

Cholera Toxin Subunit B (CT-B) Conjugates

Quick Facts

Storage upon receipt:

- ≤-20°C
- Desiccate
- · Protect from light

Abs/Em: See Table 1

Introduction

Cholera toxin from *Vibrio cholerae* is comprised of two subunits, A and B, arranged in an AB_5 configuration. The A subunit is an ADP-ribosyltransferase, which disrupts the proper signaling of G proteins and eventually leads to dehydration of the cell. The nontoxic B subunit is important to the protein complex as it allows the protein to bind to cellular surfaces via the pentasaccharide chain of ganglioside G_{M1} . At neutral pH, the 11.4 kDa B subunit exists as a 57 kDa pentamer.

The B subunit of cholera toxin (CT-B) has proven to be a powerful tool for retrograde labeling of neurons. 4,5 This tracer has been used in a variety of applications, including tracing of rat forebrain afferents,6 projections of the parabrachial region 7 and neurons of the urinary bladder wall.8 More recently, researchers have found that CT-B can be used as a marker for lipid rafts, which are membrane microdomains enriched in cholesterol and sphingolipids. Lipid rafts segregate specific groups of proteins and thereby provide a hub for cellular signaling and protein trafficking. 9, 10

Molecular Probes' CT-B conjugates (Table 1) are made from recombinant cholera toxin subunit B. Because the B-subunit source material is recombinant in origin, it is extremely pure and completely free of the toxic A subunit.

Materials

The biotin- and dye-labeled cholera toxin B subunits are supplied in unit sizes of 100 μ g and 500 μ g. When stored desiccated at \leq -20°C, these products are stable for at least six months. Solutions of 1.0 mg/mL can be prepared by dissolving the

Table 1. Conjugates of cholera toxin subunit B.

Label	Abs/Em *	Unit Size	
		100 µg	500 µg
Alexa Fluor 488	495/519	C-34775	C-22841
Alexa Fluor 555	555/565	C-34776	C-22843
Alexa Fluor 594	590/617	C-34777	C-22842
Alexa Fluor 647	650/668	C-34778	
Biotin	NA	C-34779	
Horseradish peroxidase (HRP)	NA	C-34780	

^{*} Approximate absorption (Abs) and fluorescence emission (Em) maxima, in nm. NA = Not applicable.

powder in 0.1 mL or 0.5 mL of buffer, for example, phosphate-buffered saline (PBS). With the addition of 2 mM sodium azide, solutions can be stored at $2-6^{\circ}$ C for approximately three months. For longer storage, divide the solution into aliquots and freeze at $\leq -20^{\circ}$ C.

The peroxidase conjugates of cholera toxin B are supplied in a unit size of 100 μ g. When stored desiccated at -20° C, the lyophilized powder is stable for at least six months. Solutions of 1.0 mg/mL can be prepared by dissolving the powder in 0.1 mL of PBS, pH 7.2. Store solutions at 2–6°C with the addition of thimerosal to a final concentration of 0.02%. DO NOT USE AZIDE for the HRP-conjugates. For prolonged storage after reconstitution, add glycerol to a final concentration of 50% (v/v), aliquot and store at -20° C. When stored properly, solutions are stable for approximately three months. PROTECT FROM LIGHT. AVOID REPEATED FREEZING AND THAWING OF SOLUTIONS.

For the Alexa Fluor and biotin conjugates, the lot-specific degree of labeling (typically 5–10 moles of dye per mole of the B subunit pentamer) is indicated on the product label.

Application

Due to the diversity of applications for the cholera toxin subunit B, please consult the primary literature for appropriate working concentrations.

References

1. J Biol Chem 255, 1252 (1980); **2.** Mol Microbiol 13, 745 (1994); **3.** Biochemistry 35, 16069 (1996); **4.** Brain Res 243 215 (1982); **5.** Brain Res 231, 33 (1982); **6.** Neuroscience 82, 443 (1998); **7.** Brain Res 816, 364 (1999); **8.** Neuroscience 87, 275 (1998); **9.** J Cell Biol 147, 447 (1999); **10.** J Cell Biol 141, 929 (1998).

Product List Current prices may be obtained from our Web site or from our Customer Service Department.

Cat #	Product Name	Unit Size
Cat # C-34775 C-22841 C-34776 C-22843 C-34777 C-22842 C-34778	cholera toxin subunit B (recombinant), Alexa Fluor® 488 conjugate	100 µg 500 µg 100 µg 500 µg 100 µg 500 µg
C-34779	cholera toxin subunit B (recombinant), Alexa Fluor® 647 conjugate	100 μg 100 μg
C-34780	cholera toxin subunit B (recombinant), horseradish peroxidase conjugate	100 µg

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Further information on Molecular Probes' products, including product bibliographies, is available from your local distributor or directly from Molecular Probes. Customers in Europe, Africa and the Middle East should contact our office in Leiden, the Netherlands. All others should contact our Technical Assistance Department in Eugene, Oregon.

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