# 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-One<sup>TM</sup> 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  $P_{\text{TRE3GS}}$  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: Stellar<sup>TM</sup> 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.

A<sub>260</sub>/A<sub>280</sub>: 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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### CATALOG NO.

634303

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#### **STATEMENT 42**

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