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Instructions for Use

Protein Calibration Standard I

Protein mixture for calibration of matrix-assisted laser desorption and ionization time-of-flight mass spectrometers (MALDI-TOF MS)

Product description

The protein mixture allows calibrations and testing of MALDI-TOF mass spectrometers in a mass range between ~4000 and 20000 Da. The mixture contains four standard proteins. The compounds are listed in the following table together with their molecular weights. Standard proteins are supplied in five tubes per package. The quantity of substance allows 5x250 calibration points.

The standard is for research use only. It is not for use in diagnostic procedures.

Proteins		Average m/z
Insulin	$[M+H]^+$	5734.51
Ubiquitin I	$[M+H]^+$	8565.76
Cytochrom C	$[M+H]^+$	12360.97
Myoglobin	$[M+H]^+$	16952.30
Cytochrom C	$[M+2H]^{2+}$	6180.99
Myoglobin	$[M+2H]^{2+}$	8476.65

Storage and stability

The tube box is shipped at ambient temperatures. We recommend storing the proteins on arrival at less than 0 °C. Dissolved proteins should be aliquoted and frozen. We do not recommend refreezing dissolved samples after thawing.

Note: Do not apply repeated freeze-thaw cycles to all materials.

Risk and safety information

Caution – substances not yet fully tested (EU)
The product does not have to be labeled due to the calculation procedure of the "General Classification guideline for preparations of the EU" in the latest valid version. When used and handled according to specifications, the product does not have any harmful effects according to our experience. All materials may present unknown hazards and should be used with caution. Observe the general safety regulations when handling chemicals.

Beside the kit components, we recommend further chemicals within these Instructions for Use. Please read and observe the respective Material Safety Data Sheet to be provided by your supplier. Observe the general safety regulations when handling chemicals.

Sample preparation procedure (recommendation)

Preliminary remarks
Poor sample preparation will degrade sensitivity, yield low resolution and poor reproducibility. The generation of ions through MALDI depends on the production of a suitable composite material, consisting of the matrix substance and the analyte. For best results use only chemicals of highest available purity.

Chemicals and materials required
- α-Cyano-4-hydroxycinnamic acid (HCCA) (1 g: # 8201344; 5 g: # 8203072)
  or Sinapinic acid (SA) (1 g: # 8201345; 5 g: # 8203073)
- Acetonitrile (ACN)
- 0.1% Trifluoroacetic acid (TFA) in ultra pure water
- TA solvent: mixture of ACN and 0.1% TFA in a volume ratio 1:2

: HCCA is harmful (H: 315, 319, 335; P: 261, 280, 305+351+338, 321, 405, 501), sinapinic acid is irritant (H: 315, 319, 335; P: 261, 280, 305+351+338, 321, 405, 501), acetonitrile is highly flammable and harmful (H 225, 302, 312+332, 319; P: 210, 280, 305+351+338).

Equipment and tools required: centrifuge, shaker, ultrasonic device, pipettes, pipette tips

1. Preparation of sample solution
Dissolve a protein sample (contents of tubes) in 125 µL 0.1% TFA solvent and shake for several seconds.

Alternative for dried droplet preparations only which is not compatible with thin layer or double layer preparations: Dissolve a protein mixture (contents of each tube) in 125 µL TA solvent and vortex for several seconds.

Note:	We highly recommend to aliquot the dissolved protein sample into Eppendorf Safe-Lock microcentrifuge tubes to prevent polymer contaminations.

2. Preparation of matrix solution

Solve HCCA or SA in TA solvent up to saturation at room temperature. Assist the solution process by help of ultrasonic device. Spin down excess matrix in a centrifuge (5 min at 10,000 g) and use only the homogeneous transparent phase.

3. Preparation onto MALDI-TOF-Target

According to the dried droplet method, mix equal volumes of protein sample with matrix solution (HCCA or SA). Apply 1 µL onto a standard steel target and dry at room temperature.

Note: For AnchorChip™ preparation, refer to the AnchorChip Instructions for Use (# 8215344).

Results of measurement of protein calibration standard I

The product is tested on a Bruker Daltonics MALDI-TOF mass spectrometer. Fig. 1 and 2 show typical spectra obtained from the standard on an autoflex® applying different MALDI matrices.

