V02

免染 PAGE 凝胶快速制备试剂盒（6%）

货号：G4840
规格：40T(0.75mm)/100T(0.75mm)
保存：2-8℃，避光，有效期 1 年。

产品组成：

名称		40T	100T	保存
试剂(A)：上层胶预制液	A1：SG 储备液	30mL	80mL	2-8℃，避光
	A2：SG 稀释液	30mL	80mL	2-8℃
使用前，将 A1 与 A2 等量混匀，即上层胶预制液，配好后可在 2-8℃保存 1 周。				
试剂(B)：下层胶预制液	B1：RG 储备液	80mL	2×100mL	2-8℃，避光
	B2：RG 稀释液	80mL	2×100mL	2-8℃
使用前，将 B1 与 B2 等量混匀，即下层胶预制液，配好后可在 2-8℃保存 1 周。				
试剂(C)：PAGE 胶凝固剂		0.22g	0.6g	2-8℃
使用前，配制成 10%凝固剂（即 0.22g 溶于 2.2mL 蒸馏水，0.6g 溶于 6mL 蒸馏水），配好后可在 2-8℃保存 3 个月。（见注意事项 4）				

产品介绍：

免染 PAGE 凝胶快速制备试剂盒提供了配制蛋白变性电泳凝胶所需的各种试剂，用户只需自备制胶器及蒸馏水，即可配制 SDS-PAGE 彩色凝胶进行蛋白质电泳。本试剂盒根据不同的制胶板厚度约可配制 20-40 块电泳凝胶。

本试剂盒所制备凝胶适用于 Tris-Glycine 体系电泳，不需要额外加入 TEMED 即可制备 SDS-PAGE 凝胶，其中上层胶(浓缩胶)添加有色颜料，点样孔清晰易辨，便于上样，改良后的下层胶（分离胶）可与上层胶显著分层，可在灌注下层胶后无需水或醇类试剂封闭直接灌注上层胶同时凝结，减少配胶时间。电泳或转膜结束后可将凝胶整块取下浸于缓冲液中置于凝胶成像仪中直接成像观察电泳状况和转膜效率，同时不影响后续化学发光或组学检测，有助于提高检测效率。

操作步骤：*（仅供参考）*

本产品上层胶浓度 5%，下层胶 6%，如需其他浓度请购置 G4841-G4845。SG 稀释液颜色随机发放，不同颜色 SG 稀释液可混用，如需其他颜色可购置 G4830-G4833。

1. 将制胶模具装配好，以一块 1.50 mm 厚的 mini 胶为例。（0.75mm 和 1.00mm 数据见表 1）

制胶板尺寸	0.75mm	1.00mm	1.5mm
上层胶预制液	1.4mL	2mL	2.8mL
10%凝固剂	14uL	20uL	28uL
下层胶预制液	4mL	5.4mL	8mL
10%凝固剂	40uL	54uL	80uL

表 1：不同厚度制胶板推荐配制凝胶量

2. 取等体积的 RG 储备液和 RG 稀释液各 4mL，放入配胶杯中混匀，制成下层胶预制液。
3. 取等体积的 SG 储备液和 SG 稀释液各 1.4mL，放入配胶杯中混匀，制成上层胶预制液。
4. 向下层胶预制液内加入 10%凝固剂 80uL，轻轻搅拌或摇晃混匀，避免产生气泡。（*见注意事项 2*）
5. 向制胶模具中加入步骤 4 中混匀后的下层胶溶液，使液面距离玻璃板上沿约 1.5 cm 处即可。
6. 向上层胶预制液中加入 28uL 的 10%凝固剂，轻轻搅拌或摇晃使其混匀，避免出现气泡，不需要等待下层胶凝固，可直接将混匀后的溶液缓慢灌注到下层胶溶液上面，插入梳子。（*注意：加入下层胶后要在 2 min 内将上层胶注入凝胶模具内，且在灌注上层胶时要缓慢，防止上层胶与下层胶混合在一起，若上层胶灌注不小心产生气泡，则可以用枪头轻轻扫下，赶走气泡*）
7. 室温（25℃）静置约 15 min，待上层胶完全凝固，小心地拔出梳子，即可进行常规电泳操作。
8. 可在电泳后、转膜后和曝光前对胶、膜进行免染成像，对电泳状况、转膜效率和全蛋白条带进行观察。

