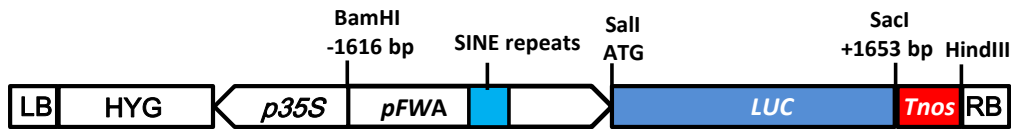


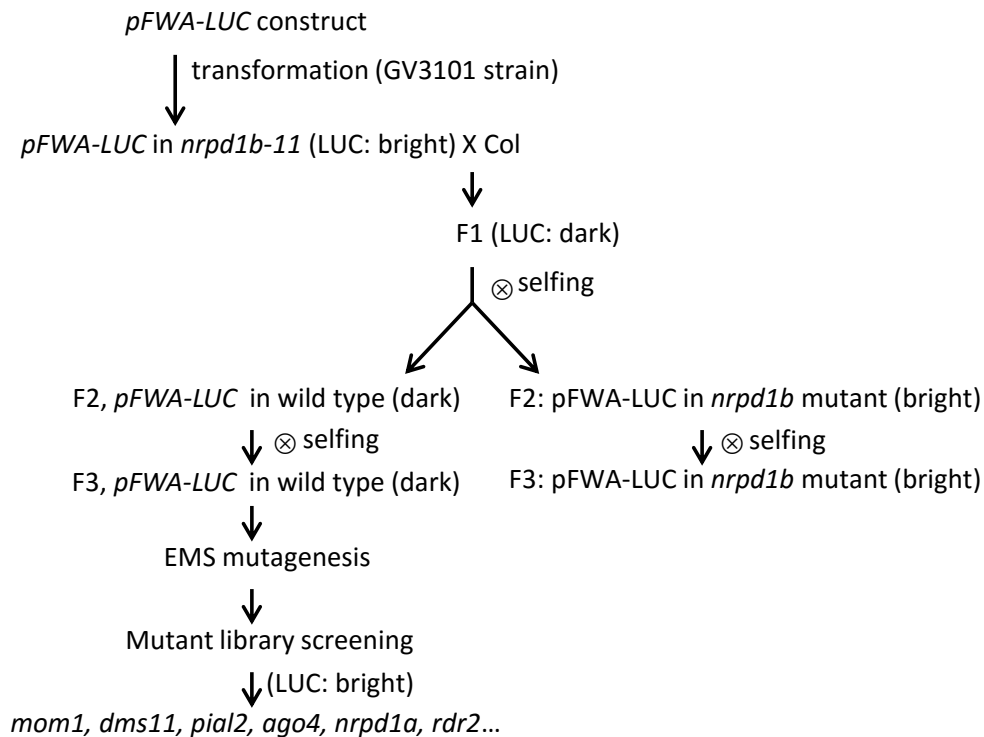
Supplemental Figure 1

A



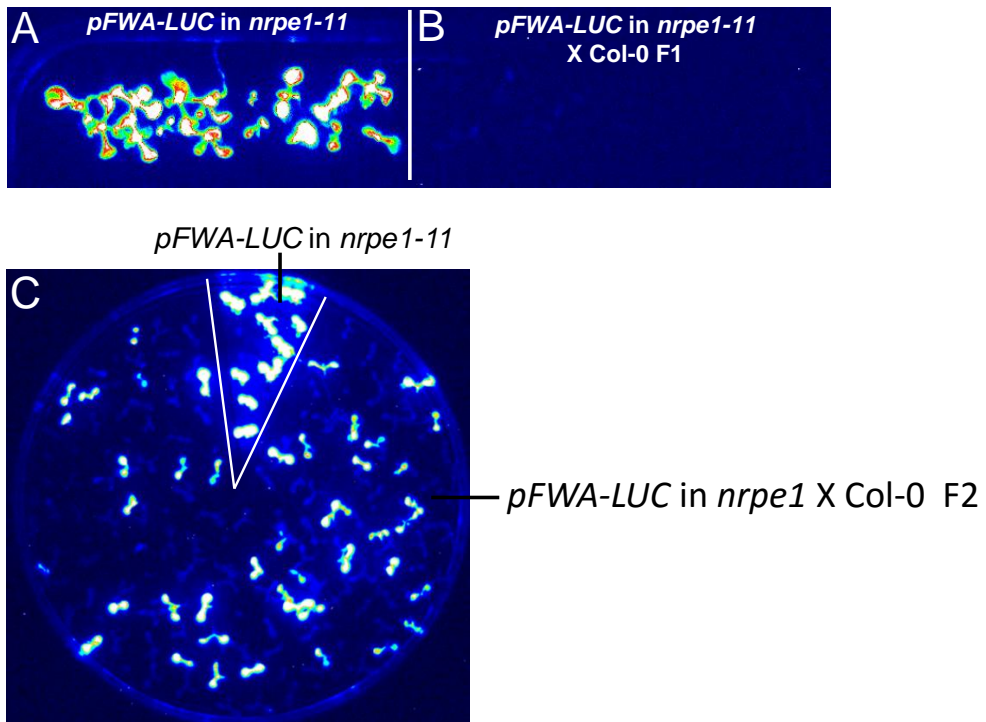
pFWA-BamHI-F: CGGGATCCGCGCCTTTCTCTTCCTCATCTGC
pFWA-Sall-R: ACGCGTCGACCATTTTCCCTCAATGCAATAACCTG

B



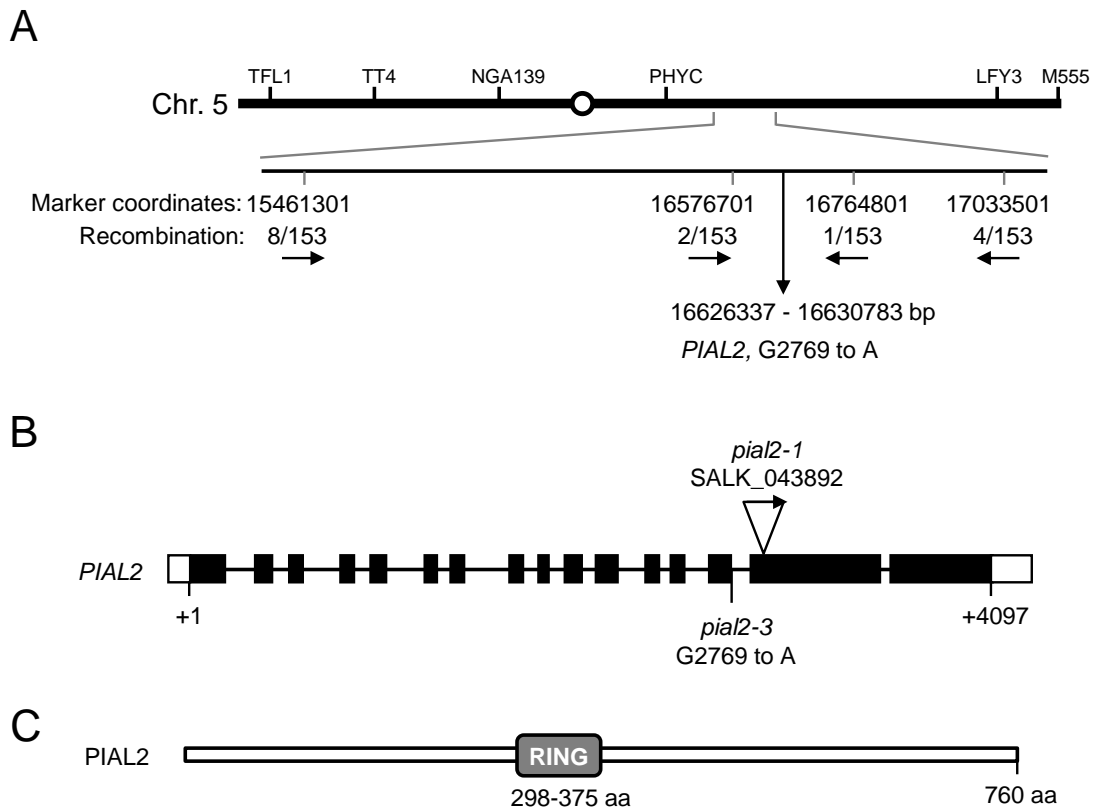
Supplemental Figure 1. The *FWA-LUC* reporter system. **(A)** Schematic representation of the *pFWA-LUC* construct. The primers used to generate the *FWA* promoter sequence are shown. The recognition sites of BamHI and Sall are shown in red in the primer sequences. **(B)** The flow diagram used for mutant library construction and map-based cloning.

