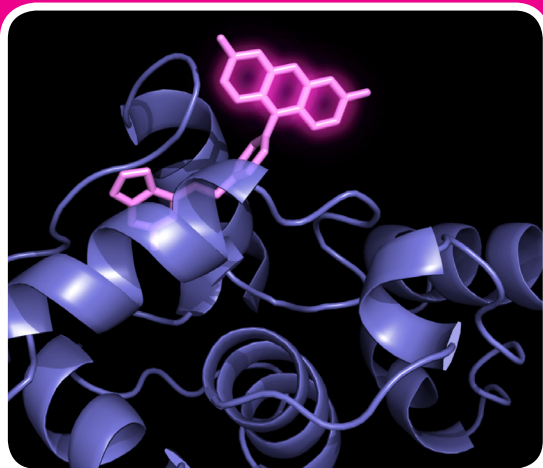


Small Ligand Labeling Kits

Featuring Mix-n-Stain™ Technology

Just Mix and then Stain - it really is that simple!



Easily conjugate fluorescent ligands for self-labeling tags or other small molecules

- Label ligands for SNAP-tag®, CLIP-tag™, HaloTag®, TMP-tag or other small molecules
- Label your own ligand for a fraction of the cost of a pre-labeled ligand
- Simple, rapid labeling with no purification step
- 15 bright & photostable CF® dyes from blue to near-IR
- Choose dyes for intracellular or surface staining of live cells
- Super-resolution compatible dye options

Mix-n-Stain™ CF® dye small ligand labeling kits are designed for rapid covalent labeling of low molecular weight (MW ~150-5,000), high affinity biological ligands. Simply mix your ligand with the buffer and CF® dye provided in the kit, a step that takes less than 2 minutes of hands-on time. After a short incubation and without further purification, you will have a covalently labeled conjugate that performs as well as synthetic fluorescent ligands from leading suppliers (Figure 1), at a fraction of the cost. Even without purification, CF® dye-labeled ligands do not show non-specific binding of cells or proteins (Figures 2-3). Suitable ligands or substrates include SNAP-tag®, CLIP-tag™, HaloTag® and TMP-tag ligands that have an aliphatic amine; other small molecules with a free amine group are also possible candidates for Mix-n-Stain™ ligand labeling.

A major advantage of the kits is our superior CF® dyes, which have exceptional brightness and photostability compared to Alexa Fluor® dyes and other next-generation dyes. Multiple CF® dyes have been verified in super-resolution and other specialized imaging applications (Table 1). We offer dyes spanning the visible and near-infrared spectra for cell surface or intracellular targets (Tables 1-2), for simple and economical multi-color detection (Figure 2).

World's Simplest Small Ligand Labeling Protocol

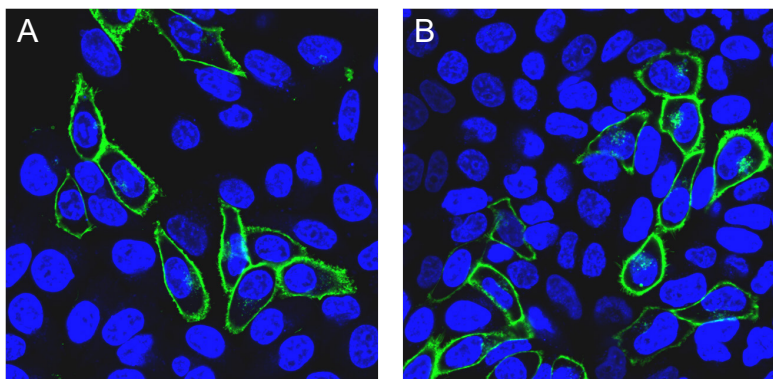
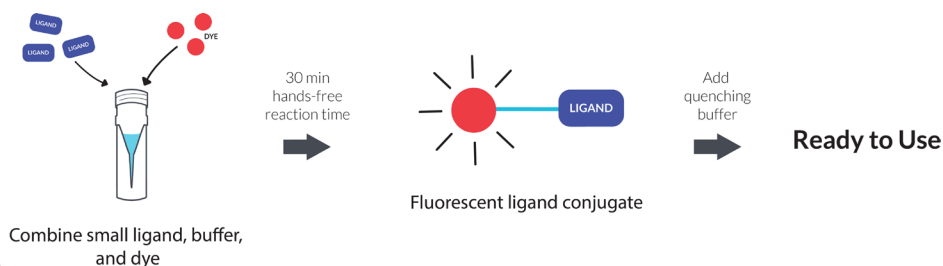


Figure 1. Live cell surface imaging of HeLa cells transfected with CLIP-tag™-NK1R fusion protein, detected with (A) CLIP-tag™ conjugated to green fluorescent CF@488A using Mix-n-Stain™, or (B) CLIP-Surface™ 488 ligand from New England Biolabs. Nuclei were stained with Hoechst.

