

Fucci (Fluorescent Ubiquitination-based Cell Cycle Indicator) series
pFucci-S/G₂/M Green (Cloning vector)

Code No.	Quantity
AM-V9014M	20 µg

VECTOR DESCRIPTION:

AM-V9014M pFucci-S/G₂/M Green is a Cloning vector encoding **CoralHue™** humanized-codon monomeric Azami-Green1 (hmAG1) fused to a part of human Geminin (hGeminin). “Fucci” stands for *Fluorescent Ubiquitination-based Cell Cycle Indicator*.

Geminin is an inhibitor of the DNA replication licensing factor. It accumulates during the S, G₂, and M phases, but is degraded during G₁ phase by ubiquitin-mediated proteolysis. A part of hGeminin (1-110) is also degradable in a cell cycle dependent manner.

CoralHue™ hmAG1 sequence is codon-optimized for higher expression in mammalian cells. **CoralHue™** monomeric AG1 (mAG1) has been generated from tetrameric **CoralHue™** Azami-Green (AG).

SOURCE: The **CoralHue™** AG gene was cloned from a stony coral (*Galaxea fascicularis*).

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

PURITY: A260/A280 > 1.5

STORAGE: Stored at -20°C

SEQUENCE LANDMARKS:

Fucci-S/G₂/M Green gene (including stop codon): bases 2264-3304

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)

For more information, please visit our web site.

<https://ruo.mbl.co.jp/>

INTENDED USE:

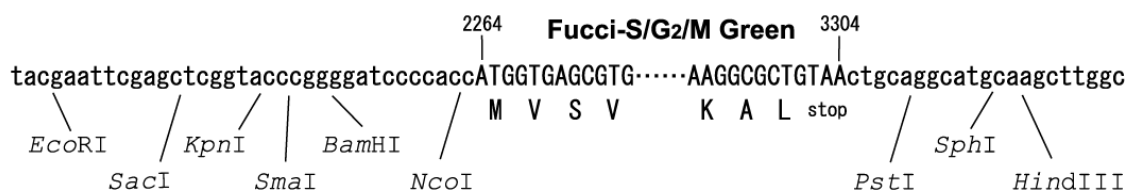
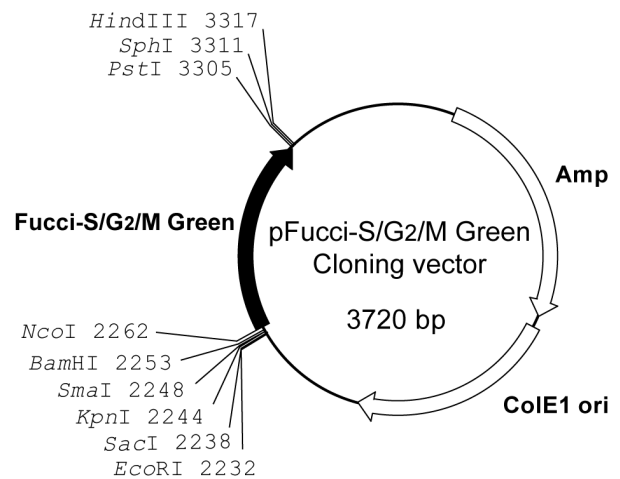
For Research Use Only. Not for use in diagnostic procedures.

GenBank:

Accession Numbers: AB370333

NOTICES:

- 1) pFucci-S/G₂/M Green (Cloning vector) is not expression vector. When pFucci-S/G₂/M Green 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™** hmAG1 to form the Kozak sequence.
- 3) It is recommended that Fucci be stably expressed.



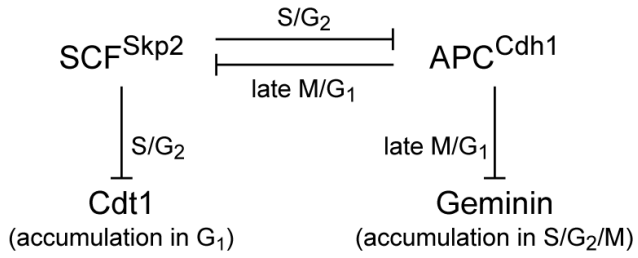


Fig 1. Cell cycle regulation by SCF^{Skp2} and APC^{Cdh1}

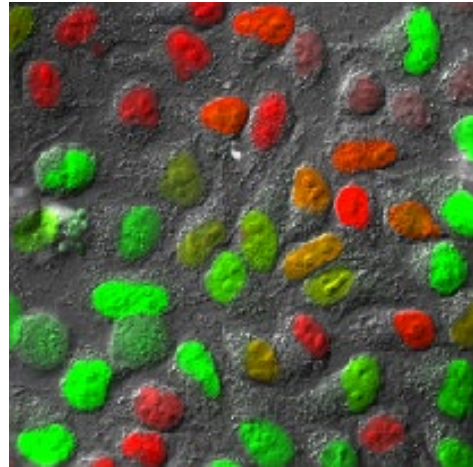


Fig 3. HeLa cells stably expressing Fucci-G₁ Orange and Fucci-S/G₂/M Green. Fucci effectively labels individual nuclei in G₁ phase orange and those in S/G₂/M phases green.

