TrueClone and TrueORF products:

The world's largest collection of expression-ready, full-length cDNA clones (human and mouse)

There are two major classes of clones offered by OriGene

- **TrueClones:** Library-based, full-length cDNA clones that usually contain native 5′ and 3′ untranslated regions; in a mammalian expression vector
- **TrueORF Clones:** Tagged open reading frame (ORF) clones in OriGene's mammalian expression vector, the PrecisionShuttle entry vector. The ORF insert can be easily shuttled with a simple digestion/ligation reaction into a wide variety of tagged destination vectors.

Human TrueClone	Authentic, full-length human cDNA delivered as 10 ug transfection-ready plasmid DNA
Mouse TrueClone	Authentic, full-length mouse cDNA delivered as 10 ug transfection-ready plasmid DNA
TrueClone Access Collection	A subset of the most popular TrueClones, priced at $95 USD for individual purchase and $5 each for bulk purchase.
Human TrueORF	Tagged human ORFs in PrecisionShuttle vector, delivered as 10 ug transfection-ready plasmid DNA
TurboFectin 8.0	Transfection reagent optimized for use with OriGene's clones for mammalian cell transfection and protein production

Both TrueClones and TrueORFs are CMV promoter-driven expression vectors suitable for transfection into mammalian cells and protein overexpression. While TrueClones express the native transcripts without any epitope tags, TrueORFs express encoded sequences as tagged proteins, facilitating detection, purification and localization with anti-tag antibodies.

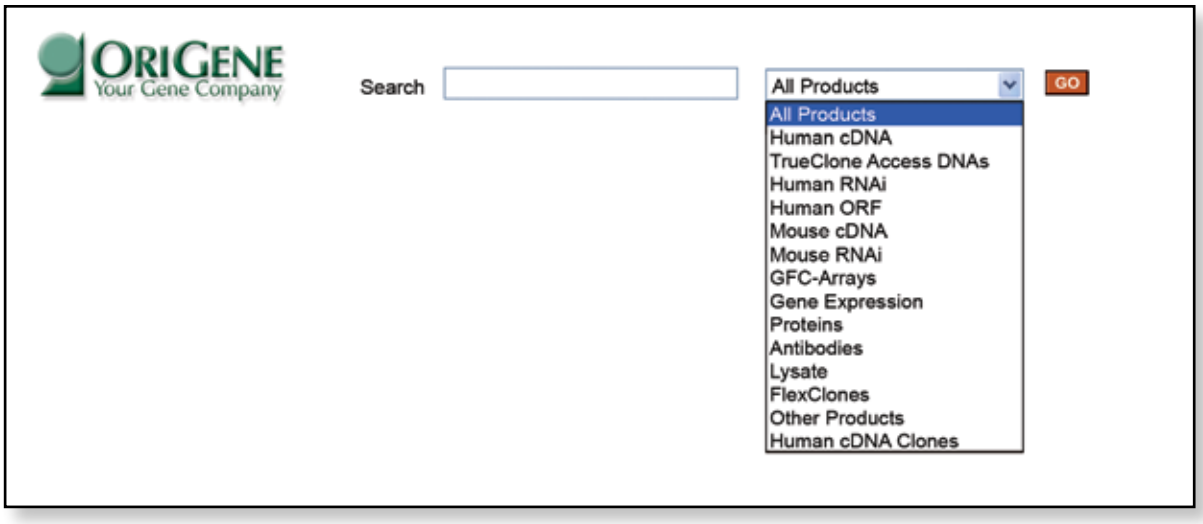
ORIGENE
Your Gene Company

How to search for a clone:

A search box is located at the top of every page of the OriGene website. Any one of three easy approaches can be used to identify the appropriate clone. Simply type into the search box:

an NCBI Accession Number (eg. NM_000044)
or
the HUGO symbol of the gene (eg. AR)
or
the gene description (eg. androgen receptor)

Another option is search the clone by BLAST. Go to the BLAST page of OriGene's website (http://blast.origene.com/blast2/index.php), paste in your target sequence, then click on the button labeled "Submit BLAST Query". On the resulting page, click on the nucleotide accession number that has the best match (lowest E value) to your sequence. You'll be directed to the product page that is the closest match to the sequence you submitted.



