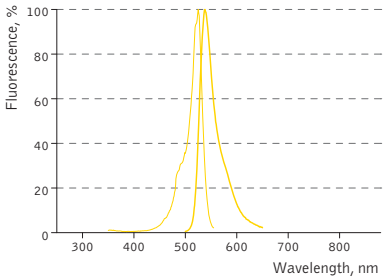
Phi-Yellow fluorescent proteins

- Bright yellow fluorescence
- Proven suitability to generate stably transfected cell lines

PhiYFP and PhiYFP-m are the mutants of a natural yellow fluorescent protein from *Phialidium* sp. (Cnidaria; Hydrozoa; Hydroida; Leptomedusae; Campanulariidae) [Shagin et al. 2004].
Phi-Yellow proteins have excitation/emission maxima at 525 and 537 nm, respectively. They exhibit lower brightness and maturation rate than TurboYFP, but are more suitable for generation of stably transfected cell lines. The emission wavelength of these proteins is ideally positioned between those of green and red fluorescent proteins, allowing easy separation of these fluorescent tags by flow cytometry using common channels of detection and a single laser excitation line.

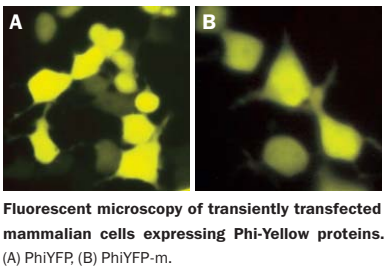


PhiYFP normalized excitation (thin line) and emission (thick line) spectra.
Complete PhiYFP spectra in Excel format can be downloaded from the Evrogen Web site at http://www.evrogen.com

Main properties of PhiYFP

Characteristic	PhiYFP / PhiYFP-m
Molecular weight, kDa	26.8
Polypeptide length, aa	234
Fluorescence color	yellow
Excitation maximum, nm	525
Emission maximum, nm	537
Quantum yield	0.40 / 0.39
Extinction coefficient, $M^{-1}cm^{-1}$	130 000 / 124 000
Brightness*	52.0 / 48.4
Brightness, % of EGFP	158 / 147
pKa	6.0
Structure	weak dimer
Aggregation	no
Maturation rate at 37°C	fast
Photostability	high
Cell toxicity	not observed

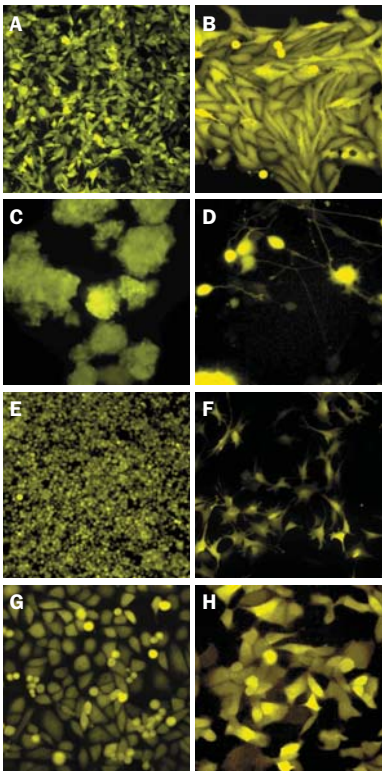
* Brightness is a product of extinction coefficient and quantum yield, divided by 1 000.



Fluorescent microscopy of transiently transfected mammalian cells expressing Phi-Yellow proteins. (A) PhiYFP, (B) PhiYFP-m.

Performance and use

Phi-Yellow proteins can be easily expressed and detected in a wide range of organisms. Mammalian cells transiently transfected with PhiYFP and PhiYFP-m expression vectors produce bright fluorescence in 10-12 hours after transfection. No cytotoxic effects or visible protein aggregation are observed.
Suitability of Phi-Yellow proteins to generate stably transfected cells has been proven by Marinpharm company. Various cell lines are commercially available.
Despite dimerization capacity, Phi-Yellow proteins demonstrate successful performance in fusions with subcellular localization signals and many cellular proteins. However, we recommend that you use TagFPs for protein labeling applications.
Important note: PhiYFP allows 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.
Phi-Yellow proteins can be used in multicolor labeling applications with blue, cyan, green, red, and far-red fluorescent dyes.



