FT-FJ5570

GelRed™ Nucleic Acid Gel Prestaining Kit

Simply the best nucleic acid gel stain.

Product Description

Product name cat.number	Components
GelRed™ Nucleic Acid Gel Prestaining Kit with 200bp and 1500-2000bp Tracking Dyes FJ5570, for at least 100 mini gels	A) 2 x 1 mL 6X GelRed™ Loading Buffer with "200bp" and "1,5Kb" tracking dyes B) 1 x 250 ml 200X GelRed™ Running Buffer (for 50 L of 1X concentrated buffer)
GelRed™ Nucleic Acid Gel Prestaining Kit with 50bp Tracking Dye FJ5580, for at least 100 mini gels	A) 2 x 1 mL 6X GelRed™ Loading Buffer with "50bp" tracking dye B) 1 x 250 ml 200X GelRed™ Running Buffer (for 50 L of 1X concentrated buffer)

Storage: Loading Buffer at room temperature (6 months). Protect from light.

Introduction

GelRed™ dye is a sensitive, stable and relatively safe fluorescent nucleic acid dye designed to replace the highly toxic ethidium bromide (EB) for staining dsDNA, ssDNA or RNA in agarose gels or polyacrylamide gels. GelRed™ is far more sensitive than EB without requiring a destaining step. GelRed™ and EB have virtually the same spectra (Figure 1), so you can directly replace EB with GelRed™ without changing your existing imaging system. If you have been using a green fluorescent gel stain such as SYBR™ Green I or GelStar™ with a UV transilluminator for viewing gels, you may replace the dye and continue to use the existing SYBR™ filter or GelStar™ filter for photographing. However, GelRed™ can not be optimally excited with a 488 nm argon laser or similar visible light and therefore is not recommended for use with a gel reader equipped with such visible light. In such cases, we recommend GelGreen™ prestaining kits (cat.# FJ5590) for visible light excitation.

The kit is provided with a 6X loading buffer conveniently containing GelRed™ dye and an electrophoresis tracking dye. The loading buffer and running buffer formulations have been especially optimized for prestaining with GelRed™ dye and running gels at high voltages without overheating. The tracking dyes run at 1,5Kb and 200bp in a 1% agarose gel.

In addition to its remarkable sensitivity and stability, toxicity test by an independent laboratory has shown that GelRed™ is non mutagenic and non-cytotoxic at concentrations used for gel staining. A key aspect of GelRed™ safety is that the dye appears to be completely cell membrane impermeable, compared to Green dye I, which enters cells rapidly. For more details on the safety of GelRed™, please contact us for the complete safety report.

Gel staining with GelRed™ is compatible with downstream DNA manipulations such as digestion with a restriction enzyme, Southern blotting techniques and cloning. GelRed™ may be removed from DNA by commercial gel extraction or ethanol precipitation.

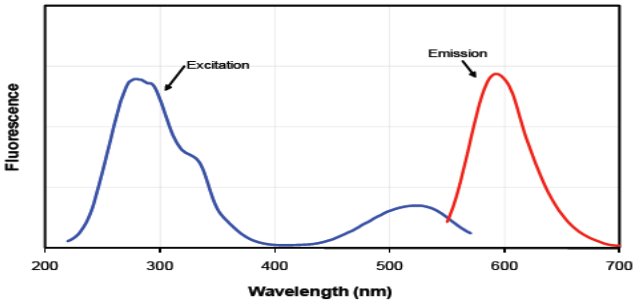

Figure 1: Excitation and emission spectra of GelRed™ dye bound to dsDNA

Directions for use

Handling and Storage

GelRed™ dye is very stable. We recommend that you store the 6X loading buffer at room temperature protected from light. It will remain stable for at least 6 months. The dye can be handled under ambient light without any problem during electrophoresis.

Staining Protocol

1- Prepare a 1X running buffer by mixing 5 mL of the 200X running buffer concentrate with 995 mL of de-ionized water for a total of 1 L of 1X running buffer. If a large amount of precipitation is seen in the 200X concentrated stock, warm up solution until clear.
2- Prepare an agarose slurry in 1X running buffer to the desired percentage. Heat until agarose is dissolved completely.
3- Cast the gel and allow enough time for the gel to completely solidify. A partially hardened gel can result in distorted bands.
4- Place the gel in the electrophoresis rig and cover sufficiently with 1X running buffer (*Note: Reusing running buffer may result in reduced performance.*)
5- Briefly vortex the 6X loading buffer vial. Add 6X loading buffer with the DNA sample at a volume ratio of 1:5 (for example, mix 10 uL sample + 2 uL 6X loading buffer).
6- Load samples and run the gels in GelRed™ running buffer at 8-10 V/cm (cm=length of gel). The optimized buffer formulation allows for twice the running voltage in half the time of standard buffers. **Unused lanes may be saved and used at a later time.**
7- Visualize bands on a UV transilluminator or other gel documentation system. Images can be taken using an ethidium bromide emission filter.

