Basic Fluorescent Proteins

Eight available colors for *in vivo* labeling, from blue to far-red

Easy detection by flow cytometry or fluorescent microscopy

No cofactors, substrate addition or chemical staining required

Special optimization for different applications, including:

- Labeling of cells, cell organelles and proteins of interest
- Gene expression analysis
- Multicolor labeling
- FRET-based studies of protein interaction
- Whole body imaging

Easy evaluation and adaptable license program for commercial use

Contents

Overview

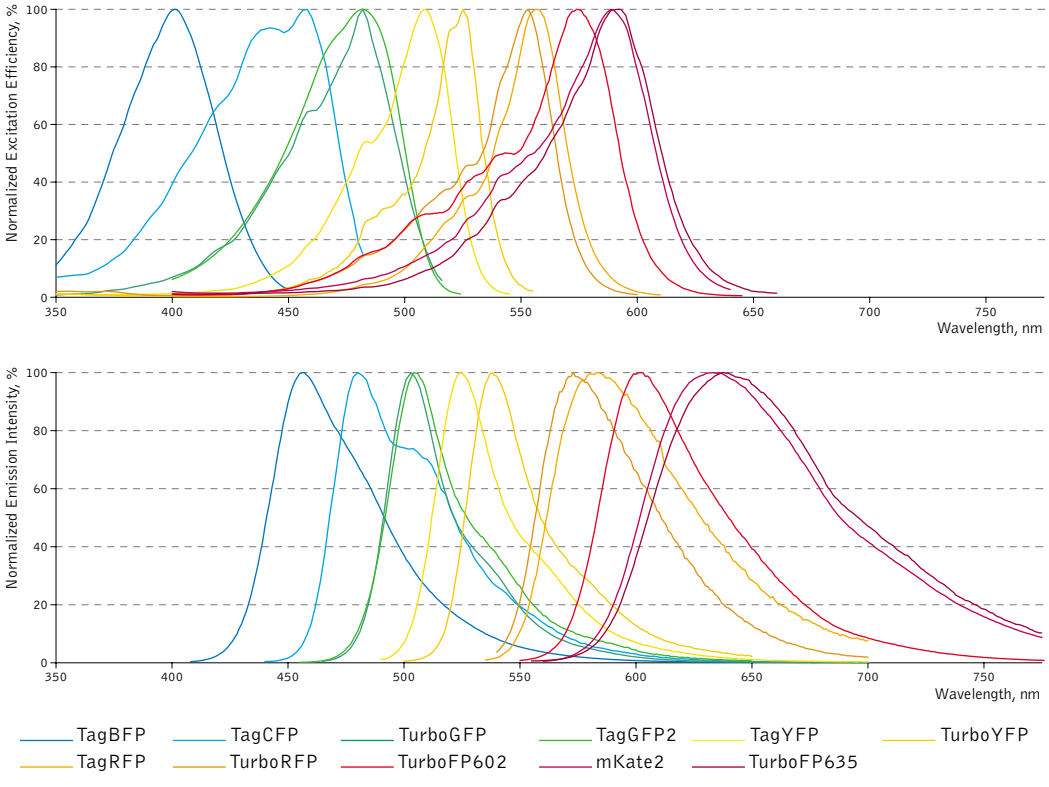
Evrogen offers a collection of bright fluorescent proteins (FPs) for wide range of applications in the field of live-cell assays. Evrogen fluorescent proteins can be used for *in vivo* protein localization and interaction studies; analysis of promoter activity in live cells; tracking subcellular organelles; labeling to identify and isolate specific populations of cells; generation of stably transfected cell lines, and more. Ranging in color from blue to far-red, Evrogen FPs allow visualization of multiple events simultaneously by both fluorescent microscopy and flow-cytometry.

All Evrogen FPs are improved by mutagenesis and codon usage optimization for high expression level in mammalian cells and fast maturation at 37°C. Variants with codon usage optimized for expression in other heterological systems are available or can be generated by request. Evrogen FPs have been used successfully in prokaryotes, yeasts, animals, and plants. The proteins possess bright stable fluorescence allowing monitoring of target cells or proteins over extended period of time. No addition of cofactors or substrates is required for FP detection.

Evrogen technologies embodied in basic FPs are available for expanded and commercial use with an adaptable licensing program.

Whole body imaging in transgenic *Xenopus laevis* using Evrogen far-red fluorescent protein TurboFP635. Photographs were kindly provided by Dr. A. Zaraisky (Institute of Bioorganic Chemistry RAS, Moscow, Russia).

Spectral diversity of Evrogen basic FPs



Basic fluorescent proteins available

Protein	Color	Ex/Em, nm	Brightness, % of EGFP	pKa	Photostability	Structure	M.W., kDa	Filter Sets
TagFPs								
TagBFP	blue	402/457	99	2.7	high	monomer	26	Omega XF119-2, QMAX-Blue; Semrock DAPI-5060B
TagCFP	cyan	458/480	64	4.7	high	monomer	27	common sets for ECFP, e.g. Omega XF114-2 and XF130-2
TagGFP2	green	483/506	105	5.0	high	monomer	27	common sets for EGFP, FITC, e.g. Omega QMAX-Green, XF100-2, XF100-3, XF115-2, and XF116-2
TagYFP	yellow	508/524	94	5.5	high	monomer	27	Omega XF104-3, XF105-2; C.T. 41028 Yellow GFP BP (10C/Topaz)
TagRFP	red (orange)	555/584	148	3.8	medium	monomer	27	TRITC filter sets, e.g. Omega QMAX-Yellow, XF108-2, XF101-2, XF111-2
mKate2	far red	588/633	74	5.4	high	monomer	26	Texas Red filter sets, e.g. Omega QMAX-Red, XF102-2
TurboFPs								
TurboGFP	green	482/502	112	5.2	high	dimer	26	common sets for EGFP, FITC, e.g. Omega QMAX-Green, XF100-2, XF100-3, XF115-2, and XF116-2
TurboYFP	yellow	525/538	169	5.9	high	dimer	26	Omega XF104-3; C.T. 42003 (ZsYellow1)
TurboRFP	red (orange)	553/574	187	4.4	high	dimer	26	TRITC filter sets, e.g. Omega QMAX-Yellow, XF108-2, XF101-2, XF111-2
TurboFP602	red	574/602	79	4.7	medium	dimer	26	Omega QMAX-Red
TurboFP635	far red	588/635	67	5.5	high	dimer	26	Texas Red filter sets, e.g. Omega QMAX-Red, XF102-2

M.W. - molecular weight; Ex/Em - excitation/emission maxima

C.T. - Chroma Technology Corp. (www.chroma.com); Omega - Omega Optical (www.omegafilters.com); Semrock (www.semrock.com)

Evrogen FPs are divided into subgroups according to their properties and recommended applications:

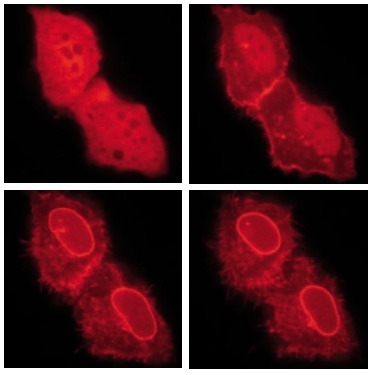
TagFPs (see page 11)

The group comprises monomeric fluorescent proteins specially optimized for protein localization and interaction studies. Successful TagFPs performance in protein labeling applications was validated in various models including highly oligomerizing cellular proteins like α-tubulin, β-actin, vinculin, zyxin, etc.

TurboFPs (see page 26)

The group comprises dimeric fluorescent proteins specially recommended for applications requiring fast appearance of bright fluorescence, including cell labeling and tracking promoter activity. Despite their dimeric structure, TurboFPs can be fused with subcellular localization signals for labeling of cellular organelles.

Evrogen offers various source and expression vectors encoding fluorescent proteins alone or in fusion with cellular proteins and localization signals. All available vector types are described in details in the section "Expression and source vectors" on page 39. Custom optimization of Evrogen vectors and proteins for your particular needs is available upon request (please see page 135 for details).



Spinning disk confocal imaging of mKate2 fused to human annexin A4 in HeLa cells during translocation from the cytoplasm to the plasma and nuclear membranes upon induction with ionomycin.

Performance of basic fluorescent proteins in different applications

Protein	Cell labeling	Fusion generation	Promoter activity testing	Whole body imaging	Acidic organelle labeling	Generation of stably transfected cell lines
TagFPs						
TagBFP	+++	++++	++++	-	++++	not tested
TagCFP	+++	+++	+++	-	+++	proved
TagGFP2	++++	++++	++++	+	++	proved**
TagYFP	+++	+++	+++	+	++	proved
TagRFP	++++	++++	+++	+++	++++	proved
mKate2	+++	++++	++++	++++	++	proved**
TurboFPs						
TurboGFP	++++	+	++++	+	++	proved
TurboYFP	++++	++	++++	+	+	not tested*
TurboRFP	++++	+	++++	+++	+++	not tested
TurboFP602	+++	+	+++	+++	+++	proved
TurboFP635	+++	+	++++	++++	++	proved

The performance is estimated basing on reporters properties. Not all reporters have been tested experimentally in each application.

* Being overexpressed in long-term culture of cells with high expression levels, TurboYFP shows slight tendency to aggregate. It might limit TurboYFP use in such experimental systems. Please use PhiYFP or PhiYFPm proteins for stable expression (see page 30)

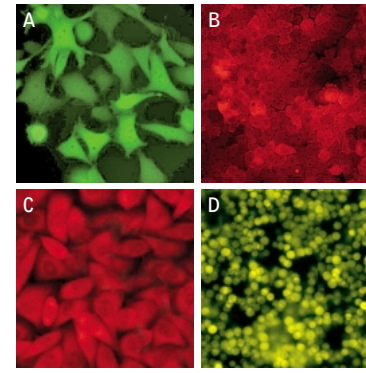
** The suitability for stable cell lines generation was proved for TagGFP and TagFP635, the parental variants of TagGFP2 and mKate2.

Cell labeling

Super bright fluorescence and absence of cytotoxic effects make Evrogen fluorescent proteins ideal for labeling living cells. Because of distinctive spectra, Evrogen FPs can be readily multiplexed, i.e. combined for the simultaneous detection of several events in a cell population. All proteins from Evrogen collection are suitable for cell labeling. We recommend TurboFPs since it is not necessary to use monomeric fluorescent proteins for this application, while the dimeric FPs often provide brighter and more stable signal.

Recommended products for cell labeling:

Product(s)	Pages
TurboFPs	26



Cell labeling using Evrogen TurboFPs.
(A) Stably transfected H-TG cells expressing TurboGFP; (B) stably transfected U-2-OS cells expressing TurboFP602; (C) stably transfected T-24 cells expressing TurboFP635; (D) stably transfected W-PY cells expressing PhiYFP*.
Photographs of stably transfected cell lines were kindly provided by Dr. Christian Petzelt (Marinpharm). *PhiYFP is a parental version of TurboYFP.

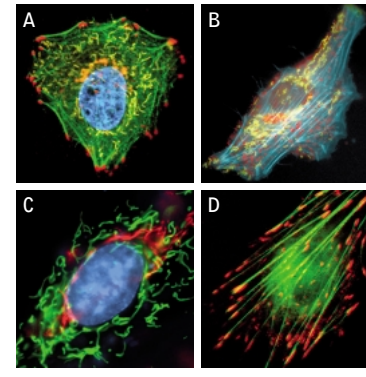
Labeling of cellular proteins and organelles

Monomeric TagFPs are the optimal choice for generation of fusions with proteins and subcellular localization signals. Successful performance of TagFPs in protein labeling applications was validated in various models including highly oligomerizing cellular proteins like cytoplasmic β-actin, α-tubulin, vinculin, zyxin, etc.

Evrogen mammalian expression vectors comprise convenient multiple cloning sites allowing easy generation of fusions of interest. Ready-to-use subcellular localization vectors for fluorescent labeling of various cellular organelles and proteins are available.

Recommended products for labeling of proteins and subcellular structures:

Product(s)	Pages
TagFPs	11
Ready-to use subcellular localization vectors	50



Multicolor labeling of subcellular structures in transiently transfected mammalian cells using Evrogen TagFPs.
(A) TagBFP-histone H2B fusion (blue), TagGFP-actin fusion (green), mitochondria-targeted PhiYFP (yellow), golgi-targeted TagRFP (orange), mKate2-zyxin fusion (red); (B) TagCFP-actin fusion (cyan), mitochondria-targeted PhiYFP (yellow), and mKate2-clathrin fusion (red) in HeLa cells; (C) TagRFP-cytokeratin 14 fusion (red) and mitochondria-targeted TagGFP2 (green) in REF3 cells with Hoechst staining (blue); (D) TagGFP2-actin fusion (green) and mKate2-zyxin fusion (red) in REF52 cells.

Generation of stably transfected cell lines

Most Evrogen fluorescent proteins have been successfully tested in stable transfection experiments. Various cell lines expressing Evrogen FPs are commercially available from Marinpharm GmbH (www.marinpharm.com).

Acidic organelle labeling

Many Evrogen fluorescent proteins are characterized by high pH stability, the most stable are TagBFP (pKa=2.7) and TagRFP (pKa=3.8). The resistance to low pH makes it possible to use these reporters for imaging in acidic organelles, such as late and recycling endosomes and lysosomes.

Recommended products for acidic organelle labeling:

Product(s)	Pages
TagBFP	12
TagRFP	20

FRET applications

Ranging in color from blue to far-red, Evrogen fluorescent proteins can be used in fluorescence resonance energy transfer (FRET) applications as donors and acceptors of fluorescence. TagFPs are perfect for *in vivo* protein interaction studies by FRET due to their improved performance in fusions. TagBFP-TagGFP2 and TagGFP2-TagRFP pairs show the highest FRET efficiency among the tested TagFPs combinations and compare favorably to other available FRET pairs of monomeric fluorescent proteins.

Recommended products for FRET application:

Product(s)	Pages
TagBFP	12
TagGFP2	16
TagRFP	20

Recommended FRET pairs are:

donor, ex/em	acceptor, ex/em
TagBFP ex/em: 402/457	TagGFP2 ex/em: 483/506
TagGFP2 ex/em: 483/506	TagRFP ex/em: 555/584

Monitoring of promoter activity

Early detection of the promoter activity onset requires reporters providing for maximally bright and fast appearing signal. All TurboFPs and some TagFPs (namely TagBFP, TagGFP2, and mKate2) perfectly meet these requirements demonstrating superior brightness and maturation speed. The monitoring of rapid changes in gene regulation can be done using the destabilized TurboFPs variants characterized by short protein half-life.

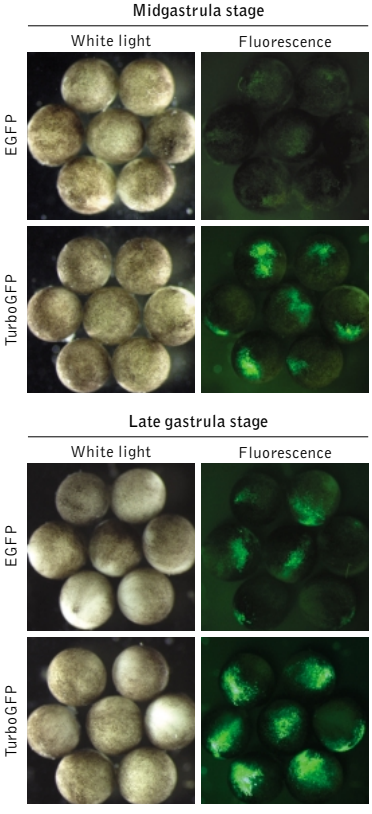
Evrogen offers promoterless vectors encoding unmodified and destabilized TurboFPs. In each vector, multiple cloning sites (MCS) located upstream of the reporter sequence can be used to clone a promoter or a promoter/enchancer combination of interest. Destabilized variants are generated by fusion of residues 422-461 of mouse ornithine decarboxylase (MODC) to the TurboFPs C-termini. MODC region contains a PEST amino acid sequence that targets the protein for degradation and provides for rapid protein turnover [Li et al. 1998]. Destabilized TurboFPs retain fluorescent properties of the native proteins and have a half-life of approximately 1-1.5 hrs, as measured by fluorescence intensity of cells treated with the protein synthesis inhibitor, cycloheximide.

Recommended products for *in vivo* testing promoter activity:

Product(s)	Pages
TurboFPs	26
TagBFP	12
TagGFP2	16
mKate2	23
Promoterless vectors	52

Whole body imaging

Deep tissue imaging using the fluorescent proteins allows direct and non-invasive observation of the biological processes inside the living organisms. Importantly, main photon absorbers within the visual spectrum in animal tissues are melanin and hemoglobin. Wavelengths longer than 1100 nm are absorbed by water. In addition, light-scattering intensity drops off as the wavelength increases.

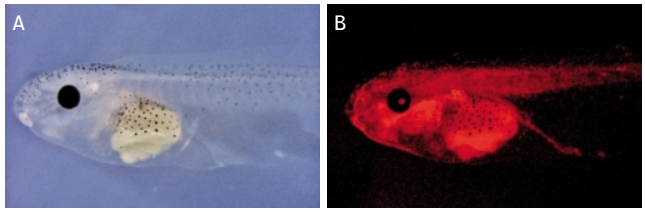


***In vivo* comparison of TurboGFP and EGFP maturation in developing *Xenopus* embryos.**
Vectors expressing the respective fluorescent proteins under the control of CMV promoter were microinjected into animal poles of *Xenopus* embryos at the stage of two blastomeres. Living embryos were then photographed from the animal pole at the middle and late gastrula stages. Experimental data were presented by Dr. A. Zaraisky (Institute of Bioorganic Chemistry RAS, Moscow, Russia).

