Instruction Manual

SERVA Proteome Markers, 5 Vials

Cat.No. 39220.01

The SERVA Proteome Marker is developed in cooperation with the German Society for Proteomic Research. The standard consists of 8 different proteins of a pI range from 5.5 to 9.8 and a molecular weight range from 11.7 to 77.0 kDa. The also enclosed Glucose-1-Dehydrogenase has a molecular weight of 113.000 Da, but will dissociate in presence of urea (8M) respectively SDS into four subunits with an apparent molecular weight of 28 kDa each. The proteins are present in equal weight proportions (each ca. 13 µg/protein).

Deutsche Gesellschaft für Proteomforschung e.V.

Application:

The single proteins are characterized as well by 2D gel electrophoresis as by LC/MS analysis. The identity of each single protein was verified by additional amino acid sequence analysis. The marker proteins are especially suitable for the calibration of 2D gel systems and as pI and molecular size marker in 2D gels. (LC-MS-Marker would be the digested standard).

SERVA Proteome Marker contains following marker proteins:

Protein	Source	pI (8M Urea)*	M_r (SDS PAGE)*
Cytochrome C	Equine heart	9.8	12.400
Myoglobin	Equine muscle	7.3, 7.5, 7.8	17.800
ß-Lactoglobulin	Bovine milk	5.5	18.400
Glucose-1-Dehydrogenase	Bac. megaterium	5.4	28.250 (4 subunits)
Lipase	Burkholderia plantarii..	7.2	33.000
Catalase	Bovine liver	7.3	58.000
Albumin	Bovine blood	6.3 – 6.5	67.000
Glucose Oxidase	Aspergillus niger	5.5	77.000

*Measured pI- und MW-values can vary depending from the gel system and experimental execution.

Storage and shelf life:

Store the lyophilized marker proteins at –80 °C. For a short period of time (a few weeks) storage at +2 °C - +8 °C is as well possible without restrictions. The lyophilized marker proteins are at least 3 years stable by storage at –80 °C.

SERVA Electrophoresis GmbH • D-69115 Heidelberg • Carl-Benz-Str. 7
Tel.: +49(0)6221 / 138 40-0 • Fax· +49(0)6221 / 138 40-10 • E-Mail: info@serva.de http://www.serva.de

Handling:

The protein mixture is directly lyophilized in glass vials. Each vial contains ca. 100 µg protein (± 20 %). According to staining method and gel size the amount of one vial is sufficient for 2 – 10 gels on 2D-gel electrophoresis.

Resuspend the lyophilized proteins in H_2O_{bidest} or in rehydration buffer for 2D-PAGE. For solving the proteins brief vortexing and then a brief centrifugation is sufficient.

Staining Method	Silver Staining	Colloidal Coomassie™ Staining
Recommended loading amount	20 µg*	50 µg*

*The data are exemplary for a 18 cm IPG-strip with a pH gradient of pH 3 – 10 and a 13 % SDS PAGE gel of 26 x 20 cm size for the 2. dimension. Chose the amount for other systems accordingly.

Apply the protein solution anodic per Cup-Loading. Alternatively, apply the proteome marker directly to the buffer during rehydration of the IPG strip. After electrophoresis the proteins can be detected by all valid staining methods (e.g. Coomassie™, silver staining, fluorescence staining) and if necessary identified by mass spectrometry.

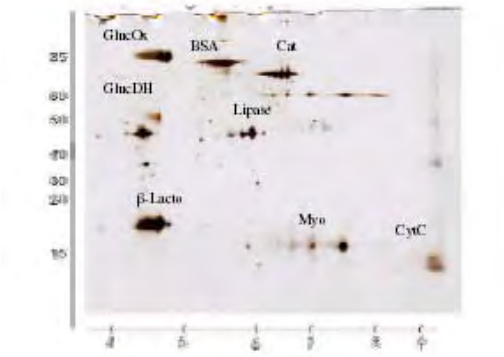

Fig. I

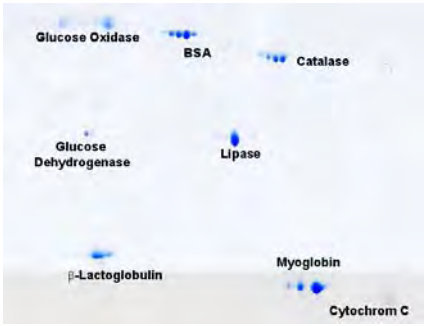

Fig. II

Legend:
2D-gel electrophoresis followed by silver staining modified according to Heukeshoven et al. [Heukeshoven, J., Dernik, R.; Electrophoresis, 6, 103-112 (1985)] (Abb I.) respectively colloidal Coomassie™ staining modified according to Neuhoff et al. [Neuhoff, V., Stamm, R., Eibl, H.; Electrophoresis, 6, 427-448 (1985)] (Abb II) of SERVA Proteome Marker (20 µg protein in Fig. I resp. 50 µg protein in Fig. II, 1. dimension: 18 cm IPG strip pH 3 – 10, non-linear , 2. dimension: 13% SDS PAGE, gel format 26 cm x 20 cm x 1 mm).
The figure is a courtesy of the Medical Proteome-Centre, Bochum.

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