

**Ag85A, Recombinant Protein Reference Standard**

**Catalog No. NR-59869**

This reagent is the tangible property of the U.S. Government.

**Product Description:**

NR-59869 is a recombinant form of the antigen 85 complex A (Ag85A) protein. The recombinant protein consists of the native protein sequence in addition to a hexa-histidine tag. The recombinant protein was expressed in *Escherichia coli* BL21 (DE3) pLysS cells transformed with BEI Resources NR-13292 lot 04.EC.09.03 and purified by immobilized-metal affinity chromatography as part of the production process for NR-49427 lot 70021268 (Manufacturing Date: 21FEB2019). Unused bulk material in 10 mM ammonium bicarbonate remaining from the production of NR-49427 lot 70021268 was frozen. NR-59869 was produced by thawing and initial evaluation of the NR-49427 lot 70021268 bulk material and vialled as lot 70069339 on 13JUN2024.

**Lot: 70069339**

**Manufacturing Date: 21FEB2019**

TEST	SPECIFICATIONS	RESULTS
<b>Appearance</b>	Report results	Clear and colorless
<b>SDS-PAGE Analysis</b>	Protein band of interest represents > 90% of total staining intensity	Dominant band of ~ 35 kDa accounts for 92.6% of total staining intensity (Figure 1)
<b>Concentration by Bicinchoninic Acid Protein Assay<sup>1</sup></b>	Report results	0.96 mg/mL
<b>Final Product</b> Quantity per vial Volume per vial	Report results Report results	192 µg 210 µL
<b>Western Blot Analysis</b> Polyclonal anti-Ag85A <sup>2</sup> Monoclonal anti-histidine tag <sup>3</sup>	Reactive Reactive	Reactive (Figure 2) Reactive (Figure 3)
<b>Endotoxin Content</b> Limulus Amoebocyte Lysate Assay <sup>4</sup>	Report results	< 5.21 EU/mg
<b>Filtration</b>	0.2 µm filter-sterilized	0.2 µm filter-sterilized

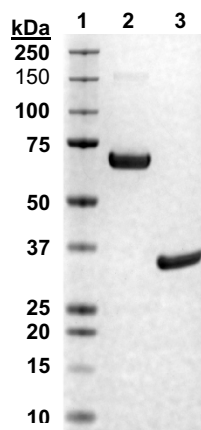
<sup>1</sup>Using bovine serum albumin (BSA) as a standard

<sup>2</sup>Using a 1:3000 dilution of rabbit polyclonal anti-*Mycobacterium tuberculosis* Ag85A (Abcam ab193499) as primary antibody and a 1:3000 dilution of HRP-conjugated anti-rabbit IgG (R&D Systems HAF008) as secondary antibody

<sup>3</sup>Using a 1:3000 dilution of mouse monoclonal anti-His (R&D Systems MAB050) as primary antibody and a 1:3000 dilution of HRP-conjugated anti-mouse IgG (R&D Systems HAF007) as secondary antibody

<sup>4</sup>Charles River Endosafe®-PTS Endotoxin Testing System (Charles River PTS200SF)

**Figure 1: SDS-PAGE Analysis**

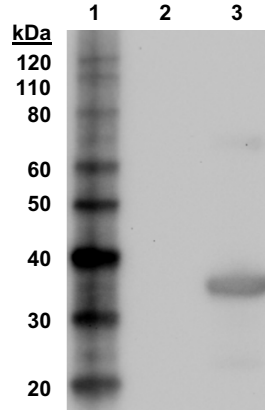


Lane 1: Precision Plus Protein Standard

Lane 2: BSA, 2 µg

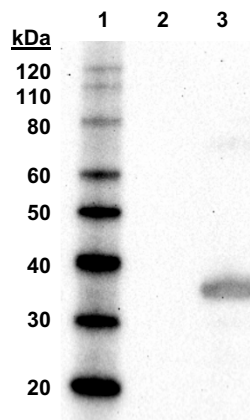
Lane 3: NR-59869, 2 µg

**Figure 2: Western Blot with Polyclonal Anti-Ag85A**



Lane 1: MagicMark™ XP Protein Standard  
 Lane 2: BSA, 1 µg  
 Lane 3: NR-59869, 1 µg

**Figure 3: Western Blot with Monoclonal Anti-Histidine Tag**



Lane 1: MagicMark™ XP Protein Standard  
 Lane 2: BSA, 1 µg  
 Lane 3: NR-59869, 1 µg

/Sonia Bjorum Brower/  
 Sonia Bjorum Brower

Technical Manager or designee, ATCC Federal Solutions

09 MAY 2025

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

ATCC® is a trademark of the American Type Culture Collection.  
 You are authorized to use this product for research use only. It is not intended for human use.