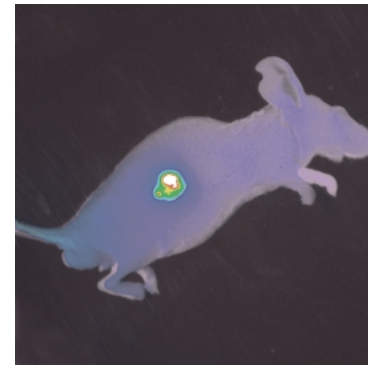
The "optical window" for the visualization in living tissues is approximately 650-1100 nm. Within this optical window, TurboFP635 and mKate2 are the brightest fluorescent proteins available so far. Experimental studies show that the signal coming from TurboFP635 located deep inside tissue is about 45 times stronger than the signal from EGFP and 2 times stronger than the signal from mRaspberry, the closest far-red fluorescent protein tested [Deliolanis et al. 2008]. Together with excellent photostability and fast maturation it makes TurboFP635 and mKate2 the proteins of choice for whole body imaging.

Recommended products for whole body imaging:

Product(s)	Pages
TurboFP635	37
mKate2	23



Expression of mKate2-zyxin in *Xenopus laevis* embryos.
To test in an embryonic model the performance of mKate2 in a targeting protein fusion, transgenic *Xenopus laevis* embryos were generated bearing a mKate2-zyxin fusion construct under the control of the CMV promoter. Despite quite extensive and ubiquitous expression of mKate2-zyxin, these embryos appear normal and healthy indicating that mKate2 exerts a low toxic effect on living cells in transgenic organisms. (A) White light; (B) fluorescence. Images from Shcherbo et al. 2009.



Melanoma implant expressing TurboFP635 in mouse xenograft model.
KODAK *In-Vivo* Imaging System FX. Image was kindly provided by ChemDiv Inc.

REFERENCES

Deliolanis et al. (2008). J. Biomed. Opt., 13 (4): 044008 / pmid: 19021336

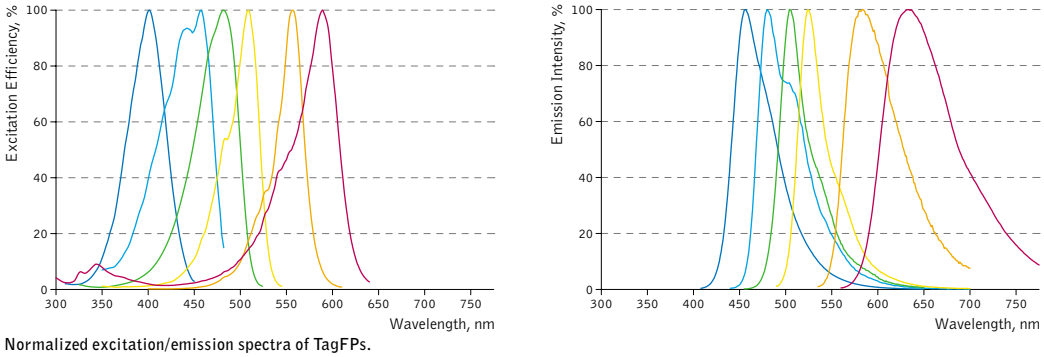
Li et al. (1998). J Biol Chem, 273 (52): 34970–34975 / pmid: 9857028

Shcherbo et al. (2009). Biochemical Journal, 418 (3): 567–574 / pmid: 19143658

TagFPs

Monomeric tags for protein labeling

TagFPs are monomeric fluorescent proteins specially designed for generation of fusions. Six available colors allow multi-color labeling of different cellular proteins for protein localization and interaction studies.



Normalized excitation/emission spectra of TagFPs.
TagBFP - blue line, TagCFP - cyan line, TagGFP2 - green line, TagYFP - yellow line, TagRFP - orange line, mKate2 - dark-red line.

Main properties of TagFPs:

Protein	TagBFP	TagCFP	TagGFP2	TagYFP	TagRFP	mKate2
Molecular weight, kDa	26	27	27	27	27	26
Polypeptide length, aa	233	239	238	239	237	232
Fluorescence color	blue	cyan	green	yellow	red (orange)	far-red
Excitation maximum, nm	402	458	483	508	555	588
Emission maximum, nm	457	480	506	524	584	633
Quantum yield	0.63	0.57	0.60	0.62	0.48	0.40
Extinction coefficient, $M^{-1}cm^{-1}$	52 000	37 000	56 500	50 000	100 000	62 500
Brightness*	32.8	21.1	33.9	31.0	48.0	25.0
Brightness, % of EGFP	99	64	105	94	148	74
pKa	2.7	4.7	5.0	5.5	3.8	5.4
Structure	monomer	monomer	monomer	monomer	monomer	monomer
Aggregation	no	no	no	no	no	no
Maturation rate at 37°C	fast	fast	fast	fast	fast	fast
Photostability	high	high	high	high	medium	high
Cell toxicity	not observed	not observed	not observed	not observed	not observed	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.

Blue fluorescent protein TagBFP

- Bright blue fluorescence
- Monomeric protein with successful performance in fusions
- Fast maturation, high photostability
- Extremely high pH-stability
- Recommended for protein labeling, acidic organelle labeling, FRET applications

TagBFP (scientific name mTagBFP) is a monomeric blue fluorescent protein generated by site-specific and random mutagenesis of TagRFP [Subach et al. 2008]. TagBFP possesses bright blue fluorescence with excitation/emission maxima at 402 and 457 nm, characterized by high photostability and extremely high pH-stability.

Compared to EBFP2 [Ai et al. 2007], TagBFP is more then 1.8 times brighter, much more pH-stable and has twice shorter maturation half-time at 37°C. Narrow fluorescence emission peak of TagBFP provides for accurate and easy spectral separation with cyan and green fluorescent proteins and makes it a preferable tag for multicolor labeling.

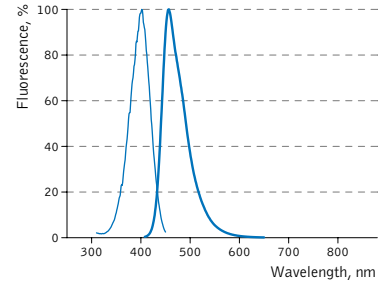
Good overlap between the emission spectrum of TagBFP and the absorbance spectra of TagGFP2 (page 16) allows using these two proteins as a FRET pair. The calculated Forster distance (R_0 = 5.25 nm) for the TagBFP / TagGFP2 pair is larger than those reported for the standard ECFP-EYFP and mCyPet-mYPet pairs (R_0 = 4.86 nm and 4.93 nm correspondingly), suggesting that TagBFP / TagGFP2 is one of the best among available FRET pairs of monomeric fluorescent proteins. High efficiency of TagBFP as a FRET donor was demonstrated in living cells by Subach et al. [Subach et al. 2008].

TagBFP can be easily expressed and detected in a wide range of organisms. Mammalian cells transiently transfected with TagBFP expression vectors

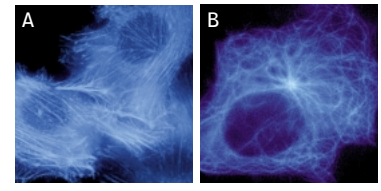
Main properties of TagBFP

Characteristic	
Molecular weight, kDa	26
Polypeptide length, aa	233
Fluorescence color	blue
Excitation maximum, nm	402
Emission maximum, nm	457
Quantum yield	0.63
Extinction coefficient, $M^{-1}cm^{-1}$	52 000
Brightness*	32.8
Brightness, % of EGFP	99
pKa	2.7
Structure	monomer
Aggregation	no
Maturation rate at 37°C	fast
Photostability	high
Cell toxicity	not observed

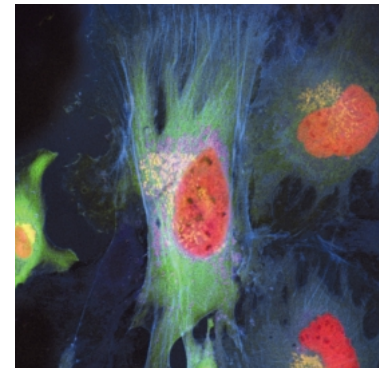
*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TagBFP normalized excitation (thin line) and emission (thick line) spectra.
Complete TagBFP spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



HeLa cells expressing TagBFP fusion proteins.
(A) Confocal microscopy of TagBFP fusion with β-actin in transiently transfected HeLa cells; (B) confocal microscopy of TagBFP fusion with cytoplasmic α-tubulin in transiently transfected HeLa cells.



TagBFP use in multicolor labeling of mammalian cells. TagYFP - tagged α-tubulin (green), TagCFP - tagged β-actin (cyan), mitochondria-targeted TagBFP (magenta), Golgi-targeted TagRFP(yellow), and TagFP635-H2B fusion (red).

give bright fluorescent signals within 10-12 hrs after transfection. No cytotoxic effects or visible protein aggregation are observed.

TagBFP performance in fusions has been demonstrated in the β-actin and α-tubulin models.

TagBFP can be used in multicolor labeling applications with other fluorescent proteins of green, yellow, red, and far-red colors.

High pH-stability with pKa=2.7 makes it possible to use TagBFP for imaging in acidic organelles, such as late and recycling endosomes and lysosomes.

Recommended filter sets and antibodies

TagBFP can be recognized using Anti-tRFP antibody (Cat.# AB231-AB232) available from Evrogen.

The protein can be detected using common fluorescence filter sets for BFP, DAPI, and other blue dyes.

Recommended filter sets are: XF119-2*, QMAX-Blue*, XF131, XF06, XF13-2, XF03, XF11, XF129-2, XF05-2 (Omega Optical); DAPI-5060B* and DAPI-1160A (Semrock); 31037, 31041, 31016*, 31021, 31000v2, 1009v2, 31013v2, 11005v2, 31047 (Chroma Technology Corp.).
* - preferred filter sets

REFERENCES

Ai et al. (2007). Biochemistry, 46 (20): 5904–5910 / pmid: 17444659

Subach et al. (2008). Chemistry & Biology, 15 (10): 1116–1124 / pmid: 18940671

TagBFP-related products

Product	Cat.#	Description	Size	Page(s)
TagBFP expression/source vectors				
pTagBFP-C	FP171	Mammalian expression vector encoding humanized TagBFP and allowing its expression and generation of fusions to the TagBFP C-terminus	20 μg	43
pTagBFP-N	FP172	Mammalian expression vector encoding humanized TagBFP and allowing its expression and generation of fusions to the TagBFP N-terminus	20 μg	45
pTagBFP-actin	FP174	Mammalian expression vector encoding humanized TagBFP fused with human cytoplasmic β-actin	20 μg	50
pTagBFP-tubulin	FP175	Mammalian expression vector encoding humanized TagBFP fused with human α-tubulin	20 μg	50
pTagBFP-H2B	FP176	Mammalian expression vector encoding humanized TagRFP fused with human histone H2B	20 μg	51
Vector sets				
Fusion Blue	FPF20	Mammalian expression vectors encoding TagBFP for its expression and fusion generation: pTagBFP-N and pTagBFP-C	20 μg each	45, 43
Antibodies against TagBFP				
Anti-tRFP	AB231 AB232	Rabbit polyclonal antibody against TurboRFP, TurboFP602, TurboFP635, TagBFP, TagRFP, TagFP635, and mKate2	100 μg 200 μg	104

Please contact your local distributor for exact prices and delivery information.

Cyan fluorescent protein TagCFP

- Bright cyan fluorescence
- Monomeric protein with successful performance in fusions
- Fast maturation, high pH-stability and photostability
- Proven suitability to generate stably transfected cell lines
- Recommended for protein labeling

TagCFP is a cyan monomeric protein generated on the basis of the wild-type GFP-like protein from jellyfish *Aequorea macrodactyla* [Xia et al. 2002]. It possesses bright fluorescence with excitation/emission maxima at 458 and 480 nm, respectively. TagCFP is significantly brighter than commonly used ECFP.

TagCFP is mainly intended for protein labeling in protein localization and interaction studies. It can also be used for cell and organelle labeling and for tracking the promoter activity.

TagCFP can be easily expressed and detected in a wide range of organisms. Mammalian cells transiently transfected with TagCFP expression vectors give bright fluorescent signals within 10-12 hrs after transfection. No cytotoxic effects or visible protein aggregation are observed.

TagCFP performance in fusions has been demonstrated in human cytoplasmic β-actin, α-tubulin, and mitochondria models.

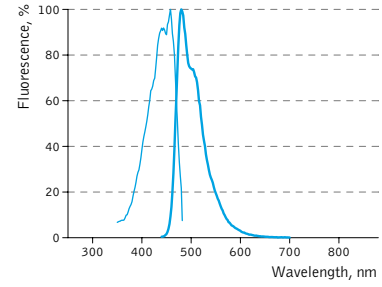
TagCFP suitability to generate stably transfected cells has been proven by Marinpharm company. A cell line expressing TagCFP fusion with mitochondrial targeting sequence (MTS) is commercially available.

TagCFP can be used in multicolor labeling applications with green, yellow, red, and far-red fluorescent dyes.

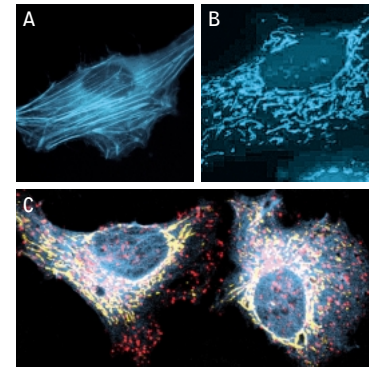
Main properties of TagCFP

Characteristic	
Molecular weight, kDa	27
Polypeptide length, aa	239
Fluorescence color	cyan
Excitation maximum, nm	458
Emission maximum, nm	480
Quantum yield	0.57
Extinction coefficient, $M^{-1}cm^{-1}$	37 000
Brightness*	21.1
Brightness, % of EGFP	64
pKa	4.7
Structure	monomer
Aggregation	no
Maturation rate at 37°C	fast
Photostability	high
Cell toxicity	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TagCFP normalized excitation (thin line) and emission (thick line) spectra.
Complete TagCFP spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



Expression of TagCFP fusions in mammalian cells. (A) Transiently transfected HeLa cells expressing TagCFP-tagged β-actin; (B) stably transfected U-205 cells expressing mitochondria-targeted TagCFP; (C) TagCFP use in multicolor labeling of HeLa cells: TagCFP-tagged α-tubulin (cyan), TagFP635-clathrin fusion (red), mitochondria-targeted TagYFP (yellow). Image was kindly provided by Michael W. Davidson (Florida State University).

Recommended filter sets and antibodies

TagCFP can be recognized using Anti-Tag(CGY)FP antibody (Cat.# AB121-AB122) available from Evrogen.

TagCFP can be detected using fluorescence filter sets for ECFP and the similar. Recommended Omega Optical filter sets are XF114-2 and XF130-2.

REFERENCES

Xia et al. (2002). Mar Biotechnol (NY), 4 (2): 155–162 / pmid: 14961275

TagCFP-related products

Product	Cat.#	Description	Size	Page(s)
TagCFP expression/source vectors				
pTagCFP-C	FP111	Mammalian expression vector encoding humanized TagCFP and allowing its expression and generation of fusions to the TagCFP C-terminus	20 μg	43
pTagCFP-N	FP112	Mammalian expression vector encoding humanized TagCFP and allowing its expression and generation of fusions to the TagCFP N-terminus	20 μg	45
pTagCFP-actin	FP114	Mammalian expression vector encoding humanized TagCFP fused with human cytoplasmic β-actin	20 μg	50
pTagCFP-tubulin	FP115	Mammalian expression vector encoding humanized TagCFP fused with human α-tubulin	20 μg	50
pTagCFP-mito	FP117	Mammalian expression vector encoding humanized TagCFP targeted to mitochondria	20 μg	47
Vector sets				
Fusion Cyan	FPF11	Mammalian expression vectors encoding TagCFP for its expression and fusion generation: pTagCFP-N and pTagCFP-C	20 μg each	43, 45
Antibodies against TagCFP				
Anti-Tag(CGY)FP	AB121 AB122	Rabbit polyclonal antibody against TagCFP, TagGFP, TagYFP, PS-CFP2, and EGFP	100 μg 200 μg	101

Please contact your local distributor for exact prices and delivery information.

Green fluorescent protein TagGFP2

- Bright green fluorescence
- Monomeric protein with successful performance in fusions
- Fast maturation, high pH-stability and photostability
- Proven suitability to generate stably transfected cell lines
- Recommended for protein labeling and FRET applications

TagGFP2 (scientific name mTagGFP) is the improved variant of TagGFP, the mutant of the *Aequorea macrodactyla* GFP-like protein [Xia et al. 2002, Subach et al. 2008]. TagGFP2 possesses bright green fluorescence with excitation/emission maxima at 483 and 506 nm, respectively.

TagGFP2 matures 1.6-fold faster than TagGFP and is characterized by the improved performance in fusions. Compared to EGFP, TagGFP2 provides about the same brightness of fluorescence but is significantly more pH stable. TagGFP2 is specially optimized for expression at 37°C.

Because of monomeric nature, TagGFP2 is mainly intended for protein localization studies and expression in long-term cell cultures. In FRET applications, TagGFP2 can be used as a donor for red fluorescent protein TagRFP (see page 20) or as an acceptor for blue fluorescent protein TagBFP (see page 12).

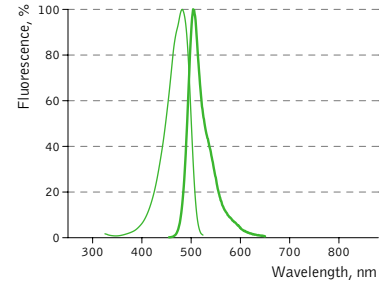
TagGFP2 can be easily expressed and detected in a wide range of organisms. Mammalian cells transiently transfected with TagGFP2 expression vectors give bright fluorescent signals within 10-12 hrs after transfection. No cytotoxic effects or visible protein aggregation are observed.

TagGFP2 performance in fusions has been demonstrated in the β-actin, α-tubulin and mitochondria-targeting signal models. It can be used in multicolor labeling applications with cyan, yellow, red, and far-red fluorescent dyes.

