

***Clostridium botulinum*, Strain VPI 1731 (Type B, Non-Proteolytic)**

**Catalog No. NR-273**

(Derived from ATCC® 25765™)

**Product Description:** *Clostridium botulinum* (*C. botulinum*), strain VPI 1731 (type B, non-proteolytic) was isolated by Mel Eklund and transferred to Dr. Louis D. S. Smith of the Virginia Polytechnic Institute (VPI) who deposited the material to the ATCC® in 1970.

**Lot<sup>1</sup>: 61176377**

**Manufacturing Date: 17OCT2014**

TEST	SPECIFICATIONS	RESULTS
<b>Phenotypic Analysis</b> Cellular morphology Colony morphology <sup>2</sup> Biochemical tests RapID™ ANA II	Gram-positive rod Report results  <i>C. botulinum</i> (>95%)	Gram-positive rod Irregular, flat, entire, opaque and gray  <i>C. botulinum</i> , Group II (>99.9%) <sup>3</sup>
<b>Genotypic Analysis</b> Sequencing of 16S ribosomal RNA gene (~ 1290 base pairs)	>99% identical to <i>C. botulinum</i> , Group II (GenBank: FR745875)	99.8% identical to <i>C. botulinum</i> , Group II (GenBank: FR745875)
<b>Confirmation of Neurotoxin Type by PCR Amplification of Extracted DNA</b> Positive control (16S ribosomal RNA gene) Negative control ( <i>Clostridium difficile</i> ) NR-273 (neurotoxin type B) Neurotoxin types A, C, D, E, F, G	Amplicon present No amplicon present Amplicon present No amplicons present	Amplicon present No amplicon present Amplicon present No amplicons present
<b>Purity (post-freeze)<sup>4</sup></b> Anaerobic atmosphere (90% N <sub>2</sub> :5% CO <sub>2</sub> :5% H <sub>2</sub> )  Aerobic atmosphere	Growth consistent with expected colony morphology No growth	Growth consistent with expected colony morphology No growth
<b>Viability (post-freeze)<sup>2</sup></b>	Growth	Growth

<sup>1</sup>NR-273 was produced by inoculation of ATCC® 25765™ (Lot 52354) into Reinforced Clostridial broth and grown 7 days at 37°C in an anaerobic atmosphere (90% N<sub>2</sub>:5% CO<sub>2</sub>:5% H<sub>2</sub>). Inoculum from this growth was added to on Tryptic Soy agar with 5% defibrinated sheep blood kolles and grown 2 days at 37°C in an anaerobic atmosphere (90% N<sub>2</sub>:5% CO<sub>2</sub>:5% H<sub>2</sub>) to produce this lot.

<sup>2</sup>2 days at 37°C in an anaerobic atmosphere (90% N<sub>2</sub>:5% CO<sub>2</sub>:5% H<sub>2</sub>) on Tryptic Soy agar with 5% defibrinated sheep blood.

<sup>3</sup>*C. botulinum* Group II consists of toxin type E and non-proteolytic strains of toxin type B and F. Toxin neutralization testing is recommended for confirmation of *C. botulinum* Group II.

<sup>4</sup>Purity of this lot was assessed for 7 days on Tryptic Soy agar with 5% defibrinated sheep blood at 37°C.

**Date:** 26 JUL 2016

**Signature:**



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