Quick Guide Protocol

1- Prepare 1X running buffer from 200X stock.
2- Cast agarose gel in 1X running buffer.
3- Briefly vortex the 6x loading buffer and add to the DNA samples at a volume ratio of 1:5 (for example, 10 uL sample + 2 uL 6X loading buffer).
4- Place the gel in the electrophoresis rig, cover sufficiently with 1X running buffer and load samples.
5- Run gel at 8-10 V/cm (cm = gel length).
6- Visualize bands on a UV transilluminator or other gel documentation system.

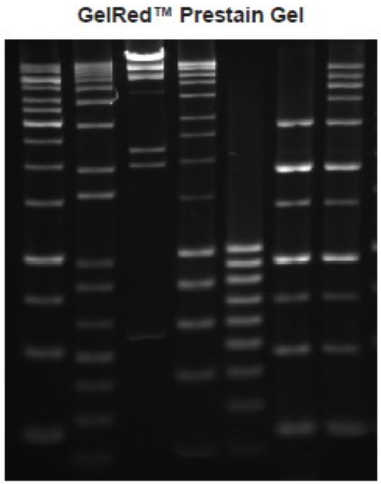

Figure 2 : DNA ladders were premixed with the GelRed™ loading buffer and electrophoresed on the gel. Samples in the lanes are as follows:

(1) Fermentas GeneRuler 1kB ladder,
(2) Invitrogen 1kB Plus ladder,
(3) New England BioLabs Lambda DNA-HindIII digest,
(4) Bioline HyperLadder I,
(5) Bioline HyperLadder IV,
(6) Axygen M-DNA-LR, and
(7) Axygen M-DNA-BR.

Images were taken on a GelDoc-It™ system (UVP).

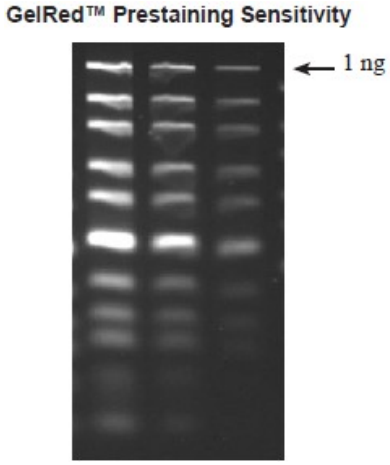

Figure 3 : GelRed™ loading buffer was premixed with GeneRuler Ultra Low Range DNA Ladder at 62.5, 31.3 and 15.6 ng per lane from left to right and ran on a 3% agarose gel.

Images were taken on a GelDoc-It™ system (UVP). The lowest band is a 10 bp DNA fragment. The uppermost band in the righthand lane is approximately 1 ng.

Troubleshooting tips for distorted/smeared resolution of DNA

1- The optimal amount of DNA is 50 - 250 ng/lane. Using more or less than this range may affect DNA migration depending on the dye to DNA ratio. If accurate sizing or concentration estimation of DNA fragments is required, we recommend using our 10,000X GelRed products with a post-staining procedure.
2- Lower running voltage (warmer running buffer may distort bands);
3- Adjust the amount of agarose in the gel (lower agarose % to separate high Mwt fragments or increase agarose % for better separation of low Mwt fragments);
4- Run a longer, slower gel for better separation;
5- Increase gel solidification time to ensure sharp well formation.

