M09E

Unique Protein Reporter Assays:
Green Fluorescent Proteins (GFP)
Multiplexed Luciferase:
SEAP

Split GFP and **SuperFolder GFP**
This innovative GFP system increases S/N signal, solves solubility/aggregation issues, and, next, protein/protein interactions.

EvoGlow GFP
This unique fluorescent reporter protein can be used in both aerobic and anaerobic biological systems

Single and Dual Luciferase assays, using Purple Cypridina, Blue Gaussia, Green Renilla and Red Firefly Luciferases
Not like current dual luciferase assays, you can not only measure 2 signals, but in one step! = do real multiplex reporter assays!

Standard Luciferase assays, Luciferins and Coelenterazines

A reporter gene assay is a widely utilized molecular technique for measurement of the activity of promoters. A target promoter sequence or response element is linked to a detectable reporter gene which encodes an enzyme like SEAP (secreted alkaline phosphatase), a fluorescent protein such as GFP, or a luciferase such as Firefly luciferase or Renilla luciferase.

Because of their speed, dynamic range and sensitivity and use of relatively inexpensivemicroplate asays, Luminescent or Fluorescent read-out instruments, the SEAP and Luciferase assay provide invaluable tools for the studies of regulatory pathways. The LightSwitch™ Luciferase Assay Reagent enables one-step reagent addition to measure luciferase signal.

See also **Protein expression systems** Products HighLight BB213a , for

LEXSY protein expression system: a unique eukaryotic protein expression system that·combines advantages of bacteria and mammalian systems (rapid growth, full eukaryotic protein folding machinery).

Pierce Protein expression systems:

UptiTherm Cloning and Expression Kit, dual-shuttle system for E.coli and Thermus thermophilus
For cloning and expressing thermophilic proteins in a host system, thermostabilization of mesophilic proteins and direct selection of thermostable mutants in a thermophilic organism.

Protein expression plasmids
A complete range of protein expression plasmids, for mammalian cells, bacteria and, phages with many options for selection (antibiotic), promoters, reporter genes (SEAP, GFP, CAT, X-Gal), peptide/protein tags, …

Protein expression plasmids, for in-vivo LEXSY expression
A unique protein expression system that combines advantages of bacteria and mammalian systems (rapid growth, full eukariotic protein folding machinery), while offering convenient restriction sites for cloning, constitutive cytosolic and secretory vectors, fusion to a C-terminal affinity tag, with four different selection markers, in-vivo expression

In-vitro Cell-Free Protein Expression (Traduction) Systems,
Protein translation systems to produce in-vitro without cells

Transient protein Expression:
Power Expression system using BPfectin, ExpressionMAX supplement, BPM cel culture media optimized for CHO and 293 cell lines.

See also **Fusion proteins detection and purifications** Product HighLight BB213b, for

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Split GFP and SuperFolder GFP, an innovative green fluorescent protein:

Split GFP *and* ***SuperFolder GFP*** allows for improved standard applications, and innovative ones!
This unique GFP system increase S/N signal, and solve solubility/aggregation issues, and next protein/protein interactions.

- **Quantify** the expression level of a target protein, more accurately
- **Localize** the expression of target protein in cells,with more natural distribution
- **Determine** a target protein's solubility, **Discover** which domains of a protein are soluble
- **Evaluate** how a protein interacts with other proteins (protein-protein interaction)
- **Reveal** the effect of a small molecule on the protein's folding

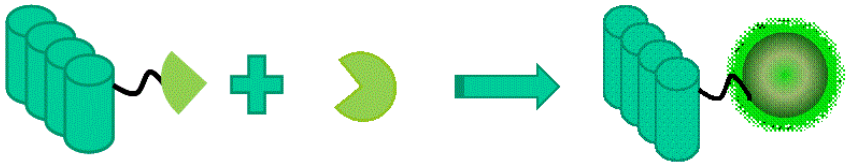
●Ordering information:

In Vitro Mammalian Optimized Split GFP Fold-N-Glow Solubility Assay - Mammalian S11 Single Plasmid	ref.22004003
In Vitro Mammalian Optimized Split GFP Fold-N-Glow Solubility Assay Kit	ref.25004001, 84 tests
In Vitro Bacterial Split GFP Fold-N-Glow Solubility Assay - Bacterial S11	ref.21004003
In Vitro Bacterial Split GFP Fold-N-Glow Solubility Assay Kit	ref.20004001, 84 tests
In Vitro Split GFP Fold-N-Glow Solubility Assay - Positive Control	ref.21004002
In Vitro Split GFP Fold-N-Glow Solubility Assay - Universal Detection Reagent	ref.21004001
Superfolder GFP Expression Plasmid	ref.23004006