Supplemental Figure 2



Supplemental Figure 2. Luminescence imaging of *pFWA-LUC* transgenic plants. **(A)** Luminescence imaging of *pFWA-LUC* transgenic plants in the *nrpe1-11* background. *pFWA-LUC* was transformed into the *nrpe1-11* background and a transgenic line highly expressed the *pFWA* reporter gene. **(B)** The expressed *pFWA-LUC* transgenic line was crossed to the wild-type Col-0 and the *pFWA-LUC* reporter gene was silenced in the F1 plants. **(C)** Luminescence imaging of the F2 plants from the cross of *pFWA-LUC* in *nrpe1* and the wild-type Col-0. The *pFWA-LUC* transgenic plants in the *nrpe1-11* background are shown as a control.

Supplemental Figure 3



Supplemental Figure 3. Map-based cloning and characterization of *PIAL2*. **(A)** Diagram for map-based cloning of *PIAL2*. A ~188-kb region on chromosome 5 was determined by map-based cloning. A mutation was identified by whole-genome DNA deep sequencing at a splice site of *PIAL2* in the ~188-kb region. A black bar with a blank circle represents chromosome 5. The centromere is indicated by the blank circle. Positions of indicated genes and markers are shown on the top of the black bar. **(B)** Schematic representation of the *PIAL1* gene and the mutations in the *pial2-1* and *pial2-3* mutants. Boxes and lines represent exons and introns, respectively. 5' and 3' UTRs are indicated by blank boxes, whereas coding regions are indicated by filled boxes. **(C)** The *PIAL2* protein contains a RING-type zinc finger motif that is conserved in SUMO E3 ligases. The blank bar represents the full-length *PIAL2* proteins. The position and size of the conserved RING-type zinc finger motif are shown.

Supplemental Figure 4

```

Arabidopsis PIAL1      1  MVIPATSR-----FGFR-----AEFNTKEFQASCTSLANEIDAAGRNEVPNGIQELAL
Arabidopsis PIAL2      1  MSTAAARPVAGTGLREKTAASLVNSFRLASVTRQLRYHIQDGAKVDPKPEQICCIIFAKGIDFAIANNDIPKKVIEFPW
Human PIAS2           1  -----
Drosophila SU (VAR) 2-10 1  -----MRKTRSGTARTQAENAATSSSPGHQSTSS

Arabidopsis PIAL1     50  ILNNVCRKRCDDVQTRAVVMALMISVKSACQLGWFPEREIQEELLAIDLMWNGFSCPENVTSCVNSPVTIISQVIERFYP
Arabidopsis PIAL2     81  LKQLCRHGTDVY--TKTALMVLMISSVKHACHLGFSDSESOELIATADEIRTCFGSSGTSSPGKISPGSTFSQIVRFRYP
Human PIAS2           1  -----MADFEELRNVMVSSFRVSELOVLGFPAGRNKSGRKHDLMLRALHLKSG-----CSPAQVIKIRRELYRR
Drosophila SU (VAR) 2-10 30  APIAVNPFDSTRKKECEQMVQMLRVVBLQKILSFLNISFAGRKTDLQSRILSLFRLT-----NLELLAPKVCQEVYAO

Arabidopsis PIAL1     130  QVKLGHILVSEFAKPESEKMMKDFHISKKBHSPKQKVGDFVVRTEILSRSNCTVHPQGVDFLLNGKGDIKRVN--ISME
Arabidopsis PIAL2     160  FVKLGHVLSSEEVKAGYTMLAHDFYISKKNPHSLQEKIRLFVAQTDNIDTSACISNPPVSVFLNGKGVKRVN--IAMD
Human PIAS2           64  RYPTLEGLSDLSTIKSSVFLDGGSPVEEDLA--VAGLHSLPSTSVTPHSPSSPVGSQLLODTKPTFEMQQPSPIPP
Drosophila SU (VAR) 2-10 101  SVOEQNATLQYIDPTR--MYSHIQLPPTVQENP---VGLVSGSQGVVPGGQMNVVGGAPFLHSH--SINSLQIP-----

Arabidopsis PIAL1     208  SQPOLPTNVTALLNLGANLQAIIGCFGGSYLIAIAFMDVIPLPNPKPLLKDYVHPVVGNSDCDIEGPSRISLSCPISR
Arabidopsis PIAL2     238  TGPOLPTNVTALQKYGTNDLQVMNGFKGNYYIIIAFTGLVVPPEKPVLDKDYLSQGVIEASPSDSIDEGPSRVLSCPISR
Human PIAS2           142  VHPDVLKNLFFYDVLVDLVIKPTSLVQSSIQRFQEKFFIFALTPQOVREICISRDFLPGGRDYTVQVQLRLQIAETSCP
