

## **Certificate of Analysis for NR-49113**

## Coli Surface Protein 3 (CS3) from Enterotoxigenic Escherichia coli

## Catalog No. NR-49113

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

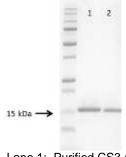
**Product Description:** NR-49113 is a preparation of coli surface protein 3 (CS3) purified from enterotoxigenic *Escherichia coli* (*E. coli*) (ETEC). CS3 was purified from *E. coli*, strain E 9034, grown in DME/F-12 (Dulbecco's Modified Eagle's medium and F-12 serum-free medium) broth in a fermenter under cGMP conditions in December 2002. The protein was purified from the culture supernatant by ammonium sulfate precipitation and tangential flow filtration and stored as a bulk preparation, at approximately -80°C, until it was vialed.

Lot<sup>1</sup>: 05062014-3 Manufacturing Date: 13DEC2002

TEST	SPECIFICATIONS	RESULTS
Appearance	Report results	Clear
SDS-PAGE (GelCode® Blue Stain Reagent)	15 kDa band represents > 95% of total staining intensity above background	15 kDa band represents > 95% of total staining intensity above background (Figure 1)
Confirmation of Identity by Western Blot <sup>2</sup> CS3 (NR-49113)	Reactive	Reactive (Figure 2)
Concentration by Bicinchoninic Acid Protein Assay	Report results	1.86 mg/mL
Endotoxin Content <sup>3</sup>	Report results	1.0 EU/μg protein
Sterility	0.22 µm filter sterilized	0.22 µm filter sterilized
Post-Purification Viability <sup>4</sup>	No growth	No growth

Production and quality control completed by Department of Subunit Enteric Vaccines and Immunology (SEVI), Bacterial Diseases Branch, Walter Reed Army Institute of Research, Silver Spring, Maryland, USA. The bulk material was thawed and vialed 06MAY2014. Quality control testing was completed at the time of vialing.

Figure 1 — SDS-PAGE of CS3



Lane 1: Purified CS3 (5 μg) Lane 2: Purified CS3 (2.5 μg)

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<sup>&</sup>lt;sup>2</sup>Using rabbit anti-CS3 serum

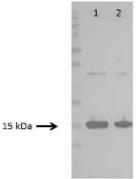
<sup>&</sup>lt;sup>3</sup>Endosafe<sup>®</sup> LAL Assay from Charles River

<sup>&</sup>lt;sup>4</sup>Presence of bacteria was tested by plating 100 μL of purified protein onto a Tryptic Soy agar plate which was incubated in an aerobic atmosphere at 37°C for 24 hours, in triplicate.



## **Certificate of Analysis for NR-49113**

Figure 2 — Western Blot of CS3



Lane 1: Purified CS3 (0.5 μg) Lane 2: Purified CS3 (0.25 μg)

Date: 16 JAN 2015 Signature:

Title: Technical Manager, BEI Authentication or designee

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