2.3. Quantification of Radiolabeled Amino Acid Incorporation

1. Whatman GF/A glass fiber filters (Maidstone, UK).
2. 1*M* NaOH, 2% H_2O_2.
3. Ice-cold 25% trichloroacetic acid (TCA), 2% casamino acids, vitamin assay grade (Difco, Detroit, MI).
4. 5% ice-cold TCA.
5. Acetone.
6. Scintillation fluid (e.g., Opti-flur O, Packard).

2.4. SDS Polyacrylamide Gel Electrophoresis (SDS-PAGE)

1. Gel apparatus (e.g., Bio-Rad vertical minigel apparatus).
2. 10X Glycine running buffer (Tris-glycine, pH 8.3): For 1 L mix 30 g Tris-base, 144 g glycine, 100 mL 10% SDS, then add distilled water to volume.
3. Acrylamide/*bis*-acrylamide solution (30:8%). For 100 mL, dissolve 30 g acrylamide and 0.8 g *bis*-acrylamide in distilled water. Store at 4°C.
4. 1.5*M* Tris-HCl, pH 8.8. For 1 L, dissolve 181.65 g of Tris-base in 800 mL of distilled water, and titrate to the correct pH with concentrated HCl. Make to 1 L with distilled water.
5. 0.5*M* Tris-HCl, pH 6.8. For 1 L, dissolve 60.55 g of Tris-base in 880 mL of distilled water, titrate to the correct pH with concentrated HCL. Make to 1 L with distilled water.
6. 10% SDS.
7. Tetramethylethylene diamine (TEMED).
8. 1% Ammonium persulfate (make fresh before use).
9. Propanol:water (1:1).
10. Sample loading buffer: 2% (w/v) SDS, 5% β-mercaptoethanol, 10% glycerol, 0.01% bromophenol blue, in 0.125*M* Tris-HCl, pH 6.8.
11. Coomassie blue stain solution. Distilled water, methanol, and glacial acetic acid at a ratio of 5:5:1, with 0.1% Coomassie blue.
12. Destain solution. As in **item 11**, without the Coomassie blue.

2.5. Autoradiography

1. Gel-drying apparatus.
2. X-ray film (e.g., Kodak's X-omat AR or Amersham's Hyperfilm).

2.6. Immunopreciptation

1. 8% SDS.
2. TNTE buffer: 1% Triton-X 100, 150 m*M* NaCl, 2 m*M* EDTA, 50 m*M* Tris-HCl, pH 7.8.
3. High-salt TNTE: 1% Triton-X 100, 0.95*M* NaCl, 2 m*M* EDTA, 50 m*M* Tris-HCl, pH 7.8.
4. Protein A Sepharose (Sigma).
5. Sample loading buffer (*see* **Subheading 2.4., item 10**).