How to find sequence information or pricing of the clones:

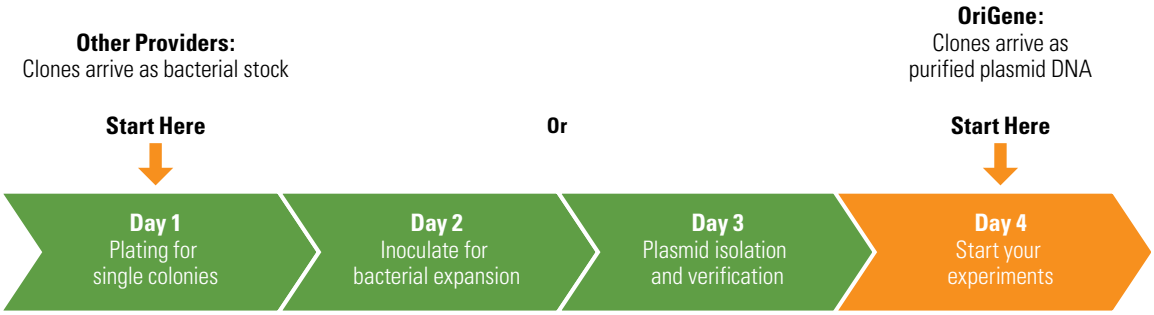
Click on the catalog number that corresponds to your product of interest, then click on the orange "Details and Pricing" button. After entering a valid email address, you'll be directed to a page containing the product's list price and availability, related products, vector identity, insert size, end sequence information, and reference sequence information.

The OriGene Clone Advantage:

Purified plasmid DNA instead of glycerol stock

A key feature that differentiates OriGene from other vendors is the format in which the final product is delivered. OriGene's clones are all provided as 10 ug of purified plasmid DNA derived from a single colony, which was end-sequence verified before shipment. This format provides unmatched convenience and industry-leading accuracy and saves at least 3 days of routine lab work and related costs.

Most clones from other providers are provided as bacterial stocks from which DNA needs to be isolated and purified for identity verification and for downstream applications, such as transfection, subcloning, protein expression in cell-free systems, etc. You, the customer, are forced to verify that you received the correct clone. OriGene realizes that quality control is not your responsibility. We put in extra effort to deliver pure plasmids so that you can start your project on the first day the clone arrives.



With 10 ug of DNA, you can start your experiments immediately, without spending precious time amplifying the DNA. Ten micrograms of DNA is sufficient for multiple reactions, regardless of what type of experiment you're performing.

Application	Typical Amount of DNA	Number of Reactions
PCR	10 ng	1000
Transfection	200 ng	50
Sequencing Reactions	125 ng	80
Enzyme Digests / Subcloning	500 ng	20
In vitro Protein Synthesis	1 ug	10
Probe Synthesis	25 ng	400

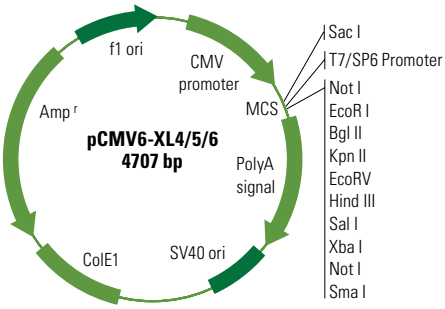
TrueClone™

Authentic, full-length cDNA clones for expression and functional studies of a native protein

All TrueClones are isolated from cDNA libraries and represent native transcripts. There is no PCR amplification during the generation of these clones; therefore, the clones are devoid of any PCR-based artifacts. The clones tend to carry 5′- and 3′-UTRs in addition to complete open reading frames (ORF). TrueClones are the clones of choice when a protein's function needs to be studied in its native condition, with its native cis-regulatory elements and without modification to the ORF.

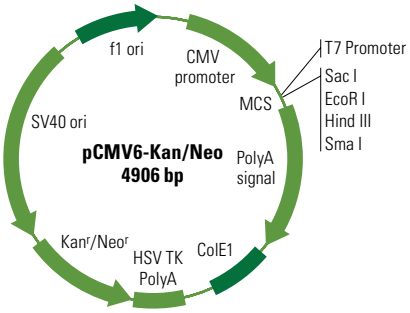
OriGene's TrueClones cover both human and mouse species. The vectors for the two species differ slightly.

Human TrueClone Vector:



All human TrueClones are unidirectionally cloned in the EcoR I and Sal I restriction sites. The Sal I site is destroyed during cloning, and cannot be reused. The insert can be liberated by a simple digestion with Not I.

Mouse TrueClone Vector:



All mouse TrueClones are cloned unidirectionally between two sites in the MCS. Please contact OriGene's Technical Support Professionals for details.