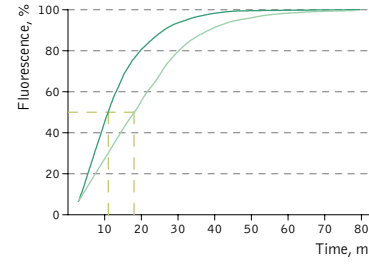
Main properties of TagGFP2

Characteristic	
Molecular weight, kDa	27
Polypeptide length, aa	238
Fluorescence color	green
Excitation maximum, nm	483
Emission maximum, nm	506
Quantum yield	0.6
Extinction coefficient, $M^{-1}cm^{-1}$	56 500
Brightness*	33.9
Brightness, % of EGFP	105
pKa	5.0
Structure	monomer
Aggregation	no
Maturation rate at 37°C	fast
Photostability	high
Cell toxicity	not observed

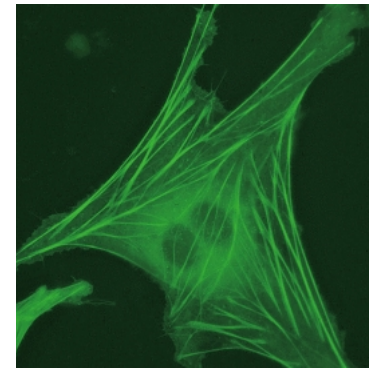
*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TagGFP2 normalized excitation (thin line) and emission (thick line) spectra.
Complete TagGFP2 spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



Maturation curves for TagGFP2 and parental TagGFP.
Color dashed lines indicate maturation half-times of 11 min and 18 min for TagGFP2 (dark green curve) and TagGFP (light green curve), respectively. Recording of protein maturation was started when about 7% from their maximal fluorescence has been detected. Time point "0" was defined using an approximation of the beginning of the maturation curves with straight lines.
Data from Subach et al. 2008.



Transiently transfected REF-52 cells expressing TagGFP2-tagged β-actin.

Recommended filter sets and antibodies

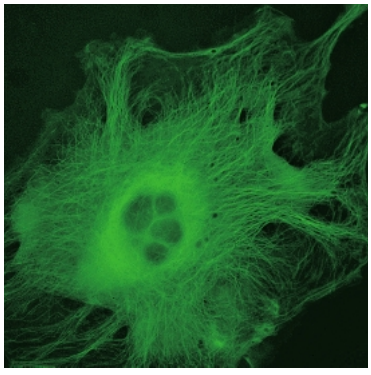
The protein can be recognized using Anti-Tag(CGY)FP antibody (Cat.# AB121-AB122) available from Evrogen.

TagGFP2 can be detected using common fluorescence filter sets for EGFP, FITC, and other green dyes. Recommended Omega Optical filter sets are QMAX-Green, XF100-2, XF100-3, (XF115-2), and XF116-2.

REFERENCES

Subach et al. (2008). Chemistry & Biology, 15 (10): 1116–1124 / pmid: 18940671

Xia et al. (2002). Mar Biotechnol (NY), 4 (2): 155–162 / pmid: 14961275



Transiently transfected REF-52 cells expressing TagGFP2-tagged α-tubulin.

TagGFP2-related products

Product	Cat.#	Description	Size	Page(s)
TagGFP2 expression/source vectors				
pTagGFP2-C	FP191	Mammalian expression vector encoding humanized TagGFP2 and allowing its expression and generation of fusions to the TagGFP2 C-terminus	20 μg	43
pTagGFP2-N	FP192	Mammalian expression vector encoding humanized TagGFP2 and allowing its expression and generation of fusions to the TagGFP2 N-terminus	20 μg	45
pTagGFP2-actin	FP194	Mammalian expression vector encoding humanized TagGFP2 fused with human cytoplasmic β-actin	20 μg	50
pTagGFP2-tubulin	FP195	Mammalian expression vector encoding humanized TagGFP2 fused with human α-tubulin	20 μg	50
pTagGFP2-mito	FP197	Mammalian expression vector encoding humanized TagGFP2 targeted to mitochondria	20 μg	47
Vector sets				
Fusion Green	FPF22	Mammalian expression vectors encoding TagGFP2 for its expression and fusion generation: pTagGFP-N and pTagGFP-C	20 μg each	43, 45
Antibodies against TagGFP2				
Anti-Tag(CGY)FP	AB121 AB122	Rabbit polyclonal antibody against TagCFP, TagGFP, TagGFP2, TagYFP, PS-CFP2, and EGFP	100 μg 200 μg	101

Please contact your local distributor for exact prices and delivery information.

Yellow fluorescent protein TagYFP

- Bright yellow fluorescence
- Monomeric protein with successful performance in fusions
- Fast maturation, high pH-stability and photostability
- Proven suitability to generate stably transfected cell lines
- Recommended for protein labeling

TagYFP is a monomeric yellow fluorescent protein developed on the basis of GFP-like protein from jellyfish *Aequorea macrodactyla* [Xia et al. 2002]. TagYFP possesses single excitation maximum at 508 nm, and emission maximum at 524 nm. TagYFP is more pH stable than EYFP.

TagYFP is mainly intended for protein labeling in protein localization and interaction studies. It can also be used for cell and organelle labeling and for tracking the promoter activity, although TurboYFP and Phi-Yellow proteins are preferable for such applications because they mature faster and give brighter fluorescent signal (see page 30).

TagYFP can be easily expressed and detected in a wide range of organisms. Mammalian cells transiently transfected with TagYFP expression vectors give bright fluorescent signals within 10-12 hrs after transfection. No cytotoxic effects or visible protein aggregation are observed.

TagYFP performance in fusions has been demonstrated in human cytoplasmic β-actin and α-tubulin models. An expected pattern of fluorescence has been obtained in each case.

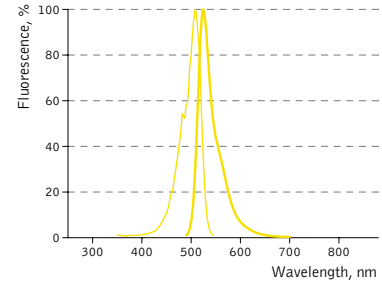
TagYFP suitability to generate stably transfected cells has been proven by Marinpharm company. Cell lines expressing TagYFP fusions are commercially available.

TagYFP can be used in multicolor labeling applications with blue, cyan, green, red, and far-red fluorescent dyes.

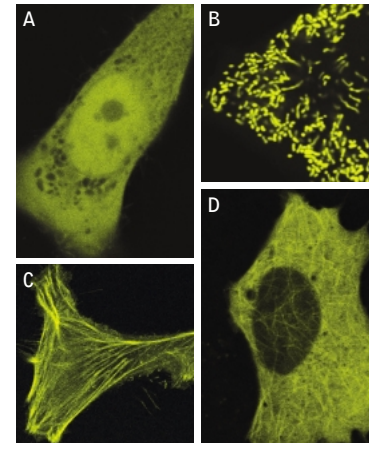
Main properties of TagYFP

Characteristic	
Molecular weight, kDa	27
Polypeptide length, aa	239
Fluorescence color	yellow
Excitation maximum, nm	508
Emission maximum, nm	524
Quantum yield	0.62
Extinction coefficient, $M^{-1}cm^{-1}$	50 000
Brightness*	31
Brightness, % of EGFP	94
pKa	5.5
Structure	monomer
Aggregation	no
Maturation rate at 37°C	fast
Photostability	high
Cell toxicity	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TagYFP normalized excitation (thin line) and emission (thick line) spectra.
Complete TagYFP spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



TagYFP expression in mammalian cells.
(A) Confocal microscopy of cytoplasmic TagYFP expression in transiently transfected human HeLa cells; (B) confocal microscopy of mitochondria-targeted TagYFP expression in transiently transfected HeLa cells; (C) confocal microscopy of TagYFP fusion with the cytoplasmic β-actin in transiently transfected 3T3 cells; (D) confocal microscopy of TagYFP fusion with the α-tubulin in transiently transfected 3T3 cells.

Recommended filter sets and antibodies

The protein can be recognized using Anti-Tag(CGY)FP antibody (Cat.# AB121-AB122) available from Evrogen.

Recommended Omega Optical filter sets for TagYFP are XF104-3 and XF105-2. It can also be detected using Chroma Technology Corp. filter set 41028 Yellow GFP BP (10C/Topaz) or the similar.

REFERENCES

Xia et al. (2002). Mar Biotechnol (NY), 4 (2): 155–162 / pmid: 14961275

TagYFP-related products

Product	Cat.#	Description	Size	Page(s)
TagYFP expression/source vectors				
pTagYFP-C	FP131	Mammalian expression vector encoding humanized TagYFP and allowing its expression and generation of fusions to the TagYFP C-terminus	20 μg	43
pTagYFP-N	FP132	Mammalian expression vector encoding humanized TagYFP and allowing its expression and generation of fusions to the TagYFP N-terminus	20 μg	45
pTagYFP-actin	FP134	Mammalian expression vector encoding humanized TagYFP fused with human cytoplasmic β-actin	20 μg	50
pTagYFP-tubulin	FP135	Mammalian expression vector encoding humanized TagYFP fused with human α-tubulin	20 μg	50
pTagYFP-mito	FP137	Mammalian expression vector encoding humanized TagYFP targeted to mitochondria	20 μg	47
Vector sets				
Fusion Yellow	FPF13	Mammalian expression vectors encoding TagYFP for its expression and fusion generation: pTagYFP-C and pTagYFP-N	20 μg each	43, 45
Antibodies against TagYFP				
Anti-Tag(CGY)FP	AB121 AB122	Rabbit polyclonal antibody against TagCFP, TagGFP, TagYFP, PS-CFP2, and EGFP	100 μg 200 μg	101

Please contact your local distributor for exact prices and delivery information.

Red fluorescent protein TagRFP

- Bright red (orange) fluorescence
- Monomeric protein with successful performance in fusions
- Fast maturation, high pH-stability
- Proven suitability to generate stably transfected cell lines
- Recommended for protein labeling, acidic organelle labeling, FRET applications

TagRFP is a monomeric red (orange) fluorescent protein generated from the wild-type RFP from sea anemone *Entacmaea quadricolor* [Merzlyak et al. 2007]. It possesses bright fluorescence with excitation/emission maxima at 555 and 584 nm, respectively. TagRFP is about three times brighter than mCherry protein [Shaner et al. 2004], which makes it the brightest monomeric red fluorescent protein available so far.

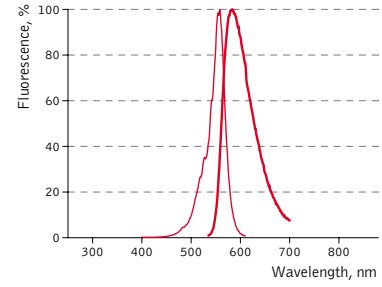
TagRFP is mainly intended for protein labeling. It can also be used for cell and organelle labeling and for tracking the promoter activity.

Another application of TagRFP is its use as an acceptor for FRET in pair with green fluorescent proteins. The traditional cyan and yellow FRET partners exhibit several substantial drawbacks limiting their utility, such as relatively low dynamic range (donor/acceptor emission ratio change) and difficulties with spectral separation. Using of TagRFP as an acceptor for Evrogen green fluorescent protein TagGFP2 (page 16) ensures higher FRET efficiency and more reliable spectral separation of the donor and acceptor fluorescence. Shifting the wavelengths towards the red part of the spectrum reduces input of cellular autofluorescence. High molar extinction coefficient of TagRFP along with high fluorescence quantum yield of TagGFP2 and excellent overlap of TagGFP2 emission and TagRFP excitation spectra result in highly effective FRET between these fluorescent proteins. The calculated Forster distance (R_0 = 5.7 nm) for the TagGFP2 / TagRFP pair is one of the largest

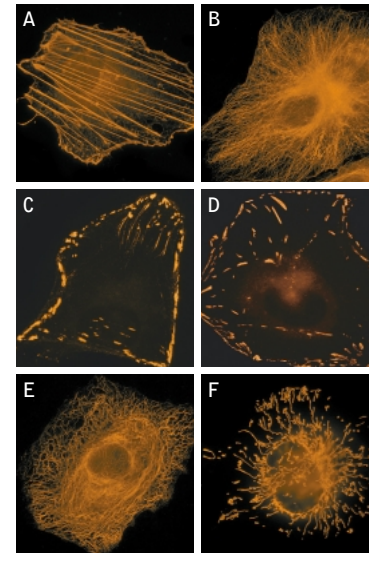
Main properties of TagRFP

Characteristic	
Molecular weight, kDa	27
Polypeptide length, aa	237
Fluorescence color	red (orange)
Excitation maximum, nm	555
Emission maximum, nm	584
Quantum yield	0.48
Extinction coefficient, $M^{-1}cm^{-1}$	100 000
Brightness*	48.0
Brightness, % of EGFP	148
pKa	3.8
Structure	monomer
Aggregation	no
Maturation rate at 37°C	fast
Photostability	medium
Cell toxicity	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TagRFP normalized excitation (thin line) and emission (thick line) spectra.
Complete TagRFP spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



TagRFP use for cell and protein labeling.
(A) HeLa cells expressing TagRFP fusion with β-actin; (B) HeLa cells expressing TagRFP fusion with α-tubulin; (C) HeLa cells expressing TagRFP fusion with zyxin; (D) HeLa cells expressing TagRFP fusion with vinculin; (E) HeLa cells expressing TagRFP fusion with keratin; (F) HeLa cells expressing TagRFP targeted to mitochondria.

among the values reported. At the same time, since TagGFP2 and TagRFP emission peaks are spaced by as much as 78 nm, the emission signal for these two proteins can be easily separated in any imaging system. High pH-stability of the both proteins allows using this pair for imaging in acidic organelles. As an additional advance, TagRFP and TagGFP2 proteins derive from different marine sources and therefore luck the ability to form heterodimers. It ensures zero background for FRET analysis that may not be the case for weakly dimerizing FRET pairs consisting of highly homological fluorescent proteins. The excellent performance of TagRFP in FRET application was demonstrated both *in vitro* and *in vivo* on the example of FRET-based apoptosis reporter CaspeR3-GR (see page 86 of this catalogue and [Shcherbo et al. 2009]).

TagRFP can be easily expressed and detected in a wide range of organisms. It becomes clearly detectable in mammalian cells as early as within 10-12 hrs after transfection. No cytotoxic effects or visible protein aggregation are observed.

TagRFP performance in protein fusions has been demonstrated in fibrillarin, vinculin, zyxin, β-actin, α-tubulin, and other models.

TagRFP suitability to generate stably transfected cells has been proven by Marinpharm company. Cell lines expressing TagRFP fusions are commercially available.

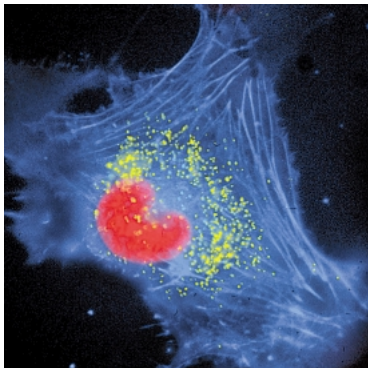
TagRFP can be used in multicolor labeling applications with other fluorescent proteins of blue, cyan, green, yellow, and far-red colors.

High pH-stability with pKa=3.8 makes it possible to use TagRFP for imaging in acidic organelles, such as late and recycling endosomes and lysosomes.

Recommended filter sets and antibodies

TagRFP can be recognized using Anti-tRFP antibody (Cat.# AB231-AB232) available from Evrogen.

Recommended Omega Optical filter sets are QMAX-Yellow, XF108-2, XF101-2, and XF111-2. TagRFP can also be detected using TRITC filter set or similar.



TagRFP application for multicolor labeling. Transiently transfected HeLa cells expressing TagBFP fusion with β-actin (blue), peroxisomes-targeted PhiYFP (yellow), and TagRFP fusion with histon H2B (red).

