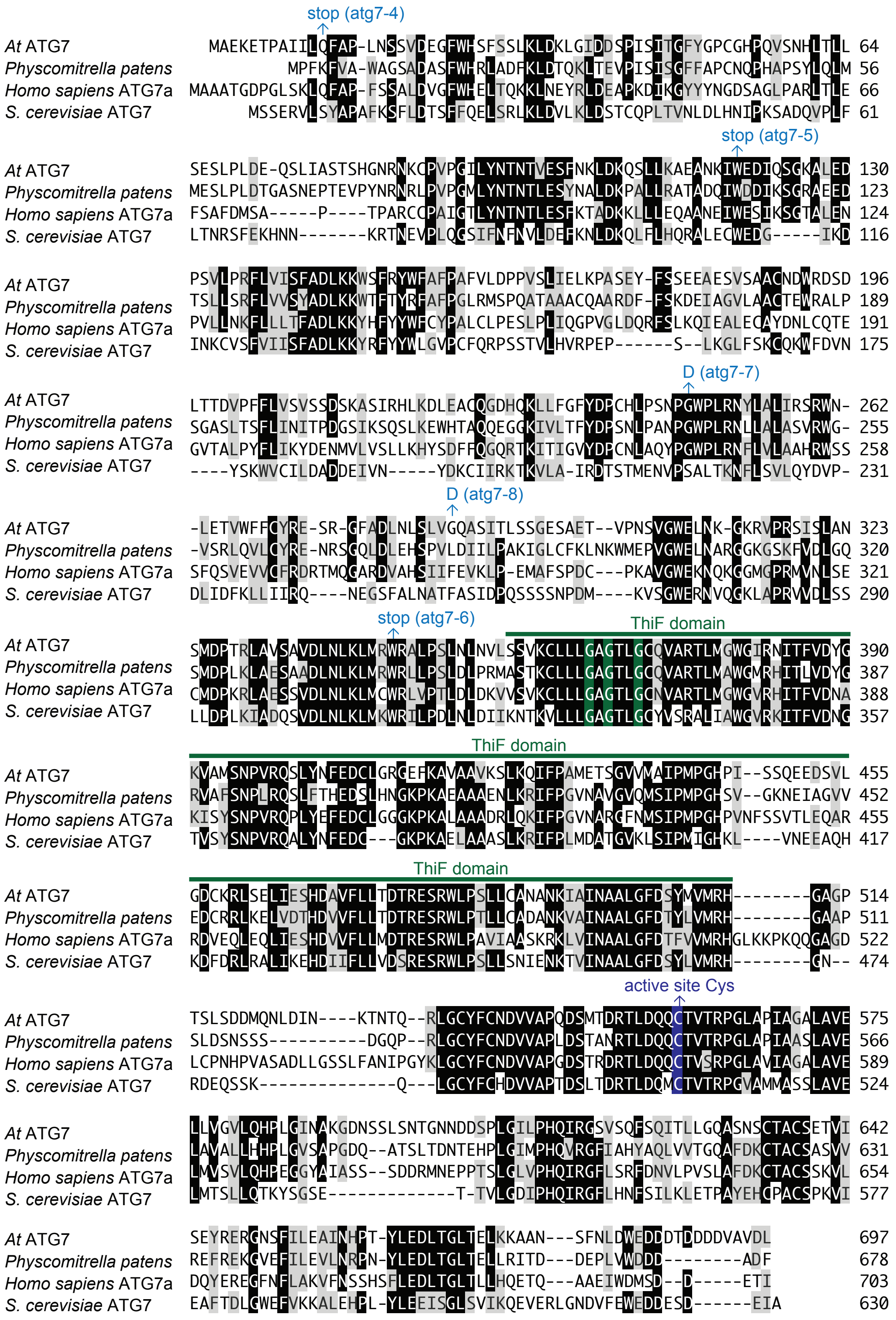

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At ATG7                     MAEKETPAIILQFAP-LNSSVDEGFWHSFSSLKLDKLGIDDSPISITGFYGPCGHPQVSNHLTLL 64
Physcomitrella patens               MPFKFVA-WAGSADASFWHRLADFKLDTQKLTEVPISISGFFAPCNQPHAPSYLQLM 56
Homo sapiens ATG7a        MAAATGDPGLSKLQFAP-FSSALDVGFWHELTQKKLNEYRLDEAPKDIKGYYYNGDSAGLPARLTLE 66
S. cerevisiae ATG7             MSSERVLSYAPAFKSFLDTSFFQELSRLKLDVLKLDSTCQPLTVNLDLHNIPKSADQVPLF 61

At ATG7                   SESLPLDE-QSLIASTSHGNRNKCPVPGILYNTNTVESFNKLDKQSLLKAEANKIWEDIQSGKALED 130
Physcomitrella patens     MESLPLDTGASNEPTEVPYNRNRLPVPGMLYNTNTLESYNALDKPALLRATADQIWDDIKSGRAEED 123
Homo sapiens ATG7a        FSAFDMSA-----P----TPARCCPAIGTLYNTNTLESFKTADKKLLLEQAANEIWESIKSGTALEN 124
S. cerevisiae ATG7        LTNRSFEKHNN-------KRTNEVPLQGSIFNFNVLDEFKNLDKQLFLHQRALECWEDG-----IKD 116

At ATG7                   PSVLPRFLVISFADLKKWSFRYWFAFPAFVLDPPVSLIELKPASEY-FSSEEAESVSAACNDWRDSD 196
Physcomitrella patens     TSLLSRFLVVSYADLKKWTFTYRFAFPGLRMSPQATAAACQAARDF-FSKDEIAGVLAACTEWRALP 189