TrueClones are excellent for:

- Overexpression of the native protein in mammalian cells
- Functional studies of native protein
- Templates for quantitative PCR
- Probes for hybridization-based detection, such as Northern blots or FISH assays
- Protein expression in cell-free systems (eg. TNT)

TrueClone Advantages:

- TrueClones are a cost-effective and time-saving alternative to de novo cloning.
- The TrueClone collection contains the largest number of full-length transcripts
- TrueClones are expression-ready and transfection-ready.
- TrueClones are authentic cDNA clones without PCR errors
- TrueClones have consistent vectors
- TrueClones are accurately prepared and delivered

1. TrueClones are a cost-effective and time-saving alternative to de novo cloning.

Although some believe that gene cloning has become molecular biology 101, it still requires substantial resources and the time to clone even the most common gene into an expression vector. It should be pointed out that all of the easy-to-clone transcripts have been isolated, and the remaining are rare, large, GC-rich, or toxic to standard cloning bacteria. Current cloning estimates of time and effort required are usually grossly underestimated.

Why spend your time, resources and energy on gene cloning when a pre-cloned gene is available and ready to be shipped to your laboratory?

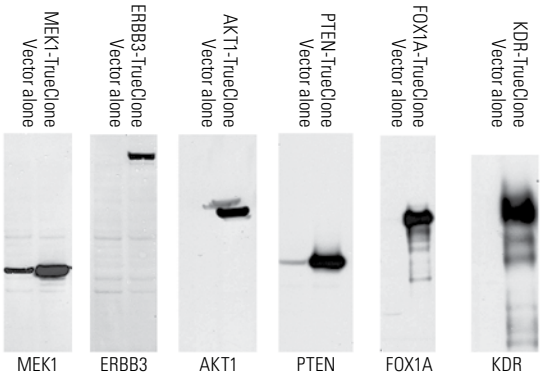
2. The TrueClone collection contains the largest number of full-length transcripts compared to other clone providers

Transcripts from over 98% of the human genome are offered. When you need a clone, you have the best chance of obtaining it from OriGene. While others provide the common clones, the TrueClone collection contains thousands of difficult-to-clone transcripts that are only available from OriGene. OriGene's dedication and technological expertise separates TrueClones from the rest of pack. Thanks to a group of dedicated and skilled molecular biologists, a steady stream of difficult-to-clone cDNAs are added to the TrueClone collection on a daily basis.

3. TrueClones are expression-ready and transfection-ready.

All TrueClones have a strong CMV promoter upstream of the cDNA for mammalian expression. The plasmid can be transfected immediately for protein overexpression. The image below is an example of the validated overexpression of several popular TrueClones in HEK293 cells.

TrueClone Overexpression Validated by Western Blots



4. Authenticity

Each TrueClone is derived from cDNA library instead of via PCR synthesis. That means that every clone represents a real transcript without PCR-introduced artifacts that might affect your downstream applications.

5. Consistent vectors

While other vendors provide clones in a conglomeration of vectors with various antibiotic selection markers and variable expression-readiness, all TrueClones are in a single series of CMV vectors. Every TrueClone is ready for immediate protein overexpression in mammalian cells or in cell-free systems utilizing T7 or SP6 promoters.

The expression-readiness and uniform vector system of TrueClones render them ideal for high-throughput screening for functional genes. Researchers from Novartis and Harvard have pioneered novel applications using arrays of over 10,000 TrueClones, and have published several interesting discoveries*.

6. Accuracy

Verified, pure plasmid DNA stocks will be delivered instead of glycerol stocks. There is no need for colony screening, plasmid preparation or restriction digestion/ sequencing, which are all necessary when ordering a clone from most suppliers.

Recent TrueClone Citations:

Activation of the JNK pathway promotes phosphorylation and degradation of BimEL—a novel mechanism of chemoresistance in T-cell acute lymphoblastic leukemia. Kam Tong Leung et. al. Carcinogenesis, Mar 2008; 29: 544 - 551.

Autoimmune Polyendocrine Syndrome Type 1 and NALP5, a Parathyroid Autoantigen. Mohammad Alimohammadi et. al. N. Engl. J. Med. 2008; 358(10): p. 1018-1028.

Cdc42- and Rac1-mediated endothelial lumen formation requires Pak2, Pak4 and Par3, and PKC-dependent signaling. Wonshill Koh et. al. J. Cell Sci., Mar 2008; 10.1242/jcs.020693.

Antisera induced by infusions of autologous Ad-CD154-leukemia B cells identify ROR1 as an oncofetal antigen and receptor for Wnt5a. Tetsuya Fukuda et. al. PNAS, Feb 2008; 105: 3047 - 3052.

* Citations using a large set of TrueClone for novel discovery:

A functional genomics approach to the mode of action of apratoxin A. Nat Chem Biol. 2006 Mar;2(3):158-67.

A genomic screen for activators of the antioxidant response element. Proc Natl Acad Sci U S A. 2007 Mar 20;104(12):5205-10.

