SDS-PAGE Gel kit

Cat. No: E-IR-R305 **Size: 10 Assays/ 25 Assays / 50 Assays**

Cat.	Products	10 Assays	25 Assays	50 Assays	Storage
E-IR-R305A	30% Acr-Bis (29:1)	45 mL	100 mL	100 mLx2	2~8°C
E-IR-R305B	Separating Gel Mix	22 mL	55 mL	55 mL x2	RT
E-IR-R305C	Stacking Gel Mix	6 mL	12 mL	22 mL	RT
E-IR-R305D	APS	170 mg	170 mg x 2	700 mg	RT
E-IR-R305E	TEMED	100 µL	250 µL	500 µL	2~8°C
Manual			**One Copy**		

Introduction

Elabscience® SDS-PAGE Gel Assay kit is the classic SDS-PAGE gel preparation kit which contains all kinds of reagents needed to prepare the SDS-PAGE gel. Users only need apparatus and ddH_2O to complete the gel preparation.

The Gel Mix in this kit is the mixture of the gel buffer such as SDS and Tris-HCl and so on, which simplify the procedure of gel preparation. There are Tris-HCl pH 8.8 and SDS in the Separating Gel Mix, Tris-HCl pH 6.8 and SDS are in the Stacking Gel Mix.

Instructions

According to the different molecular weight of the target protein, different concentration of separating gel should be prepared.

1. Comparison of protein separation linear range is shown in Schedule 1.
2. Sample volume of each component corresponding to different stacking gel volumes is shown in Schedule 2.
3. Sample volume of each component corresponding to different separation gel volumes is shown in Schedule3.

Procedure

1. Preparation of 10% APS: add distilled water or DI water (add 10µL of water to 1mg APS) to APS [E-IR-R305D] to prepare a 10% APS, For example: add 1.7mL distilled water to170mg APS, the mixture is 10% APS. 10% APS should be split into small tubes and stored at -20°C.
2. Choose the clean gel mold and complete the assembly.
3. Refer to Schedule 3, prepare the separating gel of the required concentration and volume. Add appropriate amount of ddH2O, 30% Acr-Bis (29:1)[E-IR-R305A] and Separating Gel Mix [E-IR-R305B] to a clean beaker, mix it.
4. Add appropriate amount of 10% APS and TEMED, mix gently and avoid bubbles.
5. Add the mixture gel to the assembled gel mold immediately. Then add 1~2 mL ddH2O or absolute alcohol on the separating gel to flatten the separating gel level.
6. Keep at RT for 30~60 min until the separating gel solidified completely.
7. Pour out the ddH2O or absolute ethanol, absorb the residual liquid with absorbent paper.
8. Refer to Schedule 2, prepare the stacking gel of the required volume. Add appropriate amount of ddH2O,30% Acr-Bis (29:1)[E-IR-R305A] and stacking Gel Mix [E-IR-R305C] to a clean beaker, mix it.
9. Add appropriate amount of 10% APS and TEMED, mix gently and avoid bubbles.
10. Add the mixture gel upon to the separating gel immediately. Insert comb teeth carefully, avoid bubbles.

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11. Keep at RT for 30~40 min until the stacking gel solidified completely.
12. Pull out the comb teeth carefully. Follow the SDS-PAGE experiment.

Storage

Store separately for 6 months.
30% Acr-Bis (29:1) [E-IR-R305A] and TEMED [E-IR-R305E] should be stored at 2~8°C in dark.
10% APS should be split into small tubes and stored at -20°C for 6 months, If store at 2~8°C, please do not use it after one week.
Other reagents should be stored at RT.

Cautions

1. For maximal assay performance, this reagent should be used within 6 months. Avoid freeze / thaw cycles.
2. TEMED is volatile. Please tighten the cap after use. Due to the different temperatures, the rate of gel solidification will be different. The amount of TEMED can be adjusted appropriately to adjust the rate of gelation at different temperatures.
3. Avoid bubbles to avoid affecting the experimental results.
4. Add the ddH2O or absolute alcohol on the separating gel gently to avoid uneven line pressing.
5. This kit is for research use only. For your safety and health, please wear lab clothes and gloves. Instructions should be followed strictly, changes of operation may result in unreliable results.