Homo sapiens ATG7a        PVLLNKFLLLTFADLKKYHFYYWFCYPALCLPESLPLIQGPVGLDQRFSLKQIEALECAYDNLCQTE 191
S. cerevisiae ATG7        INKCVSFVIISFADLKKYRFYYWLGVPCFQRPSSTVLHVRPEP------S--LKGLFSKCQKWFDVN 175

At ATG7                   LTTDVPFFLVSVSSDSKASIRHLKDLEACQGDHQKLLFGFYDPCHLPSNPGWPLRNYLALIRSRWN- 262
Physcomitrella patens     SGASLTSFLINITPDGSIKSQSLKEWHTAQQEGGKIVLTFYDPSNLPANPGWPLRNLLALASVRWG- 255
Homo sapiens ATG7a        GVTALPYFLIKYDENMVLVSLLKHYSDFFQGQRTKITIGVYDPCNLAQYPGWPLRNFLVLAAHRWSS 258
S. cerevisiae ATG7        ----YSKWVCILDADDEIVN-----YDKCIIRKTKVLA-IRDTSTMENVPSALTKNFLSVLQYDVP- 231

At ATG7                   -LETVWFFCYRE-SR-GFADLNLSLVGQASITLSSGESAET--VPNSVGWELNK-GKRVPRSISLAN 323
Physcomitrella patens     -VSRLQVLCYRE-NRSGQLDLEHSPVLDIILPAKIGLCFKLNKWMEPVGWELNARGGKGSKFVDLGQ 320
Homo sapiens ATG7a        SFQSVEVVCFRDRTMQGARDVAHSIIFEVKLP-EMAFSPDC---PKAVGWEKNQKGGMGPRMVNLSE 321
S. cerevisiae ATG7        DLIDFKLLIIRQ----NEGSFALNATFASIDPQSSSSNPDM----KVSGWERNVQGKLAPRVVDLSS 290

