Neuronal Probes & Reagents

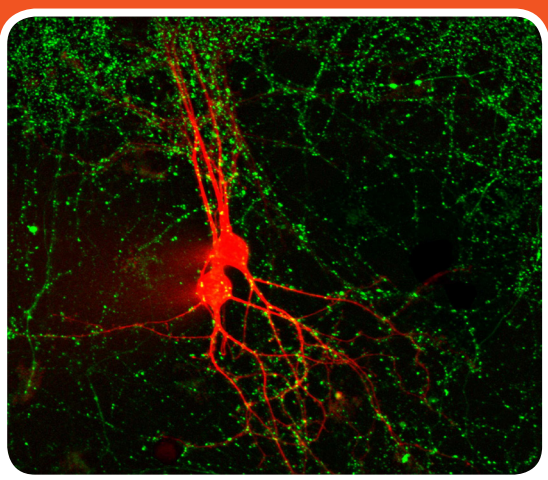

Figure 1. Cultured rat hippocampal neurons microinjected with CF®647 hydrazide (red) and stained with SynaptoGreen™. Image courtesy of Professor Guosong Liu, Tsinghua University, Beijing.

Fluorescent dyes and toxins for neuronal tracing and more

Biotium offers a wide selection of dyes, toxins, and other probes for neuronal staining, featuring our next-generation fluorescent CF® dyes. CF® dyes have advantages in brightness, photostability, and solubility, come in a wide selection of colors ranging from UV to near-infrared, and have been validated in multiple super-resolution imaging applications.

Nerve terminal dyes for activity depending vesicle tracking

- SynaptoRed™ & SynaptoGreen™ (equivalent to FM®1-43 dyes)
- Activity-dependent fluorescent vesicle tracking
- Aldehyde-fixable versions
- Nerve terminal staining kits with background reducers

Toxins & fluorescent toxin receptor probes

- Tetrodotoxin (TTX) sodium channel blocker, with or without citrate
- α-Bungarotoxin & fluorescent CF® dye conjugates
- CF® dye-labeled cholera toxin subunit B

Amyloid & neurodegeneration stains

- PathoGreen™ Histofluorescent Stain for neuronal death (Fluoro-Jade® C alternative)
- Congo Red colorimetric & fluorescent amyloid stain
- DCDAPH near-infrared stain for Aβ1-42 aggregates and β-amyloid
- Thioflavin T cell permeable fluorescent amyloid probe

Anterograde / retrograde tracers

- Fluorescent WGA, Cholera toxin, and amino-dextran CF® dye conjugates
- Hydroxystilbamidine (equivalent to Fluoro-Gold™)

Cytosolic tracers for morphology & gap junction connectivity

- Bright & photostable CF® dye hydrazides in 10+ colors
- Hydroxystilbamidine (equivalent to Fluoro-Gold™)
- Lucifer yellow derivatives
- Biocytin and biotin probes
- Membrane permeant and fixable cytoplasmic dyes

And more...

- Fluorescent ion indicators & membrane potential dyes
- Live cell microtubule, mitochondrial, and other organelle stains
- Labeled primary antibodies for neuroscience

Biotium

Nerve Terminal Dyes: SynaptoGreen™ and SynaptoRed™

Nerve terminal dyes (originally called FM® dyes) are sold by Biotium under the trademark names SynaptoGreen™ and SynaptoRed™. They have a lipophilic tail at one end and a highly hydrophilic, cationic head group at the other end. They are virtually non-fluorescent in solution, but when added to cells, insertion of the lipophilic tails into the plasma membrane causes the dyes to become intensely fluorescent.

These dyes can be used to label membranes and vesicles in many cell types, but they are commonly called nerve terminal dyes or synaptic vesicle dyes due to their utility for dynamic tracking of synaptic vesicles in cultured neurons and tissue preparations (Fig. 1, Fig. 3). When applied to neurons, the dyes are incorporated into synaptic vesicles by endocytosis (termed the "on-rate"). After extracellular dye is quenched or washed away, the fluorescent vesicles can be imaged over time. During exocytosis and neurotransmitter release, the dyes also are released from the vesicles, causing a decrease in fluorescence signal (or "off-rate").

