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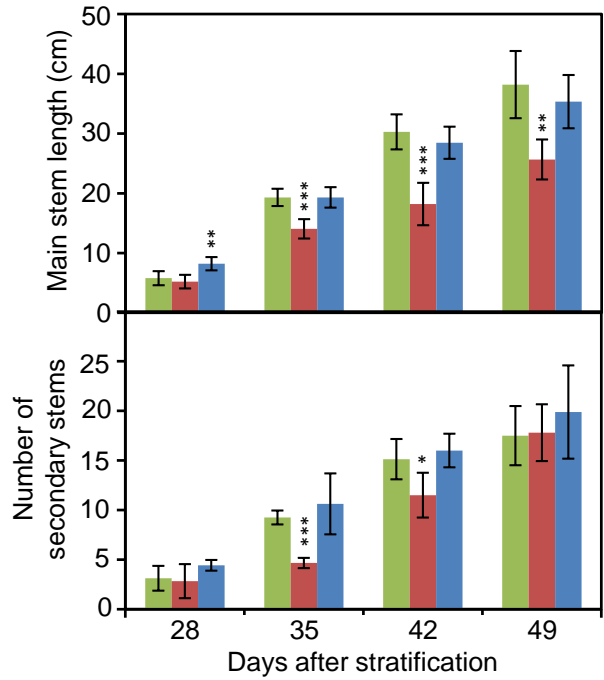
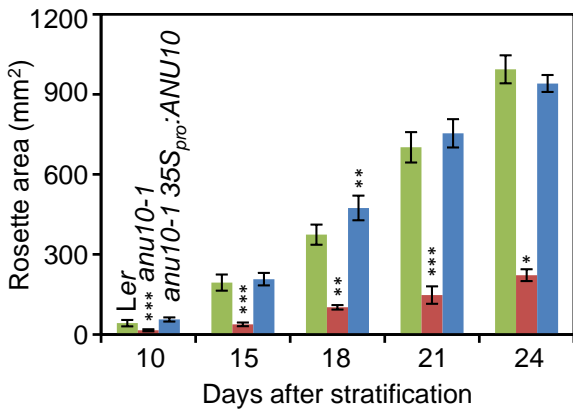
**Arabidopsis ANGULATA10 is required for
thylakoid biogenesis and mesophyll development**

Rubén Casanova-Sáez, Eduardo Mateo-Bonmatí, Saijaliisa Kangasjärvi, Héctor Candela
and José Luis Micol

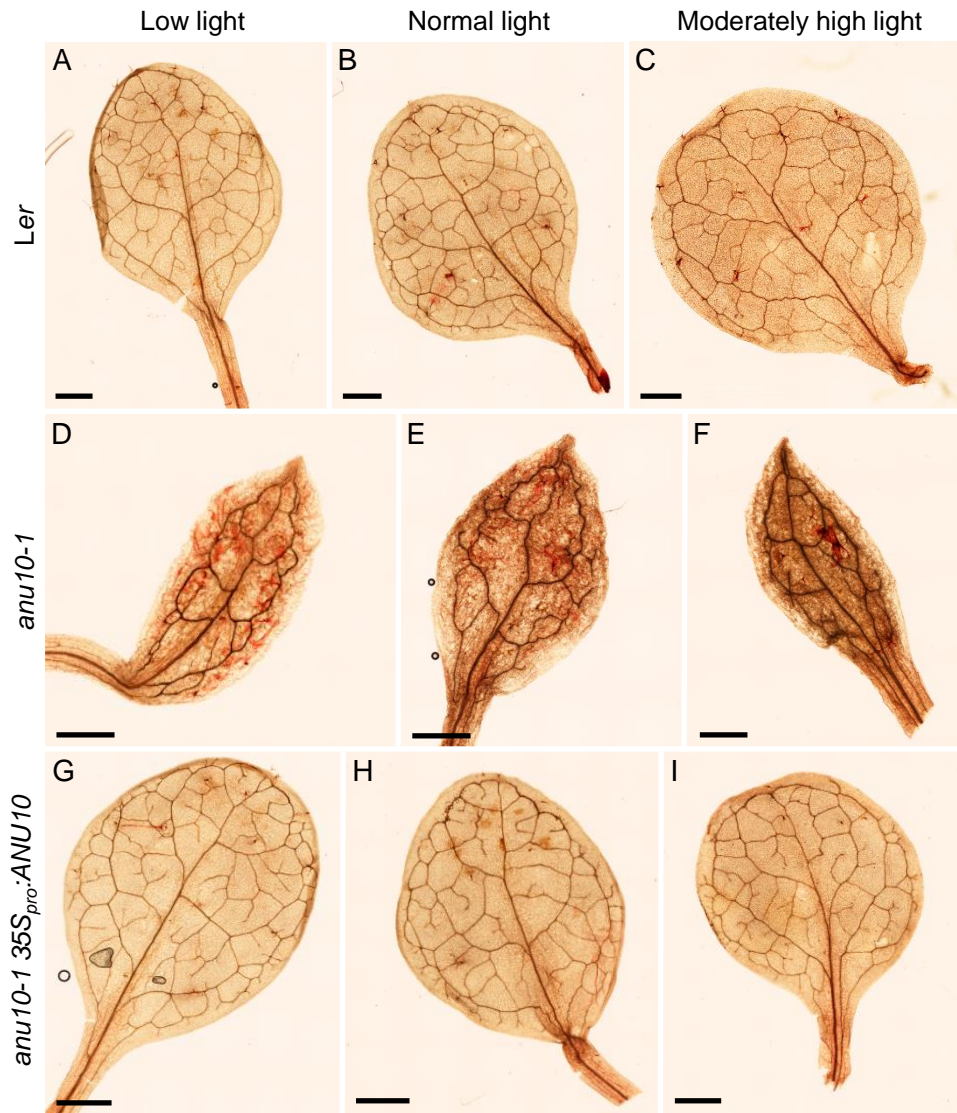
Supplementary Data

Author contributions

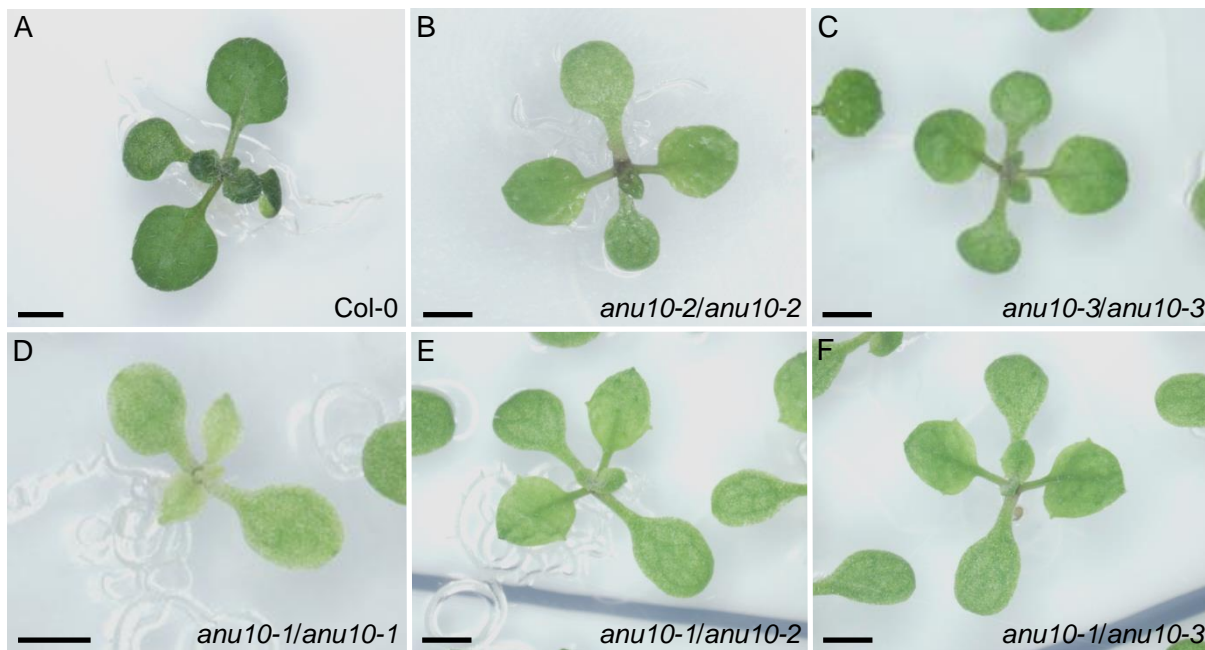
H.C. and J.L.M. conceived and designed the research. R.C.S., E.M.B., S.K. and H.C. performed the research. R.C.S, H.C. and J.L.M. wrote the article.



Supplementary Figure S1. Growth of the *anu10-1* mutant. Error bars indicate standard deviations. Asterisks indicate values significantly different from Ler in a Mann-Whitney U test (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$; $n = 8$).



Supplementary Figure S2. ROS in *anu10-1* leaves. H₂O₂ accumulation visualized by DAB staining in first-node leaves from (A-C) *Ler*, (D-F) *anu10-1* and (G-I) *anu10-1 35S_{pro}:ANU10* plants collected 16 das. Plants were grown at low ($\sim 55 \mu\text{mol m}^{-2} \text{s}^{-1}$), normal ($\sim 90 \mu\text{mol m}^{-2} \text{s}^{-1}$) or moderately high ($\sim 180 \mu\text{mol m}^{-2} \text{s}^{-1}$) light intensity. Scale bars indicate 1 mm.



Supplementary Figure S3. Rosette phenotype of homozygotes and heterozygotes for the *anu10* alleles. Pictures were taken 10 das. Scale bars indicate 2 mm.

