



## **Operating Instructions**

### 1 Product Description

Use the Sequazyme<sup>™</sup> BSA Test Standard Kit to test instrument function, optimize instrument parameters, and calibrate the mass scale using a known standard. The kit is designed for use in the Sequazyme<sup>™</sup> Peptide Mass Standards Kit with the Voyager<sup>™</sup> Biospectrometry<sup>™</sup> Workstation or with any MALDI-TOF (matrix-assisted laser desorption ionization time-of-flight) mass spectrometry system.

The standard in the kit requires minimal preparation and covers a mass range of 60,000 to 70,000 daltons (Da). The standard was chosen for this kit on the basis of high purity and consistent results when analyzed using MALDI-TOF mass spectrometry.

#### **Applications**

Using the Sequazyme BSA Test Standard Kit, you can monitor results in a mass range from 60,000 to 70,000 Da for mass assignment, resolution, and sensitivity.

#### 2 Materials

#### **Materials Provided**

The Sequazyme BSA Test Standard Kit includes —lyophilized bovine serum albumin, **BSA** (1 vial, 2 mg/vial)

#### **Matrix and Diluent**

- Matrix B:Sinapinic Acid—3,5-Dimethoxy-4-hydroxycinnamic acid, 1 vial (7 to 10 mg/vial)
- Matrix B Diluent—30% acetonitrile in 0.3% TFA, 1 vial (1 ml/vial)

After you use up the sinapinic acid matrix provided in the Sequazyme<sup>™</sup> Peptide Mass Standards Kit, you can use commercially available sinapinic acid to analyze standards.

### 3 Preparing Matrix and Standards

See Section 7, Storing the Kit, for storage and stability conditions of prepared reagents.

## A WARNING CHEMICAL HAZARD. Diluent (with

**acetonitrile)** is a flammable liquid and vapor. It may cause eye, skin, and respiratory tract irritation, central nervous system depression, and heart, liver, and kidney damage. Please read the MSDS, and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

A CAUTION CHEMICAL HAZARD. Sinapinic acid may cause eye, skin, and respiratory tract irritation. Please read the MSDS, and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

#### Reconstitute BSA

Store the BSA Test Standard at  $-20^{\circ}$  C prior to use. Sinapinic Acid Matrix from a Sequazyme kit is required to use this standard.

- 1. Add 0.5 ml HPLC-grade water to the BSA vial.
- 2. Recap and vortex for 1 minute.

You can use the standard immediately.

#### **Sinapinic Acid Matrix**

Use sinapinic acid matrix B from the Sequazyme Peptide Mass Standards Kit.

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- Add the entire contents of the Matrix B Diluent vial to the Matrix B:Sinapinic Acid vial.
- 2. Vortex for 1 minute.
- Warm the solution, if necessary, to completely dissolve the solid.
   Use the supernatant for sample preparation.

#### Standard

Final concentration of the standard after mixing with matrix is 4 pmol/µl.

- 1. Pipette 1 µl of the reconstituted BSA solution into a microcentrifuge tube.
- 2. Add 29 µl of the sinapinic acid matrix.
- 3. Vortex briefly at low speed.

# 4 Loading Matrix:Standard on Sample Plates

Each matrix:standard preparation yields 30  $\mu$ I, enough to load approximately 30 sample positions.

- 1. Pipette 1.0 µl of sample on the MALDI plate.
- Allow the mixture to air dry until all solvent is evaporated, usually less than 5 minutes.

Analyze within one day for best results.

## 5 Analyzing Standards

#### Standard methods on your system

Your Voyager<sup>™</sup> workstation includes standard methods created during factory testing of your instrument. Standard methods contain typical parameters for a given mass or mass range. Standard methods are useful as starting points when you create methods to analyze the standards in the Peptide Mass Standards Kit.

#### Analyzing standards

Analyze the standard according to the needs of your application.

Refer to Section 6, BSA Standard Spectrum and Masses, for representative spectra and mass assignments for standards and matrices in the kit.

## 6 BSA Standard Spectrum and Masses

Figure 1 shows a representative spectrum for the test mixture. Masses are included in the spectra for peak identification only. Use the precise masses listed in Table 1 for calibration.

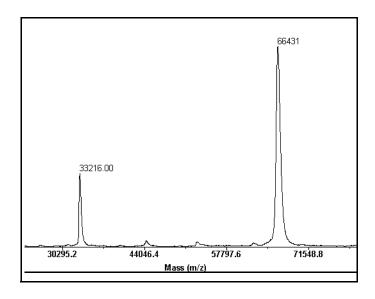


Figure 1 BSA Linear Resolution

**Table 1 Mass Assignments for the Standard** 

Standard	Charge (n)	(M+nH)n <sup>+</sup> Average
BSA (Bovine Serum Albumin)	+1	66,431
	+2	33,216

## 7 Storing the Kit

Store the Sequazyme<sup>™</sup> BSA Test Standard Kit and components of the kit under the following conditions. Avoid prolonged exposure to light.

Kit Component	Storage Temperature	Stability
Unopened kit	–20°C	1 year from date of shipment
Reconstituted standards	–20°C	6 months
Reconstituted matrix	4°C	1 week

## 8 Accessories, Spare Parts, and Ordering Information

Description	Quantity (Kits)	Part Number
Sequazyme <sup>™</sup> Peptide Mass Standards Install Kit includes:	1	4316866
<ul> <li>Sequazyme Peptide Mass Standards Kit</li> <li>BSA Test Standard Kit</li> <li>IgG1 Mass Standard Kit</li> </ul>		
Sequazyme Peptide Mass Standards Kit	1	P2-3143-00
BSA Test Standard Kit	1	2-2158-00
IgG1 Mass Standard Kit	1	GEN602151

## 9 Technical Support

Applied Biosystems is committed to meeting the needs of your research through enabling technologies like the Sequazyme BSA Test Standard Kit. Our dedicated support staff is available to answer questions about using this product to the fullest extent possible.

Applied Biosystems offers a complete line of time-of-flight (TOF) mass spectrometry products to meet your analysis needs. Please contact your Applied Biosystems representative for technical and ordering information.

Applied Biosystems publishes a continuing series of Application Notes and Technical Notes, highlighting ladder sequencing applications and strategies. Please contact Applied Biosystems directly for a publication list

For further details or for answers to questions related to other products contact Applied Biosystems. Refer to the bottom of the following page for the number of the appropriate office.

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