SynaptoGreen™ and SynaptoRed™ dyes vary in the length of the lipophilic tail and the number of double bonds linking the two aromatic rings in the dye (Fig. 2). In general, dyes with longer tails and more double bonds have a higher affinity for membrane and thus a higher on-rate and lower off-rate. AM and HM dyes possess an additional aldehyde-fixable amine or hydrazide group, which makes them more water-soluble with a higher off-rate and lower on-rate than the corresponding SynaptoGreen™/SynaptoRed™ or FM® dye.

Some nerve terminal dyes can enter cells through ion channels in addition to endocytosis; SynaptoGreen™ C18 and AM3-25 are high molecular weight dyes that cannot pass through ion channels that have been used as controls to distinguish mechanisms of dye uptake.

Background Quenchers and Nerve Terminal Staining Kits

A common problem encountered with nerve terminal dyes is background fluorescence due to residual membrane staining after washing. To reduce extracellular fluorescence, we offer three quencher or dye-clearing agents. ADVASEP-7, a sulfonated β-cyclodextrin, binds dyes and allows them to be more efficiently washed away. SCAS is a quencher that reduces dye fluorescence without the need for washing. Sulforhodamine 101 quenches SynaptoGreen™ background by fluorescent resonance energy transfer (FRET). We also offer Nerve Terminal Staining Kits that pair dyes with background reducers.

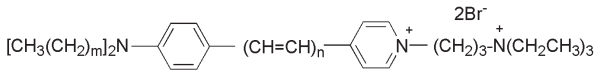

Figure 2. General structure of SynaptoGreen and SynaptoRed dyes. m = 0-17; n = 1-3.

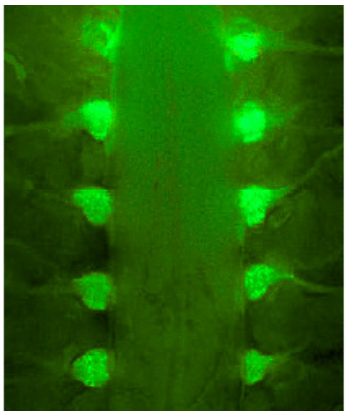

Figure 3. Neurons in mouse dorsal root ganglia (DRG) labeled with AM1-43. Image courtesy of Dr. David Corey, Harvard Medical School.

Cat. #	Size	Product name	m*	n*	Ex/Em in membranes	Fixable?
70042, 70043	5 mg, 5 x 1 mg	SynaptoGreen™ C1	0	1	~480/600 nm	No
70044, 70045	5 mg, 5 x 1 mg	SynaptoGreen™ C2 (equivalent to FM®2-10)	1	1	~480/600 nm	No
70023, 70026	5 mg, 5 x 1 mg	SynaptoGreen™ C3	2	1	~480/600 nm	No
70020, 70022	5 mg, 5 x 1 mg	SynaptoGreen™ C4 (equivalent to FM®1-43)	3	1	~480/600 nm	No
70046, 70047	5 mg, 5 x 1 mg	SynaptoGreen™ C5 (equivalent to FM®1-84)	4	1	~480/600 nm	No
70048, 70049	5 mg, 5 x 1 mg	SynaptoGreen™ C18 (equivalent to FM®3-25)	17	1	~480/600 nm	No
70024	1 mg	AM1-43	3	1	~480/600 nm	Yes
70038	1 mg	AM1-44	4	1	~480/600 nm	Yes
70036	1 mg	AM2-10	1	1	~480/600 nm	Yes
70051	1 mg	AM3-25	17	1	~480/600 nm	Yes
70053	1 mg	HM1-43	3	1	~480/600 nm	Yes
70040, 70041	5 mg, 5 x 1 mg	SynaptoRed™ C1	0	3	~510/750 nm	No
70021, 70027	5 mg, 5 x 1 mg	SynaptoRed™ C2 (equivalent to FM®4-64)	1	3	~510/750 nm	No
70019, 70028	5 mg, 5 x 1 mg	SynaptoRed™ C2M** (equivalent to FM®5-95)	1	3	~510/750 nm	No
70025	1 mg	AM4-64	1	3	~510/750 nm	Yes
70039	1 mg	AM4-65	3	3	~510/750 nm	Yes
70050	1 mg	AM4-66	4	3	~510/750 nm	Yes

*m is the number of carbons in the lipophilic tail and n is the number of double bonds linking the two aromatic rings in the dye (see Fig. 1)
**The positively-charged end of SynaptoRed C2M is a trimethylammonium group.

