V02

SDS 残留检测试剂盒

货号： G4710
规格： 100T
保存： 室温，避光保存，有效期 1 年。

产品组成：

试剂名称	100T	保存
试剂(A): 0.1%SDS 溶液	10mL	室温，避光
试剂(B): 染色液	10mL	室温，避光
试剂(C): 萃取液	2×75mL	室温，避光

产品介绍：

SDS 分子具有亲水和疏水两性特征，在基因工程蛋白质药物的提取、纯化及抗原的制备的过程中有重要的应用价值，可是 SDS 的残留会对动物和人体产生毒副作用，因此随时监测 SDS 的残留有很重要的意义。

SDS 残留检测试剂盒由 0.1%SDS 溶液、染色液和萃取液三部分组成，0.1%SDS 溶液可用水稀释配制梯度 SDS 标准溶液绘制标曲，染色液可以选择性结合样品中的 SDS 形成复合物，这种复合物可被萃取液萃取到有机相中，再通过紫外分光光度法测定 OD 值；而未结合成复合物的染料在萃取后留在水相中，这样样品中 SDS 的含量就与被萃取到有机相中复合物在特定最大吸收波长的吸收值存在一定的线性关系，经测定在 0.001%~0.01% SDS 浓度范围内，线性拟合度较好，可达到 0.99 以上。

操作步骤：*(仅供参考)*

一、 制作标准曲线

1. 取 8 支 0.5mL 离心管，按照下表配制梯度 SDS 标准溶液(以 100μL/管为例）

终浓度	0.01%	0.008%	0.006%	0.004%	0.002%	0.001%	0
试剂(A)	30μL	管 1-160μL	管 2-150μL	管 3-134μL	管 4-100μL	管 5-100μL	0
蒸馏水	270μL	40μL	50μL	66μL	100μL	100μL	100μL
编号	管 1	管 2	管 3	管 4	管 5	管 6	管 7
终体积	140μL	100μL	100μL	100μL	100μL	200μL	100μL

2. 另取 8 支 2mL 离心管，每管分别加入 50μL 梯度浓度的 SDS 标准溶液。
3. 向每管中加入 50μL 染色液混匀。
4. 向每管中加入 1.5mL 萃取液，剧烈振荡充分混匀 3 min，进行萃取。
5. 室温 5000 rpm（2000 g），离心 5 min，吸取下层液体转移到新的离心管中。（见注意事项 1）
6. 在紫外分光光度计上测每管在 A660 处的 OD 值。
7. 重复步骤 2~6，依据相同 SDS 浓度，取两次同浓度下测量 A660 处的 OD 值的平均值。
8. 以每管 A660 平均值为纵坐标，对应的 SDS 浓度为横坐标，在 Microsoft Excel 软件中绘制标准曲线。

二、 样品 SDS 浓度测定

1. 将样品作适当倍数的稀释。
2. 取 3 支 2mL 离心管，其中两管各加入 50μL 稀释后的样品，另外一管加入 50μL 蒸馏水作为空白对照。
3. 向每管中加入 50μL 染色液混匀。
4. 向每管中加入 1.5mL 萃取液，剧烈振荡充分混匀 3 min，进行萃取。
5. 室温 5000 rpm（2000 g），离心 5 min，吸取下层液体转移到新的离心管中。（*见注意事项 1*）
6. 在紫外分光光度计上测每管的 A660 处的 OD 值。
7. 计算两管样品稀释液在 A660 处的 OD 值的平均值。 在标准曲线上确定出待测稀释后样品的 SDS 浓度。
8. 根据 SDS 浓度（%）=稀释后样品 SDS 浓度×样品稀释倍数从而计算待测样品的 SDS 浓度。

注意事项：

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1. 除离心外，也可竖立静置 10min 以上，然后吸取下层有机相液体转移到新的离心管中。
2. 试剂(C): 萃取液有一定气味且容易挥发，使用时请在化学通风橱中进行并做好防护措施，使用完毕后及时拧紧盖子防止溶剂挥发或变质。
3. 待测样品稀释后检测的 A660 处 OD 值应在标准曲线范围之内，若超过此范围则需要继续加大样品稀释倍数。
4. 因为染料和 SDS-染料复合物有相同的最大吸收波长，当样品含有三氯乙酸时，会使水相中未被 SDS 结合的染料也被萃取到有机相中，增加了 A660 值从而影响实验结果的准确性。

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SDS Residue Detection Kit

Cat: G4710
Size: 100T
Storage: RT, avoid light, valid for 1 year.

