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Product Information

Cholera Toxin, B subunit
recombinant, expressed in HEK 293 cells

Product Number **SAE0069**
Storage Temperature –20 °C

CAS RN 131096-89-4
Synonyms: CTxB, ToxB, CTB, Cholera enterotoxin subunit B, Cholera enterotoxin B chain, Cholera enterotoxin gamma chain

Product Description
Cholera toxin is the virulent factor from *Vibrio cholera* that leads to severe diarrhea, followed by dehydration, in humans.[1,2] Several bacterial toxins are ADP-ribosyltransferases with protein substrates. Many substrates that are ADP-ribosylated by bacterial protein toxins are G-proteins, which are involved in signal transduction. ADP-ribosylation is one of the more significant post-translational modifications of proteins. The ADP-ribosylation activity of cholera toxin activates adenylate cyclase, resulting in the production of cyclic AMP by adenylate cyclase, which causes many metabolic alterations.[1,2]

Cholera toxin belongs to the AB_5-subunit family of toxins.[1] The native hexameric protein has a molecular mass of ~85 kDa and contains two distinct subunits:

- The single A subunit (~27.2 kDa) is responsible for the ADP-ribosylation activity.
- Five B subunits (~11.6 kDa each) are arranged as a pentameric ring with an apparent 5-fold symmetry. These are associated with the cell surface receptor binding and subsequent internalization (transmembrane transport) of the enzymatic component.[3,4]

A single isoelectric variant of the cholera toxin has been isolated, which crystallizes readily and reproducibly.[5] Cholera toxin has an isoelectric point (pI) of 6.6. Chromatographic properties, however, suggest that a cationic surface is exposed at pH 7.0, which apparently resides in the B subunit.[6]

The entire hexameric complex is required for toxic behavior. Choleragenoid, the intact pentamer of B subunits, interacts with a ganglioside G_{M1} membrane receptor, but cannot activate adenylyl cyclase, whereas the A subunit alone does not enter the cell.[7] Because of its effect on adenylate cyclase, cholera toxin and its purified A subunit are frequently used for the study of signal transduction mechanisms. In addition, cholera toxin acts as an adjuvant through the stimulation of B lymphocytes.

The cholera toxin B (CTxB) subunit alone is used for track tracing in neurological research, taking advantage of G_{M1} ganglioside binding and retrograde transport. Tissue culture cells treated with CTxB are not killed, and animal tissues treated with CTxB do not become necrotic.

CTxB is non-toxic to cells and possesses no intrinsic adenylate cyclase activity. CTxB attaches to cells by binding to ganglioside G_{M1}.[8] As a result, CTxB has been shown to be a good label for microglial cells (because of to the enrichment of ganglioside G_{M1} on their cell surface), but not for oligodendrocytes nor astrocytes.[9]

CTxB has been reported to be an excellent tracer for the study of axonal transport by immunohistochemical methods. CTxB has been widely used as a marker of membrane lipid rafts (membrane microdomains enriched with cholestrol and sphingolipids). These lipid rafts have an important role in cell signaling and protein trafficking.[10]

Recombinant CTxB is expressed in human HEK 293 cells with a C-terminal histidine-tag, with a calculated molecular mass of 13 kDa (amino acids Thr^{22}-Asn^{124}). The advantage of this product over the native CTxB is that it has no traces of *Vibrio cholera*, and no contamination of the highly toxic Cholera toxin A subunit.

This product is supplied as an aseptically-filled, lyophilized powder. When reconstituted with water to a final concentration of 1 mg of CTxB per mL, the solution will contain 1× PBS.

Purity: ≥95% (SDS-PAGE)

The activity is measured by ELISA using G_{M1}-coated plates, anti-rabbit CTxB primary antibodies, and peroxidase-labeled goat anti-rabbit IgG as the secondary antibody. Binding saturation of 50% is achieved with ≤1 μg/mL of CTxB.

Precautions and Disclaimer
This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions
Briefly centrifuge the vial before opening. Reconstitute in water to a concentration of 0.5–10 mg/mL. **Do not vortex.** Instead, swirl the vial gently during reconstitution. Avoid vigorous pipetting of solutions, which may lead to foaming. Solutions can be filtered through a 0.2 μm filter.

This solution can be stored at 2–8 °C for up to 1 week. For extended storage, it is recommended to store reconstituted solutions in working aliquots at –20 °C.

Storage/Stability
Store the lyophilized product at –20 °C. The product is stable for at least 2 years as supplied.

References

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