Cat. #	Product name	Kit Contents
70030	Nerve Terminal Staining Kit I	5 x 1 mg SynaptoGreen™ C4 (70022) + 250 mg ADVASEP-7 (70029)
70031	Nerve Terminal Staining Kit II (A)	1 mg AM1-43 (70024) + 100 mg ADVASEP-7 (70029-1)
70031-1	Nerve Terminal Staining Kit II (B)	1 mg of AM1-43 (70024) + 100 mg SCAS (70037)
70032	Nerve Terminal Staining Kit III	5 x 1 mg SynaptoGreen™ C4 (70022) + 100 mg Sulforhodamine 101 (80101)
70034	Nerve Terminal Staining Kit V	5 x 1 mg SynaptoRed™ C2 (70027) + 250 mg ADVASEP-7 (70029)

Toxins and Fluorescent Receptor Probes

Tetrodotoxin
Tetrodotoxin (TTX) reversibly blocks excitable sodium channels and has been a widely used tool for studies of excitable membranes of nerve and muscle cells. Available lyophilized in citrate buffer, or citrate-free.

Bungarotoxin and Fluorescent Conjugates
α-Bungarotoxin is a potent inhibitor for the motor endplate acetylcholine receptor. Fluorescent conjugates of a-bungarotoxin can be used to label neuromuscular junctions (Fig. 4). We offer a α-bungarotoxin conjugates of CF® dyes with colors ranging from blue to near-IR fluorescence, plus biotin and other labels. Many CF® dyes are compatible with super resolution imaging by SIM, STED, and STORM; visit biotium.com to learn more.

Cholera Toxin Subunit B Conjugates
Cholera toxin subunit B binds GM1 ganglioside in lipid rafts, and is used as a retrograde neuronal tracer. Available with a wide selection of bright and photostable CF® dyes.

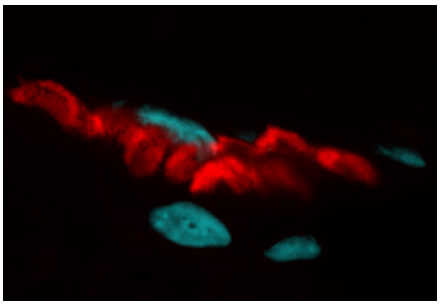

Figure 4. Rat skeletal muscle cryosection stained with CF®594 a-bungarotoxin (motor endplate, red) and DAPI (nuclei, blue).

Anterograde and Retrograde Neuronal Tracers

Wheat Germ Agglutinin (WGA) Conjugates
WGA is a glycoprotein-binding lectin that has been used for retrograde and anterograde neuronal tracing. We offer WGA CF® dye conjugates with fluorescence from UV to near-IR, plus HRP.

Dextran Conjugates
Labeled dextran amine can be used for both retrograde and anterograde tracing. CF® dye dextrans are anionic with an aldehyde-fixable free amine group, and are available with a wide selection of colors and a range of molecular weights.

Hydroxystilbamidine (equivalent to Fluoro-Gold™)
Hydroxystilbamidine (also called Fluoro-Gold™) has been used extensively as a retrograde tracer for neurons and also a histochemical stain. Fluoro-Gold™ is used for retrograde tracing and dendrite filling.

Also see Cholera Toxin Subunit B (above) and Biotin Ethylenediamine (below).

Amyloid Stains

Congo Red is commonly used to detect amyloid protein aggregates associated with Alzheimer's disease, Bovine Spongiform Encephalopathy, and related diseases. The staining can be detected by either colorimetric or fluorescence imaging (Ex/Em 497/614 nm).

DCDAPH is a far-red fluorescent probe (Ex/Em 597/665 nm) with high affinity (Kd=27 nM) to Aβ1-42 aggregates. It has been used for fluorescent staining of brain sections, as well as *in vivo* small animal near-IR imaging.