Drosophila SU (VAR) 2-10 168  IHPDVLRLKKLAFYDVLGTLIKPSTLVRPNTQRVQEVFPFYFTLTPQATEIASNRDIRNSSKVEHALQVQLRQGLVETSCD

Arabidopsis PIAL1     288  TRIKLPVKGHVCKHLQCFDFNYYNMMNTR-----RHHGAARILEVVGNAADVVISADGTMVMVETE
Arabidopsis PIAL2     318  KRILKPVKGLQCKHLQCFDFSNVYHIMRNPTWRCPHCNQPVCPDIRLDQNAKILKDVHNAADVITDAGGTVKVTKN
Human PIAS2           222  QEDNYPNSLCTKVNKGLPLPGYAPPEKNG-----IDQRKPGRPLNITSLVRLSSAVP
Drosophila SU (VAR) 2-10 248  QEDCFEPNVNKNVNNKLCQLPNIPTNRPN-----VDEKRPVPPVNVNVSVKLSPTVT

Arabidopsis PIAL1     350  NDEDVELVPEITTHDHGDPNSFIN--LGPTVKNPARD--ENEMETSTQVEHNPCLSEIQGGSND---THREASDYTML
Arabidopsis PIAL2     398  TGETPEVPRIIHHDLEDPMSLIN--SGPVVFDLTGDDDALELVFGDNKVEDRKPCMSDAQGGSNNMNTNKHPSNDDYSSI
Human PIAS2           275  NQISISWASLIGKNYSMSVYVLRQLTSAMLLQRLKMGIRNPDHSRALIKRKLTADESEIATTSLRVSLMCLGKMRIT
Drosophila SU (VAR) 2-10 301  NFIITVQWCPDYTRSCLAVYVVKLLTSTLQORMKTKGVKPADYTRGLIKRKLTEADCEIATTMLKVSINCLGLGKMKML

Arabidopsis PIAL1     421  NQS-----HTSNTLPLQPLRTLNAPDGGQFVNLPOVINTRDSEASQALPMTFSPTEPSPQDITLATAANAFGTSMSP
Arabidopsis PIAL2     476  FDISDVLALDPEILSALGNTPAQPHQASNTGTGQQVSNLSQIPIPSIDP--MPVPVFSQTEPSPDRPATSTVFTIPNP
Human PIAS2           355  IPCR-----AVTCTHLQCFDAALYLQMNKKTWICF-----VCDKKAAYESLILDGLFMEILND
Drosophila SU (VAR) 2-10 381  LPCR-----ASTCSHLQCFDASLYLQMNKKTWICF-----VCDKPAIYDNLVIDGYFQEVGLS

Arabidopsis PIAL1     490  AAQSSQFGSHVTSIGNCEGRSDLMARNHHYGRVQTFPPAPLSHHHYSMQNSPSPAQQRPVPSYIAHPQTFHVNYG
Arabidopsis PIAL2     553  SPQYSOVHASPVTPPTCYLGRITS--PRNNTQTY--QSQAP--PMTTPYTSRKYVSVPTVSQS-----
Human PIAS2           409  --CSDVDEIKFQEDG--SACP--MRPKKEMKVSSQPCTKIESSS-----
Drosophila SU (VAR) 2-10 436  SLLKSDDETIQLHQDQ-----SMTSPGLRSETCILDTESKPAKQVEVISDDIELIS-----

Arabidopsis PIAL1     570  ENADQRWMPSSIAHPQTLFVNYGGNTNQRPIPSISIAHPQTLFVNYRGNTDHRSTPYSITHLQTLNLNYGGNADQRMPSSI
Arabidopsis PIAL2     607  -----PANVSVFVQSQHVRVLSQPN-----NYG-----
Human PIAS2           448  -----VLSKPCSVTVASEASKKVDVIDIT-----
Drosophila SU (VAR) 2-10 486  -----DDAKPVKRDLSPAQDEQPTSTSNSETVIDIT-----

Arabidopsis PIAL1     650  TNLQTLPATYGGYAHQPEMSSSITHPRTSPVNYGGTDPDRMPSSIITHPQTLFVSYGGTTDQILNPGGAMGQFSSREFMN
Arabidopsis PIAL2     631  --VRGLTSSHASTSRQEPESGPTVQS-----VSRSLDLVDVD
Human PIAS2           473  --IESSSD--EEEDFPAKRKICF-----MSETCS-----
Drosophila SU (VAR) 2-10 516  --LSDSDDDMLAKRREPAKQAVAS-----STSNSSGGGQR

Arabidopsis PIAL1     730  LTPANTENWRPQSRMRGSAVPG---TGYDHMIHETRFPVHPQAQT--PPAPLSTSYDGADEIQAFIGHPSYVPSNNETQAG
Arabidopsis PIAL2     666  LTVPDTSNWRP--RMRGSLVPGSHSTALDHMIHRSQQSQTSIRLNSQCFVQTPSVQTSQAQSPFTTAAAYRT---ETVLG
Human PIAS2           498  -----SPTKGVLMYQESVSRVPSVSVDPAAIPESLTDY--SVPFHHTPEISSMSDLPGEVRRNDINNELKLGTSSTDV
Drosophila SU (VAR) 2-10 551  AYTPAQQFQSESPEQAASRQSEPKQTVSEQQLOQQQHEQPAATAAVHASLLESIAAAVADQRHFQLLDLAAVAATAATA

Arabidopsis PIAL1     806  TSSLVPAEGLGYSGSFWSMPPETW
Arabidopsis PIAL2     741  NRNHVPVAPP---GIVRPTGPTS-
Human PIAS2           571  QQ-----
Drosophila SU (VAR) 2-10 631  SSGSQNAGP-----

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Supplemental Figure 4. Alignment of PIAL1 and PIAL2 in *Arabidopsis*, PIAS2 in human, and SU(VAR)2-10 in *Drosophila*. Black and gray boxes indicate that the residues are highly and weakly conserved, respectively.

