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Protocol: DNA Staining in Gels with dsGreen or SYBR Green I

dsGreen, an analog of SYBR® Green I, is a fluorescent dye that binds specifically to double-stranded DNA. There are three variants of the staining protocol: gel soaking, gel pre-staining, and sample pre-staining.

Gel soaking

Classical method for agarose and polyacrylamide gels.

1. Run sample(s) in an agarose or polyacrylamide gel.
2. In a beaker, add 10 µL of the 10,000× dsGreen solution in DMSO to 100 mL of 1× TE, TBE, or TAE buffer (for mini gels), or 50 µL of the 10,000× dsGreen solution in DMSO to 500 mL of 1× TE, TBE, or TAE buffer (for mid-sized gels). Mix thoroughly with a spatula, rod, or magnetic stirrer.
3. Pour the diluted dsGreen solution into an appropriate tray or pan and submerge the gel.
4. Soak the gel for 5–10 min.
5. View or document the gel using available light source and an green/yellow filter. Transilluminators with blue light, or with UV low pressure mercury lamp (254 nm) can be used to visualize gels stained with dsGreen. High pressure mercury lamp (365 nm) can be used too, but this light source gives somewhat less efficient excitation.

Gel pre-staining

This method is suitable for **agarose** gels only, but not for PAAG. Note - this staining method can sometimes cause bands to warp or form smears. Use gel soaking in this case.

1. Boil the agarose in buffer to dissolution using a microwave or heating appliance.
2. While still fluid, add 1 µL of the 10,000× dsGreen solution in DMSO per each 10 mL of gel solution. Mix thoroughly.
3. Pour the gel and let it solidify.
4. For best results, add 1 µL of of the 10,000× dsGreen solution in DMSO per each 10 mL of buffer near the anode ("+", red wire).
5. Run the samples. Real-time monitoring of migrating bands under a 254 nm low-pressure mercury lamp is possible.
6. View or document the gel using available light source and an green/yellow filter. Transilluminators with blue light, or with UV low pressure mercury lamp (254 nm) can be used to visualize gels stained with dsGreen. High pressure mercury lamp (365 nm) can be used too, but

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Sample pre-staining

Least sensitive, most economical method.

1. Mix 25 µL of DMSO and 1 µL of the 10,000× dsGreen solution in DMSO.
2. Add 1 µL of the solution to each sample to be separated on an agarose or polyacrylamide gel.
3. Run the samples. Real-time monitoring of migrating bands under a 254 nm low-pressure mercury lamp is possible.
4. View or document the gel using available light source and an green/yellow filter. Transilluminators with blue light, or with UV low pressure mercury lamp (254 nm) can be used to visualize gels stained with dsGreen. High pressure mercury lamp (365 nm) can be used too, but this light source gives somewhat less efficient excitation.