

Product Information

ProteoProfile™ PTM MARKER

Post Translational Modification Marker

Product Number **P 1745**

Storage Temperature 2-8 °C

Product Description

Glycosylation and phosphorylation are the protein post-translational modifications (PTM) most frequently encountered in proteomic analysis. The ProteoProfile™ PTM Marker contains glycosylated and phosphorylated proteins and is designed as both a positive and negative control for SDS-PAGE gels and Western blots of proteins with post-translation modifications.

Component

The ProteoProfile PTM Marker is supplied as a solution of four proteins, each at 1 mg/ml, in 250 mM Tris buffer, pH 7, with 25% glycerol. Table 1 shows the phosphate and carbohydrate content of 1 µl of the marker. Properties of the four proteins contained in the marker are shown in Table 2.

Table 1.

Carbohydrate and phosphate content of 1 µl of the ProteoProfile PTM Marker

Protein	Carbohydrate (ng)	Phosphate (pmole)
Albumin	none	none
Ovalbumin	35	45
β-Casein	none	160
RNase B	200	none

Precautions and Disclaimer

This product is for laboratory research use only, not for drug, household, or other use. Consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Storage/Stability

It is recommended to store the product at 2-8 °C. The product as supplied is stable for one year when stored properly. After dilution with sample loading buffer, the marker should be used within 24 hours.

Preparation Instructions

The marker must be diluted with an appropriate sample loading buffer, such as Laemmli 2x Sample Buffer (Product Code S 3401), prior to SDS-PAGE. The dilution and loading volume of the marker is determined by the dynamic range of the detection method and the marker concentration should be similar to that of the sample.

Note: After the marker has been diluted with Laemmli 2x Sample Buffer (Product Code S 3401), incubate the solution in a boiling water bath for 3-5 minutes. For other loading buffers, incubate as appropriate for the loading buffer used.

For fluorescent detection of glycoproteins (detection limit of 5–25 ng of carbohydrate) on SDS-PAGE gels or Western blots (ProteoProfile Fluorescent Glycoprotein Detection Kit, Product code PP0300), 3-5 µl of a 6-fold dilution of the marker should be loaded.

For glycoprotein detection with the Periodic Acid/Schiff reagent (PAS, detection range of 25–100 ng of carbohydrate) on SDS-PAGE gels or Western blots (Glycoprotein Detection Kit, Product Code GlycoPro), load 10 µl of a 2-fold dilution.

For phosphoprotein detection using Methyl Green, 10 µl of a 2-fold dilution may be required. For detection with anti-phosphoserine antibodies, the dilution should be adjusted to accommodate the expected sensitivity of the method.

Note: This product is not suitable as a marker for detection with anti-phosphothreonine or anti-phosphotyrosine antibodies. The only phosphorylation sites on the marker proteins occur on serine residues.

Table 2.

Properties of Proteins in the ProteoProfile PTM Marker

Protein	MW (kDa)	Glycosylation	Phosphorylation
Albumin, bovine serum	66	none	none
Ovalbumin, chicken egg	45	1 N-linked glycan 3.2% carbohydrate ¹	2 phosphorylation sites (serine)* 0.3-0.4% phosphate by weight ¹
β-Casein, bovine milk	30	none	5 phosphorylation sites ² (serine)* 1.3-1.7% phosphate by weight
RNase B, bovine pancreas	17	2 N-linked glycans 19.5% carbohydrate ³	none

*Potential phosphorylation sites. Not all sites will be phosphorylated.

References

1. [http://www.food-allergens.de/symposium-vol1\(1\)/data/egg-white/gald2.htm](http://www.food-allergens.de/symposium-vol1(1)/data/egg-white/gald2.htm)
2. Posewitz, M., and Tempst, P., *Anal. Chem.*, **71**, 2883-2892 (1999).
3. Plummer, T., and Hirs, C., *J. Biol. Chem.*, **238**, 1396 (1963).

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