Lumiprobe
Life science solutions

DNA Staining in Gels with dsSafe®

dsSafe® is a fluorescent dye for staining nucleic acids in agarose and polyacrylamide gels. This dye is the best safe alternative to ethidium bromide (EtBr): dsSafe is non-toxic, does not have mutagenic or carcinogenic activity, and does not require special storage and disposal conditions.

DNA bands stained with dsSafe dye can be detected using a standard UV transilluminator, a visible light transilluminator, or a laser scanner. The dye is also suitable for staining RNA in gels.

In complex with DNA, the spectrum of the dsSafe dye has two excitation peaks: in the ultraviolet (280 nm) and blue ranges (502 nm), and an emission maximum in the green range of ~530 nm.

There are two nucleic acid staining protocols in gels: soaking gel after electrophoresis or gel pre-staining before electrophoresis.

Gel soaking

This method is the most useful for staining DNA in agarose and polyacrylamide gels (PAGE).

1. Perform agarose or polyacrylamide gel electrophoresis of the sample(s).
2. Take the required 1× TAE or 1× TBE volume into a plastic container. To stain a 20 mL agarose gel, 35–50 mL of dye solution will be sufficient.
3. Add dye concentrate to the buffer in a ratio of 1:10,000. Mix thoroughly.
4. Place the gel in a plastic tray (tip box lid or household storage container).

   Important! Do not use a glass container, as the dye may adsorb to the walls of the container, resulting in poor gel staining.
5. Add enough dye solution volume to cover the gel completely.
6. Cover the gel and the staining solution with aluminum foil or place them in the dark to protect from light.
7. Soak the gel in the dsSafe solution for at least 20 minutes at 37 °C and constant stirring on an orbital shaker at 50–80 rpm.

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Gel pre-staining

This method is only suitable for agarose gels and not for PAGE.

Important! This staining method can sometimes cause DNA bands to warp or form smears. In this case, the gel should be soaked in a dye solution.

1. Dilute dsSafe dye concentrate 1:10,000 in agarose gel buffer (e. g. 1× TBE or 1× TAE). Add agarose powder to the dye solution. For example, if 20 mL of molten agarose is required, add 2 µL of 10,000× dsSafe dye concentrate to 20 mL of 1× buffer, mix the solution well and add agarose.
2. Heat the agarose in the dye buffer to boiling and wait until it is completely dissolved. You can use a microwave or other heating device.

   (Optional) Alternatively, dsSafe can be added directly to the molten agarose at a ratio of 1:10,000 (1 µL dye concentrate for every 10 mL buffer).
3. Pour the resulting mixture into the gel mold and wait until it hardens.

   Important! As with pre-staining gels with ethidium bromide, the mobility of nucleic acid fragments in the gel may be slightly lower compared to their mobility in the gel without stain.
4. Run the samples. It is possible to observe migrating DNA bands in real time under a low-pressure mercury lamp with a wavelength of 254 nm.

Gel viewing and photographing

You can view stained gels using a standard 300 nm transilluminator, a 254 nm epi- or transilluminator, or a blue light transilluminator.

DNA stained with dsSafe dye can also be visualized and analyzed using imaging systems equipped with an excitation source in the UV or 470–530 nm range.

Important! If bands from the dsSafe stained gel are to be excised and used in the ligation reaction, illumination with a blue light source, but not ultraviolet light, is recommended. Sometimes, UV light sources combined with dsSafe staining may decrease cloning efficiency.

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