

HOMEMADE SDS-PAGE PROTEIN GELS

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1. Assemble the glass –plate sandwich of the electrophoresis apparatus using two clean glass plates and two 0.75mm spacers. (cleaning the glass plates with 5% alconox, distilled water and 100% ethanol before assembling)
2. Lock the sandwich to the casting stand.
3. Prepare the separating gel solution as directed in following table.(you'd better add the 10%APS and TEMED at the last two steps, after mixing the solution ,use it as soon as you can)
4. Apply the separating gel solution to the sandwich along an edge of one of the spacers until the height of the solution between the glass plates is 2~3cm to the top.
5. Slowly cover the top of the gel with a layer of butanol (1~2cm thick).
6. Allow the gel to polymerize 30 to 60 min at room temperature.
7. Pour the layer of butanol and wash the top of the gel with distilled water gently.
8. Prepare the stacking gel solution as ditected in following table.
9. Apply the stacking gel to the sandwich along an edge of one of the spacers until the height of the solution is 1cm from the top of the plates.
10. Insert a 0.75mm comb into the layer of stacking gel solution. If necessary , add additional stacking gel to fill the space in the comb completely.
11. Allow the stacking gel solution polymerize 30 to 45 min at room temperature.