



pSmart I

1. Product Description

The SMT3 protein belonging to the SUMO (Small Ubiquitin-like Modifier) family in *Saccharomyces cerevisiae*, positioned on chromosome IV, serves as a ubiquitin-like protein that enhances the soluble expression of other proteins. The SUMO protein, with a small molecular weight comprising over a hundred amino acid residues, can be recognized by ULP (Ubiquitin-Like Protein-specific Protease) enzymes based on its spatial conformation, allowing for specific excision from fusion proteins. The pSmart-I vector is a modification of the pET-28a vector, primarily achieved by the insertion of the nucleic acid sequence encoding the SUMO protein. This modification preserves the overall structure and the multiple cloning sites of the original vector.

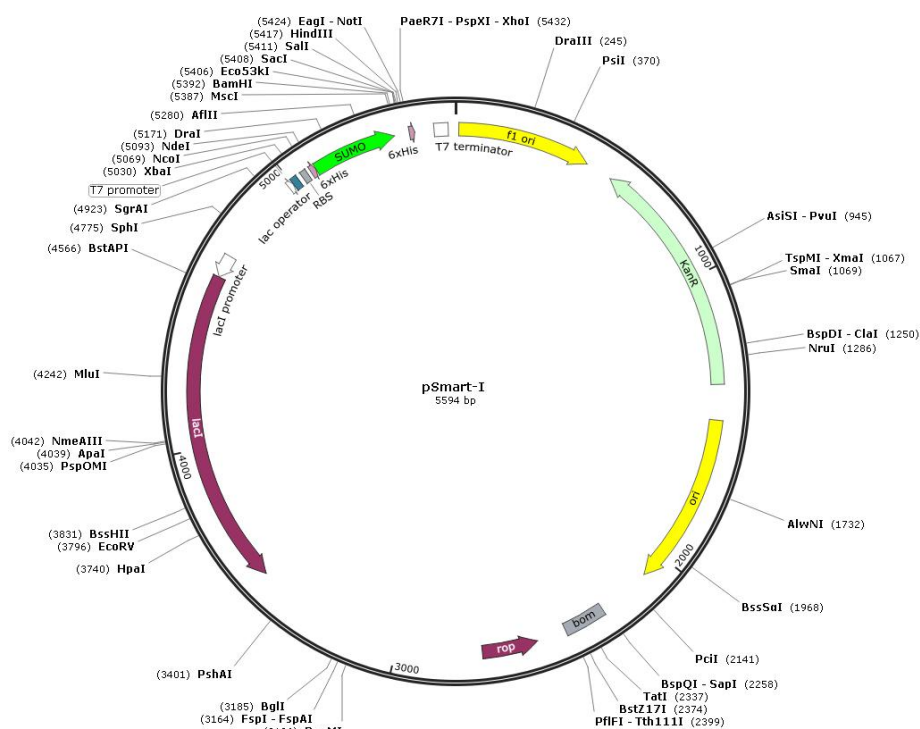


Fig.1. Schematic diagram

2. Multiple Cloning Site (MCS)

N-terminal-his6

ATGGGTCACCATCACCATCACCATATGTCGGACTCAGAAGTCAATCAAGAAGCTAAGCCAGAGGTCAAGCCAGAAGTCAAGCCTGAG
ACTCACATCAATTTAAAGGTGTCCGATGGATCTTCAGAGATCTTCTTCAAGATCAAAAAGACCACTCCTTTAAGAAGGCTGATGGAAG
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TTTGGACATGGAGGATAACGATATTATTGAGGCTCACAGAGAACAGATTGGTGGC↓CAAGGATCCGAATTCGAGCTCCGTCGACAAG
CTTGCGGCCCGCTCGAGCACCACCACCACCACCTGA multi-clonal sites(BamHI, EcoRI, SacI, SalI, HindIII, NotI, XhoI,)

C-terminal-his6

Please do not use EcoRI in polyclonal sites, because there is also an EcoRI sequence in the SUMO sequence, and it is not a single enzyme restriction site. It should also be noted that the reading frame of the access protein and the reading frame of SUMO need to coincide, and the upstream enzyme cleavage site can be selected BamHI or SacI.

3. Protease cleavage site

MGHHHHHHMSDSEVNQEAKPEVKPEVKPETHINLKVSDGSSEIFFKIKKTTPLRRLMEAFKRQKGEMDSLRFlyDGIRIQADQTPEDLD
MEDNDIEAHREQGGQGS





The N-terminal His6-tag can be used for nickel column purification, and the C-terminal His6-tag can optionally be used, if not required, by adding a stop codon in front of it. The fusion protein can be excised by SUMO protease, and the arrow position is the final cut point of the protease. If BamHI is used to access the nucleic acid sequence, the n-end of the target protein will have three more redundant amino acid residues Gln-Gly-Ser after the protease excise the label. If you want to obtain the target protein with no redundant amino acid residues at all, use seamless cloning techniques to attach the protein sequence directly downstream of the SUMO sequence.

Note: The first amino acid downstream of the cleavage site is proline (Pro), which is difficult to cut.

4. Sequencing Primer

The use of any general primers with pET-28a vectors is acceptable, but the sequencing primers T7-Pro or T7-Ter that are far away from the target protein are recommended here to obtain more reliable sequencing information.

5. Complete vector sequence

The nucleic acid sequence of SUMO and its location in the vector are marked in green.

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6. Related Products

Product	Cat. No.	Size
pSmart I	SLP023	100 ng/μl, 10 μl