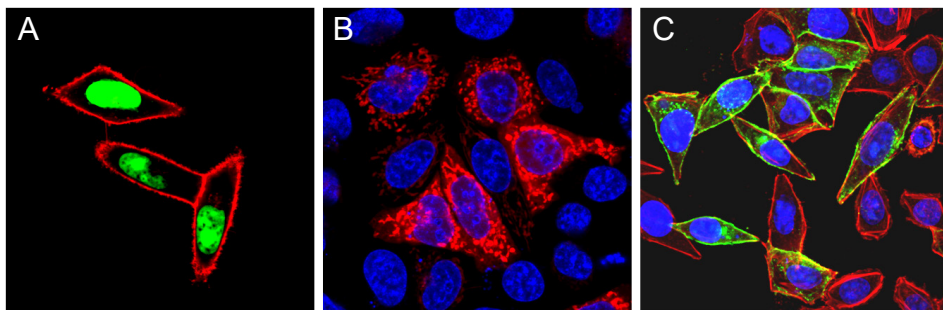


Figure 2. Versatility of Mix-n-Stain™-labeled ligands for multicolor live cell imaging. (A) CF@500-labeled CLIP-tag™ ligand was used to detect nuclear protein H2B (green), and CF@568-labeled SNAP-tag® ligand was used to detect cell surface protein ADRβ2. (B) CF@540-labeled CLIP-tag™ ligand was used to detect mitochondrial protein Cox8A in living cells (red); nuclei were stained with Hoechst 33342 (blue). (C) Three color imaging in fixed cells. CF@488A-CLIP-tag™ ligand was used to stain cell surface protein NK1R (green). Cells were then fixed and stained with CF@633 phalloidin (red) and mounted with EverBrite™ mounting medium with DAPI (blue).

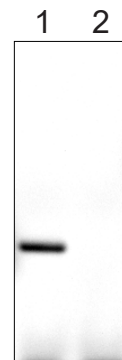


Figure 3. In-gel fluorescence analysis of CLIP-tag™ fusion protein detected with CF@488A-labeled ligand. In transfected CHO cell lysates (lane 1) the ligand-dye conjugate labels the receptor protein with high specificity. In untransfected cell lysate (lane 2) no protein labeling was detectable.

Table 1. Mix-n-Stain™ Small Ligand Labeling Kits for Cell Surface Targets

Cat. #	Dye	Abs/Em	Spectrally similar to	Specialized applications
92362	CF@405M	408/452 nm	Pacific Blue™, BD Horizon™ V450	SIM
92350	CF@488A	490/515 nm	FITC, Alexa Fluor® 488	STED, TIRF, 2-photon
92351	CF@568	562/583 nm	Rhodamine Red, Alexa Fluor® 568	STORM, SIM, TIRF
92352	CF@594	593/614 nm	Texas Red®, Alexa Fluor® 594	2-photon
92353	CF@633	630/650 nm	Cy@5, Alexa Fluor® 633	FIONA, gSHRImP, SMT, TIRF
92354	CF@640R	642/662 nm	Cy@5, Alexa Fluor® 647	FLImP, SIM, TIRF
92359	CF@647	650/665 nm	Cy@5, Alexa Fluor® 647	STORM
92360	CF@660C	667/685 nm	Alexa Fluor® 660	STORM
92361	CF@680	681/698 nm	Cy@5.5, Alexa Fluor® 660, IRDye®680LT	3D SMLM, STORM
92355	CF@680R	680/701 nm	Cy@5.5, Alexa Fluor® 660, IRDye®680LT	STORM, SMS, SMT, STED

FIONA: Fluorescence Imaging with One Nanometer Accuracy; FLImP: Fluorophore localization imaging with photobleaching; SHRImP: Single-molecule high-resolution imaging with photobleaching; SIM: Structured illumination microscopy; SMLM: Single molecule localization microscopy; SMS: Single molecule spectroscopy; SMT: Single molecule tracking; STED: Stimulated emission depletion; STORM: Stochastic optical reconstruction microscopy; TIRF: Total internal reflection fluorescence. Visit www.biotium.com for references.

Table 2. Mix-n-Stain™ Small Ligand Labeling Kits for Intracellular Targets

Cat. #	Dye	Abs/Em	Spectrally similar to
92356	CF@408	408/450 nm	Pacific Blue™, BD Horizon™ V450
92357	CF@500	500/510 nm	FITC, Alexa Fluor® 488
92358	CF@540	540/570 nm	Tetramethylrhodamine (TAMRA), TRITC, Alexa Fluor® 546, Cy@3
92364	CF@555	555/585 nm	Alexa Fluor® 555, Cy@3, TRITC
92363	CF@650	650/670 nm	Cy@5, Alexa Fluor® 647