

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 than 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 allows using these two proteins as a FRET pair.

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 1 000.

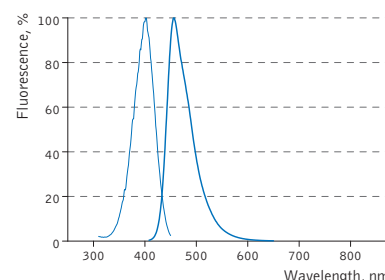
Performance and use

TagBFP can be easily expressed and detected in a wide range of organisms. Mammalian cells transiently transfected with TagBFP expression vectors produce bright fluorescence in 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. It can be used in multicolor labeling applications with cyan, green, yellow, red, and far-red fluorescent dyes.

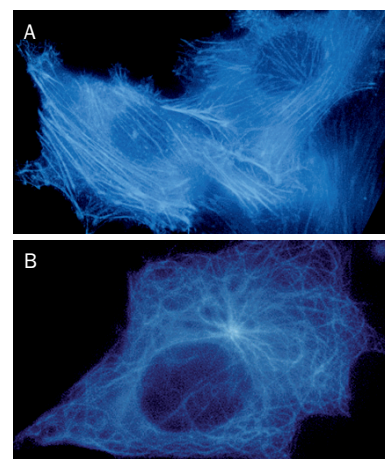
High pH-stability with $pK_a=2.7$ makes it possible to use TagBFP for imaging in acidic organelles, such as late and recycling endosomes and lysosomes.

Emission spectrum of TagBFP and absorbance spectra of GFPs demonstrate good overlap, suggesting TagBFP may be a good donor for GFP acceptor for Forster resonance energy transfer (FRET). Among available GFPs, TagGFP2 is preferable because of the small absorbance below 400 nm where TagBFP is excited. High efficiency of TagBFP-TagGFP2 FRET pair was demonstrated in living cells [Subach et al. 2008].



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 <http://www.evrogen.com>



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.

Recommended filter sets and antibodies

TagBFP can be recognized using Anti-tRFP antibody (Cat.# AB233-AB234) 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

Available variants and fusions

TagBFP mammalian expression vectors contain TagBFP coding sequence with codon usage optimized for high expression in mammalian cells, i.e. humanized [Haas et al. 1996]. Humanized TagBFP can also be expressed in *E. coli* and some other heterologous systems upon subcloning into appropriate vector.

TagBFP-AS codon usage is optimized for expression in *Arabidopsis* and *Saccharomyces*.

The available vectors encoding TagBFP variants and fusions are listed below in the section TagBFP-related products. For most updated product information, please visit Evrogen website www.evrogen.com.

If you need TagBFP codon variant or fusion construct that is not listed on our website, please contact us at product@evrogen.com.

Licensing opportunities

Evrogen technology embodied in TagBFP is available for expanded and commercial use with an adaptable licensing program. Benefits from flexible and market driven license options are offered for upgrade and novel development of products and applications. For licensing information, please contact Evrogen at license@evrogen.com.

References

- Ai, HW et al. (2007). *Biochemistry*, 46 (20): 5904–5910 / pmid: 17444659
- Haas, J. et al. (1996). *Curr Biol*, 6 (3): 315–324 / pmid: 8805248
- Subach, O.M. et al. (2008). *Chemistry & Biology*, 15 (10): 1116–1124 / pmid: 18940671

TagBFP-related products

Product	Cat.#	Description	Size
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
pTagBFP-N	FP172	Mammalian expression vector encoding humanized TagBFP and allowing its expression and generation of fusions to the TagBFP N-terminus	20 µg
pTagBFP-actin	FP174	Mammalian expression vector encoding humanized TagBFP fused with human cytoplasmic β -actin	20 µg
pTagBFP-tubulin	FP175	Mammalian expression vector encoding humanized TagBFP fused with human α -tubulin	20 µg
pTagBFP-H2B	FP176	Mammalian expression vector encoding humanized TagBFP fused with human histone H2B	20 µg
Gateway® TagBFP-AS-C	FP177	Gateway® entry clone for generation of fusions to the C-terminus of TagBFP; transfer of the construct encoding TagBFP or its fusion into Gateway® destination vectors; TagBFP codon usage is optimized for expression in <i>Arabidopsis</i> and <i>Saccharomyces</i>	20 µg
Gateway® TagBFP-AS-N	FP178	Gateway® entry clone for generation of fusions to the N-terminus of TagBFP; transfer of the construct encoding TagBFP or its fusion into Gateway® destination vectors; TagBFP codon usage is optimized for expression in <i>Arabidopsis</i> and <i>Saccharomyces</i>	20 µg
Antibodies against TagBFP			
Anti-tRFP	AB233	Rabbit polyclonal antibody against TurboRFP, TurboFP602, TurboFP635, TurboFP650,	100 µg
	AB234	NirFP, TagBFP, TagRFP, FusionRed, TagFP635, mKate2 and PA-TagRFP	200 µg

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

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TagBFP-related materials (also referred to as "Products") are intended for research use only.

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Invitrogen Gateway® Technology: please see Invitrogen Limited Use Label License No. 19: Gateway® Cloning Products.

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.

MSDS information is available at <http://www.evrogen.com/MSDS.shtml>