

CooBlue FX Protein Gel Stain

Product Description

CooBlue FX Stain for Proteins in Gel	
1X Ready-to-use Colloidal Coomassie Stain for Proteins in electrophoresis Gels	
Optimized for analytical applications	
31X Concentrate: 500mL Concentrated CooBlue FX and 387.5 ml Enhancing Buffer (# CE2080)	
UPG4562A, 500mL (for 20minigels)	
UPG4562B, 4.5L (for 225minigels)	

Storage

+4°C, stable for long term for +1 year. Shipped at RT. DO NOT FREEZE. Can be stored at Room Temperature.

Features

Hands-Off! Stain and read directly:

Ready-to-use stain reagent No fixation required (no gel shrinkage) Fast, One hour staining (12 minutes using microwave protocol) Bands can be viewed directly in the staining tray (wash with water for clear background)

- High Sensitivity, below 10 ng of Protein per Band
- Safe: no methanol or acetic acid

Applications

2D, SDS-PAGE, Native PAGE, Peptide analysis (Tricine gels with fixation step): -protein staining for analytical purposes / proteomics -protein staining to control the efficiency of protein transfer / blotting

Introduction

CooBlue FX Stain is a convenient, flexible, safer and superior alternative to traditional Coomassie Blue staining procedures.

- The simple "hands-off" staining/destaining procedure saves valuable time.
- It does not contain nor need hazardous chemicals such as methanol, acetic acid and phosphoric acid as required by conventional Coomassie stains.
 0.11.5 5 10 ^{µg}
- After electrophoresis, no prior fixation of the gel is required.
- Based on a colloidal formulation, **staining is rapid**, within 60 min, with no risk of overstaining in cases of overnight staining, and down 12min using the microwave protocol!
- The **proteins can be visualized directly**, before destaining. Washing with water is however recommended to get a clear gel with intense blue stained proteins bands, hence enhancing sensitivity and getting nicer pictures.
- CooBlue Staining exhibits **high sensitivity**, below 10 ng of protein per band (with scanning) making it ideal for most applications including SDS-PAGE, IEF and 2D gels.







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Directions for Use

Staining Procedure for SDS-PAGE gels:

Before starting: allow the CooBlue FX protein staining solution to reach RT under continuous agitation. Note: This protocol is optimised for 8 x 10cm² minigels 10% SDS-PAGE (1mm thick).

• 1. Prepare the gel: Remove stacking gel from the separating gel.

• 2. Wash the gel with 300 ml of ultra-pure water with gentle shaking.

- Notes: SDS interferes with the staining procedure; so we recommend for optimal results,
- to use 300 ml of ultra-pure water 3 x 5-10 minutes per minigels.
- to wash longer if the thickness of gel (>2mm) or acrylamide percentage ratio (>15%) are important (see the timetable below)
- for native gels (no SDS), wash can be avoided, or a simple 1-5min pre-wash is usually sufficient.
- 3. Stain for 1 hour with CooBlue Stain: Submerge the gel completely with CooBlue FX reagent. Gently shake tray.

Notes: The CooBlue stain should be well temperature-equilibrated / mixed before use for optimal performance. Close the 500ml vial immediately after use. It can be store at room temperature when used daily or weekly.

The reagent can be conveniently dispensed using appropriate pump #T34711 / CooBlue in 4.5L container.

Use 20ml of CooBlue FX per minigel 8x10cm² (more may be needed depending on the tray), and gently shake.

Stain intensity reaches a maximum within approximately 1 hour. Gels may be stained overnight without increased background.

• 4. Read the gel: Quick wash the gel with ultrapure water for immediate reading. Note : The Gel can be read also after just discarding the stain buffer, but a quick wash is easy to do and better.

(optional) Destain for 3x10 min in deionized water for optimal reading (clears the gel and enhances stain sensitivity)

Notes: Ultra fast washing can be achieved using 30% methanol and 10% acetic acid solution -but impeach protein recovery-.

If proteins destaining is required for applications such as other detections, extraction of proteins, use CooBlue UP47255A

• 5. (optional) The stained gel can be dried using Crack Free Solution #U50450 (for permanent records, autoradiography,...)

Timetable for optimizing the removal of SDS from gels

Gel %	Time (with shaking)
Protein gel <14%	3 x 10 minutes in 300mL water
Protein gel 14%-16%	3 x 30 minutes in 300mL water
Protein gel >16%	3 x 60 minutes in 300mL water

Quick Microwave Procedure

- This quick procedure (only10-15min) uses microwaves to speed the staining, while preserving sensitivity (10 ng BSA). It is optimized for 1.0mm mini-gels. (For 1.5 mm mini-gels, use the values in italic)

Caution: Use caution using the stain in a microwave oven. Do not overheat the staining solutions.

1. After electrophoresis, place the gel in 100 ml of ultra pure water in a loosely covered container and **microwave** on high power (950 to 1100 watts) for 1 minute (2 minutes) until the solution almost boils.

2. Discard the water.

3. Repeat Steps 1 and 2 two more times.

4. After the last wash, add 20 ml (30 ml) of CooBlue FX and microwave on high power for 45 seconds to 1 minute (1.5 minutes) until the solution almost boils.

5. Shake the gel on an orbital shaker for 5 minutes (10 minutes). Note: Stain intensity reaches a maximum within approximately 1 hour. Gels may be stained overnight without increased background.

6. (optional / can enhances staining sensitivity and increase staining over background.) Wash the gel in 100 ml of ultra pure water for 10 minutes on a shaker.

Troubleshooting

Problem	Cause	Solution
No band development	Gel is > 1 mm thick	Longer staining incubation
	Gel has high polyacrylamide concentration	Longer staining incubation
	SDS interference	Wash the gel with plenty of water before staining
Bands are faint	Lack of sensitivity / protein load	Check the gel has been washed properly before staining. Increase protein load
Undesired bands	Protein contamination	Check gel / buffers preparation
High background	Protein contamination	Check gel / buffers preparation
Staining not homogenous	Bubbles, uncovered edges	Check for incubation conditions, increase washing and staining volumes

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Related products and documents

* See all CooBlue products, including the

CooBlue Protein Gel stain <u>#UP47255A</u>: the original version allowing the stain non fixed gels for protein recovery after electroelution (i.e. with preparative gels)

CooBlue Blot-Membrane Protein Gel stain #20078A:

- Pump dispensor **#T34711**. This pump fits to the opening of the PP blue container of CooBlue 4.5L and delivers per push 10ml doses of reagent. It can be provided for free for your first order (please ask when ordering), or can be purchased separately.

- * Protein Electrophoresis in Agarose Gels NT-47255g
- Crack free solution #U50450: to dry your gels without hassle of cracking.
- ProSave 5min Protein gel stain #BP7121: combines high sensitivity & max. flexibility (analytical, compatible with MS, elution, WB...)
- LavaPurple Gel & Blot protein stain #67433A: the highest sensitive protein gel stain (50ng protein), by fluorescence (MS compatible).
- High quality reagents for electrophoresis (acrylamides, buffers...). See the catalog.

* Other Coomassie based reagents. I.e. <u>CooAssay protein dosage kit</u>: use the same Coomassie to quantitate proteins in solutions. See BioSciences Innovations catalogue and e-search tool.

Ordering information

Catalog size quantities and prices may be found at http://www.interchim.com.

For any information, please ask: Uptima / Interchim Hotline: +33 4 70 03 73 06

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