

Protocol: SYBR Green I Staining of DNA in Gels

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

Gel soaking

1. Run sample(s) in agarose or polyacrylamide gel.
2. In a beaker, add 10 uL of 10,000x SYBR Green I solution in DMSO to 100 mL of distilled water (for mini gel), or 50 uL of 10,000x SYBR Green I solution in DMSO to 500 mL of distilled water (for mid-sized gel). Mix thoroughly with spatula, rod, or magnetic stirrer.
3. Pour the diluted SYBR Green I solution in appropriate tray or pan.
4. Soak the gel for 5-10 min.
5. View or document the gel using 254 nm low-pressure mercury lamp and orange filter.

Gel pre-staining

This method is acceptable for agarose gels only, but not for PAAG.

1. Boil the agarose in buffer to dissolution using microwave or heating appliance. While hot, add 1 uL of 10,000x SYBR Green I solution in DMSO per each 10 mL of gel solution. Mix thoroughly.
2. Pour the gel and let it cool down.
3. For best results, add 1 uL of 10,000x SYBR Green I solution in DMSO per each 10 mL of buffer near anode ("+", red wire).
4. Run the samples. Real-time monitoring of migrating bands under 254 nm low-pressure mercury lamp possible.
5. View or document the gel using 254 nm low-pressure mercury lamp and orange filter.

Sample pre-staining

Least sensitive, most economical method.

1. Mix 25 uL of DMSO and 1 uL of 10,000x SYBR Green I solution in DMSO.
2. Add 1 uL of the solution to each sample to be separated on agarose or polyacrylamide gel.
3. Run the samples. Real-time monitoring of migrating bands under 254 nm low-pressure mercury lamp possible.
4. View or document the gel using 254 nm low-pressure mercury lamp and orange filter.