User Manual

Product Name: Coomassie Brilliant Blue G-250

Cat #: 18-446, 18-447

Staining Protocol

A. For SDS-PAGE Mini Gels

NOTE: *This protocol is written for mini gels (8-10") and if large size gels (16-20"), double the volumes used in the protocol.*

1. After the completion of electrophoresis step, remove the gel and place it in a smooth plastic tray, then add about 200 ml of deionized (DI) water into it.

2. Wash the gel 3 times in about 200 mL DI Water, 5 minutes each time on a rotary shaker to remove the SDS present in the running buffer. ***NOTE:*** *If the gel is not thoroughly washed, the presence of SDS in it will interfere with the protein staining and band intensity.*

3. Remove all water from the container to minimize the dilution of the Coomassie G-250 stain

4. ***NOTE:*** *Gently mix the Coomassie Brilliant Blue G-250 Stain bottle before opening and using (**Important**).*

Add 50 mL of Coomassie Brilliant Blue G-250 Stain to cover the gel (sufficient for a mini gels). Put the container with gel in stain on an orbital shaker and gently shake it for 1 hour at room temperature.

5. Protein bands will start appearing and most of them visible within 15-20 minutes with maximum intensity in 1 hour.

6. Pour off the stain carefully and rinse the gel with 200 mL DI water for 30 minutes to overnight, the bands will become prominent and sharp. Rinsing the gel thoroughly in DI water will reduce the background.

7. Proceed with downstream applications. Stained gels can be stored in water for 2-3 days.

B. For Peptide Gels

Fix the gel in 40% methanol, 10% acetic acid for 30 minutes, remove all free fixing agent and follow the above protocol used for SDS-PAGE starting from step 4 onward and increase the time at step 4 above to 2 hours.