At ATG7                   SMDPTRLAVSAVDLNLKLMRWRALPSLNLNVLSSVKCLLLGAGTLGCQVARTLMGWGIRNITFVDYG 390
Physcomitrella patens     SMDPLKLAESAADLNLKLMRWRLLPSLDLPRMASTKCLLLGAGTLGCQVARTLMAWGMRHITLVDYG 387
Homo sapiens ATG7a        CMDPKRLAESSVDLNLKLMCWRLVPTLDLDKVVSVKCLLLGAGTLGCNVARTLMGWGVRHITFVDNA 388
S. cerevisiae ATG7        LLDPLKIADQSVDLNLKLMKWRILPDLNLDIIKNTKVLLLGAGTLGCYVSRALIAWGVRKITFVDNG 357

At ATG7                   KVAMSNPVRQSLYNFEDCLGRGEFKAVAAVKSLKQIFPAMETSGVVMAIPMPGHPI--SSQEEDSVL 455
Physcomitrella patens     RVAFSNPLRQSLFTHEDSLHNGKPKAEAAAENLKRIFPGVNAVGVQMSIPMPGHSV--GKNEIAGVV 452
Homo sapiens ATG7a        KISYSNPVRQPLYEFEDCLGGGKPKALAAADRLQKIFPGVNARGFNMSIPMPGHPVNFSSVTLEQAR 455
S. cerevisiae ATG7        TVSYSNPVRQALYNFEDC---GKPKAELAAASLKRIFPLMDATGVKLSIPMIGHKL----VNEEAQH 417

At ATG7                   GDCKRLSELIESHDAVFLLTDTRESRWLPSLLCANANKIAINAALGFDSYMVMRH--------GAGP 514
Physcomitrella patens     EDCRRLKELVDTHDVVFLLTDTRESRWLPTLLCADANKVAINAALGFDTYLVMRH--------GAAP 511
Homo sapiens ATG7a        RDVEQLEQLIESHDVVFLLMDTRESRWLPAVIAASKRKLVINAALGFDTFVVMRHGLKKPKQQGAGD 522
S. cerevisiae ATG7        KDFDRLRALIKEHDIIFLLVDSRESRWLPSLLSNIENKTVINAALGFDSYLVMRH--------GN-- 474

At ATG7                   TSLSDDMQNLDIN----KTNTQ--RLGCYFCNDVVAPQDSMTDRTLDQQCTVTRPGLAPIAGALAVE 575
Physcomitrella patens     SLDSNSSS----------DGQP--RLGCYFCNDVVAPLDSTANRTLDQQCTVTRPGLAPIAASLAVE 566
Homo sapiens ATG7a        LCPNHPVASADLLGSSLFANIPGYKLGCYFCNDVVAPGDSTRDRTLDQQCTVSRPGLAVIAGALAVE 589
S. cerevisiae ATG7        RDEQSSK--------------Q---LGCYFCHDVVAPTDSLTDRTLDQMCTVTRPGVAMMASSLAVE 524

At ATG7                   LLVGVLQHPLGINAKGDNSSLSNTGNNDDSPLGILPHQIRGSVSQFSQITLLGQASNSCTACSETVI 642
Physcomitrella patens     LAVALLHHPLGVSAPGDQ--ATSLTDNTEHPLGIMPHQVRGFIAHYAQLVVTGQAFDKCTACSASVV 631
Homo sapiens ATG7a        LMVSVLQHPEGGYAIASS--SDDRMNEPPTSLGLVPHQIRGFLSRFDNVLPVSLAFDKCTACSSKVL 654
S. cerevisiae ATG7        LMTSLLQTKYSGSE-------------T-TVLGDIPHQIRGFLHNFSILKLETPAYEHCPACSPKVI 577