A

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Ppa XP_001764411.1 1 -----MALSSGSQIVLPYSQSSSGIPWGCFLPRHHARHSQAIRKQLQLDLI
Sbi XP_002451363.1 1 -----MPSPLLRLALPLCPSPNSHS---PRRFAPSP
Osa BAD07831.1 1 -----MPSPLLHHHHHRLSPRFAPS---R---SPSPR
Ata EMT06249.1 1 -----
Tur EMS59110.1 1 -----MEWGSLSRSP-----
Sly XP_004234099.1 1 -----MAFTLTLINSEANSISPETNSAIRKCPFLIRNLH
Cru EOA37571.1 1 -----MASTASSSLFESRNFKS-----VFLIP
Ath NP_174175.2 1 -----MASISTASSSSLLFSPRNFKSI-----PL
Aly XP_002893534.1 1 -----MASVSTACSSSSSLLFSPRNFA-----SIFLIP
Csa XP_004134942.1 1 -----MAISPMALSSYFIQYSEIQTI PARLFA---IPSIYTFKIV
Gma XP_003535421.1 1 -----MTMAIAASSLSSEVLSY---TKFS---NETPR
Gma XP_003541453.1 1 -----MAASSLFSEVLPYTKFSSST-----LR
Mtr XP_003593674.1 1 -----MASVTSTTTTLETFEFLSHRHTTKPF-----LFSPLH
Car XP_004485849.1 1 MKCPAQOEGTQHFSLSLSPMATAMATVATSTTSLETFVPLRSNKF-----LFFETLH
Rco XP_002522507.1 1 -----MASHSLSFSSVFLSPPQSLSR-----TPIKLF
Tca EOY28124.1 1 -----MAFAHSVTLSEAFPTQFPSCFP---RKLFFFETLP
Vvi CBI30631.3 1 -----MAFIHPPARIELLLTFQPRSSST---RILTFEKLL
Vvi XP_002273236.2 1 -----MAFIHPPARIELLLTFQPRSSST---RILTFEKLL
Fve XP_004295562.1 1 -----MTSAHSLLEHPELVSFTRPSRCS-----L
Ppe EMJ12593.1 1 -----MCPGERSGSSGNQNDVVPF-----
Consensus 1
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Sbi XP_002451363.1 32 TTSSPADRLRLRLTPAASRHTERTSVRAVV-----DGGDLPIARCYEGRRLARL
Osa BAD07831.1 29 TLSLHPAPRLRLRLRLRASARGGGGSTRDHD-----DGA---PIARCYEAAARL
Ata EMT06249.1 1 -----
Tur EMS59110.1 12 -----GKGASPHAVRRLHADRAAA
Sly XP_004234099.1 35 FTLYRNRNRRS-ISC SAQOKPKFKMTEENTLEA---VADFDGDEKSPCIRTYENDLARL
Cru EOA37571.1 24 LRRRSFVSFIRFSKN-----PSAEEDILRF---VAE---SDGMALPCVPTYENNSARL
Ath NP_174175.2 26 LPQQRSSVSF-VRCFSKN---SSTEEDILRF---VAE---SDGMALPCVPTYENNSARL
Aly XP_002893534.1 30 LRRSSVSFIRGYSKN-----SSTEEDILRF---VAE---SDGMALPCVPTYENNSARL
Csa XP_004134942.1 39 LKNSRHCFST-FSCSAGRPI---PTEEEVLOA---VLE---SDEKILPCVPTYENDLARL
Gma XP_003535421.1 27 FTNSKNSIFA-LSCSPKSI---PVTEKEVLOA---IAD---SDGNLPCVPTYDIDLSQL
Gma XP_003541453.1 23 FTNSKNSIFA-LSCSPKSI---PVTEKEVLOA---IAD---SDGNLPCVPTYDNDLSQL
Mtr XP_003593674.1 33 LPNTTPSIFI-LSCSPKTI---FVTEQQVLOA---ISD---SSDOKLPCVPTYENDLARL
Car XP_004485849.1 56 FPNSKYSIFV-LSCSPKTI---FVTEQQVLOA---IAD---SSDOKLPCVPTYENDLARL
Rco XP_002522507.1 30 LTNARNCIPV-LSCST-----FPQOEAILQL---VAN---ADENTLPCIRAFENDLARV
Tca EOY28124.1 34 FRNPTVPLLI-LSRSSPKPA---PSTEPEALKF---ITD---SDGITLPCVPTYENDLARL
Vvi CBI30631.3 34 FKNPRNSLYI-VSCSTPKAI---PATEQEV LDA---IAB---SDEKSLPAVRSENDLARL