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9. 免染相关成像可使用 Bio-red 系列成像仪选择凝胶成像或膜成像的 Stain-Free 模块进行免染成像，iBright 系列成像仪可选择 Protein Gels 成像下的 No-stain 模块进行免染成像。其他仪器成像条件可咨询我司技术支持。

注意事项：

1. 向制胶器中加液时一定要缓缓加入，不可急躁，避免气泡的产生。
2. 凝固剂添加量与凝固时间直接相关，操作步骤中添加比例可保证 10-20min 全胶凝胶完毕。如需同时配制多块凝胶建议适当减少凝固剂添加比例（±20%）或使用压胶试剂分层配置凝胶。
3. 蛋白条带的分辨率和电泳美观度与电泳条件相关，如果想跑出清晰，美观的条带，建议电泳时电压在 100-120V 之间，如需快速电泳可适当加压至 160-200V，高电压下建议冰浴电泳，防止烧胶。
4. 配置好的 10%凝固剂如需长期保存建议分装冻存，在-20℃可保存 1 年。
5. 彩色上层胶中含有染料，因染料本身性质，长期静置后可能会产生沉淀，使用前请小心混匀。
6. 产品中的保存条件及有效期均以未开封情况下计算，为了防止产品与空气接触发生化学反应影响产品性能，将未使用完毕的组分按存储要求保存同时建议开封后的组分尽快使用完毕。
7. 本品仅适用于变性蛋白凝胶电泳，非变性蛋白凝胶电泳请选取我司其他产品。
8. 为了您的安全和健康，请穿实验服并戴一次性手套操作。

相关产品：

A1010　30%（29：1）制胶液
D1060　10×电泳转移缓冲液（转膜液）
G4829　PAGE 胶促凝剂（非 TEMED）
G4830　PAGE 浓缩胶红色染料（200×）
G4831　PAGE 浓缩胶黑色染料（200×）
G4832　PAGE 浓缩胶蓝色染料（200×）
G4833　PAGE 浓缩胶绿色染料（200×）
G4841　免染 PAGE 凝胶快速制备试剂盒（8%）
G4842　免染 PAGE 凝胶快速制备试剂盒（10%）
G4843　免染 PAGE 凝胶快速制备试剂盒（12%）
G4844　免染 PAGE 凝胶快速制备试剂盒（15%）
T1070　5×Tris-甘氨酸电泳缓冲液

V02

Stain Free SDS-PAGE Gel Casting Kit(6%)

Cat: G4840
Size: 40T(0.75mm)/100T(0.75mm)
Storage: 2-8℃,avoid light, valid for 1 year.

Kit Components

Reagent		40T	100T	Storage
Reagent(A): Stacking Gel Casting Solution	A1:SG Stock Solution	30mL	80mL	2-8℃, avoid light
	A2:SG Diluent Solution	30mL	80mL	2-8℃
Before use, mix A1 and A2 equally to prepare Reagent(A): Stacking Gel Casting Solution which can be stored at 2-8 ℃ for 1 week.				
Reagent(B): Resolving Gel Casting Solution	B1:RG Stock Solution	80mL	2×100mL	2-8℃, avoid light
	B2:RG Diluent Solution	80mL	2×100mL	2-8℃
Before use, mix B1 and B2 equally to prepare Reagent(B): Resolving Gel Casting Solution which can be stored at 2-8 ℃ for 1 week.				
Reagent(C): PAGE Gel Initiator		0.22g	0.6g	2-8℃
Before use, prepare 10% Initiator Solution(dissolve 0.22g in 2.2mL distilled water, dissolve 0.6g in 6mL distilled water), which can be stored at 2-8 ℃ for 3 months. (See Note 4)				

Introduction

The Stain Free SDS-PAGE Gel Casting Kit provides various reagents required for preparing protein denaturing and electrophoresis gel. Users can prepare SDS-PAGE Color Gel for protein electrophoresis simply by preparing their own gel preparator and distilled water. The kit can prepare about 20-40 electrophoretic gel according to the thickness of different gel making plates.

The gel prepared by this kit is applicable to the electrophoresis of Tris glycine system. The SDS-PAGE gel can be prepared without adding TEMED. The stacking gel is added with colored pigment, and the sampling hole is clear and easy to identify, which is convenient for loading. The improved lower glue (separation glue) can be significantly layered with the upper glue, and the upper glue can be directly poured without blocking with water or alcohol reagent after pouring the lower glue, and coagulate at the same time, reducing the mixing time. After electrophoresis or film transfer, the whole gel can be taken off and immersed in buffer solution and placed in a gel imager to directly image and observe the electrophoresis condition and film transfer efficiency, without affecting the subsequent chemiluminescence or omics detection, which is helpful to improve the detection efficiency.