At ATG7                   SEYRERGNSFILEAINHPT-YLEDLTGLTELKKAAN---SFNLDWEDDDTDDDDVAVDL      697
Physcomitrella patens     REFREKGVEFILEVLNRPN-YLEDLTGLTELLRITD---DEPLVWDDD--------ADF      678
Homo sapiens ATG7a        DQYEREGFNFLAKVFNSSHSFLEDLTGLTLLHQETQ---AAEIWDMSD--D-----ETI      703
S. cerevisiae ATG7        EAFTDLGWEFVKKALEHPL-YLEEISGLSVIKQEVERLGNDVFEWEDDESD------EIA     630
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Supplemental Figure 1. Alignment of ATG7 proteins from various organisms. *Arabidopsis* (*At*) ATG7 was aligned with the corresponding proteins from moss (*Physcomitrella patens*; XP_001765080.1), humans (*Homo sapiens*; NP_006386.1), and yeast (*S. cerevisiae* ;P38862.1). Amino acid residues identical in at least three sequences are boxed in black or colors; chemically similar residues are boxed in gray. The identified atg7 lesions, the ThiF domain (GXGXXG nucleotide-binding motif highlighted in green), and the active site Cys residue are shown above the alignment.

Supplemental Table 1. Primers Used for *ATG* Sequencing.

Gene	Amplicon Size (bp)	Primer Name (5′ to 3′ Sequence)
ATG2	1089	ATG2-1L (AACCTTCACGTCACCATCCCTTAG) ATG2-2L (TGCAGCATTCATCAACATCTCA)
ATG2	1037	ATG2-3L (GCCAGCACCTCCGCCTCTA) ATG2-4L (GATTGGTAACTCCTCATT)
ATG2	1098	ATG2-5L (TTTCATCTTTCATCACAATCTGGA) ATG2-6L (AATGTGGTAGGGGGTAAACTCAAT)
ATG2	1068	ATG2-7L (GCAGGGGGCAGTGGATTTC) ATG2-8L (AAGATTTGGGACAGACATAAG)
ATG2	1147	ATG2-9L (TTCATTTGCTTCCACTTGCTATCC) ATG2-10L (TTTGCCTGTTGAATGCTGTCC)
ATG2	1195	ATG2-13L (AGGGCCGGGATACAACTTC) ATG2-14L (TGCTTCTCATCTGCCATCATACTA)
ATG2	1119	ATG2-15L (CCAGGTTACGGGTCGCTTTAC) ATG2-16L (AACAATGCCTTCTCAGTGC)
ATG2	1091	ATG2-17L (CTCTAAAGCTGGTCTCGTCAC) ATG2-18L (AAGTCAGGTTTAGTTTCTCAA)
ATG2	900	ATG2-19L (TGCTACGCCCAAAGATCAAGACGAC) ATG2-20L (CTAAAAGAAGATGCAGCTACCAGACAGG)
ATG3	914	ATG3-5 (GTATTACCAAACTCCTCGTG) ATG3-6 (ATGAAGATAATAGACAGTGA)
ATG3	1020	ATG3-7 (CGATACGATAAATTCTTCCTC) ATG3-8 (CAAGTTGAATAAGAGATAAGTGAA)
ATG3	932	ATG3-9 (CTGGCTACACATGGGAAACC) ATG3-10 (GACGATCACAAAAATACTCTACAAG)
ATG7	1139	ATG7-1 (TTGACTCTCCTCCATAATAA) ATG7-2 (TCAGAAGAAACCGAAACCAAAAAG)
ATG7	1191	ATG7-3 (GATTGGCGCGACTCAGA) ATG7-4 (TTAACAAGAACACCGCATCAT)
ATG7	1055	ATG7-5 (GTTGCCATGTCTAATCCAGTCAGG) ATG7-6 (GATTCTGCAGCGCAGTTTTCTAAG)
ATG7	844	ATG7-7 (AGTCGGAGTCCTACAACACC) ATG7-8 (GAATGGAGCTGCGTCAAC)

Supplemental Table 2. PCR-based Markers Used for Mutant Genotyping.

