SDS Polyacrylamide Gel Electrophoresis

Gel Recipes

% Acrylamide	5%	7.5%	10.%	12.5%	15%	18%	4% Stacking Gel
30% Acrylamide (ml)	5.0	7.5	10.0	12.5	15.0	18.0	1.3
1% Bisacrylamide (ml)	7.8	5.8	3.9	3.1	3.1	3.1	1.5
1.5 M Tris, pH 8.7 (ml)	8.1	8.1	8.1	8.1	8.1	8.1	-
0.5M Tris, pH 6.7 (ml)	-	-	-	-	-	-	1.25
20% SDS (ml)	0.2	0.2	0.2	0.2	0.2	0.2	65 μl
H_2O (ml)	8.9	8.5	7.9	6.2	4.2	1.2	5.725
TEMED (μl)	10	10	10	10	10	10	10
10% Ammonium persulfate (μl)	225	225	225	225	225	225	150

Makes ~30.8 ml gel solution for running gel; ~10 ml for stacking gel

Electrophoresis Buffer:

<u>5X Buffer:</u>
60 g Tris base
288 g Glycine
50 ml 20% SDS
dH_2O to 2 liters

<u>1 X Buffer</u>
9 g Tris base
43.2 g Glycine
7.5 ml 20% SDS
dH_2O to 1.5 liters

Laemmli Sample Preparation Buffer:

DTT:	123.4 mg
Glycerol or 50% sucrose	4 ml
0.2 M Tris, pH 8.0/20 mM EDTA	1 ml
20 mg% pyronine Y	1 ml
20% SDS	1 ml
dH_2O	1 ml

This comes out to:
2.5% SDS, 100 mM DTT, 25 mM Tris, 2.5 mM EDTA.
Can be diluted with twice the volume of sample (ie. 3-fold dilution)

Fixing and De-Staining Solution:
10% isopropanol
5% acetic acid

Coomassie Brilliant Blue Stain:
1 g Coomassie Brilliant Blue dye
200 ml glacial acetic acid
500 ml isopropanol
1.3 l dH_2O