Product Information

CF Dye Cholera Toxin Subunit B Conjugates

Unit Size: 100 ug

Product List

Catalog no.	Conjugate	Ex/Em (nm)
BGT-CHM-418	CF405M-Cholera Toxin Subunit B(CTB)	408/452
BGT-CHM-419	CF488A-Cholera Toxin Subunit B(CTB)	490/515
BGT-CHM-420	CF532-Cholera Toxin Subunit B(CTB)	527/558
BGT-CHM-421	CF543-Cholera Toxin Subunit B(CTB)	541/560
BGT-CHM-422	CF568-Cholera Toxin Subunit B(CTB)	562/583
BGT-CHM-423	CF594-Cholera Toxin Subunit B(CTB)	593/614
BGT-CHM-424	CF633-Cholera Toxin Subunit B(CTB)	630/650
BGT-CHM-425	CF640R-Cholera Toxin Subunit B(CTB)	642/662
BGT-CHM-426	CF647-Cholera Toxin Subunit B(CTB)	650/665
BGT-CHM-427	CF660R-Cholera Toxin Subunit B(CTB)	663/682
BGT-CHM-428	CF680R-Cholera Toxin Subunit B(CTB)	680/701

Storage and Handling

Store at -20°C upon arrival and protect from light. Product is stable for at least six months from date of receipt when stored as recommended. This product contains less than 0.5% Cholera Toxin Subunit A and should be handled and disposed of using universal laboratory safety precautions.

Product Description

Cholera toxin is the symptom-causing toxin produced by the bacteria Vibrio cholerae during cholera infection. The toxin is composed of two subunits, A and B. Subunit A is the toxic enzymatic subunit present in one copy per toxin. Cholera toxin subunit B (CT-B) is the receptor binding subunit that is found as a pentamer in each toxin and is relatively non-toxic, making it useful for cell biological studies.

CT-B has been used as a neuronal tracer and has also been shown to bind to GM1 gangliosides that are found in lipid rafts on the surface of mammalian cells. Therefore, fluorescently labeled conjugates of CT-B have been used as lipid raft markers and endocytic tracers for live imaging or on fixed cells. Please note that CT-B staining can show heterogeneity in cultured cells such as HeLa cells (1).

Our Cholera Toxin Subunit B conjugates are labeled with a selection of our CF dyes, a series of next-generation fluorescent dyes to have combined advantages in brightness, photostability, and water solubility compared to other fluorescent dyes.

References

1) Pang, H. et al. (2004). J. Cell Sci 117, 1421-1430.

Experimental Protocols

The following are protocols for labeling of GM1 ganglioside-positive lipid rafts in cultured cells. For neuronal tracing studies, please refer to the primary literature to find the appropriate method for a given application.

Materials required but not provided

- Phosphate-buffered saline (PBS)
- Bovine serum albumin (BSA)
- Hank's Balanced Salt Solution (HBSS)
- 4% Paraformaldehyde in PBS
- Hoechst dye (Optional)

Reconstitution

Reconstitute CF Dye Cholera Toxin Subunit B Conjugate in 1X PBS to a concentration of 1 mg/ml. For storage at 4°C, sodium azide can be added to a final concentration of 2 mM for storage if it is compatible with your application.

Stock solutions may be prepared in water or phosphate-buffered saline. Solutions can be aliquoted and stored protected from light at -20°C for up to six months, or store at 4°C for up to three months.

Surface labeling on live cells

1. Wash cells once with cold 1X Hank's Balanced Salt Solution (HBSS) + 0.5% (BSA).
2. Add the reconstituted CF Dye Cholera Toxin Subunit B Conjugate to a final concentration between 400 ng/ml and 1 ug/ml to cells.
3. Incubate cells at 4°C for 30 minutes in the dark.
4. Wash cells five times with cold 1X HBSS + 0.5% BSA.
5. Fix in 4% paraformaldehyde in 1X PBS for 15 minutes at room temperature (protected from light).
   Note: Hoechst may be used as a nuclear counterstain at 1 ug/mL. If cells will be permeabilized for other immunostaining, an antibody against cholera toxin subunit B may help may help in optimal preservation of the lipid raft domains and should be incubated with the cells prior to permeabilization.
6. Wash cells twice with 1X PBS and process samples for imaging or subsequent immunostaining.

Cell trafficking assay

1. Add the reconstituted CF Dye Cholera Toxin Subunit B Conjugate to a final concentration between 400 ng/ml and 1 ug/ml to cells in complete medium.
2. Incubate at 37°C for 10 minutes to 1 hour in the dark.
3. Fix in 4% paraformaldehyde in 1X PBS for 15 minutes at room temperature (protected from light).
   Note: CF Dye Cholera Toxin Subunit B Conjugate does not show cytotoxicity in HeLa cells after overnight incubation, however the low level of cholera toxin A subunit present may cause cytotoxicity in other cells types or after prolonged incubation times.
4. Wash cells twice with 1X PBS and process samples for imaging or subsequent immunostaining.