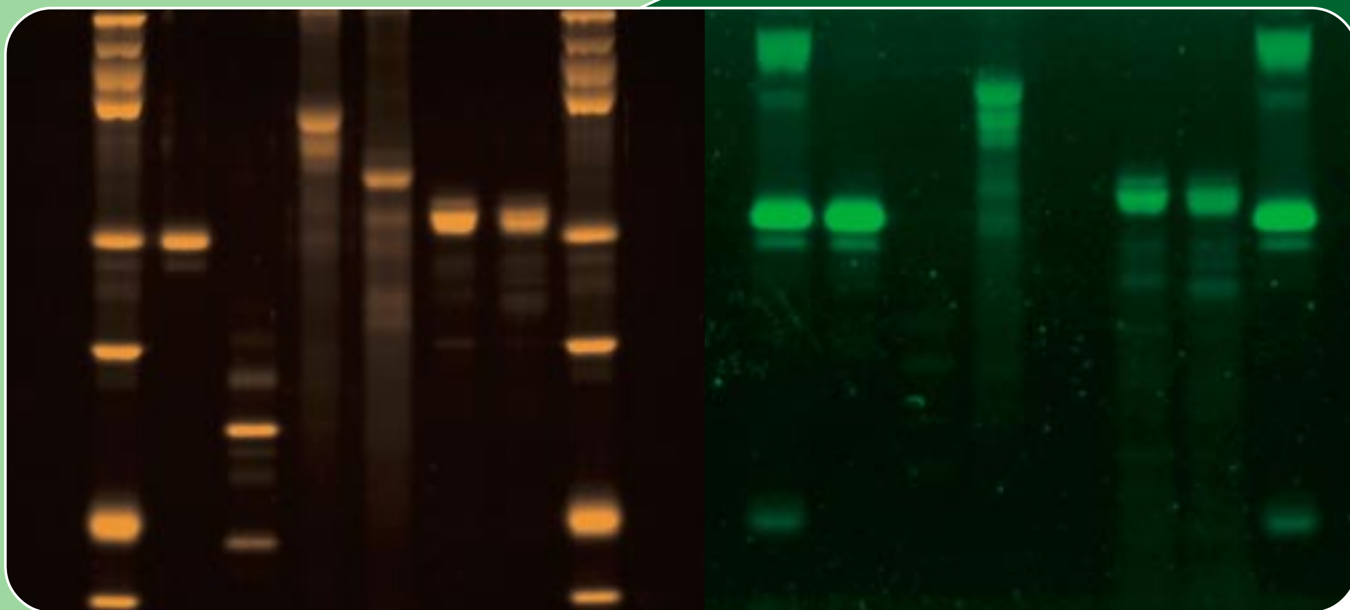
Pro-Q Emerald Glycoprotein Stain Kits

The most advanced technology for staining glycoproteins in gels or on blots



FAST	Staining is complete in less than three hours
SIMPLE	The procedure includes only three steps — fixation, oxidation and staining
SENSITIVE	More sensitive than any other nonradioactive glycoprotein staining technique
EASY TO VISUALIZE	The stain can be visualized using standard UV illumination or a laser scanner
MULTICOLOR CAPABILITIES	Compatible with SYPRO Ruby protein gel stain for dichromatic staining

Molecular Probes®

Technical Information

Molecular Probes' proprietary Pro-Q Emerald 300 and Pro-Q Emerald 488 Glycoprotein Stain Kits provide the most advanced technology available for detection of glycoproteins in gels and on blots.[1] Gel staining is rapid and very sensitive; in less than three hours, it is possible to detect as little as 300 pg of glycoprotein per band, depending on the degree of glycosylation, making these stains at least 50-fold more sensitive than standard fuchsin staining. The staining procedure is very simple, requiring just three steps — fixation, periodate oxidation of carbohydrate groups, and incubation with the Pro-Q Emerald reagent (Figure 1). Blot staining requires extra steps, but also provides excellent sensitivity (2–18 ng of glycoprotein per band) as well as an opportunity to combine glycoprotein staining with immuno-staining or other blot-based detection techniques. Pro-Q Emerald 300 and Pro-Q Emerald 488 stains can be visualized using laser scanners. Both stains exhibit bright green fluorescence.

Two of the Pro-Q Emerald 300 Glycoprotein Stain Kits include one of our fluorescent SYPRO Ruby protein stains (see *Ordering Information* below). These total-protein stains are compatible with glycoprotein staining, making it easy to compare stained glycoproteins to molecular weight standards or to localize glycoproteins in 2-D gels (Figure 2). The gel stain (in P-21855) provides a control for protease contamination in mobility shift assays (see front figure). The blot stain (in P-21856) is useful for assessing the efficiency of protein transfer to the blot; this is especially important when working with glycoproteins, which often transfer poorly to blotting membranes. Stained proteins can be visualized using either UV illumination or a laser scanner.

