

Fluorescent Protein Expression vector set

Monti-Red for Fluoppi (pMonti-Red-MNL/MCL)

Code No.  
AM-VS0802M

**Introduction:**

This product consists of two types (N- or C-terminus fusion) of tetramer red fluorescent protein expression vectors for Fluoppi analysis. pMonti-Red-MNL and pMonti-Red-MCL expresses a protein of interest fused to the N-terminus and the C-terminus of Monti-Red, respectively. To reduce the interference between a protein of interest and fluorescent protein, these vectors are designed to insert more than 22 amino acids length flexible linker peptide between them. For the details of the vectors and property of Monti-Red, please refer to the information below.

Note: Because this product does not contain the Ash-tag expression vectors, basic Fluoppi kit (e.g. Fluoppi : Ash-hAG, code no. AM-8001M) is necessary for Fluoppi analysis.

**Product components:**

Plasmids	Vial color	Form
pMonti-Red-MNL	Red	10 µg: Dry form
pMonti-Red-MCL	Red	10 µg: Dry form

\* Reconstitution in 10-50 µL of sterilized distilled water.

**Storage condition:**

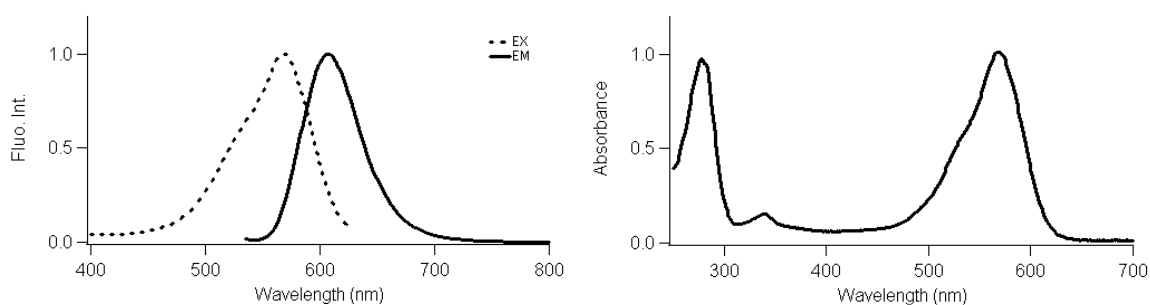
Store at -20°C. Reconstituted solution should be kept at -20°C.

**Intended use:**

For Research Use Only. Not for use in diagnostic procedures.

**Properties of red fluorescent protein “Monti-Red”:**

Monti-Red, a mutant fluorescent protein derived from Keima-Red which was originally cloned from the stony coral (Montipora sp.), forms tetramer and absorbs light maximally at 571 nm and emits red light at 607 nm. Fluorescent signal of Monti-Red can be detected by using filter sets for Texas Red or similar fluorescent dyes.



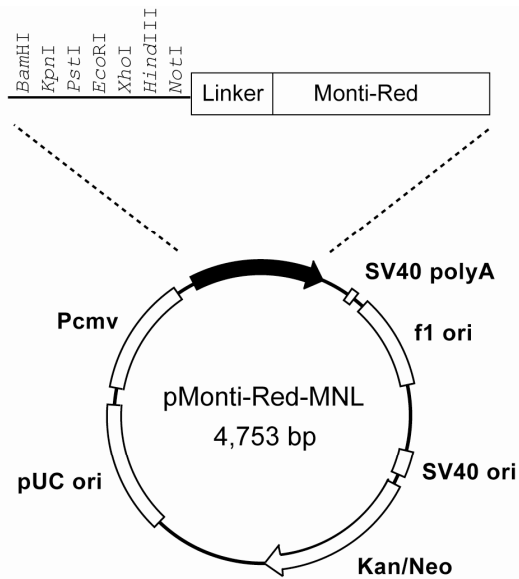
Fluorescent protein	Excitation/Emission maximum (nm)	Extinction coefficient (M <sup>-1</sup> cm <sup>-1</sup> )	Fluorescence quantum yield	pKa
Monti-Red	571/607	83,000 (571 nm)	0.3	5.5

Extinction coefficient was measured by the alkali denaturation method.

Sequence information of the two plasmids can be downloaded from our website.

<http://ruo.mbl.co.jp/product/flprotein/fluoppi.html>

## Plasmid map:



### SEQUENCE LANDMARKS:

Monti-Red gene: bases 145 - 810

peptide linker: bases 73-144

CMV promoter: bases 4163 - 4735

SV40 polyA: bases 973 - 1007

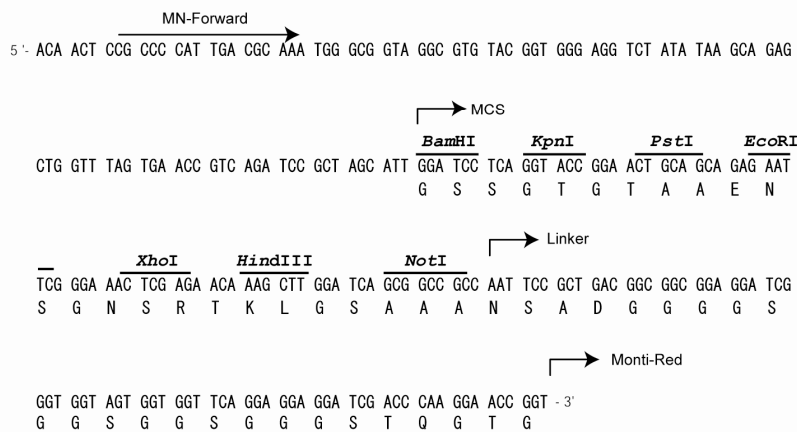
Kanamycin/Neomycin resistance gene: bases 2050 - 2841

