Visualization of proteins in SDS-PAGE gels, Coomassie blue staining

Introduction

Visualization of protein bands is carried out by incubating the gel with a staining solution. The two most commonly used methods are Coomassie and silver staining. Coomassie staining, though less sensitive, is quantitative and Coomassie-stained proteins can be used for downstream applications.

Materials

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- Coomassie staining solution (see table Coomassie staining solution (https://www.qiagen.com/fi/resources/molecular-biology-methods/protein/#Coomassie%20staining%20solution))
- Destaining solution (see table Destaining solution (https://www.qiagen.com/fi/resources/molecular-biology-methods/protein/#Destaining%20solution))
- > SDS polyacrylamide gel containing separated proteins

Procedure

1. Make Coomassie staining solution:

Table1					
K	A	В	С		
1	Composition of working solution	Component	Amount per 100 ml		
2	0.05% (w/v) Coomassie Brilliant Blue R- 250	Coomassie Brilliant Blue R- 250	50 mg		
3	40% (v/v) ethanol	Ethanol Dissolve then add:	40 ml		
4	10% (v/v) glacial acetic acid	Glacial acetic acid	10 ml		
5	50% (v/v) water	Water	50 ml		

2. Make Destaining solution:

Table2	2			
K	А		В	С
1	Compositio working solution	n of Compo	onent	Amount per 100 ml
2	40% (v/v) ethanol	Ethano	I	40 ml
3	10% (v/v) gla	acial Glacial acid	acetic	10 ml
4	50% (v/v) wa	ater Water		50 ml

- 3. Incubate the gel in Coomassie staining solution for between 30 min and 2 h with gentle shaking.
 - Tip: Coomassie Brilliant Blue R reacts nonspecifically with proteins.
- 4. Gently agitate the stained gel in destaining solution until the background becomes clear (1–2 h).
 - **Tip**: A folded paper towel placed in the destaining bath will soak up excess stain and allow the reuse of destaining solution.
- 5. After destaining the proteins appear as blue bands against a clear gel background. Typically, bands containing 50 ng protein can be visualized.