Each kit also includes our CandyCane molecular weight standards, which separate into alternating bands of glycosylated and nonglycosylated proteins (Figure 3).

Materials Supplied

- Pro-Q Emerald reagent
- Pro-Q Emerald staining buffer
- Oxidizing reagent
- SYPRO Ruby protein gel stain (in Kit P-21855 only)
- SYPRO Ruby protein blot stain (in Kit P-21856 only)
- CandyCane glycoprotein molecular weight standards
- A detailed protocol

Each kit provides sufficient materials to stain approximately ten 8 cm x 10 cm gels or blots.

Ordering Information

P-21855 Pro-Q™ Emerald 300 Glycoprotein Gel Stain Kit (with SYPRO® Ruby protein gel stain)
P-21856 Pro-Q™ Emerald 300 Glycoprotein Blot Stain Kit (with SYPRO® Ruby protein blot stain)
P-21857 Pro-Q™ Emerald 300 Glycoprotein Gel and Blot Stain Kit
P-21875 Pro-Q™ Emerald 488 Glycoprotein Gel and Blot Stain Kit

References

1. Steinberg, T.H. et al., Proteomics 1, 841 (2001).

These products are offered for research purposes only and are not available for resale or other commercial uses without a specific agreement from Molecular Probes, Inc.

For further information

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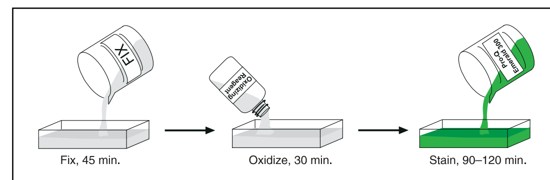

Figure 1. *Detecting glycoproteins with Pro-Q Emerald glycoprotein detection reagent is simple. After fixation, carbohydrates are oxidized and then the gel is incubated with the Pro-Q Emerald reagent.*

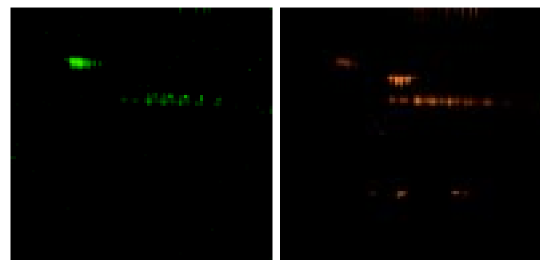

Figure 2. *2-D gel stained with SYPRO Ruby protein gel stain and Pro-Q Emerald reagent. Combined Cohn fractions II and III from cow plasma, containing primarily β- and γ-globulins, were run on a 2-D gel and stained first with Pro-Q Emerald 300 reagent (left) and then with SYPRO Ruby protein gel stain (right).*

Figure 3. *CandyCane glycoprotein molecular weight standards with four glycosylated and four non-glycosylated proteins. The standards were electrophoresed through two identical 13% polyacrylamide gels. Both lanes contain ~0.5 µg of protein in each band. The left lane is stained with Pro-Q Emerald 300 reagent. The right lane is stained with SYPRO Ruby protein gel stain (available in kit P-21855) to detect all eight marker proteins.*

Front page photo caption: *Mobility-shift gel assay using deglycosylating enzymes. Untreated glycoproteins α1-acidic glycoprotein, fetuin and horseradish peroxidase (HRP) (lanes 2, 4 and 6, respectively) and the same glycoproteins digested with glycosidases (lanes 3, 5 and 7, respectively) were loaded onto two identical SDS-polyacrylamide gels. The CandyCane molecular weight standards were loaded into lanes 1 and 8. After electrophoresis, the gels were stained with either SYPRO Ruby protein gel stain (left gel) or Pro-Q Emerald 300 reagent (right gel). Glycosidase treatment resulted in a mobility shift and loss of staining with green-fluorescent Pro-Q Emerald 300 reagent for α1-acidic glycoprotein and fetuin, indicating that carbohydrate groups had been cleaved. HRP, which contains an α-(1,3)-fucosylated asparagine GlcNac-linkage that is resistant to many glycosidases, showed neither a mobility shift nor a loss of staining with green-fluorescent Pro-Q Emerald 300 reagent. Thus, use of the Pro-Q Emerald 300 reagent together with SYPRO Ruby protein gel stain unequivocally identifies which glycoproteins are susceptible to the glycosidases used in the assay, providing important information about the glycoprotein carbohydrate moieties.*

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