Transducer of regulated CREB-binding proteins (TORCs) induce PGC-1 alpha transcription and mitochondrial biogenesis in muscle cells. Proc Natl Acad Sci U S A. 2006 Sep 26;103(39):14379-84.

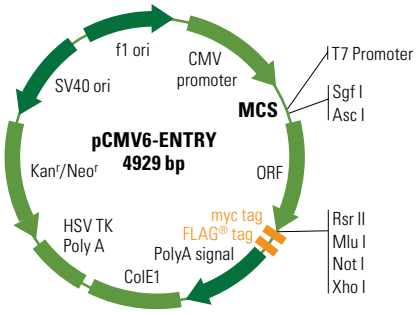
Identification of the Wnt signaling activator leucine-rich repeat in Flightless interaction protein 2 by a genome-wide functional analysis. Proc Natl Acad Sci U S A. 2005 Feb 8;102(6):1927-32.

TrueORF™:

Over 25,000 tagged ORF clones

The TrueORF product line is the newest generation of cDNA clone products from OriGene. Unlike TrueClones, a TrueORF clone enables expression of the encoded transcript as a tagged protein, which facilitates multiple downstream applications that utilize an anti-tag antibody, such as protein detection, protein purification, subcellular localization, etc.

TrueORF Vector:



All TrueORF inserts are housed in the pCMV6-Entry vector and can be easily shuttled by a simple 'cut-and-paste' mechanism into any of the PrecisionShuttle Destination Vectors (see page 8). A TrueORF clone expresses the encoded sequence as a C-terminally Myc and FLAG®-tagged protein.

FLAG® is a registered trademark of Sigma-Aldrich.

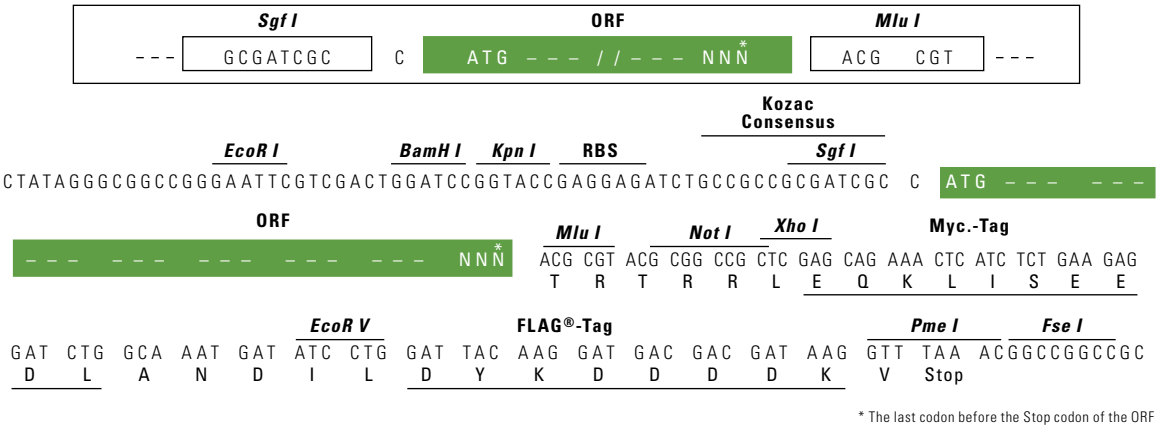
TrueORFs are excellent for:

- Mammalian overexpression of tagged proteins (20 combination of tags available)
- Detection of the exogenously introduced protein
- Purification of the overexpressed protein
- Protein interaction and localization studies
- Cellular imaging of the exogenously introduced protein
- Tagged protein expression in a cell-free system (eg. TNT)

TrueORF Advantages:

- TrueORFs provide the complete solution for tagged protein expression
- TrueORF tags can be easily switched within the PrecisionShuttle system
- TrueORFs come with verified sequences and an accuracy guarantee
- TrueORFs have been rigorously tested for expression of the target proteins and their tags

Cloning sites used for ORF Shuttling:



* The last codon before the Stop codon of the ORF

The diagram above is applicable to that majority of human ORFs that do not have internal Sgf I and Mlu I sites. Other rare restriction sites in the MCS are utilized for the ORFs with internal Sgf I or Mlu I sites.

1. Convenience: TrueORFs provide an instant solution for tagged protein expression

TrueORFs are constructed with the scientists' application of clones in mind. For those who need to use antibodies to detect and to purify the overexpressed protein, TrueORFs are a complete solution:

- Universal C-terminal tags of Myc and FLAG® for downstream experiments with anti-tag antibodies
- Strong CMV promoter and a Kozak sequence upstream of the cDNA for optimal mammalian expression.
- Provided as 10 ug purified plasmid DNA
- Additional quantities can be purchased for a nominal fee.

