



Instructions for use

Roti[®]-Mark PRESTAINED

Prestained protein molecular weight marker for SDS-PAGE

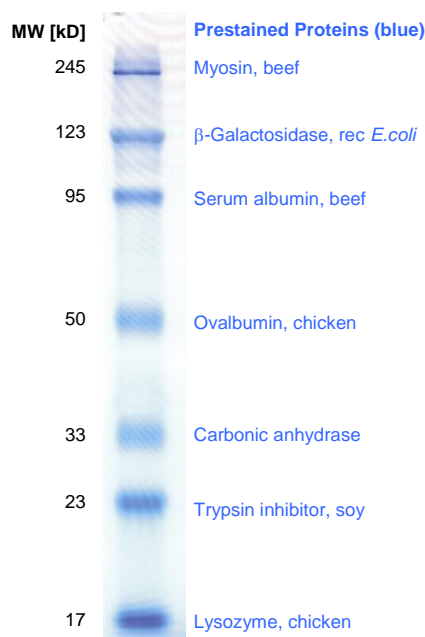


Figure:
Roti[®]-Mark PRESTAINED in a 12 % PAGE (Tris), unstained. Resolution and distinctness of bands may vary depending on the gel and buffer system used.

I. Introduction

Roti[®]-Mark PRESTAINED is composed of seven native proteins which are covalently coupled with a blue dye. Roti[®]-Mark PRESTAINED need not be stained in gel and is therefore ideal for checking transfer efficiency during Western-Blot and the gel run during electrophoresis.

The concentration of the individual proteins is set in such a way that homogeneous band intensity and optimal band sharpness can be obtained in unstained gel.

The proteins are denatured in Lämmli buffer with 0.01 % bromophenol blue and 400 mg/ml (6.67 M) urea.

Due to optimised prestaining parameters, the apparent MW of the following proteins has slightly shifted up: serum albumin, ovalbumin and carbonic anhydrase.

Table 1:

Protein	MW (kD)
Myosin, beef	245
β-Galactosidase, rec. <i>E.coli</i>	123
Serum albumin, beef*	95
Ovalbumin, chicken*	50
Carbonic anhydrase	33
Trypsin inhibitor, soy	23
Lysozyme, chicken	17

* Due to the natural origin of the proteins, a small amount of glycosylated molecules may be found.

The molecular weights of the proteins change as a result of being coupled with the dye and vary according to the coupling efficiency. Additionally, the behaviour of the prestained proteins in the gel differs slightly depending on the actual gel and buffer system used. The indicated molecular weight values are therefore only approximate. The effective molecular weights may deviate from the values indicated in Table 1. (Depending on the coupling efficiency, ovalbumin may show a second thin band at a higher molecular weight).

Please note: Roti[®]-Mark PRESTAINED, therefore, is not suitable for size determination of proteins in the gel.

II. Storage

- The marker should not be stored for a longer period at temperatures above freezing point!
- The marker will not be shipped cooled or with dry ice. This does not affect usability.
- Please store Roti[®]-Mark PRESTAINED at -20 °C. The marker can be stored at 4 °C for a short period (a few days). To avoid frequent freezing and thawing, aliquots should be frozen.
- Please heat Roti[®]-Mark PRESTAINED for 30-60 sec at 70 °C before use. Aggregate bodies may form when used for a longer period or when stored at 4 °C. These can be dissolved if heated for a short period (5 min at 80 °C).

III. Gel loading

- Recommended loading amount for mini gels: (10 %; 0.75 mm thick): approx. 5 µl
Silver-staining: approx. 1 µl
- **Important:** Loading amount required varies depending on gel thickness, C/T ratio and width of comb tooth.

IV. Trouble Shooting

Marker bands cannot/can only be seen very weakly.

- Please ensure the correct loading amount. The recommended quantity is valid for mini gels with a thickness of 0.75 mm. If thicker or larger gels are used, the loading amount must be increased.
- Few weak marker bands: Under certain conditions marker proteins may agglutinate. Resolubilise marker aliquots by incubating for 5 min at 80 °C (alt. 1 min at 95 °C). Mix carefully.

Marker bands are fuzzy.

- Avoid overloading the gel!
- Please ensure that the marker is not stored at room temperature for a longer period.
- Avoid frequent freezing/thawing of marker.
- Long-term storage should always take place at -20 °C.
- Please take care that the gel contains no air bubbles when casting.
- When casting the gel, please ensure that the gel solution is mixed thoroughly.
- Only use high quality acrylamide solutions, e.g. Rotiphorese[®] Gel 30 (Art. No. 3029) or Gel 40 (Art. No. 3030).
- Avoid overheating the gel. Reduce voltage if required.
- Check the composition and pH value of the buffer used.

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Warning H319 P305+P351+P338-P337+P313

Roti[®]-Mark PRESTAINED

T852.3	100 µl
T852.1	250 µl
T852.2	4 x 250 µl