



# Product Information FluoroStain Protein Fluorescent Staining Dye

PS2000 5 ml x 2 (Gold, 100X)

### Storage Protected from light -20°C ≥ 24 months

# Working Reagent Preparation

1:100 dilution in filtered distilled or deionized water, apply on fixated SDS-PAGE gel

#### Description

The ExcelStain Protein Fluorescent Staining Dye (Gold, 100X) is designed to substitute the Coomassie Blue protein staining method, offering greater sensitivity and ease of operation in visualizing protein separation in 1-D or 2-D SDS-PAGE analysis. The maximum emission wavelength of protein-bound ExcelStain Protein Fluorescent Staining Dye (Gold, 100x) is near 570 nm when excited by UV illumination, exhibiting high sensitivity (Fig. 1 and 3).



Fig.1

### Contents

ExcelStain Protein Fluorescent Staining Dye (Gold, 100X) is stored at 100X concentration.

### Cautions

Dispose of the stain in compliance with local regulations.

Gloves and goggles should be worn and general laboratory safety precautions be followed while handling both the undiluted and diluted products.

This product contains slightly flammable solvent (propylene glycol); keep away from heat source.

### **Experimental Protocols**

## (1) Fixation:

Prepare Fix solution: 40% (v/v) ethanol\* and 7% (v/v) acetic acid. Apply  $10^{-15}$  times the gel volume of Fix solution to the gel (Table 1) and gently agitate the gel in the fix solution for 60 minutes at room temperature\*\*.

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Gel dimension	Gel volume	Fix solution
(1mm thick)		
9 cm × 7 cm	≈ 6.5 ml	≈ 100 ml
13 cm × 9 cm	≈ 12 ml	≈ 180 ml
16 cm × 16 cm	≈ 26 ml	≈ 390 ml
26 cm × 23 cm	≈ 60 ml	≈ 900 ml

\*40% ethanol can be replaced with 50% methanol; however it's not recommended for safety and performance concerns.

\*\*Quick fixation can be done within 15 minutes agitation by microwave the Fix solution with gel to boiling (1~5 minutes for 100ml Fix solution).

Caution: Handle the boiling buffer with care. Avoid inhaling and contact with hot vapour. Proper protective wares are recommended.

### (2) Preparation for 1X Stain solution:

Dilute ExcelStain Protein Fluorescent Staining Dye (Gold, 100X) in 100X (v/v) in filtered distilled or deionized water 10 times the gel volume (Table 2).

Table 2

Gel Size	Volume of Stain	
(1mm thick)	Solution per Gel	
~ 9 cm × 7 cm	~ 60 ml	
~ 13 cm × 9 cm	~ 120 ml	
~ 16 cm × 16 cm	~ 300 ml	
~ 26 cm × 23 cm	~ 600 ml	

# (3) Staining:

Immerse gel into 1X Stain solution; gently agitate at room temperature for 45 minutes. (For protein >5 ng, the gel can be rapidly stained and visualised after staining for 15 minutes.) Stained gel can be stored in Stain solution at 2~8°C.

# (4) Destaining:

Destain is not necessary. However, it might be helpful to remove excess dye from the gel surface by quickly rinsing the gel with clean water.

#### Note:

Apply clean technique during the entire staining process; dust and contaminant such as keratins will result in smudges and speckles in the fluorescence image. Protect from light during staining and post-staining storage.

# Dynamic range of ExcelStain Protein Fluorescent Staining Dye (Gold, 100X)

ExcelStain Protein Fluorescent Staining Dye (Gold, 100X) exhibits wide dynamic range (Fig.2), optimal for  $1^{2}$  µg of individual purified protein or  $10^{20}$  µg of complex mixture on 1-D electrophoresis.



# Sensitivity

The ExcelStain Protein Fluorescent Staining Dye (Gold, 100X) is highly sensitive under UV-illumination, compatible with conventional UV DNA/RNA imaging, fluorescence imaging system. The ExcelStain Protein Fluorescent Staining Dye (Gold, 100X) is capable of detecting protein band down to ~1ng in the serially diluted unstained standard (Fig. 3).



Fig. 3

# **Trouble Shooting**

### 1. Poor staining sensitivity

- a. Follow the recommendations for fixing. Avoid over destaining in water for more than 5 minutes. For long term storage, keep the gel protected from light in stain solution at 4°C.
- b. Extend staining time to more than 45 minutes.
- c. Make sure the gel/Staining solution ratio is properly followed.
- Reusing ExcelStain Protein Fluorescent Staining Dye (Gold, 100X) is not recommended

#### 2. High staining background

- Quick rinse the gel with filtered distilled or deionised water to wash off excessive stain before taking picture.
- b. Reduce staining time or apply destaining step.

### 3. Speckles in the gel image

- Use dust-free gloves and forceps to handle gels and only by the gel's edges.
- b. Limit exposure of gels and staining solution to open air.
- c. Apply clean technique; make sure the staining trays and other equipment used are properly cleaned.

#### 4. Uneven staining

- a. Make sure the gel is well agitated and immersed during staining.
- b. Make sure the imaging system functions properly without uneven illumination and the lens is clear.
- 5. No detectable signals for bands or spots
  - Check the instrument for errors; make sure the gel is being illuminated with the correct excitation wavelength.
  - b. Verify the sample with Coomassie blue or silver staining.

# **Quality Control**

### Product Description:

The Protein Fluorescent Staining Dye enables protein analysis in SDS-PAGE gel.

### Functional Testing:

Each component is functionally tested by performing SDS-PAGE gel staining, A protein band for 5 ng carbonic anhydrase must been visible under a UV 320 nm light source, using standard staining method.

## **Other Information**

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Caution:

Not intended for human or animal diagnostic or therapeutic uses