

**Fucci (Fluorescent Ubiquitination-based Cell Cycle Indicator) series**  
**pFucci-G<sub>1</sub> Orange (Cloning vector)**

**Code No.**  
AM-V9001M

**Quantity**  
20 µg

**VECTOR DESCRIPTION:**

AM-V9001M pFucci-G<sub>1</sub> Orange is a Cloning vector encoding **CoralHue**<sup>®</sup> monomeric Kusabira-Orange 2 (mKO2) fused to a part of human Cdt1 (hCdt1: Cdc10 dependent transcript 1). “Fucci” stands for *Fluorescent Ubiquitination-based Cell Cycle Indicator*.

Cdt1 is the DNA replication licensing factor. It accumulates during G<sub>1</sub> phase in nuclei, but is degraded during S/G<sub>2</sub>/M phases by ubiquitin-mediated proteolysis. A part of hCdt1 (30-120) is also degradable in a cell cycle dependent manner.

**CoralHue**<sup>®</sup> mKO2 has been generated from dimeric **CoralHue**<sup>®</sup> Kusabira-Orange (KO). **CoralHue**<sup>®</sup> mKO2 has a feature of rapid maturation.

**SOURCE:** The **CoralHue**<sup>®</sup> KO gene was cloned from a stony coral (*Fungia concinna*).

**FORMULATION:** Dry form. Reconstitute with distilled water or TE before use.

**PURITY:** A260/A280 > 1.5

**STORAGE:** Stored at -20°C

**SEQUENCE LANDMARKS:**

Fucci-G<sub>1</sub> Orange gene (including stop codon): bases 2264-3244

Ampicillin resistance gene: bases 200-1059

ColE1 origin: bases 1062-2002

**REFERENCES:**

- 1) Sakaue-Sawano, A., *et al.*, *Cell*. **132**, 487-498 (2008)
- 2) Nakayama, K. I., *et al.*, *Nat. Rev. Cancer*. **6**, 369-381 (2006)
- 3) Blow, J. J., and Dutta, A., *Nat. Rev. Mol. Cell Biol.* **6**, 476-486 (2005)
- 4) Nishitani, H., *et al.*, *J. Biol. Chem.* **279**, 30807-30816 (2004)
- 5) Karasawa, S., *et al.*, *J. Biol. Chem.* **278**, 34167-71 (2003)
- 6) Nishitani, H., *et al.*, *Nature*. **404**, 625-628 (2000)

**INTENDED USE:**

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

**GenBank:**

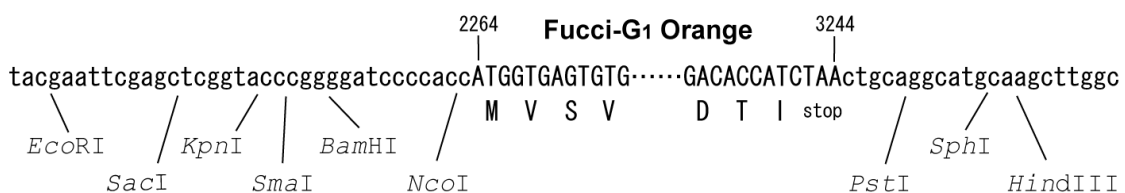
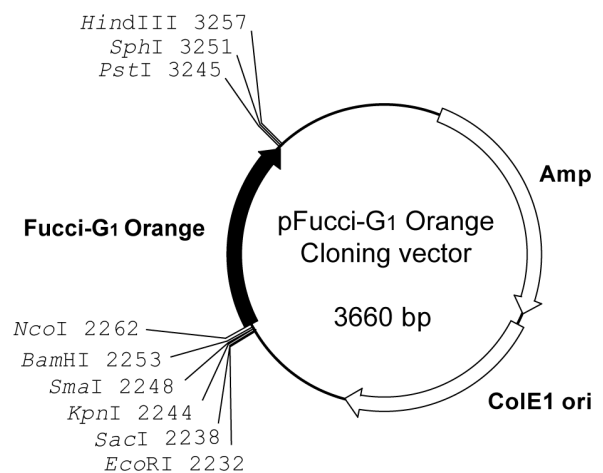
Accession Numbers: AB370332

**NOTICES:**

- 1) pFucci-G<sub>1</sub> Orange (Cloning vector) is not expression vector. When pFucci-G<sub>1</sub> Orange is expressed in any cells, the cDNA must be transferred to appropriate expression vectors by your own.
- 2) Val (encoded by GTG) is inserted as the second amino acid of **CoralHue**<sup>®</sup> mKO2 to form the Kozak sequence.
- 3) It is recommended that Fucci be stably expressed.

