

SOP-102

Coomassie Blue Staining of Polyacrylamide Gels

Procedure for detecting proteins in
polyacrylamide gels.

Materials and Reagents

Coomassie Brilliant Blue Staining Solution

(1 liter)

| | |
|-------------------------------|--------|
| Coomassie Brilliant Blue R250 | 2.5 g |
| Methanol | 400 ml |
| Glacial Acetic Acid | 100 ml |
| H ₂ O | 500 ml |

High Methanol De-stain Solution

(1 liter)

| | |
|---------------------|--------|
| Methanol | 400 ml |
| Glacial Acetic Acid | 100 ml |
| H ₂ O | 500 ml |

4 % (v/v) Glycerol Solution

(1 liter)

| | |
|------------------|--------|
| Glycerol | 40 ml |
| H ₂ O | 960 ml |

Protocol

1. Stain the gel in approximately 50 ml of staining solution. Incubate at room temperature on a rotating platform for 20 min.
2. Remove the staining solution. Used solution can be kept and reused until quality of staining decreases.
3. Rinse gel in de-stain solution. Remove excess stain by incubating gel in 100 ml of de-stain solution for 2 – 3 hr. de-staining is complete when the background is essentially clear.
4. Equilibrate gel in 4 % glycerol for 1 hr. The glycerol helps to keep gel from cracking when dried.
5. Record results electronically or by photography while gel is wet. Preserve gel by drying between two cellophane membranes. Hydrate membranes and place gel in between. Remove any air bubbles and allow gel to dry either in a frame or in a gel dryer.

References

Short Protocols in Molecular Biology. 4th Edition. Editors Frederick M. Ausubel, Roger Brent, Robert E. Kingston, David D. Moore, J.G. Seidman, John A Smith and Kevin Struhl. John Wiley & Sons, Inc. 1999