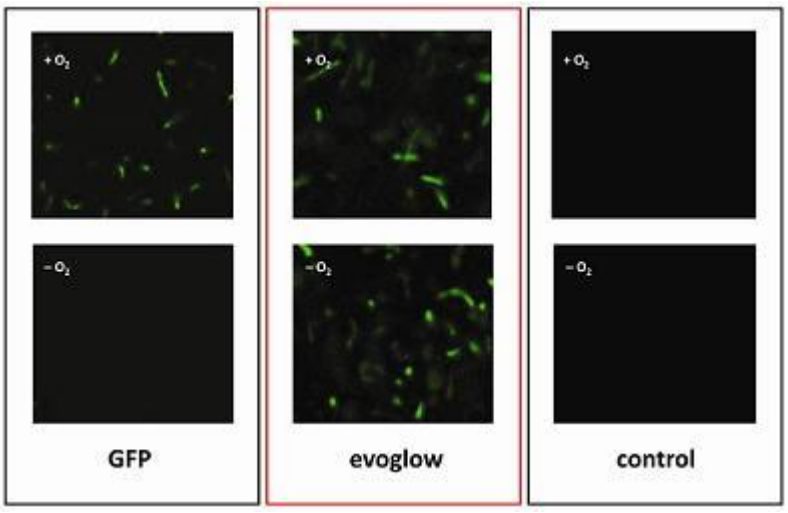
⇨ See detailled information (BB213b) | Protein Expression | Solubility | Soluble Domains | Protein-Protein Interactions | …

EvoGlow GFP ,
evocatal's anaerobic fluorescent proteins

Fluorescent reporter proteins are valuable non-invasive molecular tools for in vivo real-time imaging of living cells and tissues as well as in vitro fluorescence labeling. A major drawback of existing GFP-like reporter proteins is their strict **requirement for molecular oxygen** as a cofactor for the synthesis of their respective chromophores. Therefore, the application in anaerobic systems is not possible.
The flavin mononucleotide (FMN)-based fluorescent proteins (FbFPs) from the evoglow® series were developed to overcome these restrictions. The **evoglow® proteins can be used as fluorescent reporters in both aerobic and anaerobic biological systems**.



A properly folded protein with exposed GFP S11 fragment — Self assembles to a GFP S1-10 detection fragment — Resulting in a fluorescent GFP fusion protein



The designation evoglow® comprises a novel type of fluorescent proteins containing a Flavin-mono-nucleotide-based cofactor. They are thus capable of developing bright cyan-green fluorescence even in the complete absence of oxygen. They fold rapidly in vivo and thus enable immediate detection of expression to study anaerobic inter- and intracellular processes. Since their fluorescence intensity as well as the excitation and emission spectra of the evoglow® proteins lie within the same range as those of other fluorescent proteins, the same instrumental setup can be used for their detection and analysis as for other fluorescent proteins.

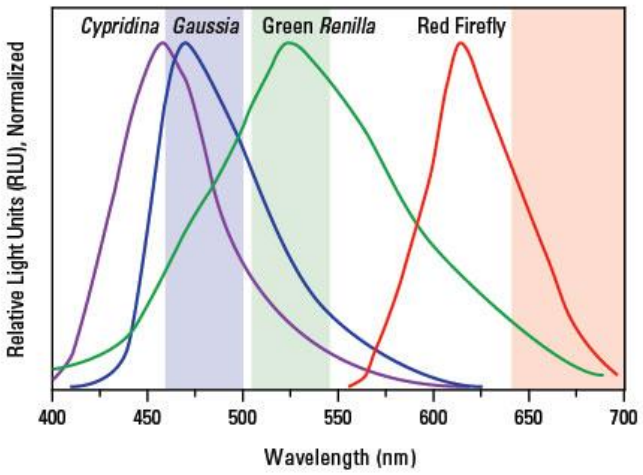
⇨ See detailled information (BB213e)

| **evoglow® Plasmid Kits** : The evoglow® Plasmid Kits allow easy cloning and expression of the evoglow® genes.

| **evoglow® Antibodies** : evoglow® antibodies are approved for the specific detection of the fluorescence reporters by Western Blot.

Single and Dual Luciferase assays,
using Purple Cypridina, Blue Gaussia, Green Renilla and Red Firefly Luciferases

The **Gaussia**, **Gaussia-Dura** (a mutant form of the Gaussia luciferase gene that confers better bioluminescent signal stability than native luciferase) and **Cypridina** luciferase proteins are secreted into the cell culture media, allowing for live cell monitoring of reporter activity. They produce a light signal considerably greater than the signal from either native Firefly or Renilla luciferases.



The **Green Renilla** luciferase is an intracellular protein with a greater serum stability as well as higher light output than native Renilla and Firefly luciferases. It allows more sensitive reporter assays.

The **Red Firefly** luciferase is an intracellular protein with an emission peak shifted into the red spectrum compared to the native Firefly luciferase. It allows multiplexing with Gaussia, Cypridina and Green Renilla luciferases.