**RELATED PRODUCTS:**

- AM-V9014M pFucci- S/G<sub>2</sub>/M Green (Cloning vector)
- AM-V9016M pFucci-S/G<sub>2</sub>/M Green (Expression vector)
- AM-V9010M pFucci-S/G<sub>2</sub>/M Green-Hyg (Expression vector)
- AM-V9003M pFucci-G<sub>1</sub> Orange (Expression vector)
- AM-V9034M pFucci-S/G<sub>2</sub>/M Green (N+C) (Cloning vector)
- AM-V9030M pFucci-S/G<sub>2</sub>/M Green (N+C)-Hyg (Expression vector)



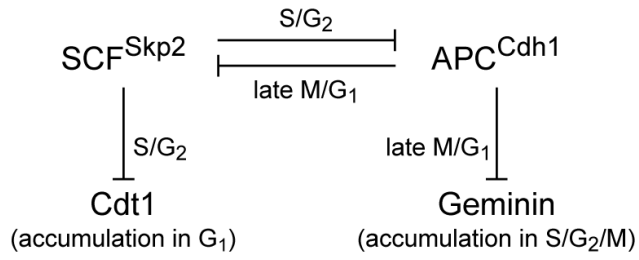


Fig 1. Cell cycle regulation by SCF<sup>Skp2</sup> and APC<sup>Cdh1</sup>.

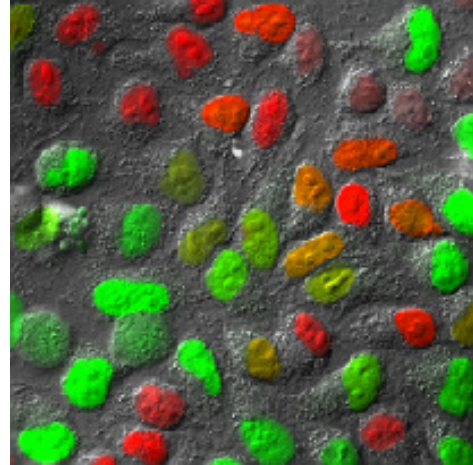


Fig 3. HeLa cells stably expressing Fucci-G<sub>1</sub> Orange and Fucci-S/G<sub>2</sub>/M Green. Fucci effectively labels individual nuclei in G<sub>1</sub> phase orange and those in S/G<sub>2</sub>/M phases green.

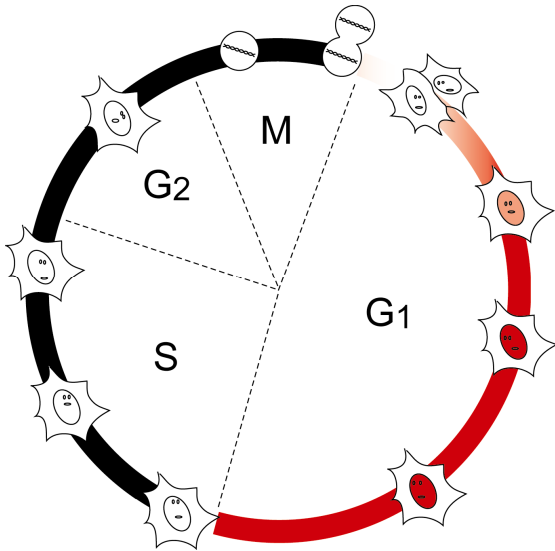
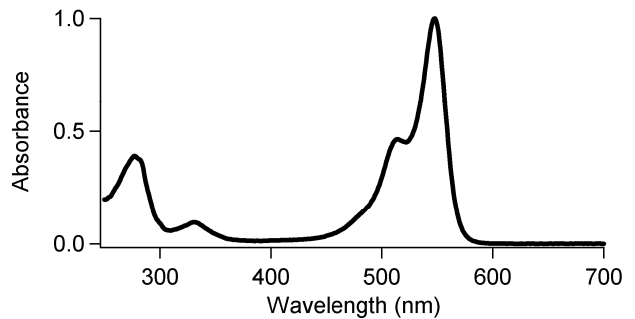
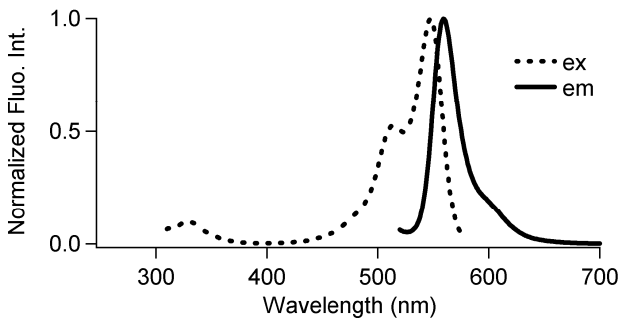


Fig 2. Schematic of the cell cycle specific fluorescence of Fucci-G<sub>1</sub> Orange.

