

Prepare 10% minigel:

1. Clean all minigel apparatus with soap and water, then with ethanol.
Assemble the glass plates and spacers, and place in a tupperware.
2. Prepare resolving gel:
 - 2.0 mL 30% acrylamide
 - 1.5 mL 4X resolving gel buffer
 - 2.5 mL ddH₂OAllow to degas for 10 min.
3. Meanwhile, use a pasteur pipette to seal the bottom of the plates by spreading 1% agarose in SDS-PAGE electrophoresis buffer along the bottom of the glass plates so that it goes up the crack by capillary action.
4. Add 60 μ L 10% APS and 2.4 μ L TEMED to degased gel solution, mix quickly, and add to apparatus with a pasteur pipette.
5. Quickly add isobutanol to the top of this until the level reaches the top of the plates.
6. Allow resolving gel to polymerize for 20-30 min.
7. Prepare stacking gel:
 - 0.65 mL 30 % acrylamide
 - 1.25 mL 4X soln* Tris pH 6.8
 - 3.1 mL ddH₂ODegas for 10 min.
8. Add Add 50 μ L 10% APS and 4 μ L TEMED. Mix quickly and add on top of resolving gel. Insert the comb straight down, then pour a little more stacking gel on the sides of the comb to fully seal it.
9. Allow the stacking gel to polymerize for 20-30 min.