

## Neodye DNA Orange (10,000 ×)

Catalogue number  
NB-79-0001

### Features

- Detects double-strand DNA and single-stranded RNA effectively.
- Offers a safe alternative to ethidium bromide staining.
- Exhibits sensitivity on par with EtBr.
- Non-toxic, non-mutagenic, and non-carcinogenic composition.
- Produces no hazardous waste, ensuring environmental friendliness.

### Description

NeoDye DNA Orange is a sensitive, stable and relatively safe fluorescent nucleic acid dye designed to replace the highly toxic ethidium bromide (EtBr) for staining dsDNA, ssDNA or RNA in agarose gels or polyacrylamide gels. The sensitivity of NeoDye DNA Orange is much higher than EB, and destaining is not required. NeoDye DNA Orange and EB have virtually the same spectral properties.

### Storage

At room temperature protected from light for two years

### Highlights

- Nontoxic: Unique lipophilic and macromolecular properties of NeoDye DNA Orange make it incapable of penetrating cell membranes. Ames test also shows the mutagenicity of NeoDye DNA Orange is far less than that of EB.
- Highly sensitive: Suitable for staining fragments of different sizes in electrophoresis gel.
- Highly stable: Suitable for using microwave or other heating methods to prepare agarose gel; extremely stable at room temperature under acidic or alkaline conditions; highly resistant to light.
- High signal-to-noise ratio: Strong fluorescence signal of the sample with low background signal
- Simple operation: Similar to ethidium bromide, the dye does not degrade during the process of preparing gel or electrophoresis. It only takes 30 minutes for staining after electrophoresis, and the fragment can be visualized by a UV-transilluminator directly without detaining or washing.
- A broad range of applications: Applicable for precast protocol (add dye during gel preparation) and post-stain protocol (submerge the gel in the staining solution); suitable for agarose gels and polyacrylamide gels electrophoresis; suitable for staining dsDNA, ssDNA or RNA.
- Changing the existing imaging system is not required: Suitable for both standard EB filter and SYBR filter; same with the standard UV-transilluminator used for EB staining observation; optimal excitation in the UV region around 300 nm.

### Components

COMPONENT	NB-79-0001-01	NB-79-0001-02	NB-79-0001-03
NeoDye DNA Orange	500 µL	1 ml	5x1 ml

## Staining Protocol:

### Method 1: Precast staining (the same as EB staining, recommended)

Add NeoDye DNA Orange to molten agarose to make 1X working concentration (e.g. add 1  $\mu$ L of NeoDye DNA Orange 10,000X stock reagent to 10 ml of agarose).

#### Note:

- Due to the high thermal stability, NeoDye DNA Orange can be added to hot agarose solution directly and mix well without waiting for the solution to cool. It is also optional to add NeoDye DNA Orange to agarose powder and electrophoresis buffer, which will be heated by microwave to prepare agarose gel. NeoDye DNA Orange is compatible with all commonly used electrophoresis buffers
- The precast gel staining protocol is not recommended for polyacrylamide gels. Polyacrylamide gels can be stained using the post-staining protocol.

### Method 2: Post-staining

1. Dilute NeoDye DNA Orange 10,000x stock solution 3,300 fold with 0.1 M NaCl to make a 3x staining solution. (e.g. add 15  $\mu$ L NeoDye DNA Orange 10000x stock reagent and 5 ml of 1 M NaCl into 45 ml H<sub>2</sub>O).
2. Place the gel carefully into an appropriate container, such as a polypropylene container.
3. Add a sufficient amount of the 3x staining solution slowly to submerge the gel.
4. Shake gently at room temperature for about 30 minutes for staining. The optimal staining time may vary depending on gel thickness and agarose concentration. For 3.5%-10% polyacrylamide gel, the staining time is usually in the range of 30 minutes to 1 hour and extends with the increase of acrylamide concentration.

#### Note:

- More dye will be used by post-staining protocol. The 3x NeoDye DNA Orange staining solution can be used for about 3 times repeatedly.
- The 3x NeoDye DNA Orange staining solution can be prepared in large quantities and should be stored at room temperature away from light until it is used up.

**FOR RESEARCH USE ONLY**