**CoralHue<sup>®</sup> mKO2:** 218 amino acids

	Excit./Emiss.Maxima (nm)	Extinction Coefficient(M <sup>-1</sup> cm <sup>-1</sup> )	Fluorescence Quantum Yield	pH sensitivity
mKO2	551/565	63,800 (551 nm)	0.62	pK <sub>a</sub> =5.5



## Fucci-G<sub>1</sub> Orange

### 1) DNA sequence

ATGGTGAGTGTGATTAACCAGAGATGAAGATGAGGTAACAT  
GGACGGCTCCGTCAATGGGCATGAGTTCACAATTGAAGGTGAAG  
GCACAGGCAGACCTTACGAGGGACATCAAGAGATGACACTACGC  
GTCACAATGGCCGAGGGCGGGCCAATGCCTTTGGCGTTTGA  
AGTGTACACAGTGTCTGTTACGGCCACAGAGTATTTACTAAAT  
ATCCAGAAGAGATACCAGACTATTTCAAACAAGCATTTCTGAA  
GGCCTGTGATGGGAAAGGTGTTGGAGTTCGAAGATGGTGGGTC  
CGCTTCAGTCAGTGCATATAAGCCTTAGAGGAAACACCTTCT  
ACCACAAATCCAAATTTACTGGGGTAACTTTCTGCGGATGGT  
CCTATCATGCAAAACCAAAGTGTGATTGGGAGCCATCAACCGA  
GAAAATTACTGCCAGCGACGGAGTTCTGAAGGGTGATGTTACGA  
TGTACCTAAAAGTTGAAGGAGGCGGCAATCACAAATGCCAAATG  
AAGACTACTTACAAGGCGGCAAAGAGATTCTTGAATGCCAGG  
AGACCATTACATCGGCCATCGCCTCGTCAGGAAAACCGAAGGCA  
ACATTACTGAGCAGGTAGAAGATGCAGTAGCTCATTACTCAATG  
TTGCCTTCCAGGGCGGATATCCATCACACTGGCGCCGCTCGA  
GCCAGCCCCGCCAGGCCCGCACTCCGCGCCCCGGCCTCCGCTA  
CCAGTGGCAGCCGCAAGCGCGCCCGCCCGCCCGCCCGCCGGA  
CGCGACCAGGCCAGGCCACCGCCCGCAGGAGACTGCGGCTGTC  
GGTGGAGGAGGTTTCCAGGCCAGTACCCCGAGGCCCGAGACA  
TCCAGCCTGCCCTTCTCCGGGCCAGAAGATAAAGAAATCCACC  
CCGGCAGCAGGTGAGCCGCCACCTGACATCCGCGCAGGACCA  
GGACACCATC

### 2) Amino acid sequence

MVSVIKPEMKMRYMDGVSNGHEFTIEEGTGRPYEGHQEMTLR  
VTMAEGGPMPPAFDLVSHVFCYGHRVFTKYPEEIPDYFKQAFPE  
GLSWERSLEFEDGGSASVSAHISLRGNTFYHKSFTGVNFPADG  
PIMQNSVDWEPSTEKITASDGVLKGDTVMYLKEGGGNHKCQM  
KTTYKAAKEILEMPGDHYIGHRLVRKTEGNITEQVEDAVAHYSM  
LPSQGGYPSHWRPLEPSARPALRAPASATSGSRKRARPPAAPG  
RDQARPPARRRLRLSVDEVSSPSTPEAPDIPACPSPGQKIKKST  
PAAGQPPHLTSAQDQDTI

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**CoralHue<sup>®</sup> mKO** is a product of co-development with Dr. Atsushi Miyawaki at the Laboratory for Cell Function and Dynamics, the Brain Science Institute, and the Institute of Physical and Chemical Research (RIKEN).

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Patent Nos. JP4258724, US7226993 and EP1700913 and patents pending.