Supplemental Figure 5

Aravidopsis thaliana 1 -----MVI PATSRFGFR-----AEFNTKEQASCTSLANEID
Arabidopsis thaliana 1 -----MSTAAARPVAGTGLRKTASLVNSFRLASVTRQLRYHIQDG-----AKVDKPEFQICQISEAKGID
Glycine max 1 -----MNSLPAFT--SDTSPSVVNDFRNKVADRLSWIAQDFG-----NRGPEYEFYINCLSLSRGID
Brassica napus 1 -----MSSASTAVRPVAGTGLPEKAAALVNSFRLASVTRQLAFHIQDG-----AKSDVKKEFQSCCTSLAKGID
Vitis vinifera 1 -----MTGATISLPTSVSNIAGVETTSYSASLANFRLNAVLERLAMVRSRHRILDQGRSTEEPHNCLSLARGID
Populus trichocarpa 1 ---MVGMMAGTMPPPPQVAEGVTIRAAGQOOSASLANFRRHVAVRLSLEHIQDG-----FDS--NPTTEFCLCLSLARGID
Oryza sativa 1 MASAAPLP--PTPPSPQQQPPQPGQKQAVAMNARRLVMIGRLRTRFRGGG---GTVLEPPDLAHVYAFARGID
Sorghum bicolor 1 MATAAATAPHTPPTFLQQQHQDQKQKQKALSMNAIRLKAIGRLKAHLRG-----MNVFPIAEFAHVVYAFARGID

Aravidopsis thaliana 33 AATGRNVEFGNIQELALILNNVCRKRCDFYQTRAVVMALMISVKSACQLGWFPERETCELLALIIDLMWNGSCPENVTSC
Arabidopsis thaliana 64 FAIANNIDPKKVEEFPFLIKQLCR-HGTDVYTKTALMVLMSVKKACHLIGWFSDESECELLALADEIRTCGSSGSTSPG
Glycine max 57 YALANGETPKKAELEPLLVKQLCO-LKNDECSQAAMVLMISLKNACEIGWFTKSEELVTTADPIRKYVSSSLGTINVG
Brassica napus 65 FAIANNIDPKKVEDLPSLLKQVCR-HRDDVYTKTAVLVMISLKHACKLIGWFSDESECELLALADOMKNGGNPENTIPV
Vitis vinifera 75 YSLANGVDPARVQDLPLLLKQLCO-RRNDLFLGGIMVLMVSVKNACKVWGFTEKDTTELLTLVMEIGSNFCNLGDNNTE
Populus trichocarpa 73 FAVANNIDLLKAQELPFLKQVCO-RKNDFVLQAAIMVLMASVKNACVWGFQKPECELLVTLAIEIGKVECTPPGIDNAG
Oryza sativa 76 FALSSGVDFTVASEIPSLIKRVYL-VGKQDFLQSSVMVLMISLKNACESEKWFQPTDCTE-LTVLEPPDLAHVYAFARGID
Sorghum bicolor 76 FAVSAGDIPQMGADIEDILRRVYD-LRREMFIQSSLMVLMISLKNACSNWFQPADSRDIFRMANELSGNFCSTSTGQAAS

Aravidopsis thaliana 113 VNSPVTILSQVIRFRYFCVKLGHILVFSFAKPEKMMKDFHISKMPHSPKQKVGIFVVRHEDLSRNCIIVHPQGVSL
Arabidopsis thaliana 143 IKSPGSTFSCIMERFYFVKCHVLSVSVKAGYMLAHDFYLSKNMPSLQEKIRLFAVQTDNIDTSACISNPPVSVFL
Glycine max 136 PRSCSTALSTIMQKFFPKFKLGPILLASDAQSGY GASVDFHITKS--EVLKDKIFLVAQTDNIDTSACILNPPQVNF
Brassica napus 144 IQSPGGTILSCIMERFYPLVKLGHVLSVSVKSGYMLAHDFHISKMPHSPQEKIRLFAVQTDNIDTSACINPPVSVFL
Vitis vinifera 154 PISFHPITSKIMARFYPRMENGQILASRVKGGYGTFLVDFHISKSTKSSQEKIRLFAVQTDNIDTSACILNPPVSVFL
Populus trichocarpa 152 TTDLSLITPTIMSRFYPLMKVQQLIVSLVWKGEGGAHVDFHISKTRNSDEKLI-----
Oryza sativa 155 DSTVIQIISTIMERFYPLKFERLVTSLEAKVGYDVLMAOFFTHKN--VPREKINLIVWQKEDLNASSCIANPPVSVFL
Sorghum bicolor 155 DSTVLELISCIIMERYPRKFERLITSLBAKVGYDILMAOFFTERN--ISRDEKIRLIVWQKEDLNASSCVSSPPVSVFL

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Arabidopsis thaliana 223 LNKGVKRRNIAMDGQPLPTNVTAQLKLYGNLLOVGMNFKGNYLIIIAFTGLVVEEKPVLKDYLLQSGVIEASDSDI
Glycine max 214 LNKGVNLRNIVQMDGQGVPTNVTMLKLGGNLLOAQVGNFRGYVVLVAVMSPTTPELDVLYQDYLPQAVTSDVSDSI
Brassica napus 224 LNKGVKRRKNISMSGQPLPTNVATLNLKYGNTLLOVGMNSNGHYIIVIAFTGLAQIPKPEVLPKDYVQSGVIEASDSDI
Vitis vinifera 234 LNKGVERRNIVVMSGQPIPTNVTMLKLYGNLLOAQVGNFRNGHYIIAAARMVAVISEPNDVLYQDYVQPAWMLHSNDEI
Populus trichocarpa 206 -----DTGQMPNTNVTMLKLYGNLLOAQVGFK-----AAAFMSVEPKPETPVLYQDYVHPCAEALDP-----
Oryza sativa 233 VNKGVDKRKNIVSMETGQVPTDITITMLKYGNITQAGYGFNANYIIAAAFINKLESFADAPLNDOYAQP--VAADPPDSDI
Sorghum bicolor 233 VNKGVDRRKNIVSMREGQVPTDITITMLKYGNITQAVGFNANYIIAAAFVNNLTSFSAKPLDOYAQP--ITVYPADSDV

Aravidopsis thaliana 273 IEGPSRISLNCPIISRTRIKLVPVKGHCKHLQCFDFWYVNMNR-----RHGGAARILIEEVGRNAA