Vvi XP_002273236.2 34 FKNPRNSLYI-VSCSTPKAI---PATEQEV LDA---IAB---SDEKSLPAVRSENDLARL
Fve XP_004295562.1 25 FSTPRNSRFTLACSSRKTA---FVTEQEVLOA---ISE---SAEKSLPCVPTYENLARL
Ppe EMJ12593.1 20 -----QQEVLOA---VAD---SDEVLGVRTYENDSACL
Consensus 61
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Sbi XP_002451363.1 82 ELGAGARRRQAVAAAAADGGARARAYLAAGSDAMVVEAFLPQSHGGGTTASSTRVILQA
Osa BAD07831.1 79 DLSGTARRRQAVAAAAADGGAAFAHLLAAVADAMVVEAFLPQDARRSAMSTRVILQA
Ata EMT06249.1 1 -----MVMEAFLPQA---GCAASTRILQA
Tur EMS59110.1 31 NFGHTEAPRMSV-LQMRKRWGREDKGSARVATRAMET-----LQA
Sly XP_004234099.1 90 TLVGAVDFOQAL-TAAAADGGEAAGEHTAAGMDAMVVETLFPGP-SDEHSTVSTRLEFLPA
Cru EOA37571.1 71 SLVGTVAFDQAL-TAAAADGGEAADDHLRENVPMVVETVFPFG-SDEKATVSTRLEFLPT
Ath NP_174175.2 75 SLVGTVAFDQAL-TAAAADGGEAADDHLRENVPMVVETVFPFG-SDEKATVSTRLEFLPT
Aly XP_002893534.1 77 SLVGTVAFDQAL-TAAAADGGEAADDHLRENVPMVVETVFPFG-SDEKATVSTRLEFLPT
Csa XP_004134942.1 90 SLVGVDFRQSV-TAAAADGGEAATEHLDSDGMSAMVVETVFPGT-SDEHSTVSTRLEFLPA
Gma XP_003535421.1 78 TLVGTVDFOQAL-TAAAADGGEVATDHDIDAGMDAMVVETVFPAP-SCDHATVSTRLEFLPA
Gma XP_003541453.1 74 TLVGTVDFOQAL-TAAAADGGEVADHDIDAGMDAMVVETVFPAP-SSDHATVSTRLEFLPA
Mtr XP_003593674.1 85 TLVGAVDFRQAV-TAAAADGGEVADHDVQAGMDAMVVETVFPAS-SSDHGTVSTRLEFLPA
Car XP_004485849.1 108 TLVGAVDFRQAV-TAAAADGGEVASEHIDASMDAMVVETVFPAS-SSDHGTVSTRLEFLPA
Rco XP_002522507.1 76 SLVGSVGFDOAV-VAAAADGGRAADHDIDSGAPAMVVETLFPGP-GDHHATVSTRLEFLPA
Tca EOY28124.1 85 TLVGAVGFEQAL-TAAAADGGRAATEHVDSGIPAMVETVFPSS-IAKNATVSTRLEFLPA
Vvi CBI30631.3 85 TMVGAIDVEQAL-TAAAADGGNTADEHTASGAMAMVVETVFPGA-SDEHSTVSTRLEFLPA
Vvi XP_002273236.2 85 TMVGAIDVEQAL-TAAAADGGNTADEHTASGAMAMVVETVFPGA-SDEHSTVSTRLEFLPA
Fve XP_004295562.1 77 GAVGAVDFOQAL-TAAAADGGEAADDHLRENVPMVVETLFPGN-SDPYSTVSTRLEFLPA
Ppe EMJ12593.1 49 ALVGAVDFOQAL-TAAAADGGQAADEHTISSGIPAMVVETLFPGH-SDEHSTVSTRLEFLPA
Consensus 121
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Ppa XP_001764411.1 166 LAVIERA-KLMAEKDAQFLRGGR-----AFAVAGQTVRLKLSKFEILCFYQPGM'ATDG
Sbi XP_002451363.1 142 REVKDKASKLEKQFGYDFFANESDSESMLAMAFQVVIQRLSNRERLEVFSPGSRVHFQD
Osa BAD07831.1 139 NEVTEKASKLKDITGTEFFSENEPDAESVLAMAFQVVMQDQLTNERLEVFSPGSEIDIQD
Ata EMT06249.1 23 REVKEKAAKIKKDFGDDIFSENEPDSSESILAMALQOVVMHKLSNRLEIFSPGSGRDFQD
Tur EMS59110.1 70 REVKEKAAEIKKDFGDDIFSENEPDSSESILAMALQOVVMOKLSNRLEIFSPGSGRNQD
Sly XP_004234099.1 148 RKVMEKAHKLRSLITIKEMLSGTT--STNILAMTFROVVLQHLWNFQLVLFIPGTERNMDD
Cru EOA37571.1 129 RKVKERAKKLRSLSEDLSSGDL--SKNILAMTFROVVLQOLWNEQLVLFQPGAEREMGD