Thioflavin T is a cell-permeable benzothiazole dye that exhibits enhanced fluorescence (Ex/Em 450/482 nm) upon binding to amyloid fibrils. Thioflavin T has also been used in histology and for protein characterization.

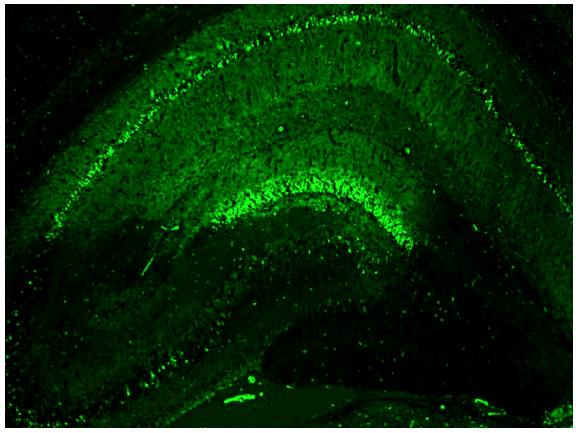

Figure 5. Degenerating neurons in a slice of mouse hippocampus stained with PathoGreen™ Histofluorescent Stain.

PathoGreen™ Histofluorescent Stain for Neurodegeneration

PathoGreen™ is an anionic green fluorescent dye functionally similar to Fluoro-Jade® dyes. These dyes stain degenerating neurons and their processes in brain sections and cell culture (Fig. 5). The mechanism of staining by this class of dyes has not been determined, but the negatively charged dyes may bind to positively charged polyamines generated in dying neurons.

We also offer a wide selection of cell viability and apoptosis assays, visit biotium.com to learn more.

Cytosolic Tracer Dyes

CF® Dye Hydrazides
Hydrazides are non-toxic, highly water soluble membrane-impermeant tracers that can be used to fill cells by microinjection (Fig. 1). We offer a large selection of bright, photostable CF® dye hydrazides.

Lucifer Yellow and Related Dyes
Lucifer Yellow is a classic cell-impermeant cytosolic and gap junction dye. We also offer Lucifer Yellow Cadaverine and Lucifer Yellow CH with aldehyde-fixable groups.

Biotin derivatives
Formaldehyde-fixable biocytin and biocytin hydrazide are widely used microinjectable polar tracers. Biocytin has been used as an anterograde tracer and gap junction probe. Biotin derivatives can be detected with labeled streptavidin or anti-biotin antibodies.

Biotin ethylenediamine is equivalent to Neurobiotin™, a useful anterograde and transneuronal tracer.

Membrane Permeant Cytosolic Stains
Calcein-AM is a membrane-permeant, non-fluorescent compound that can be loaded into cultured cells by incubation. Once inside the cytoplasm, it is hydrolyzed inside viable cells to release the green fluorescent, membrane impermeant dye calcein, which fills the entire cell. Calcein-AM can be used to assess cell viability, and for short term cytoplasmic labeling.

ViaFluor® SE Cell Proliferation Dyes are membrane-permeant compounds that are hydrolyzed in the cytoplasm to release amine-reactive fluorescent dyes. The staining fills the entire cell, is stable for several days to weeks, and can withstand fixation and permeabilization. Available with blue and green fluorescence.

Ion Indicators, Membrane Potential Dyes, Organelle Stains, & More
Biotium offers a wide selection of fluorescent calcium and ion indicator dyes, membrane potential and mitochondrial potential dyes, live cell stains for microtubules, organelles, and cell membranes, and a selection of labeled primary antibodies for neuroscience targets. Visit biotium.com to learn more.

