

Dye: Ethidium bromide (EtBr)

Ethidium bromide is commonly used in agarose gel electrophoresis to detect double-stranded DNA (dsDNA) and single-stranded DNA (ss) or RNA. When exposed to ultraviolet light, ethidium bromide will fluoresce with an orange color, after binding to DNA.

The ethidium bromide dye can either be added directly to the agarose gel before it is set or the agarose gel can be incubated for 15-30 minutes with a solution of ethidium bromide diluted in water. To reduce background staining, destaining of the gel in water for 15 minutes can be performed, if necessary.

Applications

Ethidium bromide can be used to detect DNA and RNA from PCR's, restriction enzyme digests, separation of restricted genomic DNA and many more.

Visualization

Agarose gels stained with ethidium bromide can be visualized extremely well using all Syngene image capture systems. Ethidium bromide has an excitation peak of 300nm (blue peak) and an emission peak 595nm (red peak on spectra). For UV illumination it is recommended to use a transilluminator with a UV filter.



Figure 1 – Excitation (blue line) and emission (red line) spectra of Ethidium Bromide dye

System	Lighting	Filter
Dyversity and G:BOX iChemi range using cooled cameras	Transilluminator – medium wave UV	FiltUV
G:BOX range using non cooled cameras and InGenius	Transilluminator – medium wave UV	FiltUV

Table I - Recommended lighting and filter selection for visualizing Ethidium bromide stained agarose gels using Syngene image capture systems



Ethidium Bromide stained agarose gel visualised using the InGenius image capture system

Samples of DNA were resolved on a pre-stained ethidium bromide 2% agarose gel in TAE buffer. The gel was run at 120 volts for an hour. Bands were visualized by exposure to UV light on a transilluminator and a UV filter. The gel image was captured using GeneSnap software (Syngene) for an exposure time of 400 milliseconds using the InGenius Syngene image capture system.

An ideal ethidium bromide gel image will have clearly defined bright bands as illustrated in the image above.

Syngene reserves the right to amend or change specifications without prior notice. This Application note supersedes all earlier versions.

All trademarks acknowledged.

48.05.09

UK tel: +44 (0)1223 727123 Email: <u>sales@syngene.com</u>

USA tel: 800 686 4407/301 662 2863 Email: <u>ussales@syngene.com</u>