Ath NP_174175.2 133 RKVKERAKKLRSLSEDLSSGDL--SKNILAMTFROVVLQOLWNEQLVLFQPGAEREMGD
Aly XP_002893534.1 135 RKVKERAKKLRSLSEDLSSGDL--SKNILAMTFROVVLQOLWNEQLVLFQPGAEREMGE
Csa XP_004134942.1 148 RKVTEKATKLRSLAQDFNSSTE--SKNILAMTFROVVLQOLWDFQLVVFQTPGSDRNMED
Gma XP_003535421.1 136 RKVKEKAAKLRNRFPKDLFSGSG--SKNVLAMTFROVVLQOITWSEFLVTFQPGEEERKMVD
Gma XP_003541453.1 132 RKVKEKATKLRSEFPKDAFSGSA--SKNVLAMTFROVVLQOITWSEFLVTFQPGEEERKMD
Mtr XP_003593674.1 143 RKVKEKAAKLRSESEDI FSNIT--SRNVLMTFROVVLQOVWNEFLVTFQPGEEERKMD
Car XP_004485849.1 166 RKVKEKAAKLRSLSQDMFSNIT--SRNVLAMTFROVVLQOVWNEFLVTFQPGEEERKMD
Rco XP_002522507.1 134 RKVKEKAAKLRSEFKEDIFSGTA--SENILAMTFROVVLQOLWNEFLAVFRPGTERNMED
Tca EOY28124.1 143 RKVKEKAQKLRSLSEDI LITETS--SKNILAMTFROVVLQKFWNEFLVLFERPGTERNMED
Vvi CBI30631.3 143 RKVKEKANRLRSETEDEFLLSST--SKDILAMTFROVVLQOLWNEFLVLFIPGTERNMEE
Vvi XP_002273236.2 143 RKVKEKANRLRSETEDEFLLSST--SKDILAMTFROVVLQOLWNEFLVLFIPGTERNMEE
Fve XP_004295562.1 135 RKVKEKAGKLRSLITQDMLSTTT--SRNILAMTFROVVLQOLLWNEQLVVERPGTERNMKD
Ppe EMJ12593.1 107 RKVKEKAGKLRSLITQDMLSTAT--SRNILSMTFROVVLQOLLWNEFLVIFRPGTERNMED
Consensus 181 ..*...*

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Sbi XP_002451363.1 202 FGKFKOK-GSLDCISSSNGKLISSLAEATFYCVI-----EDARANHGGVGS-----
Osa BAD07831.1 199 LSKPRK-VPADFISRSSDEKLI AALAEATFSCVI-----EDARNNLGGTGG-----
Ata EMT06249.1 83 WSKPRK-VPVDFISSSDGLKLI SALAEATFSCVI-----EDTEKSLFGGTGG-----
Tur EMS59110.1 130 WSKQRK-VPVDFISSSDGLKLI SALAEATFSCVI-----EDTEKSLFGGTGG-----
Sly XP_004234099.1 206 LETPRE-VPPSEALSSDERVISVLAEVICISAL-----ESTKHEFNGTGGGAPKKEFR
Cru EOA37571.1 187 FENPRE-VSTSFLLSSDTERVISVLAEVICISAL-----OSTEKHEFLDYLGLKAKFPFK
Ath NP_174175.2 191 FENPREQVSTSFLLSSDTERVISVLAEVICISAL-----OSTEKHEFLDYLGLKAKFPFK
Aly XP_002893534.1 193 FENPREQVSTSFLLSSDTERVISVLAEVICISAL-----OSTEKHEFLDYLGLKAKFPFK
Csa XP_004134942.1 206 LENPRE-VPISFLLSSSEERAI SVLAETVCMCAL-----QNTGKFLVDGTSRGTSTRLEG
Gma XP_003535421.1 194 LETPRE-VPASEALSSADEYLISVLAEAICISAL-----QSTQIQFLKVKKGNRGGFGR
Gma XP_003541453.1 190 LETPRE-VHASEALSSDEYLISVLAEAICISAL-----QSTQIQFLKAKGNRGGFGR
Mtr XP_003593674.1 201 LENPRE-VPASFLLSSDEYLISVLAEVVICISSL-----QSTQIQFLKSDGSRSGFGR
Car XP_004485849.1 224 LENPRE-VPASFLLSSDEYLSVLAEVVICISAL-----QNTQIQFLDKSQGSRSGFGR
Rco XP_002522507.1 192 LENPRE-VPASFLLSSDEHILISVLAEAITYIAL-----QNTESNFLYDFMGETSGGVFR
Tca EOY28124.1 201 LENPRK-VLCSFLLSSSEERVISNLEVVVICISAL-----OSTERHEFLNFLGKTSNNEFQ
Vvi CBI30631.3 201 LQDSRKQVPASFLLSSDDEGVISVLAEVICISAL-----QOTTERHEFLNLLGQTSNNEFK