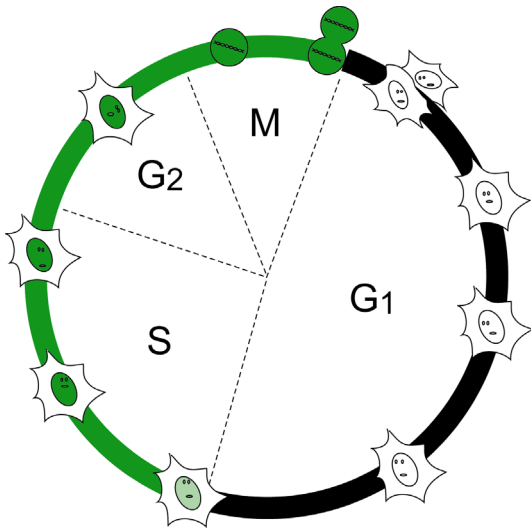
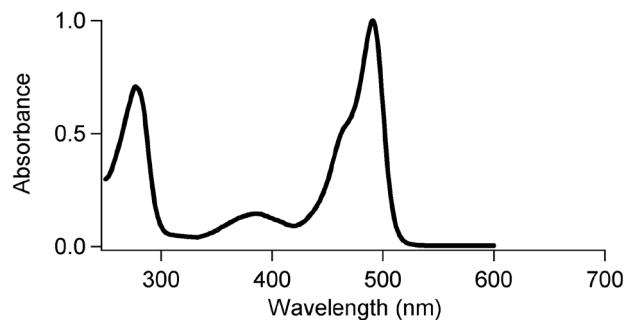
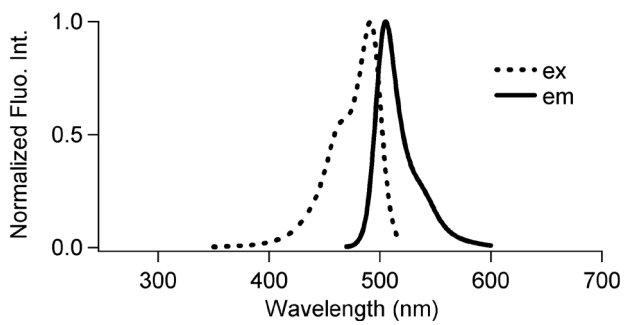


Fig 2. Schematic of the cell cycle specific fluorescence of Fucci-S/G₂/M Green.

CoralHue™ mAG1: 226 amino acids

	Excit./Emiss.Maxima (nm)	Extinction Coefficient(M ⁻¹ cm ⁻¹)	Fluorescence Quantum Yield	pH sensitivity
mAG1	492/505	55,500 (492 nm)	0.74	pK _a =5.8



Fucci-S/G₂/M Green

1) DNA sequence

ATGGTGAGCGTGATCAAGCCCGAGATGAAGATCAAGCTGTGC
ATGAGGGGCACCGTGAACGGCCACAACCTTCGTGATCGAGGGC
GAGGGCAAGGGCAACCCCTACGAGGGCACCCAGATCCTGGAC
CTGAACGTGACCGAGGGCGCCCCCTGCCCTTCGCCTACGAC
ATCCTGACCACCGTGTTCCAGTACGGCAACAGGGCCTTCACC
AAGTACCCCGCGACATCCAGGACTACTTCAAGCAGACCTTC
CCCGAGGGTACCCTGGGAGAGGAGCATGACCTACGAGGAC
CAGGGCATCTGCACCGCCACCAGCAACATCAGCATGAGGGGC
GACTGCTTCTTCTACGACATCAGGTTGACGGCACCAACTTC
CCCCCAACGGCCCGTGATGCAGAAGAAGACCCTGAAGTGG
GAGCCCAGCACCGAGAAGATGTACGTGGAGGACGGCGTGCTG
AAGGGCGACGTGAACATGAGGCTGCTGCTGGAGGGCGGGC
CACTACAGGTGCGACTTCAAGACCACCTACAAGCCAAGAAG
GAGGTGAGGCTGCCGACGCCACAAGATCGACCACAGGATC
GAGATCCTGAAGCAGACAAGGACTACAACAAGGTGAAGCTG
TACGAGAACGCCGTGGCCAGTACTCCATGCTGCCAGCCAG
GCCAAGGGATATCCATCACACTGGCGCCGCTCGAGATGAAT
CCCAGTATGAAGCAGAAACAAGAAGAAATCAAAGAGAATATA
AAGAATAGTTCTGTCCCAAGAAGAACTCTGAAGATGATTCAG
CCTTCTGCATCTGGATCTCTTGTGGAAGAGAAAATGAGCTG
TCCGCAGGCTTGCCAAAAGGAAACATCGGAATGACCACTTA
ACATCTACAACCTCCAGCCCTGGGGTTATTGTCCAGAAATCT
AGTGAATAAATAATCTTGGAGGAGTCACCCAGGAGTCATTT
GATCTTATGATTAAGAAAATCCATCCTCTCAGTATTGGAAG
GAAGTGGCAGAAAACGGAGAAAGGCGCTG

2) Amino acid sequence

MVSVIKPEMKIKLCMRGTVNGHNFVIEGEGKGNPYEGTQILDNLN
VTEGAPLPFAYDILTTVFQYGNRAFTKYPADIQDYFKQTFPEGY
HWERSMTYEDQGITATSNISMRGDCFFYDIRFDGTFNPPNGPV
MQKTLKWEKSTKMYVEDGVLKGDVNMRLLEGGGHYRCDFKT
TYKAKKEVRLPDAHKIDHRIEILKHDKDYNKVKLYENAVARYSM
LPSQAKGYPSHWRPLEMNPMSMKQKQEEIKENIKNSSVPRRTLKM
IQPSASGSLVGRENELSAGLSKRKHRNDHLTSTSSPGVIVPES
SENKNLGGVTQESFDLMIKENPSSQYWKVEAEKRRKAL

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*CoralHue*TM mAG 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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