



#### 14 SDS-PAGE

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#### **GUIDELINES**

SDS-PAGE Separation of gel samples	Scope of application
6⊠ sample gel	50-150kD
8½ sample gel	30-90kD
10½ sample gel	20-80kD
12\mathbb{\mtx\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\\	12-60kD
15½ sample gel	10-40kD

#### MATERIALS

NAME ~	CATALOG # \( \times \)	VENDOR V
TEMED	1610801	Bio-rad Laboratories
40g Acrylamide/ Bisacrylamide (29:1); Premixed Powder	786-506	G-Biosciences
Coomassie brilliant blue G-250	CB0038.SIZE.100g	Bio Basic Inc.
SDS, 10%(w/v) solution	SD8118.SIZE.100ml	Bio Basic Inc.
Tris pH 6.8		
10\( Ammonium persulfate	/	
1M Tris(PH=8.8)	/	

## SAFETY WARNINGS

Be careful of  $30\mbox{MAcr-Bis}$  M and TEMED owing to their potential harm.

# BEFORE STARTING

In general, 15 $\mbox{\ensuremath{\mbox{$\mathbb{N}$}}}$  sample gel is used, so here is only the composition of 15 $\mbox{\ensuremath{\mbox{$\mathbb{N}$}}}$  sample gel.

## Composition of the 151 separated of the gel sample(4mL):

Composition	volume  MmL  M
Distilled water	0.4
30MAcr-BisM29M1M	2
1M Tris pH=8.8	1.5
10MSDS	0.04
10\( Ammonium persulfate	0.04
TEMED	0.002

# Composition of concentrated sample gel(1mL):

Composition	volume\mL\mathbb{M}
Distilled water	0.7

30MAcr-BisM29M1M	0.165
1M Tris pH=6.8	0.125
10MSDS	0.01
10\( Ammonium persulfate	0.01
TEMED	0.001

1	Mount the	gels in	the vertica	al electrop	horesis a	apparatus.
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2	Load samples	under the	cathode buffer.	Apply 10µL	. sample volumes	to $0.7 \times 5$ mm	sample wells.

**□10** μl

- 3 Set running conditions appropriate to your type of gel. We usually set 200V to run at the beginning, after 10 minutes change the voltage into 300V for another 30 minutes.
- 4 Protein can be visualized directly in the gel by Coomassie staining or silver staining. We usually choose Coomassie staining, then transfer the gel to the decolorizing solution and remove the dye solution.
- 5 Recording with gel imager.

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