

Resource Summary Report

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DNA Separation and Detection on Agarose Gels/Post-Electrophoresis Staining Protocol

DOI:10.17504/protocols.io.q92dz8e

Type: Protocol

Proper Citation

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Protocol Information

URL: <https://dx.doi.org/10.17504/protocols.io.q92dz8e>

Authors: LI-COR Biosciences

Group: LI-COR Biosciences

Summary: The Odyssey Fc Imager, with 600 channel capabilities, can image agarose gels stained with popular DNA stains, such as ethidium bromide and SYBR Safe DNA stain, with sub-nanogram sensitivity. The Odyssey Fc Imager contains a 532 nm diffuse source with an excitation maximum of 520 nm and a detection maximum of 600 nm. These instrument parameters are within the range of the excitation and emission wavelengths of ethidium bromide (Ex/Em = 302 & 518/605 nm^{1,2}) and other visible fluorescent nucleic acid stains and provide a sensitive gel documentation option. SYBR Safe DNA stain (Ex/Em = 502/530 nm) has also been tested on the Odyssey Fc Imager (using the 600 channel) with sensitivities exceeding ethidium bromide detection. The maximum fluorescence emission wavelength of SYBR Safe is very close to the maximum excitation wavelength. However, the Odyssey Fc 600 channel collects excitation light at a wavelength 50 nm higher than the maximum excitation wavelength of SYBR Safe. These instrument properties decrease the background and improve the signal-to-noise ratio for nucleic acid detection. Specific instructions are given in this technical note for ethidium bromide and SYBR Safe use. There are a variety of commercial DNA stains that may be appropriate for fluorescent imaging with the Odyssey Fc 600 channel. SYBR Green I (Life Technologies), GelStar (FMC), Gel Red™ (Biotium), Gel Green™ (Biotium) and Nancy-520 (Sigma) stains have also been tested at LI-COR (see example images on page 7). Other nucleic acid binding stains may also be compatible with the Odyssey Fc Imager. Please check the excitation and emission spectra of each stain. Developed for: Odyssey Fc Imaging System

Affiliations: LI-COR Biosciences

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