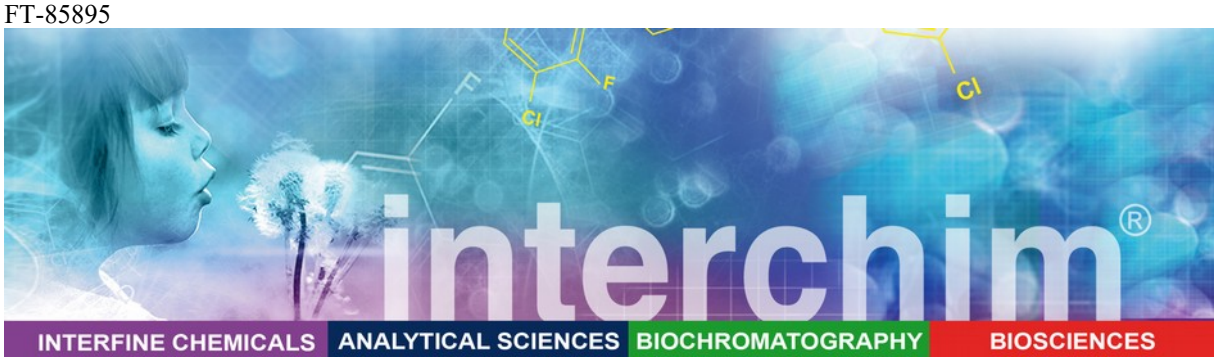
FT-85895



Bungarotoxins

Product Information

Product name cat.number	Structure	MW (g·mol^{-1})	λ_{exc}\λ_{em} ax. (nm)	Soluble in
α-Bungarotoxin FP-38034B 1mg	$C_{338}H_{529}N_{97}O_{105}S_{11}$	7984.27		Water
α-Bungarotoxin FITC FP-52482A, 500µg FP-52482B,10x50µg		~ 8400	494 / 518	Water
α-Bungarotoxin tetramethylrhodamine FP-52509A, 500µg FP-52509B, 10x50µg		~ 8600	552 / 579	Water
α -Bungarotoxin SR101 FP-22597A, 500µg FP-22597B, 10x50µg		~ 8400	593 / 613	Water
α -Bungarotoxin -xx-Biotin FP-85895A, 500µg		~ 8400		Water
β-Bungarotoxin FP-74359A, 1 mg		~ 20 000		Water

Storage: –20°C >1 year. (M) Protect from light and moisture

Introduction

α-Bungarotoxin and β-Bungarotoxin are extracted from *Bungarus multicinctus* venom.
α-Bungarotoxin has high affinity to the α-subunit of the nicotinic AchR (Nicotinic acetylcholine receptors) of neuromuscular junctions (K_d= 1 nM to 1 pM).
β-Bungarotoxin act presynaptically causing acetylcholine release and depletion.() The toxin contains a smaller subunit that blocks voltage-gated K^+ channels on the presynaptic membranes, and a larger subunit that has phospholipase A_2 activity.
It has been used to stain acetylcholine (ACh) receptors in skeletal muscle, rat myotubules, the electric organ from *Torpedo californica* and transformed *Escherichia coli*.
Unlabeled α-Bungarotoxin and β-Bungarotoxin are useful as a control, as well as for labeling, radioiodination, preparation of new conjugates, and for competitive studies.
Labeled α-bungarotoxin are used in fluorescent detection in microscopy and cell assays. It is used to image the distribution of the receptor at the neuromuscular junctions.
Furthermore, Biotin α-Bungarotoxin can be used for detection with enzyme labels and even for receptors isolation with immobilized avidins.

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Directions for use

Handling and Storage

Prepare a 1 mg/ml stock solution by reconstituting unlabeled α-bungarotoxin in 1 mL of PBS or the α-bungarotoxin conjugates in the appropriate volume of distilled water.
Store the solution at -20°C with the addition of sodium azide to a final concentration of 2mM. For longer storage, divide the solution into aliquots and freeze at -20°C protected from light.. When properly stored, these products are stable for several months. Avoid repeated freezing and thawing. A centrifugation may be required to remove aggregate that might have formed, reducing background.
The following protocol are example of use of Bungarotoxin. Each experiment should be optimized according the cells type, the assays, the bungarotoxin unlabeled and labeled…

Guidelines for use – on synaptosomes ()

1. Prepare synaptosomes on « polysine » slide. Incubate 100 nM α-Bungarotoxin-biotin Krebs buffer over slide for 20 min at room temperature. Wash slide three times in Krebs buffer
2. Fix synaptosomes by immersing slide in appropriate fixative for 10 min. Methanol fixation at -20°C is optimal as the Bungarotoxin is well fixed to the synaptosomes.
3. Place slide in humidified chamber and layer Avidin-FluoProbes® 488 over synaptosomes and incubate for 30 min in dark. Wash three times 5 minutes in PBS and view in confocal microscope.

Note : If α-Bungarotoxin -FITC or α-Bungarotoxin-tretramethylrhodamine is used, the step 5 is not required. In this case, do not use fixation in methanol at –20°C. Even if bungarotoxin are not well fixed to synaptosomes and may dissociate during later incubations, try a fixation of 4% paraformaldehyde.

Guidelines for use – on rat myotubes (Axelrod 1980) **with biotinylated α-Bungarotoxin**

1. Prepare rat myotubes primary culture.
2. Treat cells with biotinylated α-Bungarotoxin at 4µg/ml in appropriate medium for at least 10 min at +37°C. Wash them in HBSS/albumin and add Rhodamine –avidine at 4µg/ml at least 5 min at +22°C. After several washings, cells were bathed with HBSS/albumin for fluorescence microscopy.

Note: a fluorescence labeling pattern on the cell surface may be produced. A pretreatment of the cells could be realized: incubate cells with unlabeled α-Bungarotoxin at 1 µg/ml, 30min at +37°C.Therefore, Biotinylated α-Bungarotoxin bound specifically only to the same sites as unlabeled α-Bungarotoxin. Treatment of cells by unlabeled α-Bungarotoxin followed by Rhodammine Avidin produced no specific labeling. Therefore, Rhodammine Avidin bound specifically only to Biotinylated α-Bungarotoxin.

Cells could be incubated with rhodamine α-Bungarotoxin at 1µg/ml in appropriate medium for 1h at +37°C. Wash them in HBSS/albumin..

Other protocol may found in the literature.

Related products

- NeutralAvidin-HRP, 36570A
- NeutralAvidin-AP, 38592A
- NeutralAvidin-R-PhycoErythrin, 31259A
- FP Membrane Marker 1-13, FP-51254A

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