REFERENCES

Merzlyak et al. (2007). Nat Methods, 4 (7): 555–557 / pmid: 17572680

Shaner et al. (2004). Nat Biotechnol, 22 (12): 1567–1572 / pmid: 15558047

Shcherbo et al. (2009). BMC Biotechnology, 9: 24 / pmid: 19321010

TagRFP-related products

Product	Cat.#	Description	Size	Page(s)
TagRFP expression/source vectors				
pTagRFP-C	FP141	Mammalian expression vector encoding humanized TagRFP and allowing its expression and generation of fusions to the TagRFP C-terminus	20 μg	43
pTagRFP-N	FP142	Mammalian expression vector encoding humanized TagRFP and allowing its expression and generation of fusions to the TagRFP N-terminus	20 μg	45
pTagRFP-actin	FP144	Mammalian expression vector encoding humanized TagRFP fused with human cytoplasmic β-actin	20 μg	50
pTagRFP-tubulin	FP145	Mammalian expression vector encoding humanized TagRFP fused with human α-tubulin	20 μg	50
pTagRFP-mito	FP147	Mammalian expression vector encoding humanized TagRFP targeted to mitochondria	20 μg	47
pTagRFP-actinin	FP360	Mammalian expression vector encoding humanized TagRFP fused with human α-actinin	20 μg	50

Product	Cat.#	Description	Size	Page(s)
pTagRFP-integrin	FP361	Mammalian expression vector encoding humanized TagRFP fused with human α-V-integrin	20 μg	50
pTagRFP-Cx26	FP362	Mammalian expression vector encoding humanized TagRFP fused with rat connexin 26	20 μg	51
pTagRFP-Cx32	FP363	Mammalian expression vector encoding humanized TagRFP fused with human connexin 32	20 μg	51
pTagRFP-Cx43	FP364	Mammalian expression vector encoding humanized TagRFP fused with rat connexin 43	20 μg	51
pTagRFP-EB3	FP365	Mammalian expression vector encoding humanized TagRFP fused with human EB3 protein	20 μg	50
pTagRFP-FAK	FP366	Mammalian expression vector encoding humanized TagRFP fused with chicken focal adhesion kinase	20 μg	50
pTagRFP-Golgi	FP367	Mammalian expression vector encoding humanized TagRFP fused with human Golgi targeting sequence (GTS)	20 μg	47
pTagRFP-H2B	FP368	Mammalian expression vector encoding humanized TagRFP fused with human histone H2B	20 μg	51
pTagRFP-keratin	FP369	Mammalian expression vector encoding humanized TagRFP fused with human cytokeratin-18	20 μg	50
pTagRFP-laminB1	FP370	Mammalian expression vector encoding humanized TagRFP fused with human lamin B1	20 μg	51
pTagRFP-profilin	FP371	Mammalian expression vector encoding humanized TagRFP fused with mouse profilin	20 μg	50
pTagRFP-vinculin	FP372	Mammalian expression vector encoding humanized TagRFP fused with human vinculin	20 μg	50
pTagRFP-zyxin	FP373	Mammalian expression vector encoding humanized TagRFP fused with human zyxin	20 μg	51
Gateway® TagRFP-AS-C	FP148	Gateway® entry clone for generation of fusions to the C-terminus of TagRFP; transfer of the construct encoding TagRFP or its fusion into Gateway® destination vectors; TagRFP codon usage is optimized for expression in *Arabidopsis* and *Saccharomyces*	20 μg	40
Gateway® TagRFP-AS-N	FP149	Gateway® entry clone for generation of fusions to the N-terminus of TagRFP; transfer of the construct encoding TagRFP or its fusion into Gateway® destination vectors; TagRFP codon usage is optimized for expression in *Arabidopsis* and *Saccharomyces*	20 μg	41
Vector sets				
Fusion Red	FPF14	Mammalian expression vectors encoding TagRFP for its expression and fusion generation: pTagRFP-C and pTagRFP-N	20 μg each	43, 45
Antibodies against TagRFP				
Anti-tRFP	AB231 AB232	Rabbit polyclonal antibody against TurboRFP, TurboFP602, TurboFP635, TagBFP, TagRFP, TagFP635, and mKate2	100 μg 200 μg	104

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Far-red fluorescent protein mKate2

- Super bright far-red fluorescence
- Monomeric protein with successful performance in fusions
- Fast maturation, high pH-stability and photostability
- Proven suitability to generate stably transfected cell lines
- Fluorescent signal is easily distinguished from background fluorescence
- Recommended for protein labeling, multicolor applications and whole body imaging

mKate2 is the next generation of far-red fluorescent protein TagFP635 (mKate) [Shcherbo et al. 2007; Shcherbo et al. 2009]. It is almost 3-fold brighter than TagFP635 and is 10-fold brighter than mPlum at physiological pH 7.5. Within the optical window optimal for light penetration in living tissues, calculated brightness of mKate2 is at least 2-fold higher compared to any monomeric fluorescent protein reported to date.

mKate2 is characterized by complete and fast chromophore maturation at 37°C with maturation half-time <20 min (versus 40 min for mCherry). It is more photostable under both widefield and confocal illumination than other monomeric far-red proteins, including TagFP635, mRaspberry and mPlum. The high brightness, far-red emission spectrum, excellent pH resistance and photostability, coupled with low toxicity demonstrated in transgenic *Xenopus laevis* embryos, make mKate2 a superior fluorescent tag for imaging in living tissues.

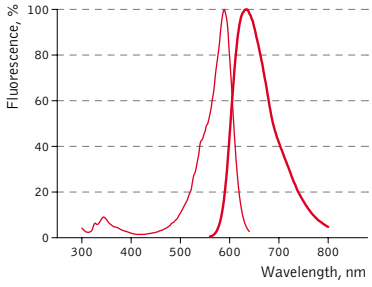
mKate2 is mainly intended for protein labeling. Its far-red fluorescence allows easy and reliable separation from standard green fluorescent labels in dual-color high-throughput assays.

mKate2 can be easily expressed and detected in a wide range of organisms. It becomes clearly detectable in mammalian cells as early as 10-12 hrs

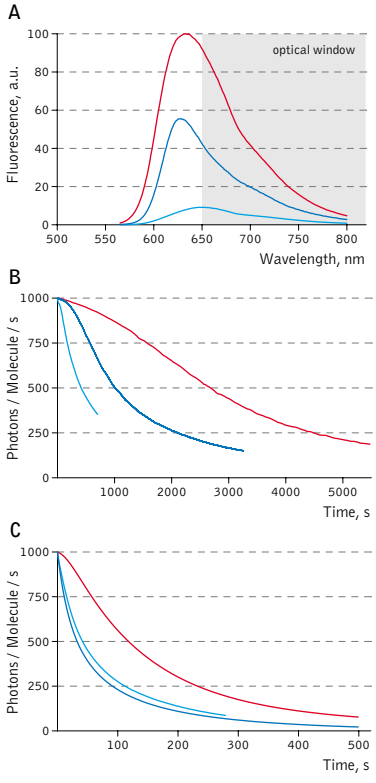
Main properties of mKate2

Characteristic	
Molecular weight, kDa	26
Polypeptide length, aa	232
Fluorescence color	far-red
Excitation maximum, nm	588
Emission maximum, nm	633
Quantum yield	0.40
Extinction coefficient, $M^{-1}cm^{-1}$	62 500
Brightness*	25.0
Brightness, % of EGFP	74
pKa	5.4
Structure	monomer
Aggregation	no
Maturation rate at 37°C	fast
Photostability	high
Cell toxicity	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



mKate2 normalized excitation (thin line) and emission (thick line) spectra.
Complete mKate2 spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml

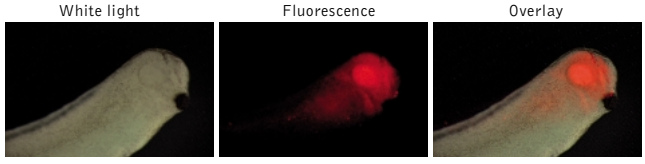


Spectral characteristics of mKate2 in comparison with selected fluorescent proteins. mKate2 - dark-red line, mRaspberry - blue line, mPlum - cyan line (A) Emission spectra of far-red monomeric fluorescent proteins given proportionally to their calculated brightness. Scaling was applied to the area of the peak. Favorable "optical window" is shaded with gray. (B) Normalized photobleaching curves for far-red monomeric fluorescent proteins, laser scanning confocal microscopy. (C) Normalized photobleaching curves, widefield fluorescence microscopy under metal halide illumination.

after transfection. No cytotoxic effects or visible protein aggregation are observed.

mKate2 performance in fusions has been demonstrated in α-actinin, zyxin, β-actin, α-tubulin, and other models.

mKate2 can be used in multicolor labeling applications with blue, cyan, green, yellow, and red (orange) fluorescent proteins.

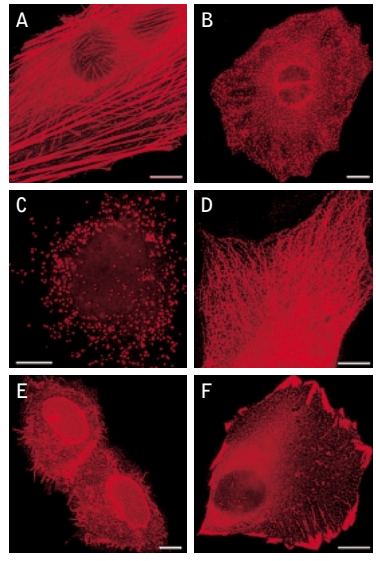


Imaging of mKate2 in *Xenopus laevis* embryos. Expression of mKate2 under the control of Xanf1 promoter in the transgenic embryos at stage 28 is specifically localized in the forehead region, including eyes, the forebrain and nasal placodes. The embryo is shown from the right side, dorsal to the top and left. Images from Shcherbo et al. 2009.

Recommended filter sets and antibodies

mKate2 can be recognized using Anti-tRFP antibody (Cat.# AB231-AB232) available from Evrogen.

Recommended Omega Optical filter sets are QMAX-Red and XF102-2. mKate2 can also be detected using Texas Red filter sets or similar.



mKate2 use for protein labeling in mammalian cells. (A) β-actin; (B) clathrin; (C) peroxisomes; (D) α-tubulin; (E) annexin (A4); (F) paxillin; Scale bar represents 10 μm. Images from Shcherbo et al. 2009.

REFERENCES

Shcherbo et al. (2007). Nat Methods, 4 (9): 741–746 / pmid: 17721542

Shcherbo et al. (2009). Biochemical Journal, 418 (3): 567–574 / pmid: 19143658

mKate2-related products

Product	Cat.#	Description	Size	Page(s)
mKate2 expression/source vectors				
pmKate2-C	FP181	Mammalian expression vector encoding humanized mKate2 and allowing its expression and generation of fusions to the mKate2 C-terminus	20 μg	43
pmKate2-N	FP182	Mammalian expression vector encoding humanized mKate2 and allowing its expression and generation of fusions to the mKate2 N-terminus	20 μg	45
pmKate2-actin	FP184	Mammalian expression vector encoding humanized mKate2 fused with human cytoplasmic β-actin	20 μg	50
pmKate2-tubulin	FP185	Mammalian expression vector encoding humanized mKate2 fused with human α-tubulin	20 μg	50
pmKate2-f-mem	FP186	Mammalian expression vector encoding membrane-targeted mKate2	20 μg	48
pmKate2-mito	FP187	Mammalian expression vector encoding humanized mKate2 targeted to mitochondria	20 μg	47
pmKate2-laminB1	FP310	Mammalian expression vector encoding humanized mKate2 fused with human lamin B1	20 μg	51
pmKate2-H2B	FP311	Mammalian expression vector encoding humanized mKate2 fused with human histone H2B	20 μg	51
pmKate2-lyso	FP312	Mammalian expression vector encoding humanized mKate2 targeted to lysosomes	20 μg	49

Product	Cat.#	Description	Size	Page(s)
pmKate2-peroxi	FP313	Mammalian expression vector encoding humanized mKate2 targeted to peroxisomes	20 μg	48
pmKate2-endo	FP314	Mammalian expression vector encoding humanized mKate2 fused with human RhoB protein	20 μg	49
pmKate2-zyxin	FP315	Mammalian expression vector encoding humanized mKate2 fused with human zyxin	20 μg	50
pmKate2-EB3	FP316	Mammalian expression vector encoding humanized mKate2 fused with human EB3 protein	20 μg	50
pmKate2-actinin	FP317	Mammalian expression vector encoding humanized mKate2 fused with human α-actinin	20 μg	50
pmKate2-vimentin	FP318	Mammalian expression vector encoding humanized mKate2 fused with human vimentin	20 μg	50
pmKate2-keratin	FP319	Mammalian expression vector encoding humanized mKate2 fused with human cytokeratin-18	20 μg	50
pmKate2-profilin	FP320	Mammalian expression vector encoding humanized mKate2 fused with mouse profilin	20 μg	50
pmKate2-annexin	FP321	Mammalian expression vector encoding humanized mKate2 fused with human annexin A4	20 μg	51
pmKate2-clathrin	FP322	Mammalian expression vector encoding humanized mKate2 fused with human clathrin light chain LCB	20 μg	51
pmKate2-paxillin	FP323	Mammalian expression vector encoding humanized mKate2 fused with chicken paxillin	20 μg	50
pTagFP635-vinculin	FP388	Mammalian expression vector encoding humanized TagFP635* fused with human vinculin	20 μg	50
pTagFP635-Cx26	FP382	Mammalian expression vector encoding humanized TagFP635* fused with rat connexin 26	20 μg	51
pTagFP635-Cx32	FP383	Mammalian expression vector encoding humanized TagFP635* fused with human connexin 32	20 μg	51
pTagFP635-Cx43	FP384	Mammalian expression vector encoding humanized TagFP635* fused with rat connexin 43	20 μg	51
Vector sets				
Fusion Far-Red	FPF25	Mammalian expression vectors encoding mKate2 for its expression and fusion generation: pmKate2-C and pmKate2-N	20 μg each	43, 45
Antibodies against mKate2				
Anti-tRFP	AB231 AB232	Rabbit polyclonal antibody against TurboRFP, TurboFP602, TurboFP635, TagBFP, TagRFP, TagFP635 and mKate2	100 μg 200 μg	104

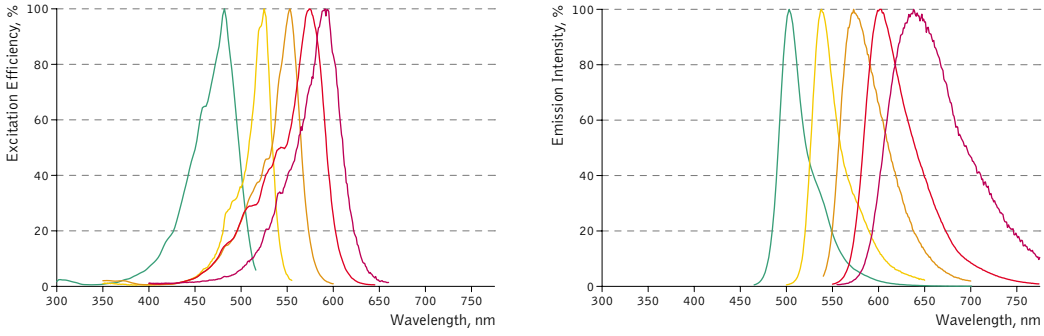
Please contact your local distributor for exact prices and delivery information.

TagFP635* - the parental variants of mKate2.

TurboFPs

Bright reporters for cell labeling

TurboFPs are proteins of different colors that are recommended for use in applications where fast appearance of bright fluorescence is crucial (e.g. for tracking the promoter activity), and for cell and organelle labeling.



Normalized excitation/emission spectra of TurboFPs.
TurboGFP - green line, TurboYFP - yellow line, TurboRFP - orange line, TurboFP602 - red line, TurboFP635 - dark-red line.

Main properties of TurboFPs:

Protein	TurboGFP	TurboYFP	TurboRFP	TurboFP602	TurboFP635
Molecular weight, kDa	26	26	26	26	26
Polypeptide length, aa	232	234	231	231	231
Fluorescence color	green	yellow	red (orange)	true-red	far-red
Excitation maximum, nm	482	525	553	574	588
Emission maximum, nm	502	538	574	602	635
Quantum yield	0.53	0.53	0.67	0.35	0.34
Extinction coefficient, $M^{-1}cm^{-1}$	70 000	105 000	92 000	74 400	65 000
Brightness*	37.1	55.7	61.6	26.0	22.1
Brightness, % of EGFP	112	169	187	79	67
pKa	5.2	5.9	4.4	4.7	5.5
Structure	dimer	dimer	dimer	dimer	dimer
Aggregation	no	at high concentrations	no	no	no
Maturation rate at 37°C	super fast	super fast	super fast	fast	super fast
Photostability	high	high	high	medium	high
Cell toxicity	not observed	at high concentrations	not observed	not observed	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000;

Green fluorescent protein TurboGFP

- Bright green fluorescence
- Fast maturation at a wide range of temperatures
- High pH-stability and photostability
- Proven suitability to generate stably transfected cell lines
- Destabilized variant is available
- Recommended for gene expression analysis and cell and organelle labeling

TurboGFP is an improved variant of the green fluorescent protein CopGFP cloned from copepod *Pontellina plumata* (Arthropoda; Crustacea; Maxillopoda; Copepoda) [Shagin et al. 2004]. It possesses bright green fluorescence (excitation/ emission max = 482/ 502 nm) that is visible earlier than fluorescence of other green fluorescent proteins.

TurboGFP is mainly intended for applications where fast appearance of bright fluorescence is crucial. It is specially recommended for cell and organelle labeling and tracking the promoter activity. Destabilized TurboGFP variant allows accurate analysis of rapid and/or transient events in gene regulation.

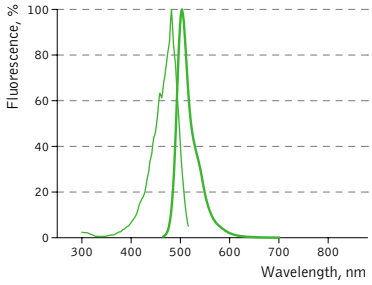
TurboGFP can be expressed and detected in a wide range of organisms including cold-blooded animals. Mammalian cells transiently transfected with TurboGFP expression vectors give bright fluorescent signals within 8-10 hrs after transfection. No cytotoxic effects or visible protein aggregation are observed.

TurboGFP suitability to generate stably transfected cells has been proven by Marinpharm company. Various cell lines expressing TurboGFP are commercially available.

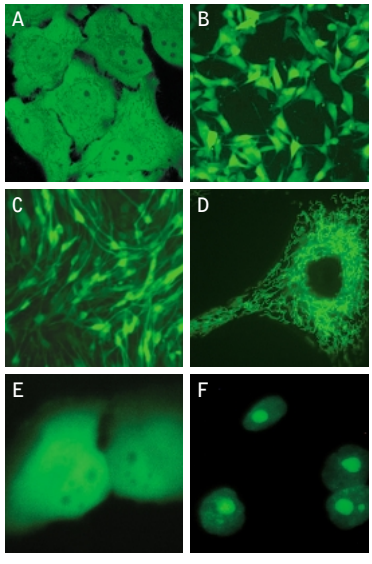
Main properties of TurboGFP

Characteristic	
Molecular weight, kDa	26
Polypeptide length, aa	232
Fluorescence color	green
Excitation maximum, nm	482
Emission maximum, nm	502
Quantum yield	0.53
Extinction coefficient, $M^{-1}cm^{-1}$	70 000
Brightness*	37.1
Brightness, % of EGFP	112
pKa	5.2
Structure	dimer
Aggregation	no
Maturation rate at 37°C	super fast
Photostability	high
Cell toxicity	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TurboGFP normalized excitation (thin line) and emission (thick line) spectra.
Complete TurboGFP spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



TurboGFP expression in mammalian cells.
(A) Transiently transfected HeLa cells expressing TurboGFP in cytoplasm; (B) stably transfected M3-mouse melanoma cells expressing TurboGFP in cytoplasm; (C) stably transfected C2C12 mouse myoblasts expressing TurboGFP in cytoplasm; (D) stably transfected HeLa cells expressing mitochondria-targeted TurboGFP; (E) stably transfected HeLa cells expressing TurboGFP-BID fusion; (F) stably transfected HeLa cells expressing TurboGFP-fibrillarin fusion.
Photographs of stably transfected cell lines were kindly provided by Dr. Christian Petzelt (Marinpharm).