Vvi XP_002273236.2 201 LQDSRK-VPASFLLSSDDEGVISVLAEVICISAL-----QOTTERHEFLNLLGQTSNNEFK
Fve XP_004295562.1 193 LENARE-VPACVLLSSSDEHILISVLAEVVICISAL-----QNTERQFLDYGKTPSDLLH
Ppe EMJ12593.1 165 LENPRE-VPPSFLLSSDERIISVLAEVVICISAL-----ENTERQFLDFMGKTKSNLVR
Consensus 241

Ppa XP_001764411.1 269 ELKRSSEGIKTVKNSIVQISSFTAEVSAHVERTVQDKKLVGTSVGLRRHRS---WVNRP
Sbi XP_002451363.1 248 LEQKROLDYS--MDSSVCIHRISEAEVVKNAKRCLETISLIKSSHVHETKNG--WVPPP
Osa BAD07831.1 245 LSHKWKSNCS--LDSSVCIDIISEEVVNSARRRLESEDLVQSSHVAGAKNG--WVPPAP
Ata EMT06249.1 129 EFQSQKLNCS--SDSTVVCVHRISAEVANNARRCLESEFNLTSSSHEVGRKKNV--WVPPAP
Tur EMS59110.1 176 EFQSQKLNCS--SDSTVVCVHRISAEVANNARRCLESEFNLTSSSHEVGRSKNA--WVPPAP
Sly XP_004234099.1 260 WEDTHKSIIVS--KDSSVTLNLMYEIVANANVLLQKESSEERANYEPREGRWGMNLTST
Cru EOA37571.1 241 WBSKHKRIAS--EDSSVVLHKLFEDE--QNTNHLLEYQOSREKFKLADTRQSRRWNMS
Ath NP_174175.2 246 WLSKRRRIAS--EDCSVVLHKLFEDE--QNTKLLLEYQOSRKENFKLADTRQSRRWNMS
Aly XP_002893534.1 248 WLNKHKRIAS--EDCSVVLHKLFEDE--PNTNLLLEYQOSRKENFKLADTRQSRRWNLS
Csa XP_004134942.1 260 WBRKSTIVAS--KDSSVVIHKLFEDEI--ADPKSLLQKENSNKESKHKRNSKSMNYWVMP
Gma XP_003535421.1 248 WBRKPESVQS--KDSAVIILSKLFEDEIVENARSLLNLYNLMKDGKPKVKIKSGHHWVKSS
Gma XP_003541453.1 244 WFOKPESVQS--KDSAVIILSKLFEDEIVENARSLLNLYNLMKDGKPKVKIKSGHHWVKFS
Mtr XP_003593674.1 255 WFOKHERIOS--KDSAVIILHKLFEDEIVENARSLLKYLHLMKDGKPKVKIKSERFWVKFS
Car XP_004485849.1 278 WFOKPERIOS--KDSAVIILHKLFEDEIVENAKSLLNLYHLMKDGKPKVKIKSGPFWVKFS
Rco XP_002522507.1 246 WFOKPKSIAS--QDSSVVIYKLFEDETAGNAKLLLEDENLSKD--KGIKLRQKYKWWTPV
Tca EOY28124.1 255 WBRKSKAIES--KDLSSVVIYKLFEDIVENAKSLLSENSSESFKMLDLRRKDYWVTPS
Vvi CBI30631.3 256 WBRKPKSVAS--KDSSVIMYELFEDEIVENAKNLLNENNSMKANKCIEKTKKYHWWTSS
Vvi XP_002273236.2 255 WBRKPKSVAS--KDSSVIMYELFEDEIVENAKNLLNENNSMKANKCIEKTKKYHWWTSS
Fve XP_004295562.1 247 SBRKPRRIVS--KDSSVVIYKLFEDIVENAKSLLNENNSAKNSKSVKTKSK--WVTPS
Ppe EMJ12593.1 219 WBRKPKRTVS--KDSSVVIYKLFEDIVENAKSLLKFNENLTGSKPKPKTKSKYYRWAT
Consensus 301 ..*...*

Ppa XP_001764411.1 326 LAEAVVKSGL--AQLVDEFI PAHRLQLN---LPSNKFEAVGLOKLSDTVCSINLTHAQM
Sbi XP_002451363.1 304 HYETLVKIGGPELVLWANEPYIPYRLOINANALENSNHE--GLFELESNRWEVLLSHSQM
Osa BAD07831.1 301 KSERLVKIGGPEMLWASEFVYTYRLOIDAKAFKNTKLG--CHELVANNKGEVLLSHAQM
Ata EMT06249.1 185 KYGRLAEIGGPEFILWAHEFVPSYRLOINANAFENTKLE--GCELVNNRWEVPISEHQL
Tur EMS59110.1 232 KYGRLAEIGGPEFILWAHEFVPSYRLOINANAFENTKLE--GCELVNNRWEVPISEHQL
Sly XP_004234099.1 318 TYEKLEQIGGPEFISWLSERVPAYLQIDAELDNKKEE--GWRKETAINTWEVFLTHSQM