Arabidopsis thaliana 303 IEGPSRISLNCPIISRTRIKLVPVKGHCKHLQCFDFSNVYVHINMRNFTWRCEHCNQPVCYEDLRLDQNLKLVHNA
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Brassica napus 304 IEGPSRISLNCPIISRTRIKLVPVKGHCKHLQCFDFWYVNMNRNBSWRCEHCNQPVSYPEIRLDQNM--ILKDAQRNAA
Vitis vinifera 314 IEGPSRISLNCPIISRTRIKVVPVKGHCKHLQCFDFNFEVINSRPSWRCEHCNQPVCYEDLRDRDNVVKLKEVGENVA
Populus trichocarpa 263 -----ATHSELLSKRGIHADIFS-----MSATQIFELIKLTLRQV---VG
Oryza sativa 312 IEGPSRISLNCPIISFRTRIKLHKGRCCKHLCQCFDFWYVNMNLRKFTWRCEHCNQPVSYPEIRLDQNM--ILKDAQRNAA
Sorghum bicolor 312 IEGPSRISLNCPIISFDVELIFSLPSLVAVLCQCFDFWYVNMNMRKFNWRCEHCNQPVSYPEIRLDQNM--ILKDAQRNAA

Aravidopsis thaliana 335 DVVLSADGTWVETENDEDELVLP-ETHDHDGPNNSFIN-----LGEFVKNPARDE-----NEMETSTQVEPHNFCI-
Arabidopsis thaliana 383 DVVLSADGTTWVETKNTGETPEPVR-EIHDLEDPMSSLN-----SGFVVDLTKDD-----DAELVEFG-DNKVEDRKPFCM-
Glycine max 374 EIVLVLANGSWKAVLEKHDHDKMOKKARANCEKEQTPQESTCP---EGTVDLTKDD-----DGLDVTGSCDIVERKPTP-
Brassica napus 382 DVVLSADGTTWVETKNTGETPEPVRDAIIEHDLEDPMSSLN-----AGFVVDLTKDDDEDDADIELFGSTSKAVPCKPHL-
Vitis vinifera 394 DVVLSADGSKWALLESNDHVDQPRVGTLSNQQKGPDLQGSTSFNSASPNVADLTKED---DEMNAFADCEIEDRKPFO-
Populus trichocarpa 301 DVVLSADGSLKALSESNDKVDQERTLHCEKGMPEQVESMTSTRALPMVVDLTKVDD---DEINGEDNIDAEDRKPFL-
Oryza sativa 392 DVVLFADGSKWALSTNDERSDRHSDVLIQSRDMDTDA-----TADDVLDLINED---NDGDVPMSPFSASDVKPFLN
Sorghum bicolor 392 DVVLFADGSKWAAAPADEKSRHRGDAIQOTCSTETDS-----PSSDVIDLNGN---DDGDMQMDVNSAPEDIKPFLN

Aravidopsis thaliana 400 ---SEIQGPSND---THRPAVDYTMNLSHTSTNTPQLPRTL-----NAFDGQGFVNLQPVIN
Arabidopsis thaliana 451 ---SDAQGSNNNNTNKHPSNDYSSIFDISVIALDEPILSALG-----NTAPQHQASNTGCGQYNSNLQSTEM
Glycine max 444 ---ASIHQSQVTPN-----STSLGMNSTGVNQVN-VATQIDDFWPG-----VCFVRSRSDPTVNGSLEP
Brassica napus 454 ---SDAQGSNNNANKASVDLYCSMFNFSVVISLDELVMDLH-----NTGTGQDYNSLQVPEM
Vitis vinifera 468 ---SNIQGHSITTK---QTMAPELNNATEVYNNAVSRVQDGFCSGILLSTYGSSTHSARSDAQFIGGTSQPSPANFLPPEV
Populus trichocarpa 375 ---ATLQNHVDTNPIPTMPSQLINANASRN-FSTLADEFWS---SPYWS---SSASDAQMVNGFSEPTTTFMTSEV
Oryza sativa 464 CQDLSVADYLSLPLMNTVSQAEDLYAGGASRGNNERGNATSTSGQ-----NSSLPSTGGLGSSSFTGLES
Sorghum bicolor 464 SQDLSVSDYLTDLPM--AVQTCGLYRG---DGNNGGSNMAFTSRQ-----NLLLPPTSGGLGSSSFTGLES

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Aravidopsis thaliana 454 TRDSFASQALPMTFSPPTSPQDILATNAANFGTSMPPAAQSSQFQGSHTVSLGNCCEGRITSDLMARWNHLYGRVYQTFQPPAP
Aravidopsis thaliana 520 SIDP---MPVVPVFFSQTPSPRDRPATTSTVFTIIPNPSFPQYSQVHASEVPTGTYLGRRTSPRWNTYQS---QAPMPTTP
Glycine max 501 LFDIVSPTFSQESAGHDNPN--VVNSAMHNQFLGPNLQ-MQMNHMNSVN--EYGRSSAPRHHITFVAVQALFVQSQAL
Brassica napus 512 PRDP---AHVPAPFSQAPSPRERPAATSTVFPSS-----SRVHASEVPTAGTYLRSSTSS-----QMSLTTTS
Vitis vinifera 544 LITDAISPALNRGTEDIRGNTHLTTSLHDQLPDPDSLQQLQAQFGHSTVSNHEYGRFPTIPRHHITRTPFAVQALPAQTQTS
Populus trichocarpa 445 ITDVSVPALNCDVGGYGNTT--TSSVMHNQLSASIYLQSLQONFVNSLANGEYGTLPPIY-HVDRSEPLAVQALPAPRQPTP
Oryza sativa 529 LLPHNLHLPVITDAVSPSLDTSNSVVLRHQVHAQGTTRSDIVFSPQPRIDPQLRLEIARPPIPRVNAVEPTGTCQALFVQPOQ--
Sorghum bicolor 524 LLPQNVLRPVITDAVSPSLETSTSTSGMQHVSHQETHCGTVQLQAQIGPVHGESELRLLRPLIPRNPREFVGVQALAVPPQNP

Aravidopsis thaliana 534 LSHHHYSMQN-----QSPSPAQQRPVPSYIAHPTTFHVNYGENADQRWMPSSIAHPQTLFVNYGGNT
Aravidopsis thaliana 594 YTSRKVSVPV-----TSQSPANV-----SSFVQSQHVPR-----