CF® Dye Labeled Toxins and Probes

Dye	Ex/Em	Alpha-Bungarotoxin 100 ug or 0.5 mg	Cholera Toxin B 100 ug	Dextran 3.5K MW 1 mg	Dextran 10K MW 1 mg	Dextran 40K MW 1 mg	Dextran 70K MW 1 mg	Dextran 150K MW 1 mg	Dextran 250K MW 1 mg	CF® Dye Hydrazide 1 mg	WGA 1 mg or 5 x 1 mg
CF®350	347/448 nm			80137						92151	29021
CF®405S	404/431 nm	00002								92183	29027
CF®405M	408/452 nm										29028
CF®488A	490/515 nm	00005	00070		80110	80126	80117	80131	80134	92152	29022
CF®532	527/558 nm		00074								29064
CF®543	541/560 nm	00026	00075		80111						
CF®555	555/565 nm	00018			80112					92153	29076
CF®568	562/583 nm	00006	00071		80113					92154	29077
CF®594	593/614 nm	00007	00072		80114					92158	29023
CF®620R	617/639 nm		00076								
CF®633	630/650 nm	00009	00077							92156	29024
CF®640R	642/662 nm	00004	00073		80115					92157	29026
CF®647	650/665 nm									92136	
CF®660R	663/682 nm		00078							96024	
CF®680	681/698 nm				80118	80127	80129	80132	80135		29029
CF®680R	680/701 nm	00003	00079		80116					96025	29025
CF®750	755/777 nm				80119	80128	80130	80133	80136		
CF®770	770/797 nm				80120	80122	80123	80124	80125	92192	29059
CF®790	784/806 nm				80121						

If you are looking for a CF® dye conjugate not listed in our catalog, please let us know. We may be able to add it as a new product, or perform a custom synthesis for you.

Additional Toxins & Toxin-Based Probes

Cat. #	Unit Size	Product name	Ex/Em
00060	1 mg	Tetrodotoxin, Citrate-Free	N/A
00061	1 mg	Tetrodotoxin, With Citrate	N/A
00010-1	1 mg	α-Bungarotoxin	N/A
00017	0.5 mg	Biotin-XX α-Bungarotoxin	N/A
00011	500 ug	Fluorescein α-Bungarotoxin	494/518 nm
00013	10 x 50 ug		
00012	500 ug	Tetramethylrhodamine α-Bungarotoxin	553/577 nm
00014	10 x 50 ug		
00015	500 ug	Sulforhodamine-101 (Texas Red®) α-Bungarotoxin	593/613 nm
00016	10 x 50 ug		

Amyloid and Neurodegeneration Stains

Cat. #	Unit Size	Product name	Ex/Em
80028	100 mg	Congo Red High Purity Grade	497/614 nm
80030	5 mg	DCDAPH	597/665 nm
80033	100 mg	Thioflavin T High Purity Grade	450/482 nm
80027-5mL	5 mL	PathoGreen™ Histofluorescent Stain	497/520 nm
80027-50mL	50 mL		

Fluorescent & Biotinylated Tracers

Cat. #	Unit Size	Product name	Ex/Em
80014	10 mg	Hydroxystilbamidine (Fluoro-Gold™)	361/536 nm
80023	200 uL	Hydroxystilbamidine (Fluoro-Gold™), 4% in H_2O	361/536 nm
80015	25 mg	Lucifer Yellow CH, lithium salt	428/536 nm
80016	25 mg	Lucifer Yellow CH, potassium salt	428/536 nm
80018	25 mg	Lucifer Yellow Cadaverine	428/536 nm
80017	10 mg	Lucifer Yellow Cadaverine Biotin-X, dipotassium salt	428/532 nm
90055	100 mg	Biocytin	N/A
90060	25 mg	Biocytin hydrazide	N/A
90057	25 mg	Biotin ethylenediamine, hydrobromide (Neurobiotin™)	N/A
90075	25 mg	Biotin ethylenediamine, hydrochloride	N/A
30026	1000 assays	Calcein AM Cell Viability Assay Kit	494/517 nm*
80011	1 mg	Calcein AM	494/517 nm*
80011-1	100 uL	Calcein AM, 4 mM in anhydrous DMSO	494/517 nm*
80011-2	1 mL	Calcein AM, 1 mg/mL in anhydrous DMSO	494/517 nm*
80011-3	20 x 50 ug	Calcein AM	494/517 nm*
30068-T	100 labelings	ViaFluor® 405 SE Cell Proliferation Kit	408/452 nm*
30068	1000 labelings		
30086-T	100 labelings	ViaFluor® 488 SE Cell Proliferation Kit	493/532 nm*
30086	1000 labelings		

* Hydrolyzed product

www.biotium.com
General Inquiries: order@biotium.com
Technical Support: techsupport@biotium.com
Phone: 800-304-5357

Biotium