TrueORF clones can be transfected immediately for protein overexpression, alleviating the labor of cloning, tagging, bacterial manipulation and plasmid preparation.

2. Flexibility: TrueORF tags can be easily switched through the Precision-Shuttle system

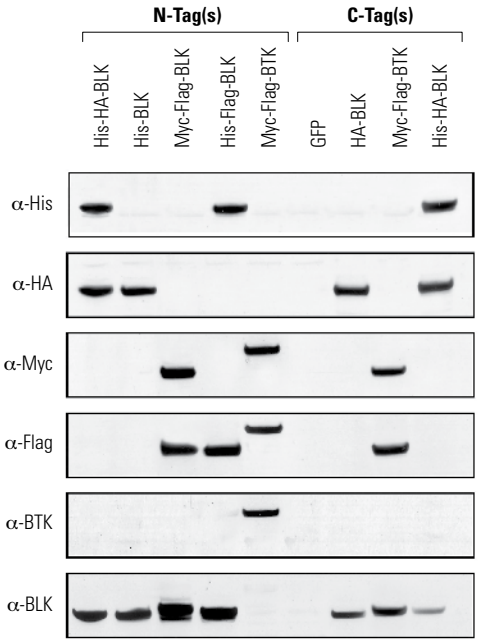
TrueORFs are cloned into the pCMV6-Entry vector. The PrecisionShuttle system includes a large selection of destination vectors, all of which have a multiple cloning site (MCS) compatible with pCMV6-Entry. Through a simple restriction digestion and ligation reaction, the insert of any TrueORF can be transferred to a destination vector, and expressed as a protein with a different epitope tag.

Coupled with the PrecisionShuttle system, TrueORFs offer great flexibility for tagged protein expression. More details of PrecisionShuttle system can be found on page 8.

3. Accuracy: TrueORFs have verified and guaranteed insert sequences

All TrueORFs are derived from full-length cDNA clones of verified sequence using a polymerase with the highest possible fidelity. To further ensure the sequence accuracy, purified plasmids were used as template and a minimal number of PCR cycles was employed to reduce potential PCR mutations.

4. Proven: TrueORFs have been rigorously tested for expression of the target proteins and their tags.



Western blot analysis of HEK293 cell lysates over-expressing BLK or BTK tagged with indicated epitopes.

PrecisionShuttle™ System:

Tagged protein expression made simple

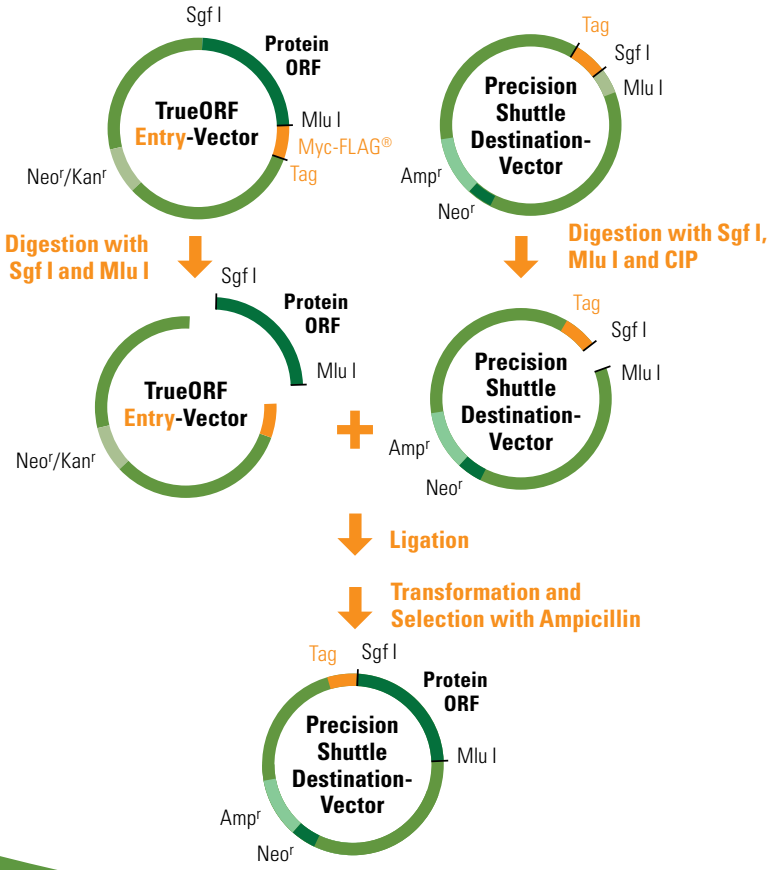
To accommodate diverse tagging needs (different tag types and/or different locations), OriGene devised the novel PrecisionShuttle™ system to allow easy subcloning of an ORF from one tagged vector to another. The TrueORF Entry Vector contains C-terminal tags of Myc and FLAG®, and is the **entry vector** for the PrecisionShuttle system. A large panel of **destination vectors** is available so that you can express the ORF with different tags, with tags at different ends of the protein, or as the native, untagged protein.