Mutant	Restriction enzyme	Amplicon size (bp)		Primer Name (5′ to 3′ Sequence)
		Wt	mutant	
atg2-1	–	none	200	LB1-SALK (CAAACCAGCGTGGACCGCTTGCTGCAACTC) ATG2-4 (TGCTACGCCCAAAGATCAAGACGAC)
ATG2	–	900	none	ATG2-4 (TGCTACGCCCAAAGATCAAGACGAC) ATG2-5 (CTAAAAGAAGATGCAGCTACCAGACAGG)
atg2-3	*Xmn*I	186	111, 75	ATG2-1 (CTCTTATCTGTGTTCTCTTCTTGTTTCCTAC) ATG2-2 (TTACCAGAAGGAAGAAGCAAAGACC)
atg2-4	*Mbo*II	136, 57	112, 57, 24	ATG2-7 (GAGGAAATTCTGGGTGGTATGATGAC) ATG2-8 (CTAGAATCATGCCAGTCAGAGCCAGAGTA)
atg3-1	*Taq*αI	151	124, 27	ATG3-TaqI* (TTGACACAATGGCTTTTACTTTTGATTCG) ATG3-12 (AATCAGAACTACAAAGCCAACAGAAATAAGC)
atg7-3	–	none	350	LB1-SAIL (GCCTTTTCAGAAATGGATAAATAGCCTTGCTTCC) ATG7-5 (GTTGCCATGTCTAATCCAGTCAGG)
ATG7	–	281	none	ATG7-5 (GTTGCCATGTCTAATCCAGTCAGG) ATG7-23 (GTTGCCATGTCTAATCCAGTCAGG)
atg7-4	*Taq*αI	112, 30	142	ATG7-12 (CATCGTCTTCTTCTTTCTCACATTCAT) ATG7-TaqI* (GTCGACGGAACTGTTCAATGGAGCGAATC)
atg7-5	*Dde*I	146	117, 29	ATG7-11 (CAGGGAACGCAAACCAATACCGAAAACTC) ATG7-DdeI* (TAACAATTTTGTTGGTGAATTTTAGACTT)
atg7-6	*Tsp*45I	107	78, 29	ATG7-10 (CAACCCAATGTACCAGCTCCAAGAAGAAGG) ATG7-Tsp45I* (GTCGATTTAAACTTAAAGTTAATGAGGTG)
atg7-7	*Hin*fI	87	56, 31	ATG7-HinfI* (TCAGTGCCAAGTAGTTTCGAAGTGGCGAG) ATG7-22 (CGAATCAGTGCCAAGTAGTTTCGAAGTGA)
atg7-8	*Bcl*I	250	174, 76	ATG7-21 (TTTGTTAAGCTCCCATCCCACTGA) ATG7-22 (CGAATCAGTGCCAAGTAGTTTCGAAGTGA)
lon2-2	–	none	248	LB1-SALK (CAAACCAGCGTGGACCGCTTGCTGCAACTC) R008 (TTACCATATGGGCCAACACAGTCC)
LON2	–	343	none	LON2-16 (AGTCTTGTTCTCGGTATTGCATTGGGGGTAG) R008 (TTACCATATGGGCCAACACAGTCC)

*The underlined nucleotide in this primer differs from the wild-type sequence to create the indicated restriction site in the mutant or wild-type PCR amplicon.

Supplemental Table 3. PCR-Based Markers Used in Recombination Mapping.

			Amplicon Size (bp)		
Marker	Nearest Gene	Enzyme	Col-0	L*er*	Primer Sequences (or reference)
K16E1	At5g42590	-	166	148	F: GGTGTATCGTCATGTGTCTC R: AACTACTAGTTCATTGTAAA
PDC2	At5g54950	*Bam*HI	360, 65	425	(Konieczny and Ausubel, 1993)
T2E22	At3g12560	-	84	74	F: ATGAGAAGCTATAATTTTTTCAATA R: CTCATATATACAAAGAACTACTATAC
MXL8	At3g21215	-	157	128	F: GTAGCCCAAAGCCGTACAG R: GAGATGCGTTTCACCTACAA
LCS341	At3g23633	*Hinf*I	142, 30	172	F: TTGTGCTTTTGCTTTTTAAAGCCGATT R: AGTTTTTCTTCTTTTTGTATTTATGTG