Certificate of Analysis



pTetOne Vector Set

Catalog No.

634303 (Not sold separately) Sold as a part of 634301 & 634302 **Amount** Each

Lot Number

Specified on product label.

Description

The pTet-One 3G Vector Set is provided as a component of the Tet-OneTM Inducible Expression System and is used to create tightly regulated and highly responsive tetracycline (Tet)-inducible mammalian expression systems that are turned on by the addition of doxycycline to the culture medium. The pTetOne Vector constitutively expresses the Tet-On® 3G transactivator from the constitutive human PGK promoter in the forward orientation and your gene of interest from the P_{TRE3GS} promoter in the reverse orientation. Also included are puromycin and hygromycin linear selection markers for creation of stable clones by cotransfection followed by antibiotic selection.

Package Contents

- 20 µl pTetOne Vector (500 ng/µl)
- 20 μl pTetOne-Luc Control Vector (500 ng/μl)
- 40 μl Linear Hygromycin Marker (50 ng/μl)
- 40 μl Linear Puromycin Marker (50 ng/μl)

Storage Conditions

- Store plasmids at -20°C.
- Spin briefly to recover contents.
- Avoid repeated freeze/thaw cycles.

Expiration Date

Specified on product label.

Storage Buffer

• 10 mM Tris-HCl (pH 8.0), 1 mM EDTA (pH 8.0)

Shipping Conditions

• Dry ice

Product Documents

Documents for our products are available for download at <u>takarabio.com/manuals</u> The following documents apply to this product:

- Tet-One Inducible Expression System User Manual
- pTetOne Vector Information
- pTetOne Vector Sequence in GenBank Format
- pTetOne-Luc Control Vector Sequence in GenBank Format

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Propagation in *E. coli*

- Recommended host strain: StellarTM Competent Cells (Cat. No. 636763)
- Selectable marker: Plasmids confer resistance to ampicillin (100 μg/ml) in *E. coli* hosts.
- E. coli replication origin: pUC

Quality Control Data

Plasmid Identity & Purity

Digestion with the indicated restriction enzymes produced fragments of the indicated sizes on a 0.8% agarose/EtBr gel:

Vector	Enzymes	Fragments
pTetOne	HindIII	4.2 kb
	HindIII & NruI	1.2 kb & 3.0 kb
pTetOne-Luc Control	HindIII	5.8 kb
_	EcoRI & PstI	1.6 kb & 4.2 kb
Linear Hygromycin Marker	HindIII & XbaI	0.5 kb, 0.6 kb, & 1.1 kb
Linear Puromycin Marker	HindIII & XbaI	0.45 kb, 0.6 kb, & 0.75 kb

Vector identity was confirmed by sequencing.

A₂₆₀/A₂₈₀: 1.8–2.0

Functional Testing of Linear Markers

• HEK 293 cells were transfected with 200 ng of either the Linear Hygromycin Marker or the Linear Puromycin Marker. After 5 hr at 37°C, the transfection solution was removed, and the cells were given fresh medium. 48 hr later, the cells were plated in two 10 cm plates. 48 hr after plating, medium containing either hygromycin or puromycin (depending on the linear marker used to transfect the cells) was added to the plates. After 2–3 weeks, >20 clones were identified.

It is certified that this product meets the above specifications, as reviewed and approved by the Quality Department.

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NOTICE TO PURCHASER:

Our products are to be used for Research Use Only. They may not be used for any other purpose, including, but not limited to, use in humans, therapeutic or diagnostic use, or commercial use of any kind. Our products may not be transferred to third parties, resold, modified for resale, or used to manufacture commercial products or to provide a service to third parties without our prior written approval.

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