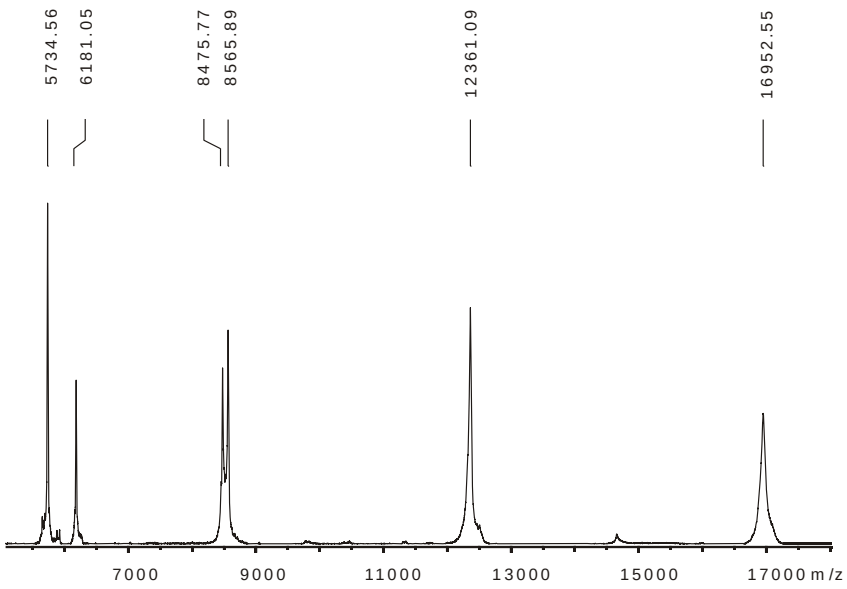

Fig. 1: MALDI mass spectrum of protein calibration standard I. Target preparation with HCCA matrix.

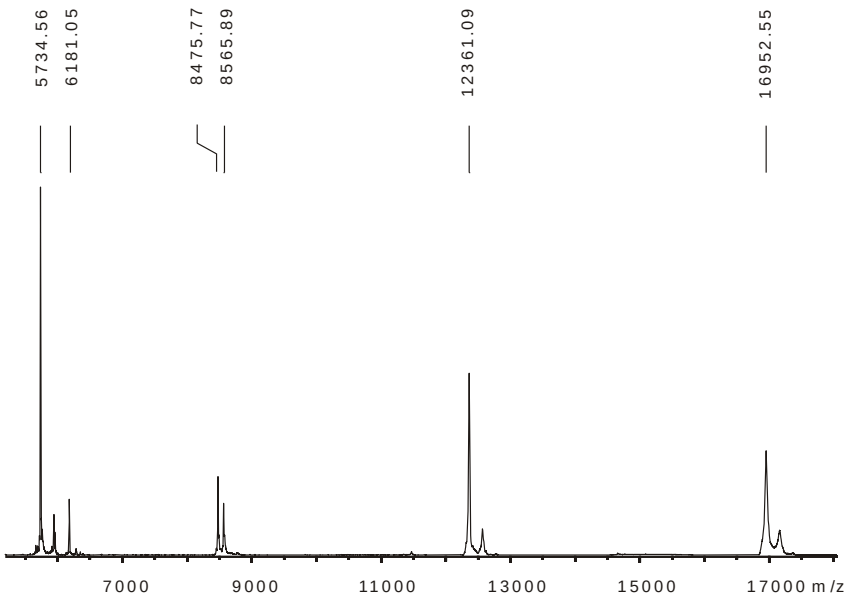

Fig. 2: MALDI mass spectrum of protein calibration standard I. Target preparation with SA matrix.

Ordering Information

Product	Part No.
Peptide Calibration Standard	8206195
Peptide Calibration Standard II	8222570
Protein Calibration Standard I	8206355
Protein Standard II	8207234
α-Cyano-4-hydroxycinnamic acid (HCCA), 1g	8201344
α-Cyano-4-hydroxycinnamic acid (HCCA), 5g	8203072
α-Cyano-4-hydroxycinnamic acid (HCCA), portioned	8255344
Sinapinic Acid, 1g	8201345
Sinapinic Acid, 5g	8203073

Bruker Daltonics' Recommendations	
Pipette tips	Eppendorf standard tips
Vials	Eppendorf Safe-Lock microcentrifuge tubes

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