TOXICITY:
GelRed™ dye was subjected to a series of tests both by us and by three independent testing services to assess the dye's safety for routine handling and disposal. These tests include: 1) glove penetration test; 2) cell membrane permeability and cytotoxicity test; 3) Ames test; and 4) environmental safety tests. Test results confirm that the dye is impenetrable to both latex gloves and cell membranes. The Ames test performed by an independent lab, Litron Laboratories (Rochester, NY), showed that GelRed™ dye is not mutagenic at concentrations used for gel staining and is only weakly mutagenic following metabolic activation at 18.5 mg/mL, which is significantly higher than the 1X (~1 mg/mL) working concentration used in our previously recommended precast gel staining protocol. It should be noted that both the concentration and the absolute amount of the DNA-binding dye used in the current prestaining protocol are far lower than before, thus offering further safety improvement. However, since these tests were not performed on humans, we still advise that researchers exercise precaution when handling the dye or any other DNA-binding molecules by wearing protective gear. For more information on the safety test result, you may download a complete report.

DISPOSAL :
Used gels generated after following this protocol contain only a negligible amount of the DNA dye and thus are generally safe to be disposed of in regular trash. Your used running buffer should be totally free of GelRed™ dye, as the DNA dye will not leak out of the gel matrix, and thus can be disposed of down the drain. However, despite the safety feature of the product, we still recommend that you check your institutional regulations for proper waste disposal.

FIRST AID: Potentially harmful. Avoid prolonged or repeated exposure. Avoid getting in eyes, on skin, or on clothing. Wash thoroughly after handling. If eye or skin contact occurs, wash affected areas with plenty of water for 15 minutes and seek medical advice. In case of inhaling or swallowing, move individual to fresh air and seek medical advice

References

- **Abouhamed M**. *et al*., Knockdown of endosomal/lysosomal divalent metal transporter 1 by RNA interference prevents cadmium-metallothionein-1 cytotoxicity in renal proximal tubule cells, *Am J Physiol Renal Physiol* 293: F705 - F712 (2007) Abstract
- **Aronica L**. *et al*., Study of an RNA helicase implicates small RNA–noncoding RNA interactions in programmed DNA elimination in Tetrahymena, *Genes & Dev*., 22: 2228 - 2241 (2008) Abstract
- **Diamond A**. *et al*., Modulation of Monocyte Chemotactic Protein-1 Expression During Lipopolysaccharide-Induced Preterm Delivery in the Pregnant Mouse, *Reproductive Sciences*, 14: 548 - 559 (2007) Abstract
- **Failor K.** *et al*., Glucocorticoid-induced degradation of GSK3 protein is triggered by Sgk and Akt signaling and controls beta-catenin dynamics and tight junction formation in mammary epithelial tumor cells, *Molecular Endocrinology,* 21: 2403 - 2415 (2007) Abstract
- **Graber H**. *et al*., Development of a Highly Sensitive and Specific Assay to Detect *Staphylococcus aureus* in Bovine Mastitic Milk, *J. Dairy Sci*.. 90:4661-4669 (2007) Abstract
- **Lotfy W.** *et al*., Evolutionary Origins, Diversification, and Biogeography of Liver Flukes (Digenea, Fasciolidae), *Am J Trop Med Hyg*, 79: 248 - 255 (2008) Abstract
- **McConnell K**. *et al*., Tolerance of Sir1p/Origin Recognition Complex-Dependent Silencing for Enhanced Origin Firing at *HMR*a, *Molecular and Cellular Biology*, p. 1955-1966, Vol. 26, No. 5 (2006) Article
- **Nikitina T.** *et al*., MeCP2-chromatin interactions include the formation of chromatosome-like structures and are altered in mutations causing Rett syndrome, *J. Biol. Chem*., 282: 28237 - 28245 (2007) Article
- **Reincke S**. *et al*., Mutation analysis of the MDM4 gene in German breast cancer patients, *BMC Cancer*,8:52 (2008) Article
- **Weber C. and King G**., Physiological, Ecological, and Phylogenetic Characterization of Stappia, a Marine CO-Oxidizing Bacterial Genus, *Appl. Envir. Microbiol*., 73: 1266 - 1276 (2007) Article

Related products

- Agarose regular uses, Molecular Biol. grade, 31272L
- GelGreen™ Nucleic Acid Gel Prestaining Kit, FJ5590
- Fast EvaGreen™ master mix for qPCR and HRM, DV7220
- AccuBlue dsDNA Quantification Kit, EV4080, EV4100
- UptiTherm™ DNA Polymerase, UPS53921
- dNTP set, UP968640
- UptiReverse Transcriptase 1-Step Kit, KB0310

Ordering information

Catalog size quantities and prices may be found at http://www.interchim.com/.
Please inquire for higher quantities (availability, shipment conditions).

For any information, please ask : FluoProbes® / Interchim; Hotline : +33(0)4 70 03 73 06