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Catalog No. Amount Lot Number
632409 20 μg Specified on product label.

Product Information

pDsRed2-ER is a mammalian expression vector designed to label the endoplasmic reticulum in living cells (1, 2). The vector encodes a fusion consisting of Discosoma sp. red fluorescent protein (DsRed2; 3, 4); the endoplasmic reticulum (ER) targeting sequence of calreticulin (5), fused to the 5' end of DsRed2; and the ER retention sequence, KDEL (6, 7), fused to the 3' end of DsRed2. DsRed2 is a human codon-optimized (8) variant of wild-type DsRed that has been engineered for faster maturation and lower non-specific aggregation.

To drive expression of DsRed2, this vector contains the immediate early promoter of cytomegalovirus (P_{CMV \text{IE}}). SV40 polyadenylation signals downstream of the DsRed2 gene direct proper processing of the 3'-end of the DsRed2 mRNA transcript. The vector also contains an SV40 origin for replication in any mammalian cell line that expresses 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—consisting of the SV40 early promoter (P_{SV40e}), the neomycin/kanamycin resistance gene of Tn5 (Neo^r/Kan^r), and polyadenylation signals from the herpes simplex virus thymidine kinase (HSV TK poly A) gene—allows stably transfected eukaryotic cells to be selected using G418 (9). A bacterial promoter (P) upstream of this cassette drives expression of the gene encoding kanamycin resistance in E. coli.

pDsRed2-ER can be introduced into mammalian cells using any standard transfection method. If required, stable transformants can be selected using G418 (9).

Package Contents

- 20 μg pDsRed2-ER 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.
Certificate of Analysis

pDsRed2-ER Vector

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)

Figure 1. pDsRed2-ER vector map.

Location of Features
- $P_{CMV\ IE}$ (human cytomegalovirus (immediate early promoter)): 1–589
- Calreticulin signal sequence: 597–647
- DsRed2: 663–1337
  - KDEL (in frame with DsRed2): 1350–1361
  - Stop codon: 1362–1364
- SV40 early polyA signals: 1576–1581 & 1605–1610
- f1 origin of replication: 1673–2128
- $P$ (promoter for Kan$^r$): 2190–2218
- SV40 origin of replication: 2469–2604
- $P_{SV40\ e}$ (SV40 early promoter and enhancer): 2302–2531
- Kan$^r$/Neo$^r$ (Kanamycin/neomycin resistance gene): 2653–3447
- HSV TK (Herpes simplex virus thymidine kinase polyadenylation signals): 3683–3688 & 3696–3701
- pUC origin of replication: 4032–4675
Certificate of Analysis
pDsRed2-ER Vector

Cat. No. 632409

Additional Information

Propagation in E. coli

- Suitable host strains: DH5α, 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 μg/ml) to E. coli hosts.
- E. coli replication origin: pUC
- Copy number: high

Excitation and Emission Maxima of DsRed2

- Excitation: 558 nm
- Emission: 583 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:

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Fragment(s) (kb)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NheI</td>
<td>4.7</td>
</tr>
<tr>
<td>EcoO109I</td>
<td>2.7 &amp; 2.0</td>
</tr>
</tbody>
</table>

- Vector identity was confirmed by sequencing.
- A260/A280: 1.8–2.0
Notice to Purchaser

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CATALOG NO.
632409

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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.