

All^{ele}ustrious pmWasabi-Lysosomes

(Lysosomes)

Catalog Number: ABP-FP-WLYS100

Size: 10ug Price: \$349.00

Introduction

All^{ele}ustrious pmWasabi-Lysosomes is a mammalian expression vector that expresses a fusion of rat LAMP1 and mWasabi. This product is a great marker for lysosomes as LAMP1, or lysosomal-associated membrane protein 1, also known as CD107a (Cluster of Differentiation 107a) associates with lysosomes.

All^{ele}ustrious mWasabi is a monomeric green fluorescent protein that can be easily detected using standard GFP filter sets. mWasabi may be used as a direct replacement for EGFP or other GFPs for superior performance, and may be co-imaged with blue and red fluorescent labels without substantial bleed-through.

Lysosomes contain enzymes that digest excess organelles, viruses, and bacteria. They are formed by the addition of hydrolytic enzymes to early endosomes from the Golgi apparatus. LAMP1 protein belongs to a family of membrane glycoproteins that localize in lysosomes. This glycoprotein provides selectins with carbohydrate ligands. It may also play a role in tumor cell metastasis.

Source

Engineered variant of mTFP1, originally derived from *Clavularia sp.* coral.

Recommended Use

mWasabi has been optimized for use with standard GFP/FITC filter sets.

Features

- About 2-fold brighter than EGFP
- Similar photostability to EGFP
- Uses standard filter sets
- Can be co-imaged with blue and red FPs or dyes
- Mammalian expression vector ready to transfect your favorite cells
- Low sensitivity to acidic pH (fluorescence pKa=4.3)
- True monomer that will not aggregate or cause nonspecific interactions

Reconstitution

10 µg provided in lyophilized powder form. Reconstitute with 10 µL of nuclease-free water for a final concentration of 1 µg/µL.

Storage

Store at -20°C or at -80°C for long-term preservation.

Human CMV Immediate-Early

Promoter (CMV Promoter).....1-589

LAMP1.....626-1846

Linker.....1847-1906

mWasabi.....1907-2617

SV40 PolyA Signal.....2769-2819

bla Promoter.....3358-3462

Ampicillin Resistance Gene.....3447-4307

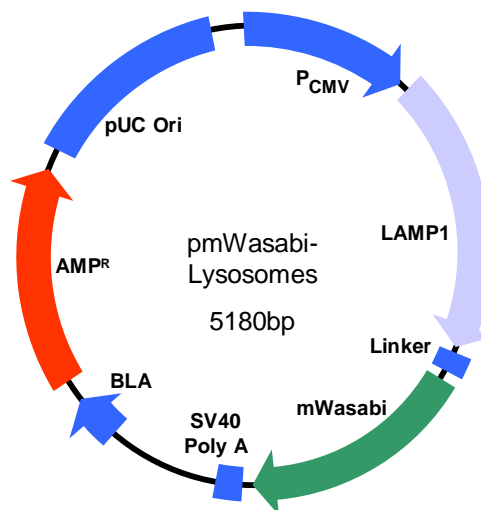
pUC Origin.....4456-5098

Upstream Sequencing Primer:

Universal CMV Promoter Primer

Downstream Sequencing Primer:

SV40 Primer:GCTTT ATTTG TGAAA TTTGT GATGC TATTG C



References: Ai H, Olenych SG, Wong P, Davidson MW, Campbell RE. Hue-shifted monomeric variants *Clavularia* cyan fluorescent protein: identification of the molecular determinants of color and applications in fluorescence imaging. *BMC Biology*. 2008 Mar; 6:13. Shaner NC, Patterson GH, Davidson MW. Advances in fluorescent protein technology. *J Cell Sci*. 2007 Dec 15;120(Pt 24):4247-60. Ai HW, Hazelwood KL, Davidson MW, Campbell RE. Fluorescent protein FRET pairs for ratiometric imaging of dual biosensors. *Nature Methods*. 2008 5(5): 401-03. Ai HW, Henderson JN, Remington SJ, Campbell RE. Directed evolution of a monomeric, bright, and photostable version of *Clavularia* cyan fluorescent protein: structural characterization and applications in fluorescence imaging. *Biochem J*. 2006. Shaner NC, Steinbach PA, Tsien RY. A guide to choosing fluorescent proteins. *Nat Methods*. 2005 2(12):905-09. Rohrer J, Schweizer A, Russell D, Kornfeld S (1996). "The targeting of Lamp1 to lysosomes is dependent on the spacing of its cytoplasmic tail tyrosine sorting motif relative to the membrane." *J. Cell Biol.* 132 (4): 565–76. Wohlgemuth, S.E., Julian, D., Akin, D.E., Fried, J., Toscano, K., Leeuwenburgh, C. and Dunn, W.A. Jr. (2007). "Autophagy in the heart and liver during normal aging and calorie restrictions" *Rejuvenation Res* 10 (3), 281-292.