Refolding and maturation kinetics of GFPs *in vitro*

	EGFP	Venus	SYFP2	TurboGFP
Refolding half-time, s	90.6	46.2	69.3	11.0
Maturation half-time, s	3915	4076	3300	1468
kox 10^{-4} s^{-1}	1.77	1.70	2.10	4.72
Reference	Evdokimov et al. 2006	Kremers et al. 2006	Kremers et al. 2006	Evdokimov et al. 2006

Samples of fluorescent proteins were heated to 95°C in denaturation solution (8 M urea, 1 mM DTT) for 4 min. Refolding reactions were initiated upon 100-fold dilution into the renaturation buffer (35 mM KCl, 2 mM $MgCl_2$, 50 mM Tris pH 7.5, 1 mM DTT). In maturation assay, 5 mM freshly dissolved dithionite was added to the denaturation solution [Reid and Flynn 1997]. Due to the instability of dithionite at high temperatures, to provide for complete chromophore reduction the sample was cooled to 25°C and the addition of 5 mM dithionite followed by heating to 5°C were repeated. Protein refolding and maturation were followed by measuring the recovery of fluorescence using Varian Cary Eclipse Fluorescence Spectrophotometer, chamber temperature maintained at 25°C. Maturation rate constants (kox) were determined by computer-fitting the kinetic data to the first order exponential decay (Origin 6.0).

Despite its dimeric structure, TurboGFP performs well in some fusions. However, for protein labeling applications we recommend using specially optimized monomeric TagFPs (see page 11).

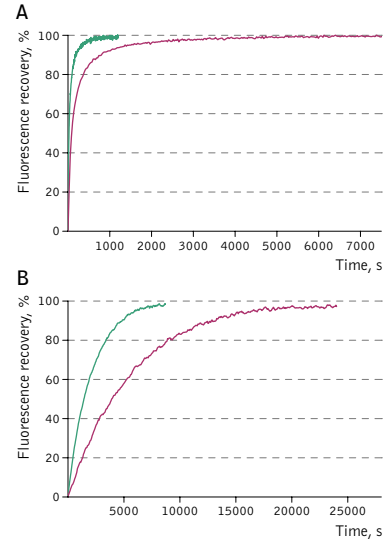
TurboGFP maturation kinetics: TurboGFP allows monitoring the activity from early promoters. It matures noticeably faster than EGFP and most other fluorescent proteins. This difference in performance is illustrated here using both *in vitro* analysis of TurboGFP and EGFP refolding and maturation kinetics (see table above) and *in vivo* examination of the developing *Xenopus* embrios expressing either TurboGFP or EGFP (see page 9).

TurboGFP can be used in multicolor labeling applications with blue, cyan, green, red, and far-red fluorescent dyes.

Recommended filter sets and antibodies

TurboGFP can be recognized using Anti-TurboGFP (Cat.# AB511-AB512) and Anti-TurboGFP(d) (Cat.# AB513-AB514) antibodies available from Evrogen.

TurboGFP can be detected using common fluorescence filter sets for EGFP, FITC, and other green dyes. Recommended Omega Optical filter sets are QMAX-Green, XF100-2, XF100-3, (XF115-2), and XF116-2.



Comparison of EGFP (violet lines) and TurboGFP (green lines) refolding and maturation speed *in vitro* [Evdokimov et al. 2006].
Normalized fluorescence recovery plots are shown. (A) Refolding kinetics; (B) chromophore maturation kinetics.

REFERENCES

Evdokimov et al. (2006). EMBO Rep, 7 (10): 1006–1012 / pmid: 16936637

Kremers et al. (2006). Biochemistry, 45 (21): 6570–6580 / pmid: 16716067

Reid and Flynn (1997). Biochemistry, 36 (22): 6786–6791 / pmid: 9184161

Shagin et al. (2004). Curr Biol, 21 (5): 841–850 / pmid: 14963095

TurboGFP-related products

Product	Cat.#	Description	Size	Page(s)
TurboGFP expression/source vectors				
pTurboGFP-C	FP511	Mammalian expression vector encoding humanized TurboGFP and allowing its expression and generation of fusions to the TurboGFP C-terminus	20 μg	43
pTurboGFP-N	FP512	Mammalian expression vector encoding humanized TurboGFP and allowing its expression and generation of fusions to the TurboGFP N-terminus	20 μg	45
pTurboGFP-B	FP513	Bacterial expression vector; source of the TurboGFP coding sequence	20 μg	42

Product	Cat.#	Description	Size	Page(s)
pTurboGFP-PRL	FP515	Promoterless vector encoding humanized TurboGFP and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 µg	52
pTurboGFP-PRL-dest1	FP518	Promoterless vector encoding destabilized TurboGFP and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 µg	52
pTurboGFP-dest1	FP519	Mammalian expression vector encoding destabilized TurboGFP for its expression and generation of fusions to the TurboGFP-dest1 N-terminus	20 µg	45
Gateway® TurboGFP-C	FP521	Gateway® entry clone for generation of fusions to the C-terminus of humanized TurboGFP; transfer of the construct encoding TurboGFP or its fusion into Gateway® destination vectors	20 µg	40
Gateway® TurboGFP-N	FP522	Gateway® entry clone for generation of fusions to the N-terminus of humanized TurboGFP; transfer of the construct encoding TurboGFP or its fusion into Gateway® destination vectors	20 µg	41
pTurboGFP-mito	FP517	Mammalian expression vector encoding humanized TurboGFP targeted to mitochondria	20 µg	47
Vector sets				
Promoter-tracker 3-colors	FPP15	Promoterless vectors pTurboYFP-PRL, pTurboGFP-PRL, and pTurboRFP-PRL	20 µg each	52
Promoter-tracker Green	FPP03	Promoterless vectors pTurboGFP-PRL, pTurboGFP-PRL-dest1, control vector pTurboGFP-dest1	20 µg each	52,45
Mito-tracker	FPM01	Mammalian expression vectors for fluorescent labeling of mitochondria: pTurboGFP-mito, pPhi-Yellow-mito, and pKindling-Red-mito	20 µg each	47,69
Recombinant protein				
rTurboGFP	FP552	Purified recombinant TurboGFP	100 µg	54
Antibodies against TurboGFP				
Anti-TurboGFP	AB511 AB512	Rabbit polyclonal antibody against non-denatured TurboGFP	100 µg 200 µg	98
Anti-TurboGFP(d)	AB513 AB514	Rabbit polyclonal antibody against denatured TurboGFP	100 µg 200 µg	99

Please contact your local distributor for exact prices and delivery information.

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Yellow fluorescent protein TurboYFP

- Super bright true-yellow fluorescence
- Emission wavelength is ideally positioned between those of green and red fluorescent proteins
- Fast maturation, high pH-stability and photostability
- Destabilized variant is available
- Recommended for gene expression analysis and cell and organelle labeling

TurboYFP is an enhanced variant of the yellow fluorescent protein PhiYFP from jellyfish *Phialidium sp.* [Shagin et al. 2004]. It possesses super-bright yellow fluorescence with emission maximum at 538 nm and is ideally positioned between green and red fluorescent proteins, allowing easy separation of these markers by flow cytometry using common channels of detection and a single laser excitation line. Compared with PhiYFP, TurboYFP matures faster in mammalian cells.

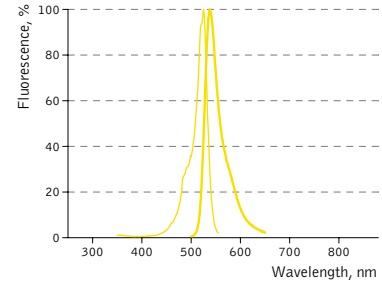
TurboYFP is mainly intended for applications where fast appearance of bright fluorescence is crucial. It is specially recommended for cell labeling and tracking the promoter activity. Destabilized TurboYFP variant allows accurate analysis of rapid and/or transient events in gene regulation.

Mammalian cells transiently transfected with TurboYFP expression vectors give bright fluorescense within 8-10 hrs after transfection. Being overexpressed in long-term culture of cells with high expression levels, TurboYFP shows slight tendency to aggregate. Therefore we recommend that you use parental PhiYFP and PhiYFP-m proteins for long-term expression and organelle labeling. Suitability of these proteins for stable transfection was demonstrated by Marinpharm (Germany).

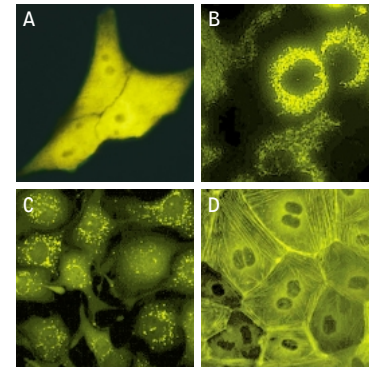
Main properties of TurboYFP

Characteristic	TurboYFP	PhiYFP	PhiYFP-m
Molecular weight, kDa	26	26	26
Polypeptide length, aa	234	234	234
Fluorescence color	yellow	yellow	yellow
Excitation maximum, nm	525	525	525
Emission maximum, nm	538	537	537
Quantum yield	0.53	0.40	0.39
Extinction coefficient, $M^{-1}cm^{-1}$	105 000	130 000	124 000
Brightness*	55.7	52.0	48.4
Brightness, % of EGFP	169	158	147
pKa	5.9	6.0	6.0
Structure	dimer	dimer	dimer
Aggregation	at high concentrations	no	no
Maturation rate at 37°C	super fast	fast	fast
Photostability	high	high	high
Cell toxicity	not observed	not observed	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TurboYFP normalized excitation (thin line) and emission (thick line) spectra.
Complete TurboYFP spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



TurboYFP and PhiYFP expression in mammalian cells. (A) Whole-cell expression in HeLa cells; (B-D) stably transfected mammalian cells expressing PhiYFP-tagged fusions: (B) mitochondria-targeted PhiYFP in 3T3 mouse fibroblasts; (C) T24 human bladder carcinoma cells expressing peroxisome-targeted PhiYFP-m; (D) PhiYFP-m fusion with β-actin in PtK rat kangaroo cells.
Images (B-D) were kindly provided by Dr. Christian Petzelt (Marinpharm).

Despite there dimeric structure, TurboYFP and PhiYFPs perform well in some fusions. Please note, that PhiYFP can be used for generation of fusions to its N-terminus, whereas PhiYFP-m is optimized to generate fusions to its C-terminus. PhiYFP can not be used to generate C-terminal fusions. For protein labeling applications we recommend using specially optimized monomeric TagFPs (see page 11).

TurboYFP can be used in multicolor labeling applications with blue, cyan, green, red, and far-red fluorescent dyes.

Recommended filter sets and antibodies

TurboYFP, PhiYFP and PhiYFP-m can be recognized using Anti-PhiYFP (Cat.# AB601-AB602) and Anti-PhiYFP(d) (Cat.# AB603-AB604) antibodies available from Evrogen.

TurboYFP can be detected using Omega Optical filter set XF104-3 or Chroma Technology Corp. filter set 42003 ("ZsYellow1").

REFERENCES

Shagin et al. (2004). Curr Biol, 21 (5): 841–850 / pmid: 14963095

TurboYFP and PhiYFP-related products

Product	Cat.#	Description	Size	Page(s)
TurboYFP expression/source vectors				
pTurboYFP-C	FP611	Mammalian expression vector encoding humanized TurboYFP and allowing its expression and generation of fusions to the TurboYFP C-terminus	20 μg	43
pTurboYFP-N	FP612	Mammalian expression vector encoding humanized TurboYFP and allowing its expression and generation of fusions to the TurboYFP N-terminus	20 μg	45
pTurboYFP-B	FP613	Bacterial expression vector; source of the TurboYFP coding sequence	20 μg	42
pTurboYFP-PRL	FP615	Promoterless vector encoding humanized TurboYFP and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 μg	52
pTurboYFP-PRL-dest1	FP618	Promoterless vector encoding destabilized TurboYFP and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 μg	52
pTurboYFP-dest1	FP619	Mammalian expression vector encoding destabilized TurboYFP for its expression and generation of fusions to the TurboYFP-dest1 N-terminus	20 μg	45
PhiYFP expression/source vectors				
pPhi-Yellow-C	FP601	Mammalian expression vector encoding humanized PhiYFP-m and allowing its expression and generation of fusions to the PhiYFP-m C-terminus	20 μg	43
pPhi-Yellow-N	FP602	Mammalian expression vector encoding humanized PhiYFP and allowing its expression and generation of fusions to the PhiYFP N-terminus	20 μg	45
pPhi-Yellow-B	FP603	Bacterial expression vector; source of the PhiYFP coding sequence	20 μg	42
pPhi-Yellow-PRL	FP604	Promoterless vector encoding humanized PhiYFP and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 μg	52
pPhi-Yellow-PRL-dest1	FP605	Promoterless vector encoding destabilized PhiYFP-m and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 μg	52

Product	Cat.#	Description	Size	Page(s)
pPhi-Yellow-peroxi	FP606	Mammalian expression vector encoding humanized PhiYFP-m targeted to peroxisomes	20 μg	48
pPhi-Yellow-mito	FP607	Mammalian expression vector encoding humanized PhiYFP targeted to mitochondria	20 μg	47
pPhi-Yellow-dest1	FP608	Mammalian expression vector encoding destabilized PhiYFP-m for its expression and generation of fusions to the PhiYFP-m-dest1 N-terminus	20 μg	45
Vector sets				
Promoter-tracker 3-colors	FPP15	Promoterless vectors pTurboYFP-PRL, pTurboGFP-PRL, and pTurboRFP-PRL	20 μg each	52,52,52
Promoter-tracker Yellow	FPP14	Promoterless vectors pTurboYFP-PRL, pTurboYFP-PRL-dest1, control vector pTurboYFP-dest1	20 μg each	52,52,45
Mito-tracker	FPM01	Mammalian expression vectors for fluorescent labeling of mitochondria: pTurboGFP-mito, pPhi-Yellow-mito, and pKindling-Red-mito	20 μg each	47,47,69
Recombinant protein				
rPhiYFP	FP651	Purified recombinant PhiYFP	100 μg	54
Antibodies against TurboYFP and PhiYFP				
Anti-PhiYFP	AB601 AB602	Rabbit polyclonal antibody against non-denatured PhiYFP, PhiYFP-m, and TurboYFP	100 μg 200 μg	102
Anti-PhiYFP(d)	AB603 AB604	Rabbit polyclonal antibody against denatured PhiYFP, PhiYFP-m, and TurboYFP	100 μg 200 μg	103

Please contact your local distributor for exact prices and delivery information.

Red (orange) fluorescent protein TurboRFP

- Super bright red (orange) fluorescence
- Fast maturation, high pH-stability
- Destabilized variant is available
- Recommended for gene expression analysis and cell and organelle labeling

TurboRFP is a red (orange) fluorescent protein derived from sea anemone *Entacmaea quadricolor* [Merzlyak et al. 2007]. TurboRFP is more than twice brighter than DsRed2. Fast TurboRFP maturation makes it clearly detectable in mammalian cells as early as within 8-10 hrs after transfection. In addition, unlike DsRed proteins TurboRFP shows no abnormal Golgi-like localization in long-term cell culture.

TurboRFP is mainly intended for applications where fast appearance of bright fluorescence is crucial. It is specially recommended for cell and organelle labeling and tracking the promoter activity. Destabilized TurboRFP variant allows accurate analysis of rapid and/or transient events in gene regulation.

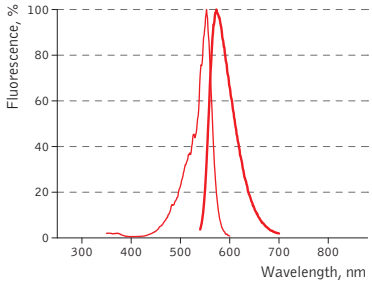
TurboRFP can be expressed and detected in a wide range of organisms. Mammalian cells transiently transfected with TurboRFP expression vectors give bright fluorescent signals within 8-10 hrs after transfection. No cytotoxic effects or visible protein aggregation are observed.

Despite its dimeric structure, TurboRFP performs well in some fusions. However, for protein labeling applications we recommend using specially optimized monomeric TagFPs (see page 11).

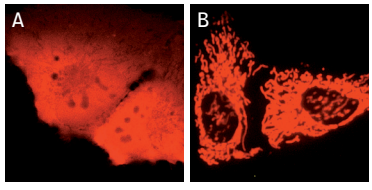
Main properties of TurboRFP

Characteristic	
Molecular weight, kDa	26
Polypeptide length, aa	231
Fluorescence color	red (orange)
Excitation maximum, nm	553
Emission maximum, nm	574
Quantum yield	0.67
Extinction coefficient, $M^{-1}cm^{-1}$	92 000
Brightness*	61.6
Brightness, % of EGFP	187
pKa	4.4
Structure	dimer
Aggregation	no
Maturation rate at 37°C	super fast
Photostability	high
Cell toxicity	not observed

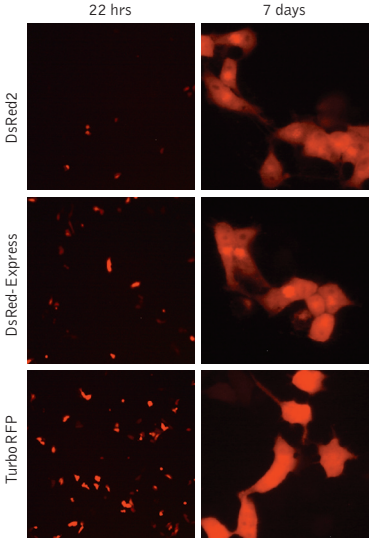
*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TurboRFP normalized excitation (thin line) and emission (thick line) spectra.
Complete TurboRFP spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



TurboRFP use for cell and organelle labeling.
(A) Fluorescent microscopy of mammalian cells expressing cytoplasmic TurboRFP; (B) Fluorescent microscopy of mammalian cells expressing TurboRFP fusion with mitochondrial targeting signal.
Images made from HeLa cells 24 hrs after transfection.



