) if the pH is lower than 7.4, until the correct pH is obtained.
- 8. Remove the stir bar and pour the paraformaldehyde solution into a graduated cylinder.
- 9. Add PBS to the solution until the volume reaches 200 ml.
- 10. Place the solution in a container such as a glass bottle with screw cap.
- 11. Cover the bottle with aluminum foil to protect the contents from light.
- 12. Label the bottle with content information, date, and your initials.
- 13. Store refrigerated at 0-5°C. Expiration date is one month from date of preparation.

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