The key in the PrecisionShuttle system is the utilization of two rare-cutting restriction endonucleases, Sgf I and Mlu I. The entry vector and all destination vectors have identical multiple cloning sites (MCS) so that they can exchange the inserts through simple restriction digestion and ligation. In the TrueORF Entry Vector, the ORF is flanked by Sgf I (5'-end) and Mlu I (3'-end) sites. Digestion with these two enzymes releases the untagged ORF, which can then be ligated into a destination vector digested with the same enzymes. As all destination vectors carry the ampicillin resistance marker instead of the kanamycin resistance marker, the successfully shuttled product can be easily selected with ampicillin. In the rare cases in which Sgf I or Mlu I cuts in the ORFs, alternative strategies are provided on OriGene's website.

PrecisionShuttle advantages over a recombination shuttling system (e.g. Gateway system):

- The 25,000 clones in the entry vector can be used for tagged expression in mammalian cells and in cell-free systems. For many applications, there is no need to shuttle the ORF to a destination vector.
- The shuttling is accomplished with a simple restriction enzyme digestion/ligation without the need for an expensive, recombination specific enzyme.
- There are no intellectual property restrictions.
- Large ORFs up to 18Kb can be readily transferred using the PrecisionShuttle system while ORFs larger than 4Kb are unstable in recombination-based systems.
- The PrecisionShuttle vectors precisely add the intended tag to its desired location. There is no appendage of multiple amino acids on both ends of the protein.

Schematic of the PrecisionShuttle system



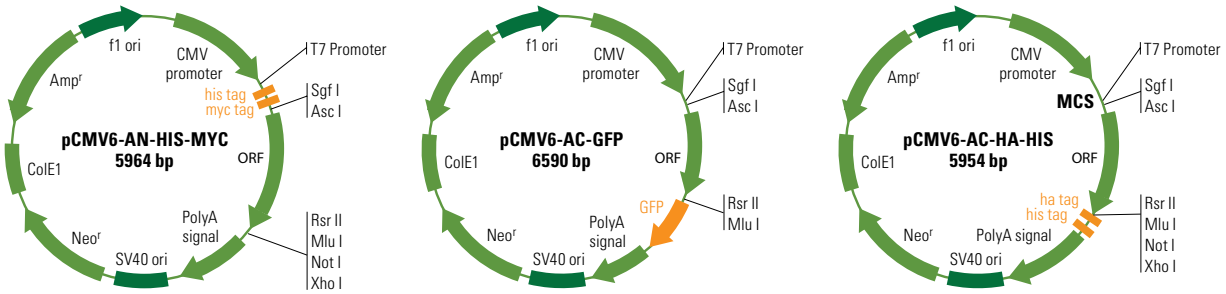
PrecisionShuttle™ Vectors:

Entry and Destination

PrecisionShuttle Entry Vector	SKU	*E. coli* Selection	Cell Selection	Expression	N-Terminal Tag	C-Terminal Tag
pCMV6-Entry (C-terminal Myc and FLAG® Tagged)	PS100001	Kanamycin	Neomycin	Mammalian	–	Myc-FLAG®

PrecisionShuttle Destination Vectors	SKU	*E. coli* Selection	Cell Selection	Expression	N-Terminal Tag	C-Terminal Tag
pCMV6-AC-His	PS100002	Ampicillin	Neomycin	Mammalian	–	His
pCMV6-AC-Myc	PS100003	Ampicillin	Neomycin	Mammalian	–	Myc
pCMV6-AC-HA	PS100004	Ampicillin	Neomycin	Mammalian	–	HA
pCMV6-AC-FLAG®	PS100005	Ampicillin	Neomycin	Mammalian	–	FLAG®
pCMV6-AC-Myc-His	PS100006	Ampicillin	Neomycin	Mammalian	–	Myc-His
pCMV6-AC-Myc-FLAG®	PS100007	Ampicillin	Neomycin	Mammalian	–	Myc-FLAG®
pCMV6-AC-HA-His	PS100008	Ampicillin	Neomycin	Mammalian	–	Ha-His
pCMV6-AC-FLAG®-His	PS100009	Ampicillin	Neomycin	Mammalian	–	FLAG®-His
pCMV6-AC-GFP	PS100010	Ampicillin	Neomycin	Mammalian	–	T-GFP
pCMV6-AN-His	PS100011	Ampicillin	Neomycin	Mammalian	His	–
pCMV6-AN-Myc	PS100012	Ampicillin	Neomycin	Mammalian	Myc	–
pCMV6-AN-HA	PS100013	Ampicillin	Neomycin	Mammalian	HA	–
pCMV6-AN-FLAG®	PS100014	Ampicillin	Neomycin	Mammalian	FLAG®	–
pCMV6-AN-His-Myc	PS100015	Ampicillin	Neomycin	Mammalian	Myc-His	–
pCMV6-AN-Myc-FLAG®	PS100016	Ampicillin	Neomycin	Mammalian	Myc-FLAG®	–
pCMV6-AN-His-HA	PS100017	Ampicillin	Neomycin	Mammalian	Ha-His	–
pCMV6-AN-His-FLAG®	PS100018	Ampicillin	Neomycin	Mammalian	FLAG®-His	–
pCMV6-AN-GFP	PS100019	Ampicillin	Neomycin	Mammalian	T-GFP	–
pCMV6-A	PS100020	Ampicillin	Neomycin	Mammalian	–	–