Protocols(*for reference only*)

The stacking gel concentration of this product is 5%, and the resolving gel concentration is 6%. If you need other concentrations, please purchase G4841-G4845. The color of SG diluent is distributed randomLy. SG diluents of different colors can be mixed. If other colors are needed, G4830-G4833 can be purchased.

1. Assemble the glue making mold. Take a piece of 1.50 mm thick mini glue as an example for the following steps. (See Table 1 for 0.75 mm and 1.00 mm data).

Gel Plate Size	0.75mm	1.00mm	1.5mm
Stacking Gel Casting Solution	1.4mL	2mL	2.8mL
10% Initiator Solution	14uL	20uL	28uL
Resolving Gel Casting Solution	4mL	5.4mL	8mL
10% Initiator Solution	40uL	54uL	80uL

Table 1 Recommended amount of gel for rubber making plates with different thicknesses

2. Take 4mL of RG Stock Solution and 4mL of RG Diluent Solution , put them into the dispensing cup and mix them evenly to make the Resolving Gel Casting Solution.
3. Take 1.4mL of SG Stock Solution and 1.4mL of SG Diluent Solution, put them into the dispensing cup and mix them well to prepare the Stacking Gel Casting Solution.
4. Add 10% Initiator Solution 80uL to the Resolving Gel Casting Solution. Gently stir or shake the mixture to avoid bubbles. (See Note 2)
5. Add the Resolving Gel Casting Solution mixed in step 4 into the gel making mold, so that the liquid level is about 1.5 cm away from the upper edge of the glass plate.
6. Add 28uL of 10% Initiator Solution to Stacking Gel Casting Solution, gently stir or shake it to mix it, so as to

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avoid bubbles. It is not necessary to wait for the lower layer of glue to solidify, but slowly pour the mixed solution onto the lower layer of glue solution, and insert the comb. (*Note: after the Resolving Gel Solution is added, the Stacking Gel Solution should be slowly injected into the gel mold within 2 minutes to prevent the Stacking gel and the Resolving Gel from mixing together. If bubbles are generated by careless filling of the stacking gel, it can be gently swept with the gun head.*)

7. Let it stand at room temperature (25℃) for about 15 min, wait for the stacking gel to completely solidify, carefully pull out the comb, and then carry out the routine electrophoresis operation.
8. Non staining imaging can be performed on the gel and film after electrophoresis, film transfer, and before exposure to observe the electrophoresis status, film transfer efficiency, and total protein bands.
9. Dye free related imaging can be observed by the Stain-Free module of gel imaging or membrane imaging can be directly selected for Bio-red series instruments to image, and the No-stain module under Protein Gels imaging can be selected for iBright series instruments to image. For other instrument imaging conditions, please consult our technical support.

Note

1. When adding liquid to the glue maker, it must be added slowly to avoid the generation of bubbles.
2. The addition amount of Initiator Solution is directly related to the setting time. The addition proportion in the operation steps can ensure that the full gel gel is completed in 10-20min. If it is necessary to prepare multiple pieces of gel at the same time, it is recommended to appropriately reduce the proportion of Initiator Solution added (±20%)or to use a pressure agent to configure the gel layer by layer.
3. The resolution and electrophoresis aesthetics of protein bands are related to the electrophoresis conditions. If you want to produce clear and beautiful bands, it is recommended that the voltage be 100-120V during electrophoresis. If you need fast electrophoresis, you can properly pressurize it to 160-200V. At high voltage, it is recommended that you use ice bath electrophoresis to prevent gel burning.
4. If the prepared 10% Initiator Solution needs to be stored for a long time, it is recommended to store it in separate packages at - 20 ℃ for 1 year.
5. The colored upper layer glue contains dye. Due to the nature of the dye itself, sediment may occur after long-term standing. Please be careful to mix well before use.
6. The storage conditions and validity period of the product are calculated based on the unopened condition. In order to prevent the chemical reaction between the product and the air from affecting the product performance, the unused components are stored according to the storage requirements, and it is recommended that the unsealed components be used as soon as possible.
7. This product is only applicable to denatured protein gel electrophoresis. Please select other products of our company for non denatured protein gel electrophoresis.
8. For your safety and health, please wear experimental clothes and disposable gloves.