SEQUENCE

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661 GTTGTGCTG GCAGGCCTTG CACACAGCGC CCCAGCACTG TTCGAGGTGA AAGACAACAA
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2581 CAAGTCCACC GACGGCATGG ACGAGCTGTA CAAGTAAGCG GCCCGACTC TAGATCATAA
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Enzyme	Cuts	Positions	Sequence					
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AatII	5	125 178 261 447 2403	gacgt/c	BstYI	10	585 609 1888 2520 3587 3604 4372	r/gatcy	
Acc113I	1	3753	agt/act			4384 4470 4481		
Acc16I	1	4011	tgc/gca	BstZI	2	647 2619	c/gggcc	
AccB1I	6	465 638 1406 2039 3050 4281	g/gyrcc	CciNI	2	647 2619	gc/gggccg	
AccB7I	2	1208 2346	ccannnn/ntgg	Cfr10I	7	1862 1871 1880 1894 2342 2986	r/ccggy	
AccBSI	2	2950 3395				4149		
AccI	1	1092	gagcgg	Cfr9I	1	634	c/ccggg	
AcsI	4	1850 2789 3274 3285	gt/mkac	CfrI	6	647 738 1990 2431 2619 3841	y/ggcer	
AcyI	8	122 175 258 444 639 2352 2400	r/aatty	DraI	4	2677 3656 4348 4367	ttt/aaa	
		3694	gr/cgyc	DraII	3	1013 1764 2130	rg/gnecy	
AfeI	1	596	agc/gct	DraIII	3	729 2503 3094	cacnnn/gtg	
AflIII	5	781 935 954 2861 5122	a/crygt	DrdI	3	1088 3138 5020	gacnnn/ngtcc	
AgeI	1	1894	a/ccggt	DsaI	5	360 624 1905 2256 2571	c/crygg	
AhdI	1	4234	gacnnn/ngtcc	EaeI	6	647 738 1990 2431 2619 3841	y/ggcer	
Alw21I	5	1119 1623 3566 3651 4812	gwgwc/c	EagI	2	647 2619	c/gggccg	
Alw44I	2	3562 4808	g/tgca	Eam1104I	6	1174 1526 1580 1717 1821 3445	ctcttc	
AlwNI	2	818 4713	cagnnn/ctg	Eam1105I	1	4234	gacnnn/ngtcc	
Ama87I	3	613 634 2183	c/yggrg	EaRI	6	1174 1526 1580 1717 1821 3445	ctcttc	
Aor51HI	1	596	agc/gct	EclHKI	1	4234	gacnnn/ngtcc	
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ApaLI	2	3562 4808	g/tgca	Eco105I	1	340	tac/gta	
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BalI	2	740 1992	tgg/cca			2395 3552 4600		
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BanI	6	465 638 1406 2039 3050 4281	g/gyrcc	Eco88I	3	613 634 2183	c/yggrg	
BanII	6	873 1017 1542 1768 2077 3020	grgcy/c	Eco91I	2	1417 2551	g/gtnacc	
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		3694		EcoRI	1	1850	g/aatc	
BbsI	1	1660	gaagac	EcoT14I	4	360 624 1905 2256	c/cwvvg	
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BglI	7	90 212 283 626 1307 2474 4116	gcennn/nggc	FauNDI	1	234	ca/tatg	
BglII	1	609	a/gatcc	FriOI	6	873 1017 1542 1768 2077 3020	grgcy/c	
BpiI	1	1660	gaagac	FspI	1	4011	tgc/gca	
BpmI	4	1456 2062 2425 4149	ctggag	GsuI	4	1456 2062 2425 4149	ctggag	
BpuAI	1	1660	gaagac	HaeII	8	598 621 642 690 1643 2936 2944	rgcgc/y	
BsaAI	3	340 2374 3091	yac/gtr			4882		
BsaBI	1	2637	gatnn/nnttc	HinI	8	122 175 258 444 639 2352 2400	gr/cgyc	
BsaHI	8	122 175 258 444 639 2352 2400	gr/cgyc			3694		
		3694		HincII	2	2446 2738	gty/rac	
BsaI	4	798 1930 2341 4167	ggdctc	HindII	2	2446 2738	gty/rac	
BsaMI	5	864 1369 1476 2725 2824	gaatgc	HpaI	1	2738	gtt/aa	
BsaOI	6	650 1899 2622 3716 3865 4788	cgry/cg	Hsp92I	8	122 175 258 444 639 2352 2400	gr/cgyc	
BsaWI	5	600 1894 3938 4769 4916	w/ccggg			3694		
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Bse8I	1	2637	gatnn/nnttc	MamI	1	2637	gatnn/nnttc	
BseRI	4	1621 1824 1927 2266	gaggag	MfeI	1	2725	c/aattg	
Bsh1285I	6	650 1899 2622 3716 3865 4788	cgry/cg	MflI	10	585 609 1888 2520 3587 3604 4372	r/gatcy	
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Bsp1407I	2	1720 2607	t/gtaca	Msp17I	8	122 175 258 444 639 2352 2400	gr/cgyc	
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BspHI	2	3394 4402	t/catga	Mva1269I	5	864 1369 1476 2725 2824	gaatgc	
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BsrDI	3	1755 3998 4180	gcaatg	NgoAIV	1	2986	g/ccgpc	
BsrFI	7	1862 1871 1880 1894 2342 2986	r/ccggy	NgoMI	1	2986	g/ccgpc	
		4149		NheI	1	591	g/ctagc	
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BstMCI	6	650 1899 2622 3716 3865 4788	cgry/cg	Psp1406I	2	3632 4005	aa/cgtt	
BstPI	2	1417 2551	g/gtnacc	PspAI	1	634	c/ccggg	
BstSFI	7	819 1457 2461 2868 3988 4666	c/tryag	PspALI	1	636	ccc/ggg	
		4857		PspBI	2	1417 2551	g/gtnacc	
BstSNI	1	340	tac/gta	PspOMI	2	1013 1764	g/gggcc	
BstX2I	10	585 609 1888 2520 3587 3604 4372	r/gatcy	PstI	2	823 1461	ctgca/g	
		4384 4470 4481		PstNHI	1	591	g/ctagc	
				PvuI	1	3865	cgat/cg	
				RcaI	2	3394 4402	t/catga	