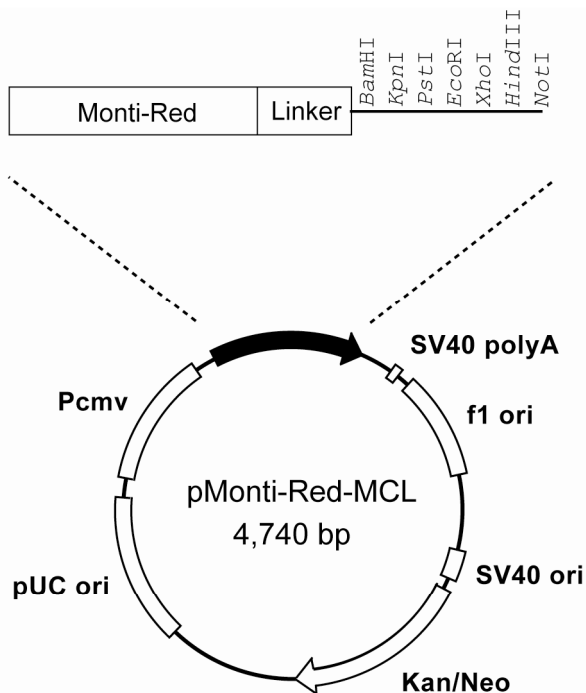
pUC origin: bases 3429 - 4072

f1 origin: bases 1070 - 1525

SV40 origin: bases 1866 - 2001

Linker, MCS and the annealing site of recommended sequence primer (MN-Forward)





**SEQUENCE LANDMARKS:**

- Monti-Red gene: bases 1 - 666
- peptide linker: bases 667-738
- CMV promoter: bases 4144-4716
- SV40 polyA: bases 954-988
- Kanamycin/Neomycin resistance gene: bases 2031-2822
- pUC origin: bases 3410-4053
- f1 origin: bases 1051-1506
- SV40 origin: bases 1847-1982

Linker, MCS and the annealing site of recommended sequence primer (MC-Reverse)

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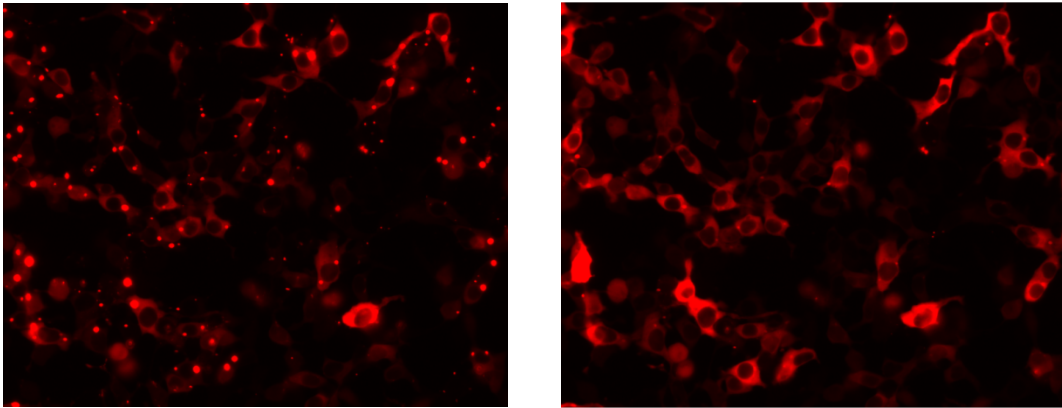
    ┌───┐ Linker
5'- ACC GGT AAT TCC GCT GAC GGC GGC GGA GGA TCG GGT GGT AGT GGT GGT TCA GGA GGA GGA TCG ACC
  T  G  N  S  A  D  G  G  G  G  S  G  G  S  G  G  S  G  G  S  G  G  G  S  T

    ┌───┐ MCS
      BamHI   KpnI     PstI     EcoRI     XhoI     HindIII
CAA GGA GGA TCC TCA GGT ACC GGA ACT GCA GCA GAG AAT TCG GGA AAC TCG AGA ACA AAG CTT GAA
Q  G  G  S  S  G  T  G  T  A  A  E  N  S  G  N  S  R  T  K  L  E

      NotI
TAA GCG GCC GCG ACT CTA GAT CAT AAT CAG CCA TAC CAC ATT TGT AGA GGT TTT ACT TGC TTT AAA
*

      MC-Reverse
AAA CCT CCC ACA CCT CCC -3'
  
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══ : stop codon

**Example of Fluoppi assay:**

HEK293 cells transiently expressing both Ash/p53 and Monti-Red/MDM2 were observed at 0 minute (left) and 10 minutes (right) after addition of 20  $\mu$ M Nutlin-3. The interactions were observed as fluorescent foci (left), and disruptions of the PPI by Nutlin-3 resulted in a cytoplasmic evenly distribution of fluorescence (right).

**Related products:**

AM-8001M	Fluoppi : Ash-hAG (Ash-MNL/MCL + hAG-MNL/MCL)
AM-8002M	Fluoppi : Ash-Red (Ash-MNL/MCL + Monti-Red-MNL/MCL)
AM-8201M	Fluoppi : Ash-hAG [ p53-MDM2 ]
AM-8202M	Fluoppi : Ash-hAG [ mTOR-FKBP12 ]
AM-VS0801M	humanized Azami-Green for Fluoppi (phAG-MNL/MCL)
AM-VS0802M	Monti-Red for Fluoppi (pMonti-Red-MNL/MCL)