pDsRed-Express-1 Vector

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Catalog No. Amount Lot Number
632413 20 μg Specified on product label.

Product Information
pDsRed-Express-1 is a promoterless mammalian expression vector that can be used to monitor transcription from different promoters and promoter/enhancer combinations inserted into the multiple cloning site (MCS). It encodes DsRed-Express, a variant of Discosoma sp. red fluorescent protein (DsRed; 1). DsRed-Express contains nine amino acid substitutions which improve the solubility of the protein, reduce the time from transfection to detection of red fluorescence, and decrease the level of residual green emission (2). When DsRed-Express is expressed in mammalian cell cultures, red-emitting cells can be detected by either fluorescence microscopy or flow cytometry 8–12 hours after transfection. Although DsRed-Express most likely forms the same tetrameric structure as wild-type DsRed, DsRed-Express displays a reduced tendency to aggregate (2). The DsRed-Express coding sequence is human codon-optimized for high expression in mammalian cells (3).

The sequence upstream of the DsRed-Express gene has been converted to a Kozak consensus sequence (4) to enhance translation efficiency in eukaryotic cells. SV40 polyadenylation signals downstream of the DsRed-Express gene direct proper processing of the 3' end of the DsRed-Express mRNA. The vector backbone contains an SV40 origin for replication in mammalian cells expressing the SV40 T antigen, a pUC origin of replication for propagation in E. coli, and an f1 origin for single-stranded DNA production. A neomycin-resistance cassette (Neo') allows stably transfected eukaryotic cells to be selected using G418. This cassette consists of the SV40 early promoter, the neomycin/kanamycin resistance gene of Tn5, and polyadenylation signals from the Herpes simplex virus thymidine kinase (HSV TK) gene. A bacterial promoter upstream of the cassette expresses kanamycin resistance in E. coli.

Promoters should be cloned into the pDsRed-Express-1 MCS upstream from the DsRed-Express coding sequence. Without addition of a functional promoter, this vector will not express DsRed-Express. The recombinant DsRed-Express vector can be transfected into mammalian cells using any standard transfection method. If required, stable transfectants can be selected using G418 (5).

Package Contents
- 20 μg pDsRed-Express-1 Vector

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

**Shelf Life**
- 1 year from date of receipt under proper storage conditions.

**Storage Buffer**
- 10 mM Tris-HCl (pH 8.0), 1 mM EDTA (pH 8.0)

**Concentration**
- 500 ng/μl

**Shipping Conditions**
- Dry ice (–70°C)

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**Location of Features**
- MCS (multiple cloning site): 12–83
- Kozak consensus sequence: 90–100

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**Figure 1.** pDsRed-Express-1 vector map. The NotI site is part of the DsRed-Express stop codon.

**Figure 2.** pDsRed-Express-1 multiple cloning site.
Certificate of Analysis

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pDsRed-Express-1 Vector

- DsRed-Express: 97–774
- SV40 early polyA signals: 926–931 & 955–960
- F1 origin of replication: 1023–1478
- P (promoter for Kan\(^\text{R}\)): 1540–1568
- SV40 origin of replication: 1819–1954
- P\(_{SV40}\), SV40 early promoter and enhancer: 1650–1881
- Kan\(^{\text{R}}\)/Neo\(^{\text{R}}\) (kanamycin/neomycin resistance gene): 2003–2797
- HSV TK polyA (herpes simplex virus thymidine kinase polyadenylation signals): 3033–3038 & 3046–3051
- pUC origin of replication: 3382–4025

**Additional Information**

**Recommended Sequencing Primer Location:**

- 297–277

**Propagation in E. coli**

- Suitable host strains: DH5\(\alpha\), HB101 and other general purpose strains. Single-stranded DNA production requires a host containing an F plasmid such as JM109 or XL1-Blue.
- Selectable marker: plasmid confers resistance to kanamycin (50 \(\mu\)g/ml) to E. coli hosts.
- E. coli replication origin: pUC
- Copy number: High

**Excitation and Emission Maxima of DsRed-Express**

- Excitation: 557 nm
- Emission: 579 nm

**References**


**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:
Certificate of Analysis

pDsRed-Express-1 Vector

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Fragment(s) (kb)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NotI</td>
<td>4.1</td>
</tr>
<tr>
<td>Stul</td>
<td>2.7 &amp; 1.4</td>
</tr>
</tbody>
</table>

- Vector identity was confirmed by sequencing.
- $A_{260}/A_{280}$: 1.8–2.0
Notice to Purchaser

pDsRed-Express-1 Vector

CATALOG NO.
632413

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LICENSING STATEMENTS:
The RCFP’s (including DsRedExpress and DsRedExpress2) are covered by one or more of the following U.S. Patent Nos. 7,166,444; 7,157,565; 7,217,789; 7,338,784; 7,338,783; 7,537,915 6,969,597, 7,150,979 and 7,442,522.

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This document has been reviewed and approved by the Clontech Quality Assurance Department.