SapI	1	1580	gctcttc	Van91I	2	1208 2346	ccannnn/ntgg
ScaI	1	3753	agt/act	VneI	2	3562 4808	g/tgcac
SfcI	7	819 1457 2461 2868 3988 4666	c/tryag	VspI	2	7 4059	at/taat
		4857		XbaI	1	2629	t/ctaga
Sfr274I	1	613	c/tcgag	XcmI	1	2593	ccannnnn/nnntgg
SgrAI	1	1880	cr/ccggyg	XhoI	1	613	c/tcgag
SmaI	1	636	ccc/ggg	XhoII	10	585 609 1888 2520 3587 3604 4372	r/gatcy
SnaBI	1	340	tac/gta			4384 4470 4481	
SseBI	2	675 1139	agg/cct	XmaI	1	634	c/ccggg
SspBI	2	1720 2607	t/gtaca	XmaIII	2	647 2619	c/ggccg
SspI	2	3299 3429	aat/att	XmnI	1	3634	gaann/nnttc
StuI	2	675 1139	agg/cct	Zsp2I	1	5179	atgca/t
StyI	4	360 624 1905 2256	c/cwggg				More info

The following enzymes do not cut:

Acc65I, AccII, AclNI, AfIII, Aocl, Ascl, Asp718I, Aspl, Atsl, AvriI, BanIII, BbrPI, Bbul, BclI, BfrI, BlnI, Bipl, Bpu1102I, Bpu14I, Bsa29I, BscI, Bse21I, BseAI, BseCI, BsePI, BsgI, BsiMI, BsiWI, BsmBI, Bsp106I, Bsp119I, Bsp13I, Bsp1720I, Bsp68I, BspDI, BspEI, BspTI, BspXI, BssHII, Bst1107I, Bst98I, BstBI, Bsu15I, Bsu36I, CellI, Cfr42I, Clal, Cpol, Csp45I, Cspl, CvnI, Ecl136II, Eco32I, Eco72I, Eco81I, EcoCRI, EcoNI, EcoRV, Esp3I, Fbal, Fsel, HindIII, Kpn2I, KpnI, Ksp22I, Kspl, Lspl, MroI, MspCI, Nrul, NspV, PacI, Pael, Pfi23II, PmaCI, Pmel, PmlI, PpuMI, PshAI, Psp124BI, Psp5II, PspLI, Pvull, RsrII, SacI, SacII, Sall, SbfI, SexAI, Sfil, Sfr303I, Sful, Sgfi, Smil, Spel, SphI, SpII, SrfI, Sse8387I, SstI, SstII, SunI, Swal, Tth111I, Vha464I



Related products:

Current Alle^{ele}ustious Fluorescent Protein Family Members:

The founding member is mTFP1.

mTFPG3 is a green FP with 3 amino difference from mTFP1. It has a slightly red-shifted emission spectrum and is 1.5 fold brighter compared to EGFP. While being very bright, mTFPG3 can be photobleached within ~5 sec, about 30 times faster than EGFP, suitable for certain cell-based assays that require a bright FP with very short half-life.

mTFP0.7 is a precursor during the evolution of mTFP1. It has photo-switchable properties like Dronpa that cycles between fluorescent and nonfluorescent states. It may be developed into components in PALM/SIM applications.

Basic Vectors

Three vectors are available: pNCS-mWasabi, pmWasabi-N and pmWasabi-C.

Subcellular Marker Vectors

Twenty six vectors are available.

Vectors in Viral Vectors

All plasmid format vectors in Allele's Phoenix Retroviral vector or HiTiter Lentiviral Vectors.

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