Cru EOA37571.1 297 ANSKLEKFGGPEFSTWASEYIPAYRLEIDTTILADAKLE--GWRKSSSENKWEVLLTHSQM
Ath NP_174175.2 302 ANSKLEKIGGPEFSSWASEYIPAYRLEMDSTILADAKLE--GWRKSSSENKWEVLLTHSQM
Aly XP_002893534.1 304 ANSKLEKIGGPEFSSWASEYIPAYRLEIDTIIILADAKLE--GWRKSSSENKWEVLLTHSQM
Csa XP_004134942.1 317 ELTRLEKIGGPEFCAWVSEYVPAAYRLOIDAHQFNGLKKEG--GWRFEVENRWEVLLTHSQM
Gma XP_003535421.1 306 CYEKLEKIGGPEFSAWASEYVPAAYRLEIDTKILGDSKIG--GWRKSAENRWEVLLTHSQM
Gma XP_003541453.1 302 CYEKLEKIGGPEFSAWASEYVPAAYRLEIDTKILGDSKIE--GWRKSAENRWEVLLTHSQM
Mtr XP_003593674.1 313 SYEKLEKIGGPEFSAWTSEYVPAAYRLEIDPKILGDSKEQ--GWRKSSSENKWEVLLTHSQM
Car XP_004485849.1 336 CYEKLEKIGGPEFSAWTSEYVPAAYRLEVDTKILGDAKFO--GWRKSSSENKWEVLLTHSQM
Rco XP_002522507.1 302 ALSKLETIGGPEFSAWVSEYVPAAYRLOIDADIVKDAKME--GWRFECDNRWEVLLTHSQM
Tca EOY28124.1 313 MHSKLDKIGGPEFSSWASEYIPAYRLEINANTLKDKEE--GWRKSSSENKWEVLLTHSQM
Vvi CBI30631.3 314 ALSKLEKIGGPEFSTWTSEYIPAYRLOIDPDKLKSVKFE--GWRKSAENRWEVLLTHSQM
Vvi XP_002273236.2 313 ALSKLEKIGGPEFSTWTSEYIPAYRLOIDPDKLKSVKFE--GWRKSAENRWEVLLTHSQM
Fve XP_004295562.1 303 ALSKLEKIGGPEFSCWTSEYVPAAYLQIDANRHKDKKEE--GWRKCEETSWEVLLTHSQM
Ppe EMJ12593.1 277 ALSKLEKIGGPEFSSWTSEYVPAAYRLOIDANQHKDKKEE--GWRKSAENWCEVFLTHSQM
Consensus 361*.....*.....*.....*.....*.....*.....*.....*.....*.....*

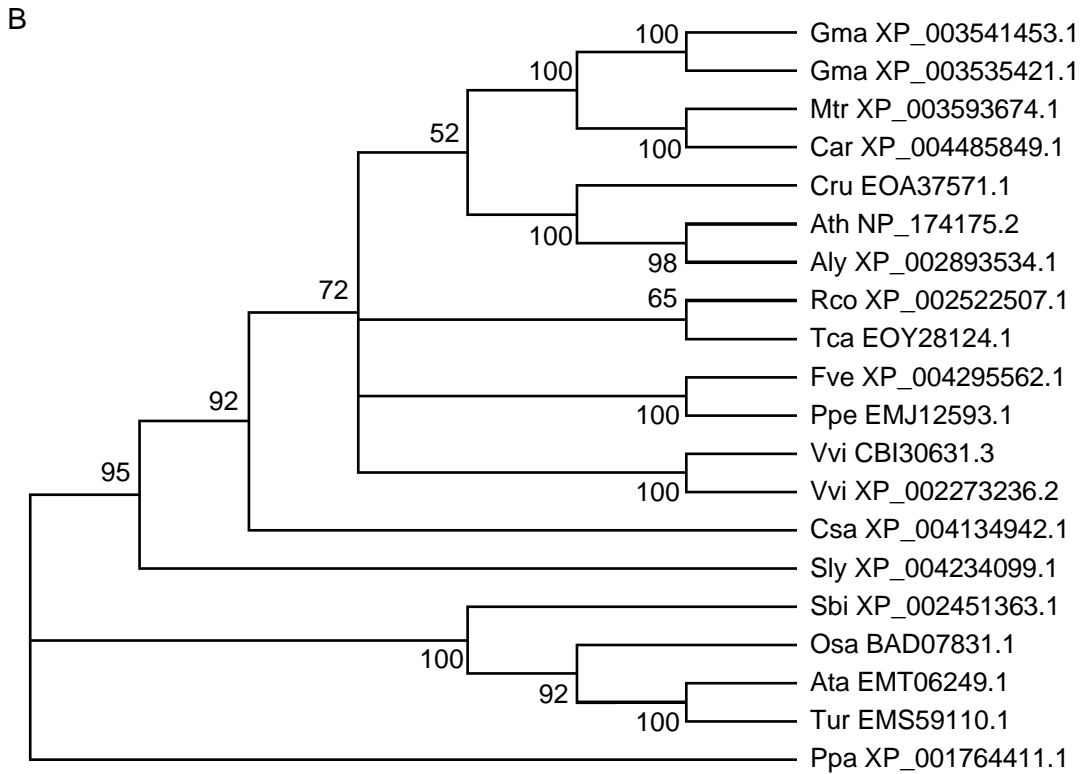
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Sbi XP_002451363.1 362 VELGNILDMYEDQFTLPCKTF-----HPHNSDPSKIKKNNGYLNNLFTFLAGSCIEFIF
Osa BAD07831.1 359 VELANILDMYEDQFTLPCKTF-----YSNNSEPSKIKKNNGYLNNLFFALLAGSSIVFL
Ata EMT06249.1 243 VELGNVVDMYEDQFTIPCKTF-----RSHNAEPSKIRRNNGYLNNLFSFLAGCSVIFFI
Tur EMS59110.1 290 VELGNVVDMYEDQFTIPCKTF-----RSHNAEPSKIRRNNGYLNNLFSFLAGCSVIFFI
Sly XP_004234099.1 376 VGLSDVLDMYEDYIYLPYKQLSCGV-VAKSFNSPSS-KRSISMSRATSMVLAGSIFLVA
Cru EOA37571.1 355 VGLAEALDIYEDYISLPKQLPCDV-PCNYANLPNE-KRGLSLKFTISVTMASGIFLLA
Ath NP_174175.2 360 VGLAEALDIYEDYISLPKQLPCDV-PCNYANLPNE-KKGLSLKFTISVTMASGIFLLA
Aly XP_002893534.1 362 VGLAEALDIYEDYISLPKQLPCDV-PCNYANLPSE-KRGLSLKFTISVTMASGIFLLA
Csa XP_004134942.1 375 VGLANILDMYEDYISLPDKQLCCGA-NVHSANLSE-KRNYSSWGLSKTLGAGFLVA
Gma XP_003535421.1 364 VQLAETLDTIYYVDPYSLPNKQLSCGV-AAKFAVNSNK--KGNSFPKFLSALASGIFLVA
Gma XP_003541453.1 360 VQLAETLDTIYYVDPYSLPYKQLSCGV-AAKFAVNSNK--KGNSFPKFLSALASGIFLVA
Mtr XP_003593674.1 371 VGLAETLDMYEDYIYLPDKELSYGV-AAKFAVNSNK--KGSSSKILSVSLASGIFLVA
Car XP_004485849.1 394 VSLAETLDMYEDYIYLPDKQLSCGV-VAEYVNVSSR-KVRLYLSKFLSVTLATGMFLVA
Rco XP_002522507.1 360 AGLAETLDMYEDYISLPKQLSCRA-TANETINANT-KKSSSWLRELSVSLISGIFLLA