Glycine max 577 GPQONSITNLNSSLPSNSSATPHISLNSPTSVDTLNAILSDTERQCHFSRTPMNLPOVSGVNSPAFQHTATQNRGLPI
Brassica napus 570 SQRVHVPVQV-----TSQSLGNG-----SSLAQSPRIPR-----
Vitis vinifera 624 GPHHRSTT-----LISMVP--NGPNIVGSDMERFQQFSRSIFNPVQISDISASALQHSMSQNNWNOV
Populus trichocarpa 522 APQQRSTRPN--PAISSGASLSSHGTLPAA--NGLSPVSGNMDRQQQFARSLNTNSSSSQNNWMDHPFMHQSSAQQQA
Oryza sativa 606 ---RVRPNLYN-----CPPFPQSSP-ASAYQVHOVTNADSVITAMSTGIGLSLRAPDAAPLLQHQST
Sorghum bicolor 604 GSSTRLQPNILN-----CPPPIPLSSPSSSTYQTHOVTNPDSVIAPMNSGSGPLPRT-----

Aravidopsis thaliana 596 NQRPIPSSIAHPQTLFVNYRGNTDHRSTPYSITHLQTLNLYGGNADQRPMPSSTINLQTLPATYGGYAHQRPMSSTIHP
Aravidopsis thaliana 622 ---VLS-CPNNYG---VRLTIS-----
Glycine max 657 NTSAPTQPNQ--YRANAFSEFRNLHLQALNLRPPPPRSSNAQWPRIQQGVQSGNFGCAAARG-ASVAAGQGS SHARN
Brassica napus 598 ---VLABCPNSYF---ARSLNS-----
Vitis vinifera 686 AGHPTTSQRPGPGAYRTSSGLPTEPQLQQQ---QSPQARTHNSNLLRSSAHHSRSVQVCGGAQGRATHAVGTGISQNAQ
Populus trichocarpa 598 VTLPSSSQLAG--AHRASS-----PNLLYQQP-LRVPQSRSHSPNVVRSLSLFLAPAQTCGAAQVGVGNSAGATNSQQSR
Oryza sativa 666 QQEIRATQNYH-----QQQFGLTAPQNFMGTRPPPGVPGQAIGANAHGAPPQQSSHVHRLVSNLNMN-
Sorghum bicolor 655 ---AASFPAASDSL-----

Aravidopsis thaliana 676 RTS-----PVNYGTPDORPMPSSITHPQTLFVSYGG-----TTDQILNPGGAMQFSSREFMNLTP
Aravidopsis thaliana 637 ---SHASTSRQHS-----GPTVQVSR-----LSDLVDVLDIV
Glycine max 732 ---V-PTSGATTHSHQARGMVANQPAP--PSVLVQNSITVAGTPFHG-----LTTEQRGNTA---QSVSRPEELFSS
Brassica napus 614 ---NHLTTQTORP-----SPPVLSVSR-----TSDLMDVDSAT
Vitis vinifera 762 ---P-MVA-----AQRAAQTRMP--LPVQNTSRTGSAFPVNANGGRSTAGEQRGNIEGMVQAVSRPELVDL
Populus trichocarpa 669 ---L-MVA-----AQLAAQRARQPPSPVPVQIQTSAGASYLTSADGIRAPATQRGNAGGALPAVSGTEGLVDL
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Sorghum bicolor 668 -----

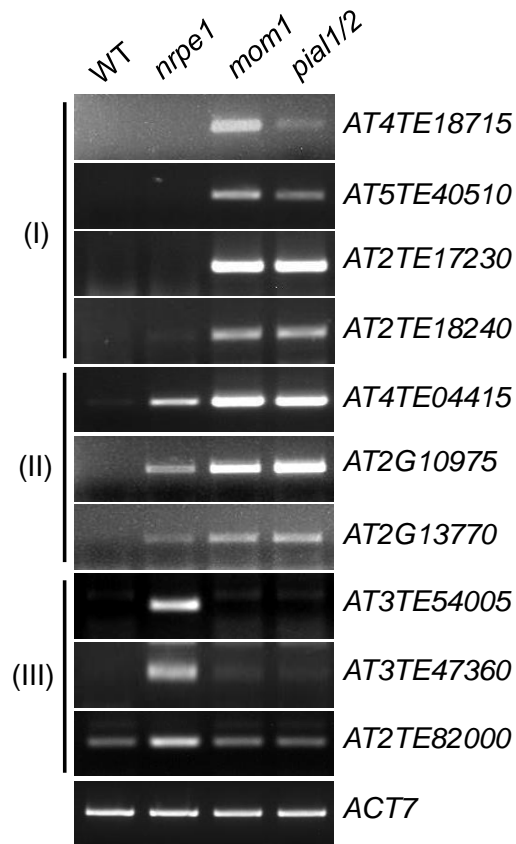
Aravidopsis thaliana 733 ANTBNWRPQSRMRGSAVPG--TGYDHMLIHPTRPVHPQAQTPPAPLSTSYDGADETCAFIGHPSYPVSNNETQAGTSSL
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Glycine max 796 QSEONWRPRTGRMRGSHDLSQLNDESIARIIITPTQG-QNSRPPGPQPIRRTGIS-SLOPATT-QQDVLIANNRNANAHNR
Brassica napus 646 PDTSNWRP--RMRGSHTPGSYSPALDHMIIRPTQ---QSQ--TRLQVSPGQTPPVQTSQALPPFST-----
Vitis vinifera 826 ASEONWRPTGLMRGSHVG-RAYNSALNQLVIOPTQPTQSTRPPTPTSPPPGFPFHICALLTNIRTPLVQAPNYPMTQP
Populus trichocarpa 735 ASEONWRPTGRMRGSHSG-RAYSAALEFVQPTQQTQTPRPPPNLPPSQSSMPPHICFLFA--RNAQVQQAQSSPVTGS
Oryza sativa 788 PRAQSPFLPATAPPPSTPPPIGTSDDLQELPVDESWRPTGMRGSLTGEAYSVAIGRYNPSVNIAGQQTSHVTSQARPAG
Sorghum bicolor 668 -----

Aravidopsis thaliana 810 PVAEGLGYSGSFWSMPPETW--
Aravidopsis thaliana 743 NHPVPAPPGIVRPTGPTS----
Glycine max 873 SSSR-----
Brassica napus 705 ----APPTFTRPSGPTAPWGI
Vitis vinifera 905 ASTTGSGGILPERSLGLH----
Populus trichocarpa 812 AISNGSSSILP-----
Oryza sativa 868 PDARR-----
Sorghum bicolor 668 -----

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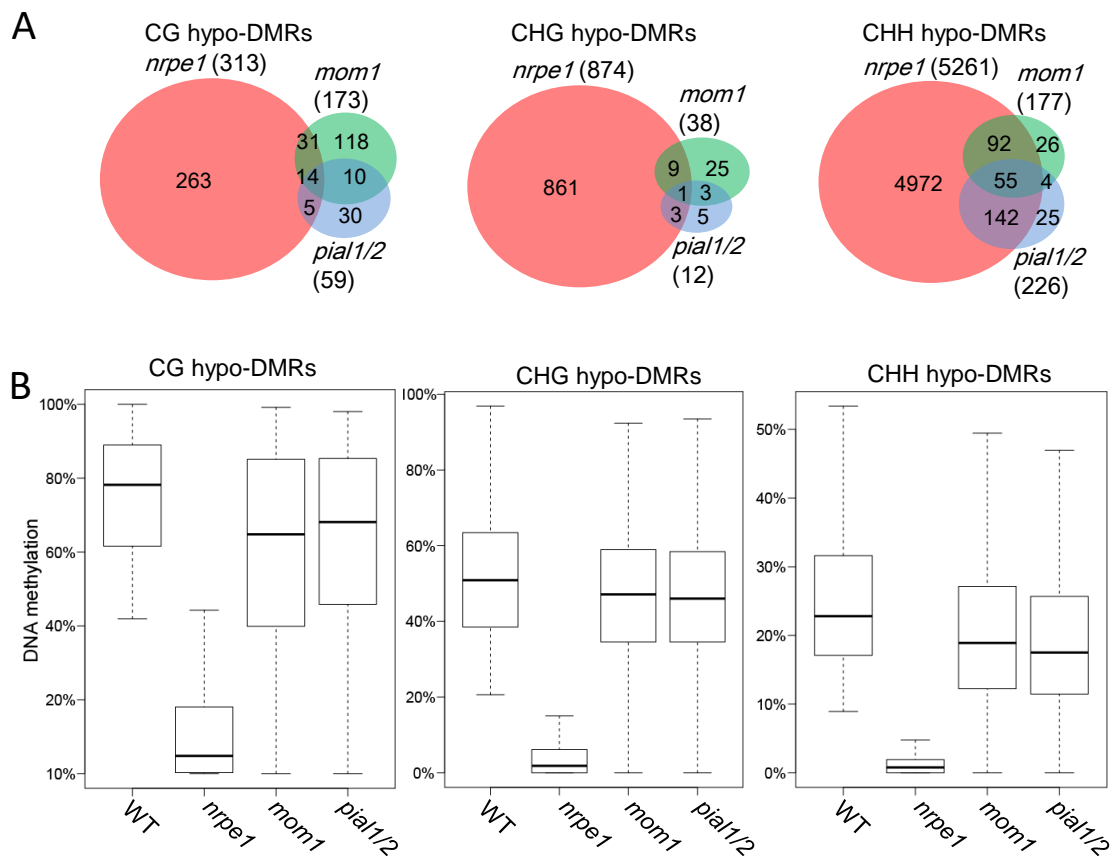
Supplemental Figure 5. Alignment of Arabidopsis PIAL1 and PIAL2 and their homologs in other plants including *Glycine max*, *Brassica napus*, *Vitis vinifera*, *Populus trichocarpa*, *Oryza sativa*, and *Sorghum bicolor*. Black and gray boxes indicate that the residues are highly and weakly conserved, respectively.

