SOP: R008

Preparation of SDS-PAGE 10X Tris-Glycine Running Buffer

Materials and Reagents: per liter

1. Milli-Q water, 1000 ml
2. Tris Base, electrophoretic grade, 30.2 g
3. Glycine, electrophoretic grade, 144.2 g
4. SDS (lauryl sulfate), electrophoretic grade, 10 g
5. 1 liter graduated cylinder
6. 2 liter pyrex beaker
7. 1 liter polypropylene or glass bottle
8. Magnetic stir bar
9. Magnetic stir plate

Protocol:

1. _____ Measure 800 ml of Milli-Q water using the graduated cylinder and pour into pyrex beaker.

2. _____ Add stirbar, place on stirplate, and begin stirring.

3. _____ Add 30.2 g Tris-Base, 144.2 g glycine, and 10 g SDS (note 1).

4. _____ Stir 20 minutes to ensure that all reagents are completely dissolved.

5. _____ Check pH. It should be ~8.3 (note 2).

6. _____ Pour contents into a clean polypropylene or glass 1 liter bottle. Store at room temperature until needed.

7. _____ Dilute from 10X to 1X before use.

Notes:

1. Always wear a mask when weighing SDS to avoid inhalation.
2. The pH should almost exactly 8.3. If the pH is not at 8.3, discard the buffer. **Do not** titrate the buffer with acid or base, as this will throw off the electrolytic balance and the buffer will be unusable.

References:

Laemmli, U. K. 1970. Cleavage of structural proteins during the assembly of the head of bacteriophage T4. Nature 227:680-5.