Fluorescent microscopy of Hela cells expressing TurboRFP, DsRed2, and DsRed-Express. TurboRFP gives the brightest signal 22 hrs after transfection; DsRed2 and DsRed-Express show abnormal Golgi-like localization 7 days after transfection, whereas TurboRFP localizes evenly in the cytosol.

TurboRFP can be used in multicolor labeling applications with blue, cyan, green, yellow, and far-red fluorescent dyes.

Recommended filter sets and antibodies

TurboRFP can be recognized using Anti-tRFP antibody (Cat.# AB231-AB232) available from Evrogen.

Recommended Omega Optical filter sets are QMAX-Yellow, XF108-2, XF101-2, and XF111-2. TurboRFP can also be detected using TRITC filter set or similar.

REFERENCES

Merzlyak et al. (2007). Nat Methods, 4 (7): 555–557 / pmid: 17572680

TurboRFP-related products

Product	Cat.#	Description	Size	Page(s)
TurboRFP expression/source vectors				
pTurboRFP-C	FP231	Mammalian expression vector encoding humanized TurboRFP and allowing its expression and generation of fusions to the TurboRFP C-terminus	20 μg	43
pTurboRFP-N	FP232	Mammalian expression vector encoding humanized TurboRFP and allowing its expression and generation of fusions to the TurboRFP N-terminus	20 μg	45
pTurboRFP-B	FP233	Bacterial expression vector; source of the TurboRFP coding sequence	20 μg	42
pTurboRFP-PRL	FP235	Promoterless vector encoding humanized TurboRFP and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 μg	52
pTurboRFP-mito	FP237	Mammalian expression vector encoding humanized TurboRFP targeted to mitochondria	20 μg	47
pTurboRFP-PRL-dest1	FP238	Promoterless vector encoding destabilized TurboRFP and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 μg	52
pTurboRFP-dest1	FP239	Mammalian expression vector encoding destabilized TurboRFP for its expression and generation of fusions to the TurboRFP-dest1 N-terminus	20 μg	45
Vector sets				
Promoter-tracker 3-colors	FPP15	Promoterless vectors pTurboYFP-PRL, pTurboGFP-PRL, and pTurboRFP-PRL	20 μg each	52,52,52
Antibodies against TurboRFP				
Anti-tRFP	AB231 AB232	Rabbit polyclonal antibody against TurboRFP, TurboFP602, TurboFP635, TagBFP, TagRFP, TagFP635, and mKate2	100 μg 200 μg	104

Please contact your local distributor for exact prices and delivery information.

Red fluorescent protein TurboFP602

- Bight true-red fluorescence
- Fast maturation, high pH-stability
- Proven suitability to generate stably transfected cell lines
- Recommended for gene expression analysis and cell and organelle labeling in an autofluorescent environment

TurboFP602 is a red-shifted variant of the red fluorescent protein TurboRFP from sea anemone *Entacmaea quadricolor* [Merzlyak et al. 2007]. TurboFP602 possesses true-red fluorescence, optimal for detection via most popular filter sets, and is easily distinguished from background signals. TurboFP602 exhibits fast maturation and high pH stability.

TurboFP602 is mainly intended for applications where fast appearance of true-red fluorescence is crucial. It is specially recommended for cell and organelle labeling and for tracking the promoter activity in autofluorescent tissues.

TurboFP602 can be expressed and detected in a wide range of organisms. Mammalian cells transiently transfected with TurboFP602 expression vectors give bright fluorescent signals within 8-12 hrs after transfection. No cytotoxic effects or visible protein aggregation are observed.

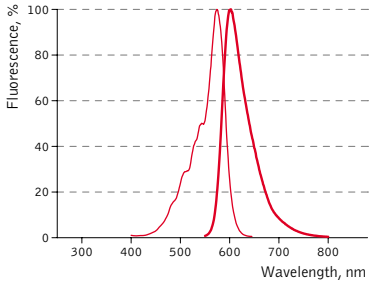
TurboFP602 suitability to generate stably transfected cells has been proven by Marinpharm company. Cell lines expressing TurboFP602 are commercially available.

Despite its dimeric structure, TurboFP602 performs well in some fusions. However, for protein labeling applications we recommend using specially optimized monomeric TagFPs (see page 11).

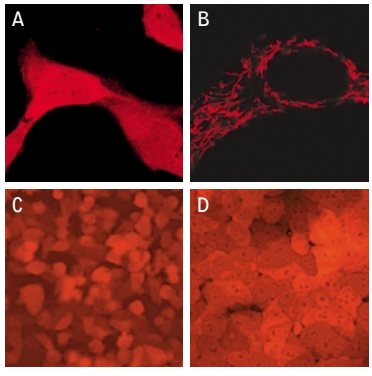
Main properties of TurboFP602

Characteristic	
Molecular weight, kDa	26
Polypeptide length, aa	231
Fluorescence color	true-red
Excitation maximum, nm	574
Emission maximum, nm	602
Quantum yield	0.35
Extinction coefficient, $M^{-1}cm^{-1}$	74 400
Brightness*	26.0
Brightness, % of EGFP	79
pKa	4.7
Structure	dimer
Aggregation	no
Maturation rate at 37°C	fast
Photostability	medium
Cell toxicity	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TurboFP602 normalized excitation (thin line) and emission (thick line) spectra.
Complete TurboFP602 spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml



TurboFP602 expression in mammalian cells.
(A) Transiently transfected HeLa cells; (B) transiently transfected HeLa cells expressing mitochondria-targeted TurboFP602; (C) stably transfected human melanoma MelJuso cell line; (D) stably transfected human osteosarcoma U-2-OS cell line.
Photographs of stably transfected cell lines were provided by Dr. Christian Petzelt (Marinpharm).

TurboFP602 can be used in multicolor labeling applications with blue, cyan, green, and yellow fluorescent dyes.

REFERENCES

Merzlyak et al. (2007). Nat Methods, 4 (7): 555–557 / pmid: 17572680

TurboFP602-related products

Product	Cat.#	Description	Size	Page(s)
TurboFP602 expression/source vectors				
pTurboFP602-C	FP711	Mammalian expression vector encoding humanized TurboFP602 and allowing its expression and generation of fusions to the TurboFP602 C-terminus	20 μg	43
pTurboFP602-N	FP712	Mammalian expression vector encoding humanized TurboFP602 and allowing its expression and generation of fusions to the TurboFP602 N-terminus	20 μg	45
pTurboFP602-B	FP713	Bacterial expression vector; source of the TurboFP602 coding sequence	20 μg	42
pTurboFP602-PRL	FP715	Promoterless vector encoding humanized TurboFP602 and designed for monitoring activity of different promoters and promoter/enhancer combinations	20 μg	52
pTurboFP602-mito	FP717	Mammalian expression vector encoding humanized TurboFP602 targeted to mitochondria	20 μg	47
Antibodies against TurboFP602				
Anti-tRFP	AB231 AB232	Rabbit polyclonal antibody against TurboRFP, TurboFP602, TurboFP635, TagBFP, TagRFP, TagFP635, and mKate2	100 μg 200 μg	104

Please contact your local distributor for exact prices and delivery information.

Far-red fluorescent protein TurboFP635

- Super bright far-red fluorescence
- Fast maturation, high photostability
- Proven suitability to generate stably transfected cell lines
- Fluorescent signal is easily distinguished from background fluorescence
- Recommended for cell and organelle labeling in autofluorescent environment, multicolor applications and whole body imaging

TurboFP635 (scientific name Katushka) is a far-red mutant of the red fluorescent protein from sea anemone *Entacmaea quadricolor* [Shcherbo et al. 2007]. Possessing excitation/emission maxima at 588/635 nm, TurboFP635 is 7 to 10-fold brighter compared to the spectrally close HcRed [Gurskaya et al. 2001] or mPlum [Wang et al. 2004]. TurboFP635 is characterized by fast maturation and a high pH-stability and photostability. The unique characteristics of TurboFP635 make it the protein of choice for visualization within living tissues and dual-color high-throughput assays.

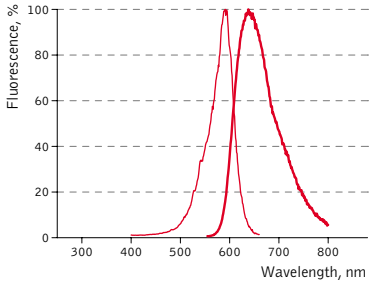
TurboFP635 is mainly intended for applications where fast appearance of far-red fluorescence is crucial. It is specially recommended for whole body imaging, cell and organelle labeling, and for tracking the promoter activity in auto-fluorescent tissues.

TurboFP635 can be easily expressed and detected in a wide range of organisms. It can be easy visualised within living tissues. Mammalian cells transiently transfected with TurboFP635 expression vectors give bright fluorescent signals within 10-12 hours after transfection. No cytotoxic effects or visible protein aggregation are observed.

Main properties of TurboFP635

Characteristic	
Molecular weight, kDa	26
Polypeptide length, aa	231
Fluorescence color	far-red
Excitation maximum, nm	588
Emission maximum, nm	635
Quantum yield	0.34
Extinction coefficient, $M^{-1}cm^{-1}$	65 000
Brightness*	22.1
Brightness, % of EGFP	67
pKa	5.5
Structure	dimer
Aggregation	no
Maturation rate at 37°C	super fast
Photostability	high
Cell toxicity	not observed

*Brightness is a product of extinction coefficient and quantum yield, divided by 1000.



TurboFP635 normalized excitation (thin line) and emission (thick line) spectra.
Complete TurboFP635 spectra in Excel format can be downloaded from the Evrogen Web site at www.evrogen.com / support / FP-tech.shtml

DsRed-Express and TurboFP635 expression in transgenic *Xenopus laevis*.
Transgenic 2.5 months living animals expressing TurboFP635 and DsRed-Express under the control of cardiac actin promoter are shown from the dorsal side. TurboFP635 (on the right) is excellently visible in the whole body, while DsRed-Express (on the left) can be hardly visualized. This experiment clearly demonstrates the advantage of longer wavelength emission of TurboFP635 for the whole body imaging. Leica MZFLIII fluorescent stereomicroscope, excitation filter 546/10; emission filter 565LP. Image from Shcherbo et al. 2007.

Despite its dimeric structure, TurboFP635 performs well in some fusions. However, for protein labeling applications we recommend using specially optimized monomeric TagFPs (see page 11).

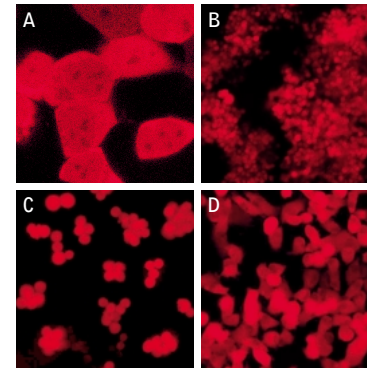
TurboFP635 suitability to generate stably transfected cells has been proven by Marinpharm company. Various cell lines expressing TurboFP635 are commercially available.

TurboFP635 can be used in multicolor labeling applications with blue, cyan, green, yellow and red (orange) fluorescent proteins.

Recommended filter sets and antibodies

TurboFP635 can be recognized using Anti-tRFP antibody (Cat.# AB231-AB232) available from Evrogen.

Recommended Omega Optical filter sets are QMAX-Red and XF102-2. TurboFP635 can also be detected using Texas Red filter sets or similar.



TurboFP635 expression in mammalian cells.
(A) Transiently transfected Phoenix cells; (B) stably transfected WALKER 256 rat tumor cells; (C) stably transfected mouse Ehrlich-Ascites cells; (D) stably transfected metastasizing melanoma MelJuSo cells.
Photographs of stably transfected cell lines were provided by Dr. Christian Petzelt (Marinpharm).

REFERENCES

Gurskaya et al. (2001). FEBS Lett, 507 (1): 16–20 / pmid: 11682051

Shcherbo et al. (2007). Nat Methods, 4 (9): 741–746 / pmid: 17721542

Wang et al. (2004). Proc Natl Acad Sci U S A, 101 (48): 16745–16749 / pmid: 15556995

TurboFP635-related products

Product	Cat.#	Description	Size	Page(s)
TurboFP635 expression/source vectors				
pTurboFP635-C	FP721	Mammalian expression vector encoding humanized TurboFP635 and allowing its expression and generation of fusions to the TurboFP635 C-terminus	20 μg	43
pTurboFP635-N	FP722	Mammalian expression vector encoding humanized TurboFP635 and allowing its expression and generation of fusions to the TurboFP635 N-terminus	20 μg	45
Antibodies against TurboFP635				
Anti-tRFP	AB231 AB232	Rabbit polyclonal antibody against TurboRFP, TurboFP602, TurboFP635, TagBFP, TagRFP, TagFP635, and mKate2	100 μg 200 μg	104

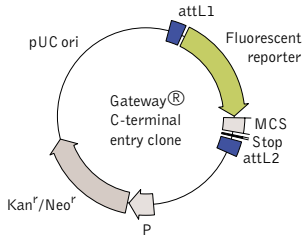
Please contact your local distributor for exact prices and delivery information.

Basic FPs: related products

Expression/source vectors

Vector group	Description	Subtypes	Details	Page
Gateway® entry clones	The vectors allow generation of fusions with the fluorescent proteins and easy transfer of the reporter sequence into Gateway® destination vectors (Invitrogen) for expression in various heterological systems	Gateway® C-terminal entry clone	MCS are located downstream of the fluorescent protein sequence allowing fusion generation to the reporter C-terminus	40
		Gateway® N-terminal entry clone	MCS are located upstream of the fluorescent protein sequence allowing fusion generation to the reporter N-terminus	41
Bacterial expression vectors	The vectors allow easy excision of the fluorescent protein sequence for subcloning and can be also used for fluorescent protein expression in prokaryotic cells			42
Basic mammalian expression vectors	The vectors allow generation of fusions with the fluorescent proteins and expression of these fusions or fluorescent proteins alone in eukaryotic (mammalian) cells under the control of early CMV promoter ($P_{CMV\ IE}$)	C-terminal mammalian expression vectors	MCS are located downstream of the fluorescent protein sequence allowing fusion generation to the reporter C-terminus	43
		N-terminal mammalian expression vectors	MCS are located upstream of the fluorescent protein sequence allowing fusion generation to the reporter N-terminus	45
Subcellular localization vectors	Ready-to-use vectors for labeling of cellular organelles and proteins	Mitochondria localization	Vectors for fluorescent labeling of mitochondria	47
		Golgi apparatus localization	Vectors for fluorescent labeling of Golgi apparatus	47
		Peroxisome localization	Vectors for fluorescent labeling of peroxisomes	48
		Plasma membrane localization	Vectors for fluorescent labeling of plasma membrane	48
		Endosomes localization	Vectors for fluorescent labeling of endosomes	49
		Lysosomes localization	Vectors for fluorescent labeling of lysosomes	49
		Protein localization vectors	Vectors for labeling of cytoskeletal and adhesion proteins (α-actinin, α-V-integrin, α-tubulin, β-actin, EB3 protein, focal adhesion kinase, cytokeratin-18, profilin, vinculin, zyxin), nuclear proteins (histone H2B, lamin B1), gap junction proteins (connexin 26, connexin 32, connexin 43), and vesicular transport protein clathrin	50
Promoterless vectors	The vectors comprise fluorescent reporter coding sequence with multiple cloning sites (MCS) at the 5'-end allowing cloning of a functional promoter			52

Gateway® C-terminal entry clone



For vector sequence, please visit our Web site at
http://www.evrogen.com/support/vector-info.shtml

Vector type	Gateway® entry clone
Reporter(s)	TurboGFP, TagRFP-AS
Promoter	No
Host cells	prokaryotic
Selection	kanamycin
Replication	pUC ori
Use	Generation of fusions to the C-terminus of the fluorescent protein; transfer of the construct encoding fluorescent protein or its fusion into Gateway® destination vectors

Product	Cat.#	Reporter	Codon usage	Color	Size
Gateway® TagRFP-AS-C	FP148	TagRFP	*Arabidopsis* and *Saccharomyces*	red (orange)	20μg
Gateway® TurboGFP-C	FP521	TurboGFP	mammalian	green	20μg

Please contact your local distributor for exact prices and delivery information.

Multiple cloning sites (MCS)

Gateway® TagRFP-AS-C vector MCS

TagRFP → . . . TCC.GGA.CTC.AGA.TCT.CGA.GCT.CAA.GCT.TCG.AAT.TCT.GCA.GTC.GAC.GGT.ACC.GCG.GGC.CCG.GGA.TCC.ACC.GGA.TCT.AGG.TAA.CTG.AAC.C . . .

Bgl II (BspE I); Sac I (Xho I); EcoR I (Hind III); Sal I (Pst I); Sac II (Kpn I); Sma I/Xma I (Apa I); BamH I; STOPs; AttL 2 site

Gateway® TurboGFP-C vector MCS

TurboGFP → . . . AGA.TCT.CGA.GCT.CAA.GCT.TCG.AAT.TCT.GCA.GTC.GAC.GGT.ACC.GCG.GGC.CCG.GGA.TCC.ACC.GGA.TCT.AGG.TAA.CTG.AAC.C . . .

Sac I (Xho I); EcoR I (Hind III); Sal I (Pst I*); Sac II (Kpn I); Sma I/Xma I (Apa I*); BamH I; STOP; AttL 2 site

* — not unique sites.

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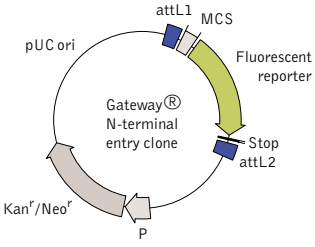
Gateway® N-terminal entry clone



For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

Vector type	Gateway® entry clone
Reporter(s)	TurboGFP, TagRFP-AS
Promoter	No
Host cells	prokaryotic
Selection	kanamycin
Replication	pUC ori
Use	Generation of fusions to the N-terminus of the fluorescent protein; transfer of the construct encoding fluorescent protein or its fusion into Gateway® destination vectors

Product	Cat.#	Reporter	Codon usage	Color	Size
Gateway® TagRFP-AS-N	FP149	TagRFP	*Arabidopsis* and *Saccharomyces*	red (orange)	20μg
Gateway® TurboGFP-N	FP522	TurboGFP	mammalian	green	20μg

Please contact your local distributor for exact prices and delivery information.