Plasmid maps of a few representative destination plasmids



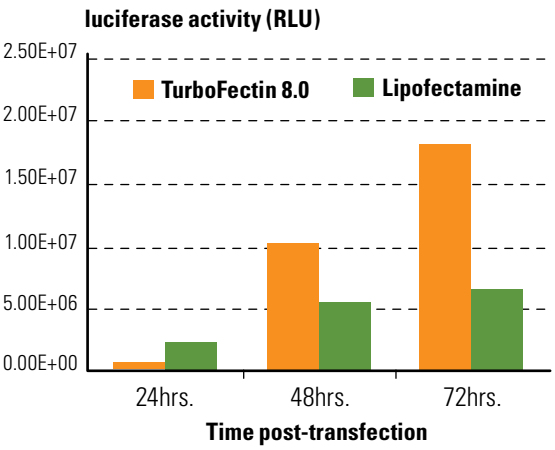
TurboFectin 8.0™:

Drive your transfection to full throttle

TurboFectin 8.0 is a new generation of transfection reagent optimized for nucleic acid delivery into eukaryotic cells. Its proprietary formulation of lipid/histone blend is supplied in 80% alcohol. TurboFectin 8.0 is the recommended transfection reagent for delivery of TrueClones (for overexpression) and HuSH-29 constructs (shRNA for expression knockdown) in many different cell types. TurboFectin 8.0 has also been optimized for and is the preferred reverse transfection reagent for GFC-Transfection Arrays.

- **Optimal performace:** Demonstrated high efficiency and low toxicity
- **Higher protein expression levels:** 180% more protein expression after 72 hrs than Lipofectamine
- **Greater RNAi effect:** Double knockdown efficiency in shRNA-mediated RNAi compared to Lipofectamine
- **Simplified use:** Works well in media containing antibiotic and antimycotic agents. Suitable for serum-containing media; no requirement for media changes.
- **Wide spectrum:** Over 100 cell lines and primary cell types have been successfully transfected with TurboFectin 8.0