Supplemental Figure 6



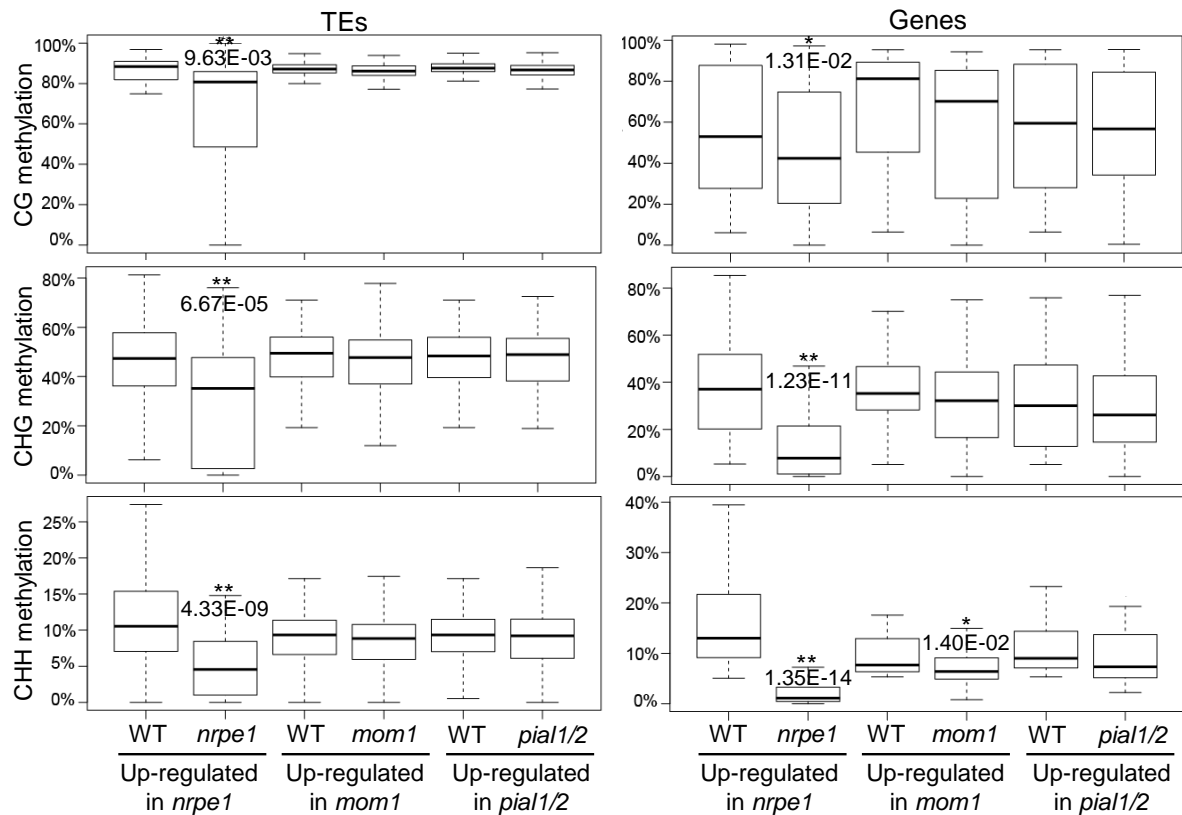
Supplemental Figure 6. Validation of the transcript levels of the MOM1, PIAL1/2, and/or NPRED1 target loci in the wild type, *nrpe1*, *mom1*, and *pial1/2*. Indicated loci were randomly selected from up-regulated loci identified by the RNA deep sequencing analyses. The transcript levels of the indicated loci were determined by RT-PCR. The loci that are transcriptionally up-regulated in *nrpe1*, *mom1*, and/or *pial1/2* were placed into three classes based on the transcript patterns of the loci. I, loci up-regulated in *mom1* and *pial1/2* but not in *nrpe1*; II, loci up-regulated in *nrpe1*, *mom1*, and *pial1/2*; III, loci up-regulated in *nrpe1* but not in *mom1* or *pial1/2*. The actin gene *ACT7* was used as an internal control. The RT-PCR experiment was biologically repeated for at least two times and the result of a representative experiment is shown.

Supplemental Figure 7



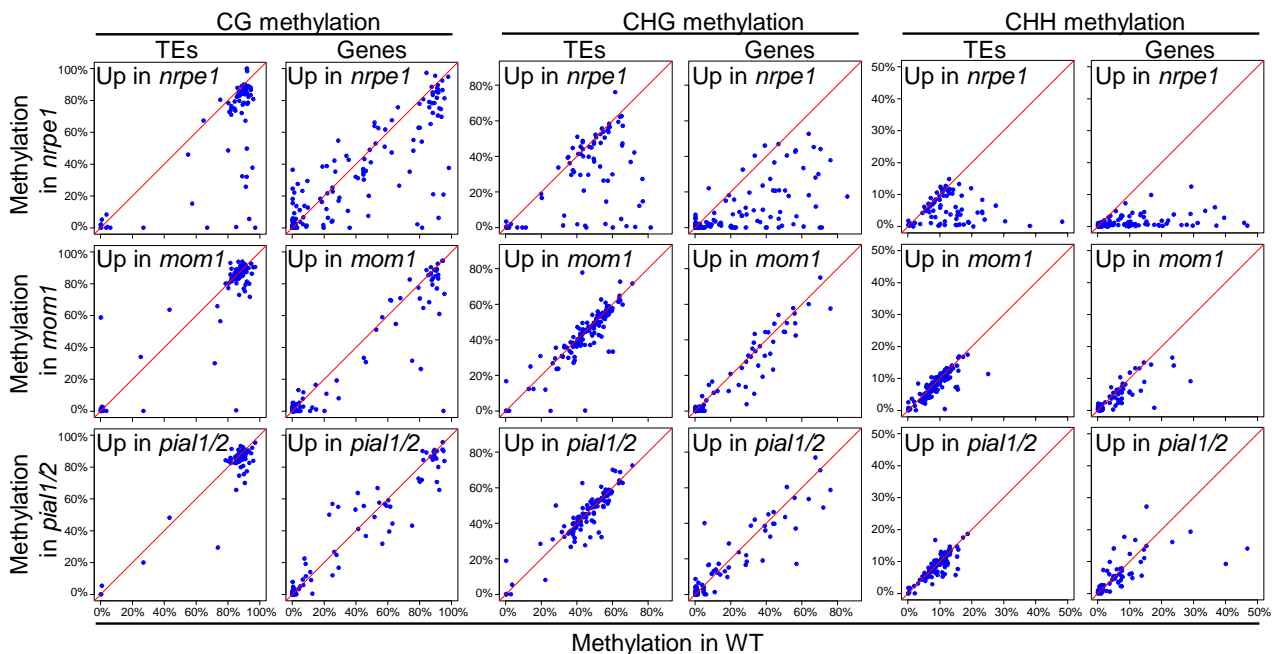
Supplemental Figure 7. Analyses of hypo-DMRs in *nrpe1*, *mom1*, and *pial1/2*. **(A)** Overlaps of hypo-DMRs in *nrpe1*, *mom1*, and *pial1/2* relative to the wild type at CG, CHG, and CHH sites. Hypo-DMRs were separately defined based on the DNA methylation levels of CG, CHG, and CHH sites in the mutants relative to the wild type. Bins were recognized as DMRs when their absolute DNA methylation change is more than 40%, 20%, and 10% for CG, CHG, and CHH, respectively. **(B)** DNA methylation levels of hypo-DMRs in the wild type, *nrpe1*, *mom1*, and *pial1/2*. CG, CHG, and CHH hypo-DMRs in *nrpe1* were separately analyzed.