They are available as:

- Vectors
- Single Reporter Assay Kits, in both flash and glow formats.

The **Flash assay** reagents may require the use of a luminometer with reagent injectors for the greatest signal strength and reduced variability.

Glow-type assay provide a less-intense but longer-duration signal that does not require reagent injectors, and is amenable to HTS.

- Dual Reporter Assay Kits

The assay kits enable **simultaneous** monitoring of an experimental reporter activity (Cypridina, Gaussia or Renilla Luciferase) and a Red Firefly luciferase reporter activity, in a single-read assay without the need for two-step addition of substrate reagents or quenching. The assay kit contains substrates for both luciferases, and the reactions occur simultaneously with flash-type kinetics. The resulting luminescent signals are spectrally resolvable using filters. The kit can be used as a reporter-and-control or assay (Red Firefly is then constitutively expressed), or for multiplexe assays (both luciferases are experimental reporters).

	Cypridina	**Gaussia**	**Gaussia-Dura**	**Green Renilla**	**Red Firefly**
Signal color	Purple	Blue		Green	Red
Substrate	Vargulin	Coelenterazine		Coelenterazine	Luciferin
Sample type	10^7 (media) 10^6 (lysate)	10^7 (media) 10^6 (lysate)		10^7 (lysate)	10^5 (lysate)
Half-life – flash Half-life - glow	30min 1hr	<5min n/a	n/a 1hr	5min 3hr	>30min 2hr
Vector & controls	16149 (pMCS) 16151(pCMV) 16151(pTK)	16146 (pMCS) 16147(pCMV) 16148(pTK)	16190 (pMCS) 16191(pCMV) 16192(pTK)	16152 (pMCS) 16153(pCMV), 16154(pTK)	16155 (pMCS) 16156(pCMV), 16157(pTK)
Co-transfection	• Gaussia Luc • Gaussia-Dura Luc • Green Renilla Luc • Red Firefly Luc		• Cypridina Luc • Red Firefly Luc		• Gaussia Luc • Gaussia-Dura Luc • Green Renilla Luc • Cypridina Luc
Single reporter kits	16174 & 16175 (Glow)	16158 & 16159 (Flash)	16160 & 16161 (Glow)	16164 & 16165 (Flash) 16166 & 16167 (Glow)	16174 & 16175 (Flash) 16176 & 16177 (Glow)
Dual reporter kits	16183 (with RedFirefly) 460nm BP/ 615nm	16181 (with RedFirefly) 470nm BP/ 615LP	-	16185 (with RedFirefly) 525nm BP/640 LP	N/A

Inquire for more information.

Associated products for Luciferase assays:
-Transfection reagents: UptiFectinON DNA transfection agent
-Cell Lysis Buffers:
-Firefly Luciferase Signal Enhancer: LV5870-16180, 0.5ml (1000tests)

Luciferins & Coelenterazines substrates, and Luciferase assays kits

●See **plasmids expressing GFP** in the Genomics chapter.

●Selected **FluoProbes Luciferin substrates** for Luciferase assays:
Over 9 salt forms and analogs available (including Ethyl- and Methyl-esters, Amino-, Galactos- derivatives).

●Selected **FluoProbes Coelenterazine substrates** for Aequaporin assays and calcium assays:
Over 12 analogs available (Coelenterazines sampler kit UP42176A include 8 analogs)

●See **Standard Luciferase Assay kits**:
Firefly & Renilla Luciferase Assay kit, FP-BE7810
Firefly Luciferase Assay Kit, Bright Glow, FP-JQ6811
Renilla Luciferase Assay Kit, FP-BE7930

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Protein expression plasmids OXG

Protein expression plasmids, for mammalian cells, bacteria and phages, with many options for antibiotic selection, promoters, reporter genes (SEAP, GFP, CAT, X-Gal), peptide/protein tags, ...
See Product Highlight Products HighLight BB215a .

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Related products/documents from Interchim - BioSciences Innovations:

- RNA transfection reagents. , i.e. **UptiFectinOff**, **Pre-Design** chimera **RNAi** products.
- Recombinant protein production, i.e. **LEXSY2 protein expression system** (combines scalability with full-eucaryotic machinery)
- Molecular Biology reagents:
  - **RealTime PCR**
  - **GelRed**, safe alternative to BET: simply the best nucleic acids stain: non-carcinogen, highly stable, more sensitive
  - **Ribozol** RNA extraction reagent: purify high yields of RNA – free of any DNA !
- Products HighLights **Overview** and on-line BioSciences Innovations at http://www.interchim.com

For ordering information please search at http://www.interchim.eu
For more information, please ask interbiotech@interchim.com

Information inquire

Reply by Fax : +33 (0) 4 70 03 82 60 or email at interbiotech@interchim.com

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