Optimal expression over time

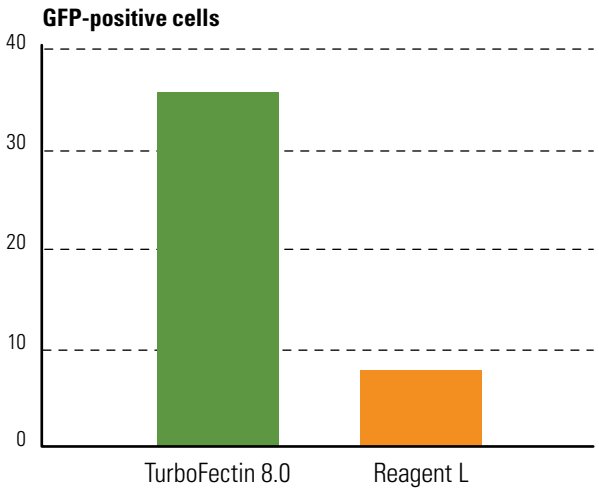


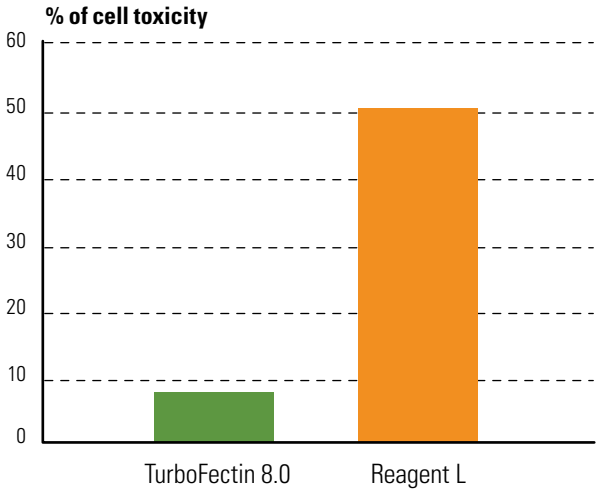
HEK293T cells were plated at 1x10^4 cells/well in a 96-well plate. Two ug of pCMV-Luciferase was transfected using optimal ratios of TurboFectin 8.0 or Lipofectamine. Luciferase activity was measured at 24, 48, and 72 hours post-transfection using BriteLite luciferase substrate (Perkin Elmer).

Cell lines that have been successfully transfected with TurboFectin 8.0

- A1847
- A549
- AR42ce4
- ARP1
- BHK-21
- BNL.CL2
- BRL-3A
- C2C12
- C6
- chicken embryo fibroblast
- CHO-K1
- Clone9
- corneal endothelium
- COS-1
- COS-7
- CV-1
- CVEC
- Daoy
- DBTRG-05MG
- DI-TNC1
- DU145
- ECV304
- H1299
- HCT-116
- HEC-1A
- HEC-1B
- HEK293
- HeLa
- Hepa1-6
- Hepa1cLc7
- HepG2
- HLF-a
- Hs578T
- Huh-7
- human bladder carcinoma
- HUVEC
- Jurkat
- K562
- KB
- KLN205
- LL/2(LLC1)
- LLC-PK
- LNCaP-FGC
- M19
- MA-10
- MC3T3-E1
- MCF-7
- MDA-MB-231
- MDA-MB-435
- MDCK
- MEL
- melanocyte
- Mv1Lu
- myometrial
- Neuro-2a
- neuroepithelial
- NIH3T3
- OV-1063
- ovarian cancer
- OVCAR10
- OVCAR3
- OVCAR4
- OVCAR7
- OVCAR8
- PC-12
- PC3
- PE01
- PE04
- primary human astrocytes
- primary human chondrocytes
- primary human keratinocytes
- primary human melanoma cells
- primary human skin fibroblasts
- primary mouse embryo niccells
- primary mouse fibroblasts
- primary mouse hepatocytes
- primary mouse myotubes
- primary mouse thymocytes
- primary rat hepatocytes
- primary rat osteoblasts
- rat smooth muscle cells
- RAW264.7
- RBL
- RBL-2H3
- SJPL
- SK-BR-3
- SK-N-MC
- SKOV3
- SVGp12
- SW900
- THP-1
- U20S
- U266
- UPN251
- Vero
- WB rat liver epithelial cells
- WiDr
- WRL-68

TurboFectin 8.0 extends the duration of your experiments so that you can maximize data aquisition from every transfection. It is even gentle enough to be used with primary cells.





High potency and low toxicity are key features to TurboFectin 8.0 transfections. Primary human chondrocytes were plated at 100,000 cells/well in a 24-well plate and transfected with 500ng of pGFP using the optimal ration of TurboFectin 8.0 or a leading transfection reagent. Potency (top panel) and toxicity (bottom panel) were determined by counting viable GFP-positive cells/field at 48 hours post-transfection. Data provided courtesy of David Hum via CedarLane Laboratories.

Catalog No.	Description
TF81001	TurboFectin 8.0 (1 vial @ 1ml each)
TF81005	TurboFectin 8.0 (5 vials @ 1ml each)

25,000 Tagged ORF Clones

including the ones you want

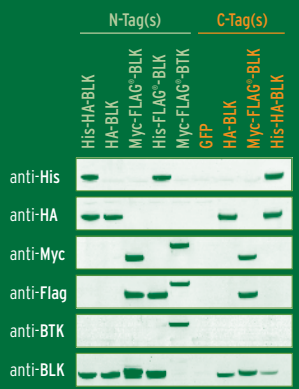
TrueORF™

for tagged protein expression

TrueORF enables the expression of the encoded transcript as a C-terminally tagged protein with Myc and FLAG® epitopes, facilitating multiple applications that utilize an anti-tag antibody, such as protein detection, protein purification, subcellular localization, etc.

- Genome-wide coverage
- Sequence verified and guaranteed
- The C-terminal dual tag of Myc and FLAG®
- Transfection-ready: Provided as 10 ug of purified plasmid
- Easy shuttling into 20 tagged vectors using PrecisionShuttle™ system

FLAG® is a registered trade mark of Sigma-Aldrich



The Western blot analysis of HEK293 cell lysate over-expressing BLK or BTK tagged with indicated epitopes.

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