Fluorescent microscopy of stably transfected mammalian cells expressing PhiYFP in cytosol. (A) M3 mouse melanoma; (B) T-406 human glioma; (C) PC-12 rat phaeochromocytoma cells; (D) PC-12 cells after the addition of nerve growth factor; (E) Walker 256 rat tumour cells; (F) BC3H1 cells; (G) T24 human bladder carcinoma cells; (H) T24 cells expressing destabilized variant PhiYFP-m-dest1. Images were kindly provided by Dr. Christian Petzelt (Marinpharm).

Recommended filter sets and antibodies

The proteins can be recognized using Anti-TurboYFP (Cat. # AB605) antibody available from Evrogen.
Phi-Yellow proteins can be detected using Omega Optical filter set XF104-3 or Chroma Technology Corp. filter set 42003 ("ZsYellow1").

Available variants and fusions

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

PhiYFP allows generation of fusions . PhiYFP allows generation of fusions to its N-terminus but cannot be used for generation of C-terminal fusions.

PhiYFP-m variant is a mutant of PhiYFP. It is suitable for generation of fusions to its C-terminus.

The available vectors encoding PhiYFP variants and fusions are listed below in the section PhiYFP-related products.

For most updated product information, please visit Evrogen website www.evrogen.com.

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

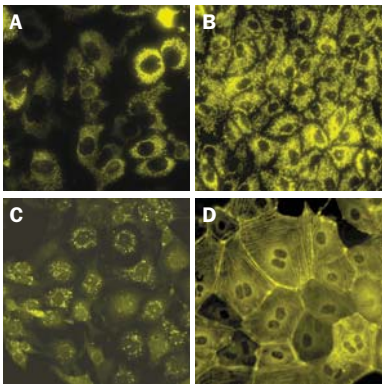
Licensing opportunities

Evrogen technology embodied in PhiYFP 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.

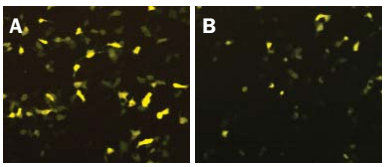
References

Haas, J. et al. (1996). Curr Biol, 6 (3): 315–324 / pmid: 8805248

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



Fluorescent microscopy of stably transfected mammalian cells expressing Phi-Yellow-tagged fusions. (A-B) Mitochondria-targeted PhiYFP in (A) 3T3 mouse fibroblasts; (B) PtK rat kangaroo cells; (C) T24 human bladder carcinoma cells expressing peroxisome-targeted PhiYFP-m; (D) PhiYFP-m fusion with β-actin in PtK rat kangaroo cells. Images were kindly provided by Dr. Christian Petzelt (Marinpharm).



Fluorescence intensities of cells expressing PhiYFP-m-dest1 rapidly decrease in response to cycloheximide (CHX). Mammalian cells expressing PhiYFP-mP-dest1 under the control of CMV promoter were treated with CHX. After 1.5 hours CHX treatment, fluorescence intensity of cells was greatly reduced. (A) – control; (B) – after 1.5 hours from CHX treatment.

PhiYFP-related products

Product	Cat.#	Description	Size
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
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
pPhi-Yellow-B	FP603	Bacterial expression vector; source of the PhiYFP coding sequence	20 μg
pPhi-Yellow-PRL	FP604	Promoterless vector encoding humanized PhiYFP and designed for monitoring of activity of different promoters and promoter/enhancer combinations	20 μg
pPhi-Yellow-peroxi	FP606	Mammalian expression vector encoding humanized PhiYFP-m targeted to peroxisomes	20 μg
pPhi-Yellow-mito	FP607	Mammalian expression vector encoding humanized PhiYFP targeted to mitochondria	20 μg
Recombinant protein			
rPhiYFP	FP651	Purified recombinant bright yellow fluorescent protein	100 μg
Antibodies against PhiYFP			
Anti-TurboYFP	AB605	Rabbit polyclonal antibody against PhiYFP, PhiYFP-m, and TurboYFP	100 μg

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