EUROMEDEX

CERTIFICATE OF ANALYSIS

Prestained Protein Ladder Plus

#06P-0211 2x 250 µl

(for 100 mini-gel applications 5µl per well or 50 large gel applications 10µl per well)

Lot Exp. date:

Store at -20°C (or at +4°C for 3 months)

Description

Prestained Protein Ladder Plus is a mixture of prestained 9 recombinant proteins covering a wide range molecular weights from 10 kDa to 250 kDa. The reference bands at ~70 kDa ladder contains two orange and 25 kDa and one green reference band of 10 kDa.

Lot-to-lot variation of the apparent molecular weight of prestained proteins is ~5%.

The ladder is supplied in gel loading buffer and is ready-to-use: no heating, further dilution or addition of a reducing agent is required.

Applications

- Monitoring of protein separation during SDS-PAGE (1-3).
- Verifying Western transfer efficiency (1-3).
- Approximate sizing of proteins on SDS-polyacrylamide gels and Western blots.
- Locating a protein of interest for excision from an unstained preparative gel.

Storage Buffer

62.5mM Tris- H_3PO_4 (pH 7.5 at 25°C), 1 mM EDTA, 2% (w/v) SDS, 10 mM DTT, 1 mM NaN₃ and 33% (v/v) glycerol.

Recommendations for Loading

- 1- Thaw the ladder either at room temperature or at 37-40°C for a few minutes to dissolve precipitated solids. **Do not boil!**
- 2- Mix gently, but thoroughly, to ensure that the solution is homogeneous.
- 3- Load the following volumes of the ladder on an SDS polyacrylamide gel:
- 5µl per well for mini-gels,
- 10µl per well for large gels.

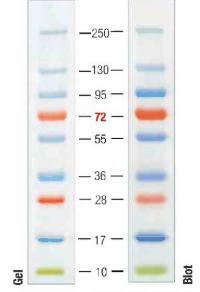
Use the same volumes for Western blotting.

The loading volumes listed above are recommended for gels with a thickness of 0.75-1.0 mm. The loading volume should be doubled for 1.5 mm thick gels.

Note

- Prestained proteins can have different mobilities in various SDS-PAGE-buffer systems. However, they are suitable for approximate molecular weight determination when calibrated against unstained standards in the same system.
- In 8 or 10% gels low molecular weight proteins may migrate with the dye front.
- Longer transfer times or higher transfer voltages may be required for Western blotting of large (>100 kDa) proteins.

Representative lot of Prestained Protein Ladder Plus, apparent MW, kDa



4-20% Tris-glycine SDS-PAGE

Quality control

5µI of Prestained Protein Ladder Plus provide 9 bands of equal intensities in SDS-PAGE (Tris-glycine buffer) and after Western blotting onto a PVDF membrane.

References

- 1. Laemmli, U.K., Cleavage of structural proteins during the assembly of the head of bacteriophage T4, Nature, 227, 680-685, 1970.
- 2. Burnette, W.N., "Western blotting": electrophoretic transfer of proteins from sodium dodecyl sulfate polyacrylamide gels to unmodified nitrocellulose and radiographic detection with antibody and radio iodinated protein A, Anal. Biochem., 112 (2), 195-203, 1981.
- 3. Towbin, H., et al., Electrophoretic transfer of proteins from polyacrylamide gels to nitrocellulose sheets: procedure and some applications, Proc. Natl. Acad. Sci. USA, 76, 4350-4354, 1979.
- 4. Kurien, B.T. and Scofield, R.H., Protein blotting: a review, J. Imm. Meth., 274, 1-15, 2003.

This product is manufactured under the license for **Streptag®** technology covered by US patent 5,506,121, UK patent 2272698 and French patent 93 13 066.

Migration Patterns of Prestained Protein Ladder Plus

Gel type Gel concentration Running buffer		Tris-Glycine													Tris-Acetate*				Bis-Tris*												
		4-20	4-20% 8-16% 10-20%			89	8% 10%		12	2%	% 15%		3-8	3%	7	%	4-1:		2%		10%			12%							
			Tris-Glycine														Tris-Acetate			МО	PS	MES		MOPS		ME	ES	МС)PS	MES	
			Apparent Molecular Weights, kDa																												
% lenght of gel	10						250			_	250	=	250	Ξ	250 130												100		105		100
	20	_	250	-	250	=	130		250	_	130	_	100 70		70 55	_	205	-	205	_	185	_	190 115	_	185 115	=	115 80	_	115	=	115
	30		130 100	-	130	_	70		100	_	70	_	55		00		200	_	120	_	115 สก	=	80 70	_	80 65	-	70	_	65	_	80 70
	40	_	70	_	100 70	-	55	_	70	-	55	_	35	-	35	_	120	_	85	_	65		50	_	50	-	50	-	50	_	50
	50	-	55	=	55	_	35	_	55		25	_	25	_	25	_	85	_	65		50	_	50			_	30			_	30 25
	60	_	35			_	25									_	65												30		
	70	-	25	-	35			_		_	25	_	15	_	15	_	50	_	50	_	30	_	25					_	25	_	15
	80	_	15	-	25	_	15	_	25											-	25	_	15	-	25	_	15				
	90	_		_						_	15	_	10	-	10	_	30	_	30	_	15		. 5					_	15	_	10
	100				10	_	10	_	15							_	25		25	_	10	_	10	_	15	_	10				

^{*} migration patterns were determined using respective NuPAGE® precast gels.

Product Use Limitation

This product is developed, designed and sold exclusively for research purposes and in vitro use only. The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals.

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