Multiple cloning sites (MCS)

Gateway® TagRFP-AS-N vector MCS

attL 1 site — Bgl II — Sac I — EcoR I — Sal I — Sac II — Sma I/Xma I — Age I — TagRFP

... AGG.CTG.CTA.GCG.CTA.CCG.GAC.TCA.GAT.CTC.GAG.CTC.AAG.CTT.CGA.ATT.CTG.CAG.TCG.ACG.GTA.CCG.CGG.GCC.CGG.GAT.CCA.CCG.GTC.GCC.ACC.ATG.G...

Afe I — Xho I — Hind III — Pst I — Kpn I — Apa I — BamH I — Nco I*

Gateway® TurboGFP-N vector MCS

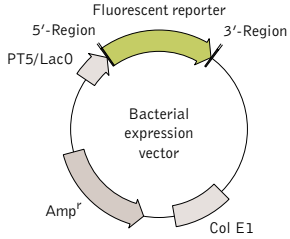
attL 1 site — Bgl II — Sac I — EcoR I — Sal I — Sac II — Sma I/Xma I — Age I — TurboGFP

... AGG.CTG.CTA.GCG.CTA.CCG.GAC.TCA.GAT.CTC.GAG.CTC.AAG.CTT.CGA.ATT.CTG.CAG.TCG.ACG.GTA.CCG.CGG.GCC.CGG.GAT.CCA.CCG.GTC.GCC.ACC.ATG.G...

Afe I — Xho I — Hind III — Pst I* — Kpn I — Apa I* — BamH I — Nco I*

* — not unique sites.

Bacterial expression vectors



For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

Vector type	bacterial expression vector
Reporter(s)	TurboGFP, TurboYFP, PhiYFP, TurboRFP, TurboFP602
Reporter codon usage	mammalian
Promoter	T5 promoter/lac operator
Host cells	prokaryotic
Selection	ampicillin
Replication	ColE1 ori
Use	Source of the reporter coding sequence; reporter expression in bacterial cells

Product	Cat.#	Reporter	Color	Size
pTurboRFP-B	FP233	TurboRFP	red (orange)	20 μg
pTurboGFP-B	FP513	TurboGFP	green	20 μg
pPhi-Yellow-B	FP603	PhiYFP	yellow	20 μg
pTurboYFP-B	FP613	TurboYFP	yellow	20 μg
pTurboFP602-B	FP713	TurboFP602	red	20 μg

Please contact your local distributor for exact prices and delivery information.

pTurboGFP-B vector 5' Region

RBS ATG.AGA.GGA.TCG.GGA.TCC.GAG.A... (TurboGFP)
BamH I

pTurboGFP-B vector 3' Region

...TGA.AGC.TT... (STOP)
Hind III

pTurboYFP-B vector 5' Region

RBS ATG.AGA.GGA.TCG.GGA.TCC.ATG.A... (TurboYFP)
BamH I

pTurboYFP-B vector 3' Region

...TGA.AAG.CTT... (STOP)
Hind III

pPhi-Yellow-B vector 5' Region

RBS ATG.AGA.GGA.TCG.GGA.TCC.A... (PhiYFP)
BamH I

pPhi-Yellow-B vector 3' Region

...TGA.AGC.TT... (STOP)
Hind III

pTurboRFP-B vector 5' Region

RBS ATG.AGA.GGA.TCG.GGA.TCC.ATG.A... (TurboRFP)
BamH I

pTurboRFP-B vector 3' Region

...TGA.AGC.TT... (STOP)
Hind III

pTurboFP602-B vector 5' Region

RBS ATG.AGA.GGA.TCG.GGA.TCC.ATG.G... (BamH I; TurboFP602)
Nco I*

pTurboFP602-B vector 3' Region

...TGA.AGC.TT... (STOP)
Hind III

* — not unique sites.

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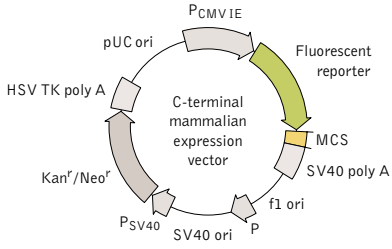
C-terminal mammalian expression vectors



For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

Vector type	mammalian expression vector
Reporter(s)	TagBFP, TagCFP, TagGFP2, TagYFP, TagRFP, mKate2, TurboGFP, TurboYFP, PhiYFP-m, TurboRFP, TurboFP602, TurboFP635
Reporter codon usage	mammalian
Promoter	$P_{CMV\ IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Reporter expression in mammalian cells; generation of fusions to the reporter C-terminus

Product	Cat.#	Reporter	Color	Size
pTagCFP-C	FP111	TagCFP	cyan	20μg
pTagYFP-C	FP131	TagYFP	yellow	20μg
pTagRFP-C	FP141	TagRFP	red (orange)	20μg
pTagBFP-C	FP171	TagBFP	blue	20μg
pmKate2-C	FP181	mKate2	far-red	20μg
pTagGFP2-C	FP191	TagGFP2	green	20μg
pTurboRFP-C	FP231	TurboRFP	red (orange)	20μg
pTurboGFP-C	FP511	TurboGFP	green	20μg
pPhi-Yellow-C	FP601	PhiYFP-m	yellow	20μg
pTurboYFP-C	FP611	TurboYFP	yellow	20μg
pTurboFP602-C	FP711	TurboFP602	red	20μg
pTurboFP635-C	FP721	TurboFP635	far-red	20μg

Please contact your local distributor for exact prices and delivery information.

Multiple cloning sites (MCS)

pTagCFP-C vector MCS

TagCFP → ... TCC.GGA.CTC.AGA.TCT.CGA.GCT.CAA.GCT.TCG.AAT.TCT.GCA.GTC.GAC.GGT.ACC.GCG.GGC.CCG.GGA.TCC.ACC.GGA.TCT.AGA.TAA.CTG.ATC.A ...
Xho I | Hind III | Pst I | Kpn I | Apa I | BamH I | STOPs
BspE I | Bgl II | Sac I | EcoR I | Sal I | Sac II* | Sma I/Xma I | Xba I# | Bcl I#

pTagYFP-C vector MCS

TagYFP → ... TCC.GGA.CTC.AGA.TCT.CGA.GCT.CAA.GCT.TCG.AAT.TCT.GCA.GTC.GAC.GGT.ACC.GCG.GGC.CCG.GGA.TCC.ACC.GGA.TCT.AGA.TAA.CTG.ATC.A ...
Xho I | Hind III | Pst I* | Kpn I | Apa I | BamH I | STOPs
BspE I | Bgl II | Sac I | EcoR I | Sal I | Sac II* | Sma I/Xma I | Xba I# | Bcl I#

pTagRFP-C vector MCS

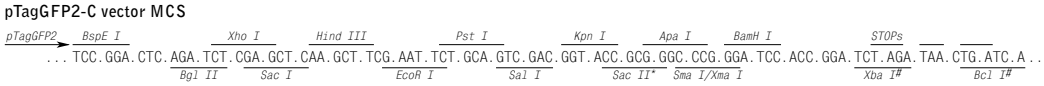
TagRFP → ... TCC.GGA.CTC.AGA.TCT.CGA.GCT.CAA.GCT.TCG.AAT.TCT.GCA.GTC.GAC.GGT.ACC.GCG.GGC.CCG.GGA.TCC.ACC.GGA.TCT.AGA.TAA.CTG.ATC.A ...
Xho I | Hind III | Pst I | Kpn I | Apa I | BamH I | STOPs
BspE I | Bgl II | Sac I | EcoR I | Sal I | Sac II | Sma I/Xma I | Xba I# | Bcl I#

pTagBFP-C vector MCS

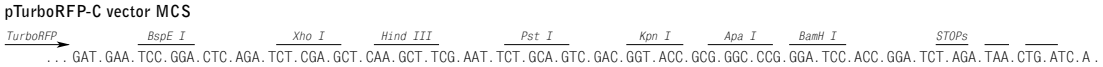
TagBFP → ... TCC.GGA.CTC.AGA.TCT.CGA.GCT.CAA.GCT.TCG.AAT.TCT.GCA.GTC.GAC.GGT.ACC.GCG.GGC.CCG.GGA.TCC.ACC.GGA.TCT.AGA.TAA.CTG.ATC.A ...
Xho I | Hind III* | Pst I | Kpn I | Apa I | BamH I | STOPs
BspE I | Bgl II | Sac I | EcoR I | Sal I | Sac II | Sma I/Xma I | Xba I# | Bcl I#

pmKate2-C vector MCS

pTagGFP2-C vector MCS

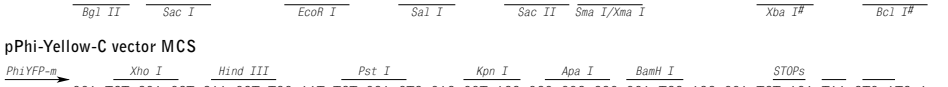


pTurboRFP-C vector MCS

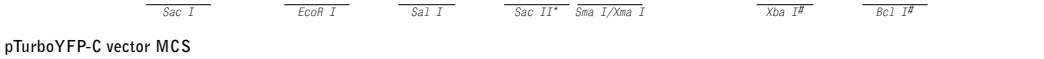


pTurboGFP-C vector MCS

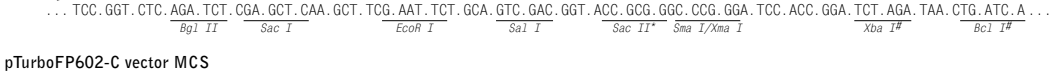
pPhi-Yellow-C vector MCS



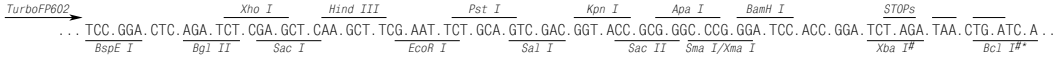
pTurboYFP-C vector MCS



pTurboFP602-C vector MCS



pTurboFP635-C vector MCS



* — not unique sites.

\# — sites are blocked by *dam* methylation. If you wish to digest the vector with these enzymes, you will need to transform the vector into a *dam*$^-$ host and make fresh DNA.

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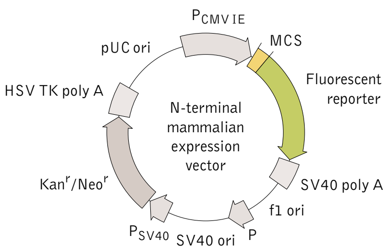
The CMV promoter is covered under U.S. Patents 5,168,062 and 5,385,839, and its use is permitted for research purposes only. Any other use of the CMV promoter requires a license from the University of Iowa Research Foundation, 214 Technology Innovation Center, Iowa City, IA 52242.

MATERIAL SAFETY DATA SHEET INFORMATION: To the best of our knowledge, these products do not require a Material Safety Data Sheet. However, all the properties of these products (and, if applicable, each of their components) have not been thoroughly investigated. Therefore, we recommend that you use gloves and eye protection, and wear a laboratory coat when working with these products.

N-terminal mammalian expression vectors



For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

*-dest1 - fluorescent proteins fused with protein degradation sequence.

Vector type	mammalian expression vector
Reporter(s)	TagBFP, TagCFP, TagGFP, TagYFP, TagRFP, mKate2, TurboGFP, TurboYFP, PhiYFP, TurboRFP, TurboFP602, TurboFP635, TurboGFP-dest1*, TurboYFP-dest1*, PhiYFP-m-dest1*, TurboRFP-dest1*
Reporter codon usage	mammalian
Promoter	$P_{CMV IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Reporter expression in mammalian cells; generation of fusions to the reporter N-terminus

Product	Cat.#	Reporter	Color	Size
pTagCFP-N	FP112	TagCFP	cyan	20μg
pTagYFP-N	FP132	TagYFP	yellow	20μg
pTagRFP-N	FP142	TagRFP	red (orange)	20μg
pTagBFP-N	FP172	TagBFP	blue	20μg
pmKate2-N	FP182	mKate2	far-red	20μg
pTagGFP2-N	FP192	TagGFP2	green	20μg
pTurboRFP-N	FP232	TurboRFP	red (orange)	20μg
pTurboRFP-dest1	FP239	TurboRFP-dest1	red (orange)	20μg
pTurboGFP-N	FP512	TurboGFP	green	20μg
pTurboGFP-dest1	FP519	TurboGFP-dest1	green	20μg
pPhi-Yellow-N	FP602	PhiYFP	yellow	20μg
pPhi-Yellow-dest1	FP608	PhiYFP-dest1	yellow	20μg
pTurboYFP-N	FP612	TurboYFP	yellow	20μg
pTurboYFP-dest1	FP619	TurboYFP-dest1	yellow	20μg
pTurboFP602-N	FP712	TurboFP602	red	20μg
pTurboFP635-N	FP722	TurboFP635	far-red	20μg

Please contact your local distributor for exact prices and delivery information.

Multiple cloning sites (MCS)

pTagCFP-N vector MCS

Above: *Afe I*, *Xho I*, *Hind III*, *Pst I*, *Kpn I*, *Apa I*, *BamH I*, *TagCFP* →

... GCT. AGC. GCT. ACC. GGA. CTC. AGA. TCT. CGA. GCT. CAA. GCT. TCG. AAT. TCT. GCA. GTC. GAC. GGT. ACC. GCG. GGC. CCG. GGA. TCC. ACC. GGT. CGC. CAC. *CAT. GA* ...

Below: *Nhe I*, *Bgl II*, *Sac I*, *EcoR I*, *Sal I*, *Sac II**, *Sma I/Xma I*, *Age I*

pTagYFP-N vector MCS

Above: *Nhe I*, *Bgl II*, *Sac I*, *EcoR I*, *Sal I*, *Sac II**, *Sma I/Xma I*, *Age I*, *TagYFP* →

... GCT. AGC. GCT. ACC. GGA. CTC. AGA. TCT. CGA. GCT. CAA. GCT. TCG. AAT. TCT. GCA. GTC. GAC. GGT. ACC. GCG. GGC. CCG. GGA. TCC. ACC. GGT. CGC. CAC. *CAT. GG* ...

Below: *Afe I*, *Xho I*, *Hind III*, *Pst I**, *Kpn I*, *Apa I*, *BamH I*, *Nco I**

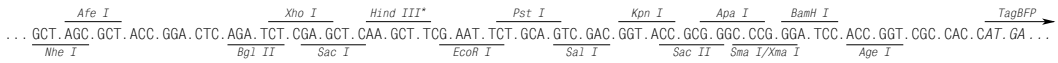
pTagRFP-N vector MCS

Above: *Nhe I*, *Bgl II*, *Sac I*, *EcoR I*, *Sal I*, *Sac II*, *Sma I/Xma I*, *Age I*, *TagRFP* →

... GCT. AGC. GCT. ACC. GGA. CTC. AGA. TCT. CGA. GCT. CAA. GCT. TCG. AAT. TCT. GCA. GTC. GAC. GGT. ACC. GCG. GGC. CCG. GGA. TCC. ACC. GGT. CGC. CAC. *CAT. GG* ...

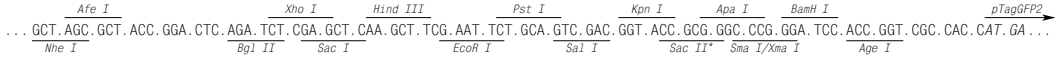
Below: *Afe I*, *Xho I*, *Hind III*, *Pst I*, *Kpn I*, *Apa I*, *BamH I*, *Nco I**

pTagBFP-N vector MCS

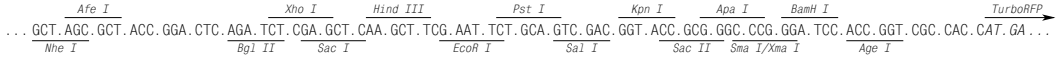


pmKate2-N vector MCS

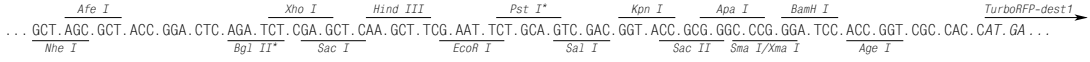
pTagGFP2-N vector MCS



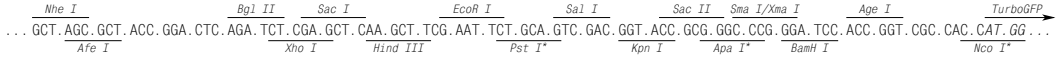
pTurboRFP-N vector MCS



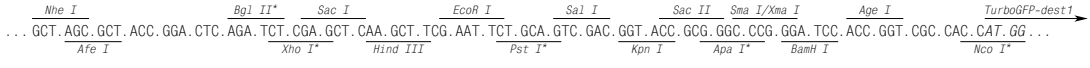
pTurboRFP-dest1 vector MCS



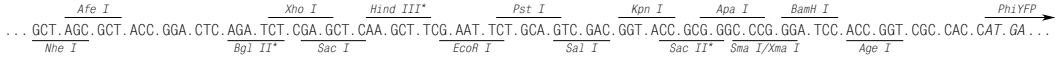
pTurboGFP-N vector MCS



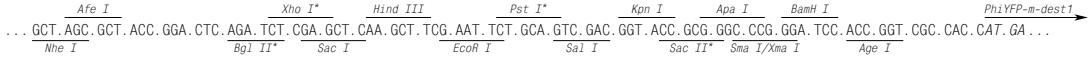
pTurboGFP-dest1 vector MCS



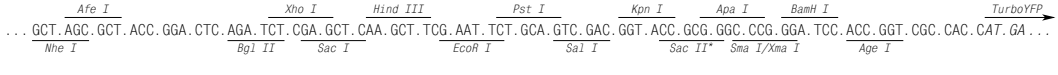
pPhi-Yellow-N vector MCS



pPhi-Yellow-dest1 vector MCS



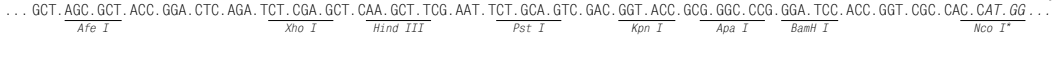
pTurboYFP-N vector MCS



pTurboYFP-dest1 vector MCS

pTurboFP602-N vector MCS

pTurboFP635-N vector MCS



* — not unique sites.

