

**SOP: R003**

**Preparation of Second Fix for SDS-PAGE**

**Materials and Reagents:** per liter

1. 1 liter glass beaker
2. Stir bar
3. Magnetic stir plate
4. 500 ml graduated cylinder
5. Milli-Q water, 880 ml
6. Methanol, 50 ml
7. Glacial acetic acid, 70 ml
8. Glass or polypropylene storage container

**Protocol:**

1. \_\_\_\_\_ Combine Milli-Q water, methanol, and acetic acid in beaker and stir on stir plate.
2. \_\_\_\_\_ Decant into appropriate storage container (note 1).

**Notes:**

1. Second fix is stable for at least a month at room temperature.

**References:**

Laemmli, U. K. 1970. Cleavage of structural proteins during the assembly of the head of bacteriophage T4. *Nature (London)* 227:680-685.

Morrissey, J.H. 1981. Silver stain for proteins in polyacrylamide gels; a modified procedure with enhanced uniform sensitivity. *Anal. Biochem.* 117:307-310.