Kit Components

Reagent	100T	Storage
Reagent(A): 0.1%SDS Solution	10mL	RT, avoid light
Reagent(B): Staining Soltuion	10mL	RT, avoid light
Reagent(C): Extraction Solution	2×75mL	RT, avoid light

Introduction

SDS molecules have both hydrophilic and hydrophobic characteristics, and have important application value in the extraction, purification, and antigen preparation of genetically engineered protein drugs. However, SDS residues can have toxic side effects on animals and humans, so monitoring SDS residues at any time is of great significance.

The SDS Residue Detection Kit consists of three parts: 0.1% SDS solution, staining solution, and extraction solution. The 0.1% SDS solution can be diluted with water to prepare a gradient SDS standard solution and draw a standard curve. The staining solution can selectively bind with SDS in the sample to form a complex, which can be extracted into the organic phase by the extraction solution and then measured for OD value by UV spectrophotometry; The dye that has not been combined into a complex remains in the aqueous phase after extraction, so there is a certain linear relationship between the content of SDS in the sample and the absorption value of the complex extracted into the organic phase at a specific maximum absorption wavelength. It has been determined that the linear fit is good within the concentration range of 0.001% to 0.01% SDS, and can reach above 0.99.

Protocol(*for reference only*)

Create standard curves

1. Take 8 of 0.5mL centrifuge tubes and prepare gradient SDS standard solution according to the following table (taking 100μL/vial as an example).

Final concentration	0.01%	0.008%	0.006%	0.004%	0.002%	0.001%	0
0.1%SDS Solution	30μL	Tube1 -160μL	Tube2 -150μL	Tube3 -134μL	Tube4 -100μL	Tube5 -100μL	0
Distilled water	270μL	40μL	50μL	66μL	100μL	100μL	100μL
Mark	Tube 1	Tube 2	Tube 3	Tube 4	Tube 5	Tube 6	Tube 7
Final volume	140μL	100μL	100μL	100μL	100μL	200μL	100μL

2. Take 8 of new 2mL centrifuge tubes and add 50μL of gradient SDS standard solution to each tube.
3. Add 50μL Staining Solution to each tube and mix well.
4. Add 1.5mL Extraction Solution into each tube, shake vigorously and mix thoroughly for 3mins for extraction.
5. Centrifuge at room temperature of 5000 rpm (2000 g) for 5mins, and transfer the lower liquid to a new centrifuge tube. (*See Note 1*)
6. Measure the OD value of each tube at A660 on a UV spectrophotometer.
7. Repeat steps 2-6 and take the average of the OD values at A660 measured twice at the same SDS concentration.
8. Plot a standard curve in Microsoft Excel software with the average A660 value of each tube as the y-axis and the corresponding SDS concentration as the x-axis.

Determination of sample SDS

1. Dilute the sample by an appropriate multiple.
2. Take 3 of 2mL centrifuge tubes, add 50μL of diluted sample into two tube and add 50μL of distilled water into the other one tube as a blank control.
3. Add 50μL Staining Solution to each tube and mix well.

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4. Add 1.5mL Extraction Solution into each tube, shake vigorously and mix thoroughly for 3mins for extraction.
5. Centrifuge at room temperature of 5000 rpm (2000 g) for 5mins, and transfer the lower liquid to a new centrifuge tube. (*See Note 1*)
6. Measure the OD value at A660 of each tube on a UV spectrophotometer.
7. Calculate the average OD value of two sample dilutions at A660. Determine the SDS concentration of the diluted sample on the standard curve.
8. Calculate the SDS concentration of the sample based on the SDS concentration (%)=diluted sample SDS concentration × sample dilution factor.

Note

1. In addition to centrifugation, it is also possible to stand upright for more than 10mins and then transfer the lower organic phase liquid to a new centrifuge tube.
2. Reagent (C): Extraction Solution has a certain odor and is easy to evaporate. Please use it in a chemical fume hood and take protective measures. After use, tighten the lid promptly to prevent solvent evaporation or deterioration.
3. The OD value at A660 of diluted sample should be within the range of the standard curve. If it exceeds this range, the dilution factor of the sample needs to be further increased.
4. Because dyes and SDS dye complexes have the same maximum absorption wavelength, when the sample contains trichloroacetic acid, dyes that have not been bound by SDS in the aqueous phase will also be extracted into the organic phase, increasing the A660 value and affecting the accuracy of experimental results.