

## Introduction

**LiGreen™ Plus Nucleic Acid Gel Stain** is a highly sensitive fluorescent stain for detecting nucleic acids in agarose and polyacrylamide gels. This single stain gives high sensitivity detection of double-stranded or single-stranded DNA and RNA. Gels can be post-stained or alternatively the stain can be added to agarose gels during gel casting. LiGreen™ Plus is compatible with a standard 300 nm transilluminator, a 254 nm transilluminator, a blue-light transilluminator, or a gel reader equipped with visible light excitation such as a 488 nm laser-based gel scanner.

LiGreen™ Plus Nucleic Acid Gel Stain, 10,000× is a concentrated LiGreen™ Plus solution that can be diluted 10,000 times for use in precast gel staining or 5,000 times for use in post gel staining according to the procedures described below. One vial of 10,000× solution can be used to prepare at least 100 precast minigels or post-stain at least 100 minigels.

Gel staining with LiGreen™ Plus is compatible with downstream applications such as gel extraction and cloning. LiGreen™ Plus is efficiently removed from DNA by phenol/chloroform extraction and ethanol precipitation.

## Package Information

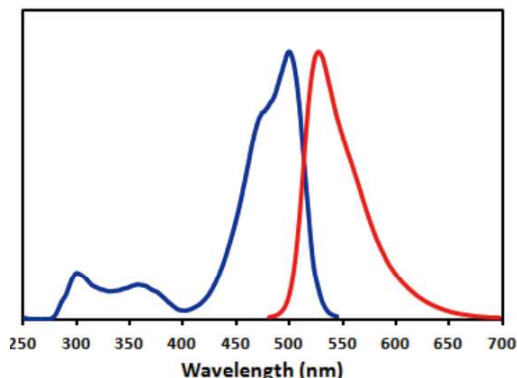
Components	M0050
LiGreen™ Plus DNA Gel Stain (10,000× in DMSO)	500 µl

Ex/Em: 500/530 nm, bound to nucleic acid

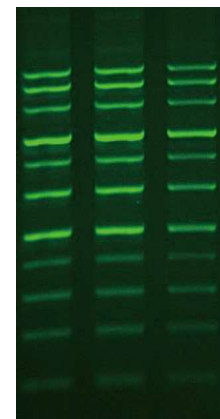
## Storage

Store at 2-25°C and protect from light.

## Spectral Characteristics



Excitation (blue) and emission spectra (red) of LiGreen™ Plus bound to dsDNA in TBE buffer



LiGreen™ Plus  
in pre-cast gel staining

## Staining Protocols

### Post-staining Protocol

1. Run gels as usual according to your standard protocol.
2. Dilute the LiGreen™ Plus 10,000× stock reagent 5,000 fold to make a 2× staining solution in TE, TBE, or TAE buffer.
3. Carefully place the gel in a suitable polypropylene container. Gently add a sufficient amount of the 2× staining solution to submerge the gel.
4. Agitate the gel gently at room temperature for 30 min.
5. Wash the gel with DI water to remove excess dye. Image the stained gel with a transilluminator, or a laser-based gel scanner using a long path green filter such as a SYBR Filter or GelStar filter.

### Pre-cast Protocol

1. Prepare molten agarose gel solution using your standard protocol.
2. Dilute the LiGreen™ Plus 10,000× stock reagent into the molten agarose gel solution at 1:10,000 and mix thoroughly.
3. Cast the gel and allow it to solidify.
4. Load samples and run the gels using your standard protocol.
5. Image the stained gel with a transilluminator, or a laser-based gel scanner using a long path green filter such as a SYBR Filter or GelStar filter.

**Note:** The pre-cast protocol is not recommended for polyacrylamide gels. Use the post staining protocol for acrylamide gels.

## Troubleshooting

### **Smeared DNA bands in precast gel**

1. Reduce the amount of DNA loading. Smeared bands can be caused by overloading.
2. Perform post-staining instead of pre-casting.
3. Prepare a lower percentage agarose gel for better resolution of large fragments.
4. Change the running buffer. TBE buffer has a higher buffering capacity than TAE.

### **Discrepant DNA migration in precast gel**

1. Reduce the amount of DNA loading.
2. Reduce the amount of dye used, i.e. use 0.5× in precast gels.
3. Perform post-staining instead of pre-casting.

### **Weak fluorescence signal**

1. The dye may be precipitated out of solution. Vortex to redissolve.
2. Increase the amount of dye used, i.e. use 2× in precast gels.