Supplemental Figure 8



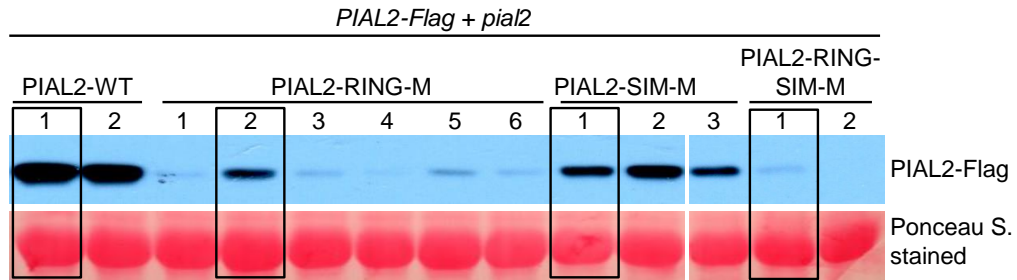
Supplemental Figure 8. Box plots showing CG, CHG, and CHH methylation of transcriptionally up-regulated TEs and genes in *nrpe1*, *mom1*, and *pial1/2* relative to the wild type. * $p < 0.05$ or ** $p < 0.01$ was determined by t test. In the box plots, gene promoters were included for DNA methylation analysis only when their DNA methylation levels in the wild type were higher than 5%.

Supplemental Figure 9



Supplemental Figure 9. Scatter plots showing CG, CHG, and CHH methylation of transcriptionally up-regulated TEs or genes in *nrpe1*, *mom1*, and *pial1/2* relative to the wild type. The DNA methylation levels of TEs and genes that are transcriptionally up-regulated in the mutants were analyzed. Diagonal red lines indicate that the DNA methylation levels of mutants are the same as those of the wild-type.

Supplemental Figure 10



Supplemental Figure 10. The expression levels of the wild-type and mutated *PIAL2* transgenes in the *pial2* mutant. The expression levels were determined by immunoblot assays with anti-Flag antibody. Rubisco proteins stained by Ponceau S. are shown as a loading control. Boxed transgenic lines were used for complementation assays by quantitative RT-PCR and the results are shown in Figure 6C. The expression of *PIAL2-Flag* in the boxed transgenic lines is also shown in Figure 6B.

Supplemental Table 1. Full list of mutants identified by map-based cloning in this study.

Mutant No.	Mutant gene	Mutation position related to ATG	Mutation	Code change	Mutation type
84-3	<i>PIAL2</i>	2769	C→T		splice site
4-1	<i>MORC6</i>	548	G→A	TGG→TGA	premature stop codon
22-1-1	<i>MORC6</i>	3049	G→T	GAG→TAG	premature stop codon
59-1	<i>MORC6</i>	3031	C→T	CAG→TAG	premature stop codon
138-2	<i>MORC6</i>	4123	C→T	CAA→TAA	premature stop codon
215-3	<i>MORC6</i>	2904	G→A		splice site
249-1	<i>MORC6</i>	3170	G→A	GGA→GAA	G→E
250-1	<i>MORC6</i>	3169	G→A	GGA→AGA	G→R
200-1	<i>MORC6</i>	315	G→A		splice site
294-1	<i>MORC6</i>	1800	C→T	GCT→GTT	A→V
31-2	<i>MOM1</i>	4782	G→A		splice site
38-1	<i>MOM1</i>	400	C→T	CGA→TGA	premature stop codon
270-2	<i>MOM1</i>	1036	C→T	CAA→TAA	premature stop codon
310-9	<i>MOM1</i>	2350	G		deletion
13-2	<i>MOM1</i>	6181	G→A		splice site
241-1	<i>MOM1</i>	3007	G→A	GAA→AAA	E→K
78-2	<i>AGO4</i>	2457	C→T		splice site
91-1	<i>AGO4</i>	4213	C→T		splice site
139-3	<i>AGO4</i>	469	C→T	CAA→TAA	premature stop codon
136-3	<i>AGO4</i>	3256	G→A		splice site
62-1	<i>AGO4</i>	2555	G→A	GGA→GAA	G→E
227-2	<i>AGO4</i>	3893	G→A	TGG→TGA	premature stop codon
310-17	<i>AGO4</i>	3370	G→A	TGG→TGA	premature stop codon
190-1	<i>AGO4</i>	3785	G→A		splice site
24-1	<i>AGO4</i>	199	C→T	CAA→TAA	premature stop codon
144-1	<i>AGO4</i>	3148	G→A	GAG→AAG	E→K
178-1	<i>AGO4</i>	1789	G→A	TGC→TAC	C→Y
324-2	<i>AGO4</i>	3199	G→A	GGG→AGG	G→R
131-1	<i>NRPD1</i>	2839	C→T	CAG→TAG	premature stop codon
45-1	<i>NRPD1</i>	3412	C→T		splice site
120-3	<i>NRPD1</i>	3169	C→T	CAG→TAG	premature stop codon
200-2	<i>NRPD1</i>	2752	C→T	CAA→TAA	premature stop codon
268-2	<i>NRPD1</i>	225	G→A	GGA→AGA	G→R
95-1	<i>NRPD1</i>	1828	C→T	CCT→TCT	P→W
40-1	<i>DRD1</i>	2014	C→T	CCA→TCA	P→S
226-1	<i>DRD1</i>	3127	G→A	GAA→AAA	E→K
231-1	<i>DRD1</i>	3209	G→A	GGG→GAG	G→E
272-5	<i>RDR2</i>	2548	C→T	CAG→TAG	premature stop codon
33-1	<i>RDR2</i>	3120	C→T	CTT→TTT	L→F
218-2	<i>NRPD4</i>	199,200	CC→GG		splice site
139-2	<i>RDM4</i>	49	C→T	CCT→TCT	P→S
17-4	<i>NRPD2</i>	4022	G→A	GGG→AGG	G→R
25-1	<i>MET1</i>	4596	C→T	CCT→CTT	P→L
218-1	<i>MET1</i>	3016	G→A	GAG→AAG	E→K
28-6	<i>MET1</i>	4557	G→A	CGA→CAA	R→Q
68-2	<i>FPGS1</i>	3803	G→A	TGG→TGA	premature stop codon
25-9	<i>FPGS1</i>	3403	G→A	TGG→TGA	premature stop codon
104-1	<i>FAS1</i>	455	G→A	TGG→TAG	premature stop codon
112-2	<i>BRU1</i>	5091	G→A		splice site