Silver Staining Kit

Catalogue #: MPK007
Storage: 4—8 ºC
Size: 20 mini gels

Description:

Silver staining made use of the principle of a reduction reaction. In silver staining, the gel is preserved with soluble silver ions and developed by treatment with reducing reagents such as formaldehyde, which reduces the Ag+ to elemental silver that appears as brown precipitate. The Ag+ of silver nitrate binds to amino acid residues of proteins and acrylamide matrix. This reduction is promoted by protein. The sodium thiosulfate present in sensitizing steps and some silver nitrate solutions keep the Ag+ in solution and diminish unspecific staining of background. It is a highly sensitive method for permanent staining of proteins in polyacrylamide gels compared with the Coomassie Brilliant Blue staining.

Kit Components:

Kit Components	*Chemicals*	*Size*	*Cat #*
I	Oxidizer, 10X	100 mL	MP023
II	Silver Staining Solution, 10X	100 mL	MP004
III	Developer A, 10X	100 mL	MP024
IV	Developer B	10 mL	MP025
V	Stop Solution, 10X	100 mL	MP026

Reagents needed, but not provided:

Fixation Solution I	*Fixation Solution II*
40% methanol	10% ethanol
10% acetic acid	5% acetic acid

Procedures:

The following protocol is appropriate for a 0.5 - 1.0 mm thick polyacrylamide gel. Prepare. Transfer the gel to fixation solution immediately after electrophoresis. The gel may be stored indefinitely in this fixative prior to staining. The volume of working solutions depends on the size of the gel (50 mL per 100 mm^2)

1. Gel fixation
   - Fix the gel in Fixation Solution I for 30 min.
   - Fix the gel in Fixation Solution II for 30 min.
2. Gel staining
   - Immerse the gel in 1X Oxidizer (dilute to 1X before use) and incubate with gentle shaking for 5 min.
   - Wash the gel with sufficient de-ionized water (>100 mL) until yellow color is removed from gel (it normally takes about 10 min with 2-3 changes of water).
   - Apply the Silver Staining Solution (dilute to 1X before use) to cover the gel for 20 min with gentle shaking.
   - Quickly rinse the gel with de-ionized Water twice.

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3. Developing
   - Apply the Developer (freshly made) until a smoky brown precipitate appears (it normally takes less than 1 min).
   - Replace with new Developer for 5 min (until desired stain intensity develops).
   - . Rinse with deionized water
   - Stop the developing process by adding Stop Solution.

Notes

- Do not use the plastic tray because most plastic containers have softeners (sulphur-compunds) which reduce silver nitrate to metallic silver.
- Avoid direct contact of the gel with hands and gloves, which will leave “finger prints” on your gel after developing.
- Prepare the developing solution freshly prior to use.

References:

1. Swain and Ross. Electrophoresis. 1995 Jun; 16(6):948-51.
2. Blum et al. Electrophoresis. 1987; 8:93-99.

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