

For plasmid sequence, please visit [www.chromotek.com](http://www.chromotek.com).

### Location of features

- T7 promoter: 1-19
- Lac operon: 19-46
- RBS: 64-80
- MCS: 90-140
- Spot-Tag®: 141-176
- Stop codon (TAA): 177-179
- rrnB terminator: 361-518
- Kanamycin resistance gene: 798-1613
- pRB322 replication origin: 1781-2400

| Product | Code | Size    |
|---------|------|---------|
| pSpot2  | ev-2 | 1.25 µg |

|             |   |
|-------------|---|
| Vector type | bacterial expression vector   |
| Tag         | Spot-Tag® (PDRVRAVSHWSS)<br>C-terminal  |
| MCS         | BamHI, EcoRI, SacI, Sall, HindIII, NotI, XhoI, NheI                                     |
| Promoter    | T7  |
| Induction   | IPTG, lactose   |
| Host cells  | <i>Escherichia coli</i> DE3 strains   |
| Selection   | kanamycin   |
| Replication | pBR322  |
| Use         | Expression of a protein of interest fused to Spot-Tag® (C-terminal) in <i>E. coli</i> . |

### Vector description

The plasmid pSpot2 is an expression vector for Spot-Tag® fusion proteins in *E. coli*. After cloning the protein of interest (POI) into the multiple cloning site (MCS) provided by pSpot2, the Spot-Tag® (sequence: PDRVRAVSHWSS) will be fused to the C-terminus of the POI.

### Expression in *E. coli*

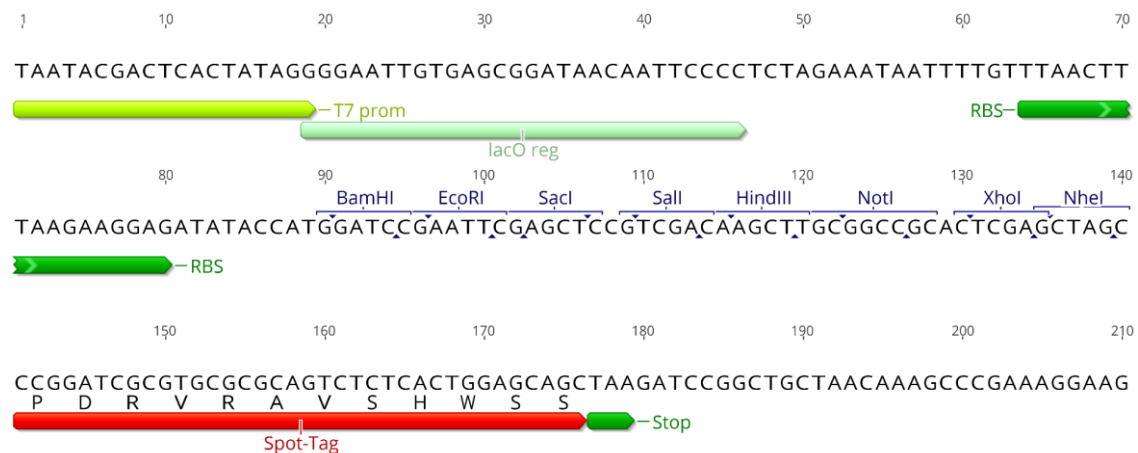
Protein expression can be induced using lactose or IPTG, but requires the use of DE3 prophage-positive *E. coli* expression strains. The resulting Spot-Tag® fusion protein can be purified using the ChromoTek Spot-Trap®.

### Propagation in *E. coli*

Suitable host strains for propagation in *E. coli* include DH5alpha, HB101, XL1-Blue, and other general purpose strains. The vector confers resistance to kanamycin (50 µg/ml) to *E. coli* hosts.

Note: The plasmid DNA was isolated from dam<sup>+</sup>-methylated *E. coli*. Therefore some restriction sites are blocked by methylation. If you wish to digest the vector using such sites you will need to transform the vector into a dam<sup>-</sup> host and make fresh DNA

### Multiple cloning site (MCS)



### Notice to Purchaser:

This plasmid was designed and generated by Dr. Philipp Kaiser from the Naturwissenschaftliches und Medizinisches Institut (NMI) at the University of Tübingen, Germany and is distributed by ChromoTek GmbH. Please acknowledge Dr. Philipp Kaiser (NMI, Tübingen, Germany) and ChromoTek GmbH (Marinsried, Germany) when using or redistributing this vector. The development of this plasmid was supported by a grant "Zentrales Innovationsprogramm Mittelstand" (ZIM) from the Federal Ministry for Economic Affairs and Energy of Germany.