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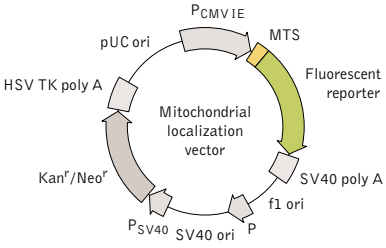
Mitochondria localization vectors



For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

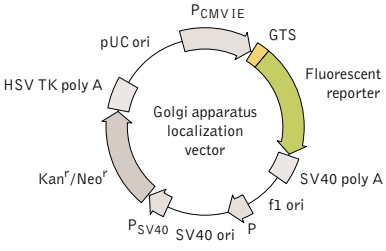
MTS - Mitochondrial targeting sequence

Vector type	mammalian expression vector
Reporter(s)	TagCFP, TagGFP2, TagYFP, TagRFP, TurboGFP, PhiYFP, TurboRFP, TurboFP602
Reporter codon usage	mammalian
Promoter	$P_{CMV\ IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Fluorescent labeling of mitochondria

Product	Cat.#	Reporter	Color	Size
pTagCFP-mito	FP117	TagCFP	cyan	20μg
pTagYFP-mito	FP137	TagYFP	yellow	20μg
pTagRFP-mito	FP147	TagRFP	red (orange)	20μg
pmKate2-mito	FP187	mKate2	far-red	20μg
pTagGFP2-mito	FP197	TagGFP2	green	20μg
pTurboRFP-mito	FP237	TurboRFP	red (orange)	20μg
pTurboGFP-mito	FP517	TurboGFP	green	20μg
pPhi-Yellow-mito	FP607	PhiYFP	yellow	20μg
pTurboFP602-mito	FP717	TurboFP602	red	20μg

Please contact your local distributor for exact prices and delivery information.

Golgi apparatus localization vectors



For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

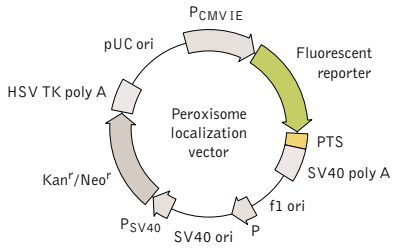
GTS - Golgi targeting sequence

Vector type	mammalian expression vector
Reporter(s)	TagRFP
Reporter codon usage	mammalian
Promoter	$P_{CMV\ IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Fluorescent labeling of Golgi apparatus

Product	Cat.#	Reporter	Color	Size
pTagRFP-Golgi	FP367	TagRFP	red (orange)	20μg

Please contact your local distributor for exact prices and delivery information.

Peroxisome localization vectors



For vector sequence, please visit our Web site at
http://www.evrogen.com/support/vector-info.shtml

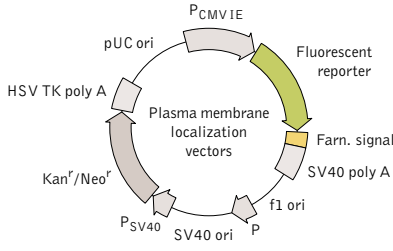
PTS - Peroximal targeting signal

Vector type	mammalian expression vector
Reporter(s)	PhiYFP-m
Reporter codon usage	mammalian
Promoter	$P_{CMV\ IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Fluorescent labeling of peroxisomes

Product	Cat.#	Reporter	Color	Size
pmKate2-peroxi	FP313	mKate2	far-red	20 μg
pPhi-Yellow-peroxi	FP606	PhiYFP	yellow	20 μg

Please contact your local distributor for exact prices and delivery information.

Plasma membrane localization vectors



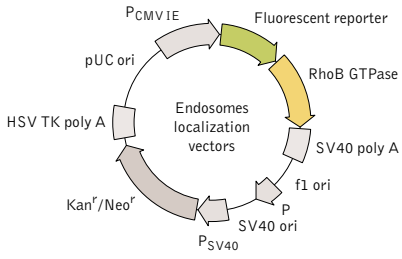
For vector sequence, please visit our Web site at
http://www.evrogen.com/support/vector-info.shtml

Vector type	mammalian expression vector
Reporter	mKate2
Reporter codon usage	mammalian
Promoter	$P_{CMV\ IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori eukaryotic - SV40 ori
Use	Far-red fluorescent labeling of plasma membrane

Product	Cat.#	Reporter	Color	Size
pmKate2-f-mem vector	FP186	mKate2	far-red	20 μg

Please contact your local distributor for exact prices and delivery information.

Endosome localization vectors



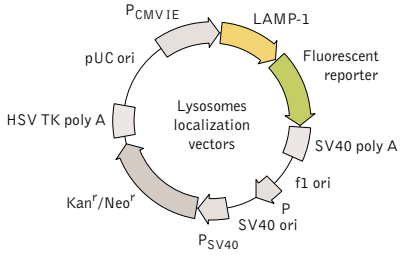
For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

Vector type	mammalian expression vector
Reporter	mKate2
Reporter codon usage	mammalian
Promoter	$P_{CMV\ IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Far-red fluorescent labeling of vesicles of the endocytic pathway

Product	Cat.#	Reporter	Color	Size
pmKate2-endo vector	FP314	mKate2	far-red	20 μg

Please contact your local distributor for exact prices and delivery information.

Lysosome localization vectors



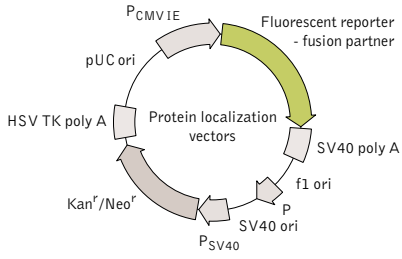
For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

Vector type	mammalian expression vector
Reporter	mKate2
Reporter codon usage	mammalian
Promoter	$P_{CMV\ IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Far-red fluorescent labeling of lysosomes

Product	Cat.#	Reporter	Color	Size
pmKate2-lyso vector	FP312	mKate2	far-red	20 μg

Please contact your local distributor for exact prices and delivery information.

Protein localization vectors



For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

TagFP635* - the parental variant of mKate2.

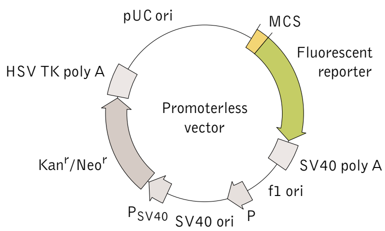
Vector type	mammalian expression vector
Reporter(s)	TagBFP, TagCFP, TagGFP2, TagYFP, TagRFP, mKate2, TagFP635*
Reporter codon usage	mammalian
Promoter	$P_{CMV\ IE}$
Host cells	mammalian
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Fluorescent protein labeling in living cells

Fusion partner	Product	Cat.#	Reporter	Color	Size
Cytoskeletal and adhesion proteins					
β-actin	pTagCFP-actin	FP114	TagCFP	cyan	20μg
	pTagYFP-actin	FP134	TagYFP	yellow	20μg
	pTagRFP-actin	FP144	TagRFP	red (orange)	20μg
	pTagBFP-actin	FP174	TagBFP	blue	20μg
	pmKate2-actin	FP184	mKate2	far-red	20μg
	pTagGFP2-actin	FP194	TagGFP2	green	20μg
α-tubulin	pTagCFP-tubulin	FP115	TagCFP	cyan	20μg
	pTagYFP-tubulin	FP135	TagYFP	yellow	20μg
	pTagRFP-tubulin	FP145	TagRFP	red (orange)	20μg
	pTagBFP-tubulin	FP175	TagBFP	blue	20μg
	pmKate2-tubulin	FP185	mKate2	far-red	20μg
	pTagGFP2-tubulin	FP195	TagGFP2	green	20μg
focal adhesion kinase	pTagRFP-FAK	FP366	TagRFP	red (orange)	20μg
paxillin	pmKate2-paxillin	FP323	mKate2	far-red	20μg
profilin	pTagRFP-profilin	FP371	TagRFP	red (orange)	20μg
	pmKate2-profilin	FP320	mKate2	far-red	20μg
vimentin	pmKate2-vimentin	FP318	mKate2	far-red	20μg
vinculin	pTagRFP-vinculin	FP372	TagRFP	red (orange)	20μg
	pTagFP635-vinculin	FP388	TagFP635	far-red	20μg
α-actinin	pTagRFP-actinin	FP360	TagRFP	red (orange)	20μg
	pmKate2-actinin	FP317	mKate2	far-red	20μg
α-V-integrin	pTagRFP-integrin	FP361	TagRFP	red (orange)	20μg
EB3 protein	pmKate2-EB3	FP316	mKate2	far-red	20μg
	pTagRFP-EB3	FP365	TagRFP	red (orange)	20μg
cytokeratin-18	pmKate2-keratin	FP319	pmKate2	far-red	20μg
	pTagRFP-keratin	FP369	TagRFP	red (orange)	20μg
zyxin	pmKate2-zyxin	FP315	mKate2	far-red	20μg

	pTagRFP-zyxin	FP373	TagRFP	red (orange)	20μg
Gap junction proteins					
connexin 26	pTagRFP-Cx26	FP362	TagRFP	red (orange)	20μg
	pTagFP635-Cx26	FP382	TagFP635	far-red	20μg
connexin 32	pTagRFP-Cx32	FP363	TagRFP	red (orange)	20μg
	pTagFP635-Cx32	FP383	TagFP635	far-red	20μg
connexin 43	pTagRFP-Cx43	FP364	TagRFP	red (orange)	20μg
	pTagFP635-Cx43	FP384	TagFP635	far-red	20μg
Vesicular transport proteins					
clathrin light chain LCB	pmKate2-clathrin	FP322	pmKate2	far-red	20μg
Nuclear proteins					
histone H2B	pTagBFP-H2B	FP176	TagBFP	blue	20μg
	pTagRFP-H2B	FP368	TagRFP	red (orange)	20μg
	pmKate2-H2B	FP311	mKate2	far-red	20μg
lamin B1	pmKate2-laminB1	FP310	mKate2	far-red	20μg
	pTagRFP-laminB1	FP370	TagRFP	red (orange)	20μg
Other					
annexin	pmKate2-annexin	FP321	mKate2	far-red	20μg

Please contact your local distributor for exact prices and delivery information.

Promoterless vectors



For vector sequence, please visit our Web site at http://www.evrogen.com/support/vector-info.shtml

*-dest1 - fluorescent proteins fused with protein degradation sequence.

Vector type	promoterless vector
Reporter(s)	TurboGFP, TurboGFP-dest1*, TurboYFP, TurboYFP-dest1*, PhiYFP-m, PhiYFP-m-dest1*, TurboRFP, TurboRFP-dest1*, TurboFP602
Reporter codon usage	mammalian
Promoter	No
Host cells	mammalian, prokaryotic
Selection	prokaryotic - kanamycin; eukaryotic - neomycin (G418)
Replication	prokaryotic - pUC ori; eukaryotic - SV40 ori
Use	Monitoring of the activity of different promoters and promoter/enhancer combinations introduced to the vector MCS

Product	Cat.#	Reporter	Color	Size
pTurboRFP-PRL	FP235	TurboRFP	red (orange)	20μg
pTurboRFP-PRL-dest1	FP238	TurboRFP-dest1	red (orange)	20μg
pTurboGFP-PRL	FP515	TurboGFP	green	20μg
pTurboGFP-PRL-dest1	FP518	TurboGFP-dest1	green	20μg
pPhi-Yellow-PRL	FP604	PhiYFP	yellow	20μg
pPhi-Yellow-PRL-dest1	FP605	PhiYFP-m-dest1	yellow	20μg
pTurboYFP-PRL	FP615	TurboYFP	yellow	20μg
pTurboYFP-PRL-dest1	FP618	TurboYFP-dest1	yellow	20μg
pTurboFP602-PRL	FP715	TurboFP602	red	20μg

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Multiple cloning sites (MCS)

pTurboRFP-PRL vector MCS

Afe I — Xho I — Hind III — Pst I — Kpn I — Apa I — BamH I — TurboRFP →

. . . ACT. AGC. GCT. ACC. GGA. CTC. AGA. TCT. CGA. GCT. CAA. GCT. TCG. AAT. TCT. GCA. GTC. GAC. GGT. ACC. GCG. GGC. CCG. GGA. TCC. ACC. GGT. CGC. CAC. *CAT. GA* . . .

Bgl II — Sac I — EcoR I — Sal I — Sac II — Sma I/Xma I — Age I

pTurboRFP-PRL-dest1 vector MCS

Afe I — Xho I — Hind III — Pst I* — Kpn I — Apa I — BamH I — TurboRFP-dest1 →

. . . ACT. AGC. GCT. ACC. GGA. CTC. AGA. TCT. CGA. GCT. CAA. GCT. TCG. AAT. TCT. GCA. GTC. GAC. GGT. ACC. GCG. GGC. CCG. GGA. TCC. ACC. GGT. CGC. CAC. *CAT. GA* . . .

Bgl II* — Sac I — EcoR I — Sal I — Sac II — Sma I/Xma I — Age I

pTurboGFP-PRL vector MCS

Bgl II — Sac I — EcoR I — Sal I — Sac II — Sma I/Xma I — Age I — TurboGFP →

. . . ACT. AGC. GCT. ACC. GGA. CTC. AGA. TCT. CGA. GCT. CAA. GCT. TCG. AAT. TCT. GCA. GTC. GAC. GGT. ACC. GCG. GGC. CCG. GGA. TCC. ACC. GGT. CGC. CAC. *CAT. GG* . . .

Afe I — Xho I — Hind III — Pst I* — Kpn I — Apa I* — BamH I — Nco I*

pTurboGFP-PRL-dest1 vector MCS

Bgl II* — Sac I — EcoR I — Sal I — Sac II — Sma I/Xma I — Age I — TurboGFP-dest1 →

. . . ACT. AGC. GCT. ACC. GGA. CTC. AGA. TCT. CGA. GCT. CAA. GCT. TCG. AAT. TCT. GCA. GTC. GAC. GGT. ACC. GCG. GGC. CCG. GGA. TCC. ACC. GGT. CGC. CAC. *CAT. GG* . . .

Afe I — Xho I* — Hind III — Pst I* — Kpn I — Apa I* — BamH I — Nco I*

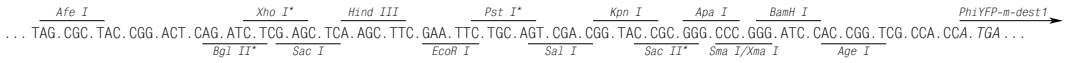
pPhi-Yellow-PRL vector MCS

Afe I — Xho I — Hind III* — Pst I — Kpn I — Apa I — BamH I — PhiYFP →

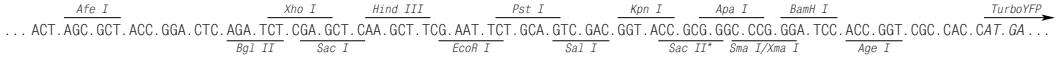
. . . TAG. CGC. TAC. CGG. ACT. CAG. ATC. TCG. AGC. TCA. AGC. TTC. GAA. TTC. TGC. AGT. CGA. CGG. TAC. CGC. GGG. CCC. GGG. ATC. CAC. CGG. TCG. CCA. CCA. *TGA* . . .

Bgl II* — Sac I — EcoR I — Sal I — Sac II* — Sma I/Xma I — Age I

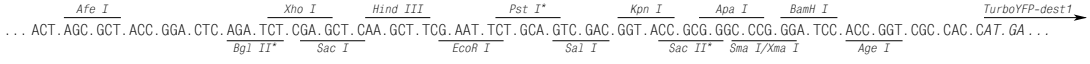
pPhi-Yellow-PRL-dest1 vector MCS



pTurboYFP-PRL vector MCS



pTurboYFP-PRL-dest1 vector MCS



pTurboFP602-PRL vector MCS

* — not unique sites.

\# — sites are blocked by *dam* methylation. If you wish to digest the vector with these enzymes, you will need to transform the vector into a *dam*⁻ host and make fresh DNA.

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Recombinant proteins

Product	Cat.#	Reporter	Color	Size
rTurboGFP	FP552	TurboGFP	green	100μg
rPhiYFP	FP651	PhiYFP	green	100μg

Please contact your local distributor for exact prices and delivery information.

Use

- Standard on protein gels and Western blots
- Control for fluorescence microscopy
- Calibration of fluorimeters and FACS machines
- Microinjection into cells and tissues

rTurboGFP

Recombinant TurboGFP (rTurboGFP) is a 26-kDa green fluorescent protein. It has excitation and emission spectra identical to those of the expressed TurboGFP. rTurboGFP is suitable as control reagent for TurboGFP expression using the TurboGFP expression vectors. rTurboGFP is purified from transformed *E. coli* using organic extraction and hydrophobic chromatography or metal-ion affinity chromatography (methods vary for different lots). Both methods ensure high purity of the recombinant protein and maintenance of fluorescence. The protein concentration is measured by chromophore absorption. rTurboGFP may contain 6xHis tag at its N-terminus (varying in different lots).

rPhiYFP

Recombinant PhiYFP (rPhiYFP) is a 26-kDa yellow fluorescent protein. It has excitation and emission spectra identical to those of the expressed PhiYFP. rPhiYFP is suitable as control reagent for PhiYFP expression using the PhiYFP expression vectors. rPhiYFP is purified from transformed *E. coli* using organic extraction and hydrophobic chromatography or metal-ion affinity chromatography (methods vary for different lots). Both methods ensure high purity of the recombinant protein and maintenance of fluorescence. The protein concentration is measured by chromophore absorption. rPhiYFP may contain 6xHis tag at its N-terminus (varying in different lots).