Schedule

Schedule 1 Comparison of protein separation linear range

Concentration of separating gel	Linear separation range
6%	50~150kDa
8%	30~90kDa
10%	20~80kDa
12%	12~60kDa
15%	10~40kDa

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Schedule 2 Preparation of stacking gel

Components	Sample volume of each component corresponding to different stacking gel volumes							
	1mL	2mL	3mL	4mL	5mL	6mL	8mL	10mL
5% Stacking Gel								
H_2O	0.68	1.4	2.1	2.7	3.4	4.1	5.5	6.8
30% Acr-Bis	0.17	0.33	0.5	0.67	0.83	1.0	1.3	1.7
Stacking Gel Mix	0.14	0.27	0.41	0.54	0.68	0.81	1.08	1.35
10% APS	0.01	0.02	0.03	0.04	0.05	0.06	0.08	0.1
TEMED	0.001	0.002	0.003	0.004	0.005	0.006	0.008	0.01

Schedule 3 Preparation of separating gel

Components	Sample volume of each component corresponding to different separation gel volumes							
	5mL	10mL	15mL	20mL	25mL	30mL	40mL	50mL
6% Gel								
ddH_2O	2.6	5.3	7.9	10.6	13.2	15.9	21.2	26.5
30% Acr-Bis	1.0	2.0	3.0	4.0	5.0	6.0	8.0	10.0
Separating Gel Mix	1.35	2.6	3.95	5.2	6.55	7.8	10.4	13.0
10% APS	0.05	0.1	0.15	0.2	0.25	0.3	0.4	0.5
TEMED	0.004	0.008	0.012	0.016	0.02	0.024	0.032	0.04
8% Gel								
ddH_2O	2.3	4.6	6.9	9.3	11.5	13.9	18.5	23.2
30% Acr-Bis	1.3	2.5	4.0	5.3	6.7	8.0	10.7	13.3
Separating Gel Mix	1.35	2.6	3.95	5.2	6.55	7.8	10.4	13.0
10% APS	0.05	0.1	0.15	0.2	0.25	0.3	0.4	0.5
TEMED	0.003	0.006	0.009	0.012	0.015	0.018	0.024	0.03
10% Gel								
ddH_2O	1.9	4.0	5.9	7.9	9.9	11.9	15.9	19.8
30% Acr-Bis	1.7	3.3	5.0	6.7	8.3	10.0	13.3	16.7
Separating Gel Mix	1.35	2.6	3.95	5.2	6.55	7.8	10.4	13.0
10% APS	0.05	0.1	0.15	0.2	0.25	0.3	0.4	0.5
TEMED	0.002	0.004	0.006	0.008	0.01	0.012	0.016	0.02
12% Gel								
ddH_2O	1.6	3.3	4.9	6.6	8.2	9.9	15.9	16.5
30% Acr-Bis	2.0	4.0	6.0	8.0	10.0	12.0	13.3	20.0
Separating Gel Mix	1.35	2.6	3.95	5.2	6.55	7.8	10.4	13.0
10% APS	0.05	0.1	0.15	0.2	0.25	0.3	0.4	0.5
TEMED	0.002	0.004	0.006	0.008	0.01	0.012	0.016	0.02
15% Gel								
ddH_2O	1.1	2.3	3.4	4.6	5.7	6.9	9.2	11.5
30% Acr-Bis	2.5	5.0	7.5	10.0	12.5	15.0	20.0	25.0
Separating Gel Mix	1.35	2.6	3.95	5.2	6.55	7.8	10.4	13.0
10% APS	0.05	0.1	0.15	0.2	0.25	0.3	0.4	0.5
TEMED	0.002	0.004	0.006	0.008	0.01	0.012	0.016	0.02

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