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COOMASSIE COLLOIDAL STAIN - CHEM 584

Forked from VISUALIZING PROTEINS ON PAGE GEL WITH COOMASSIE COLLOIDAL STAIN - CHEM 584

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ABSTRACT

Staining polyacrylamide gels using a colloidal Coomassie Blue stain. Adapted from a JoVE article.

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1 Wash gel with ~50 mL ddH_2O 3x for ~ **00:10:00** for each wash on a shaker

NOTE: insufficient washing causes poor sensitivity because the remaining SDS on the gels disturbs the bounding of the dye to the protein

2 Shake the colloidal Coomassie solution before use to disperse particles evenly.

3 Incubate the gels with the Coomassie solution by agitation on a shaker up to **12:00:00** . This staining generates marginal background so you can observe the staining progress.

Note: after 10 minutes you should see some protein bands appearing, within 2 hours of incubation about 80% maximum staining is completed. For nearly maximum sensitivity, an overnight incubation is recommended. In some cases, when the amount of protein to be stained is large, the solution turns a bright blue color. If this happens, you

should refresh the staining solution as necessary.

4 After staining, remove the Coomassie solution and rinse the gels twice with ddH_2O.

Note: you can reuse the staining solution as long as particles still remain, otherwise discard it appropriately.

5 Remove any dye particles from the staining dish with a lint-free paper towel and destain for up to **01:00:00** in ddH_2O or a Coomassie destain solution (10% (v/v) 96% ethanol, 2% (v/v) orthophosphoric acid 85% in ddH_2O.)

6 Finally rinse the gels twice with ddH_2O: the gels will reach their original thickness (the alcohol-acid media makes the gels shrink). Neutralization in water also enhances the Coomassie stain's color intensity.

7 You can record your gel using the gel documentation system or scan it on the Licor using the 680/700 nm channel.

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