Tca EOY28124.1 371 VGLADILDYIYEDYISLPKQLRCGV-VANVNSLSKA-KRSSSFSRMVSATVASGIFLIT
Vvi CBI30631.3 372 VALANILDMYEDYIYLPDKQLCCGAGFANFTNSKN-KRISSLKFLTISTIASGIFLVA
Vvi XP_002273236.2 371 VALANILDMYEDYIYLPDKQLCCGAGFANFTNSKN-KRISSLKFLTISTIASGIFLVA
Fve XP_004295562.1 361 VGLADILDYIYEDYIYLPDKELSCGD-IASETINNSNK-KTGSFLKFLSVSLASGIFLVA
Ppe EMJ12593.1 335 VGLADILDYIYEDYIYLPDKQLSCGV-VANSTINNSNK-KMGSFILRLMSVTLASGIFLIT
Consensus 421 ..*.....*.....*.....*.....*.....*.....*.....*.....*.....*

Ppa XP_001764411.1 430 IG--CALLAGFSSRRARLGQMDAPIESIFDMAFSQKIREIGRSD----IVDEQMSVNO
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Osa BAD07831.1 414 VGALAQLCWEPQSLKDKRLATVSSVSSSQSYCSDIHSLDSSLEQAVCVSVVEKIKDSVGC
Ata EMT06249.1 298 VGVVAQLCWEPQSLKDKRLFMGSSPTSSSQSYCSDVHSLDNSEVRGVCISVVKIKDSCCC
Tur EMS59110.1 345 VGVVAQLCWEPQSLKDKRLFMGSSPTSSSQSYCSDVHSLDNSEVRGVCISVVKIKDSCCC
Sly XP_004234099.1 434 TRILGQRYLPYPIRKHYHPGVNPNSSDMISIQPSSMESCKLEDVSVVRRIKELFCW
Cru EOA37571.1 413 VSAAAQFCFPQ-KSERKYPGRSQDLWSENELLSHSSDSSKLEDFCGLLVVKIKDASVW
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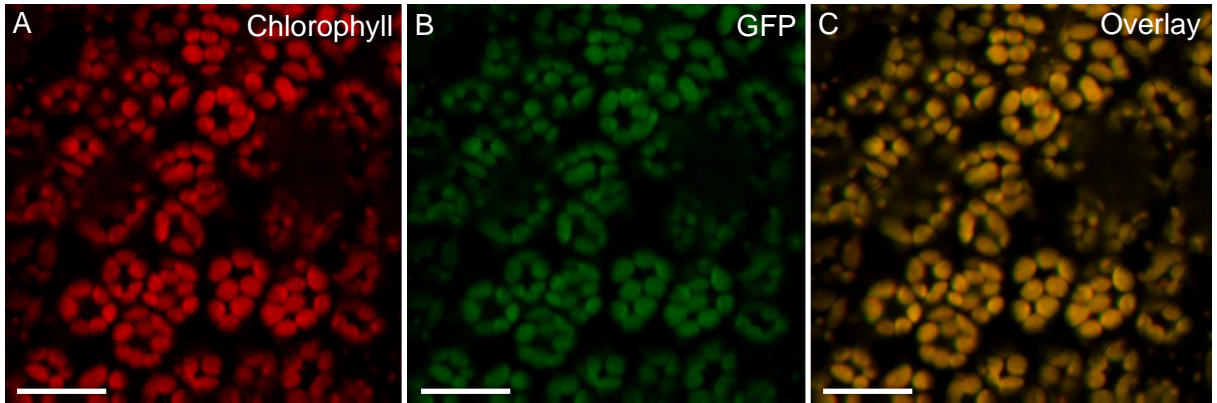
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Sly XP_004234099.1 494 PGDVVMGSGSSAWV GELPIYLNK---EMDSEVLDLNSSTPSEG-----
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Ath NP_174175.2 477 VGEITLESSIGAWV GEPDYLYKETS-RAKSVDDHVT SSSSLEEI-----
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Csa XP_004134942.1 493 HGDVRTGKRVGAWV GADPYLTVV--ESDIRSEDAPS GTIGEEN-----
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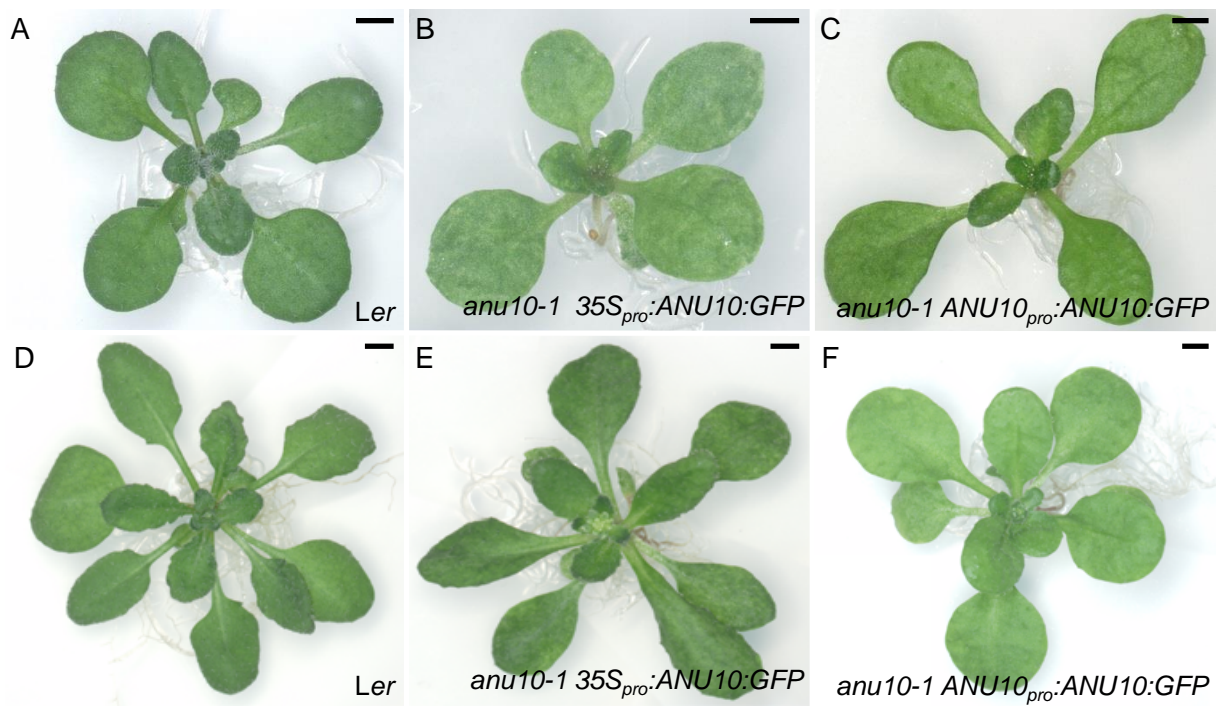
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Ata EMT06249.1 478 EPRLKISR-----PAKVVPHELLMSVNSPSEF-----ALARPQDPC-----
Tur EMS59110.1 525 EPRLKISR-----PAKVVPHELLMSVNSPSEF-----ALARPQDPC-----
Sly XP_004234099.1 594 EQGLKRSR-----GSDAVVHELLMSINPOSSEF-----ALVRPWTENSR-----
Cru EOA37571.1 574 EPGLKSRP-----PTKVVVHELLMSVNSDRPE-----ALVRPPLPQ-----
Ath NP_174175.2 579 EPGLKSHP-----PKKVVVHELLMSVNSDRPE-----ALVRPPLPQ-----
Aly XP_002893534.1 581 EPGLKSHP-----PKKVVVHELLMSVNSDRPE-----ALVRPPLPQ-----
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Gma XP_003535421.1 574 EPGLKVSL-----PEKVTVHELLMSINPDAYE-----ALARFR-----
Gma XP_003541453.1 570 EPGLKVPL-----PEKVTVHELLMSINPDAYE-----ALARFR-----
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Rco XP_002522507.1 579 EPGLKIRP-----PSETVVHELLMSLWADWHYRNLTASLEGSHECAPGSKFIHL
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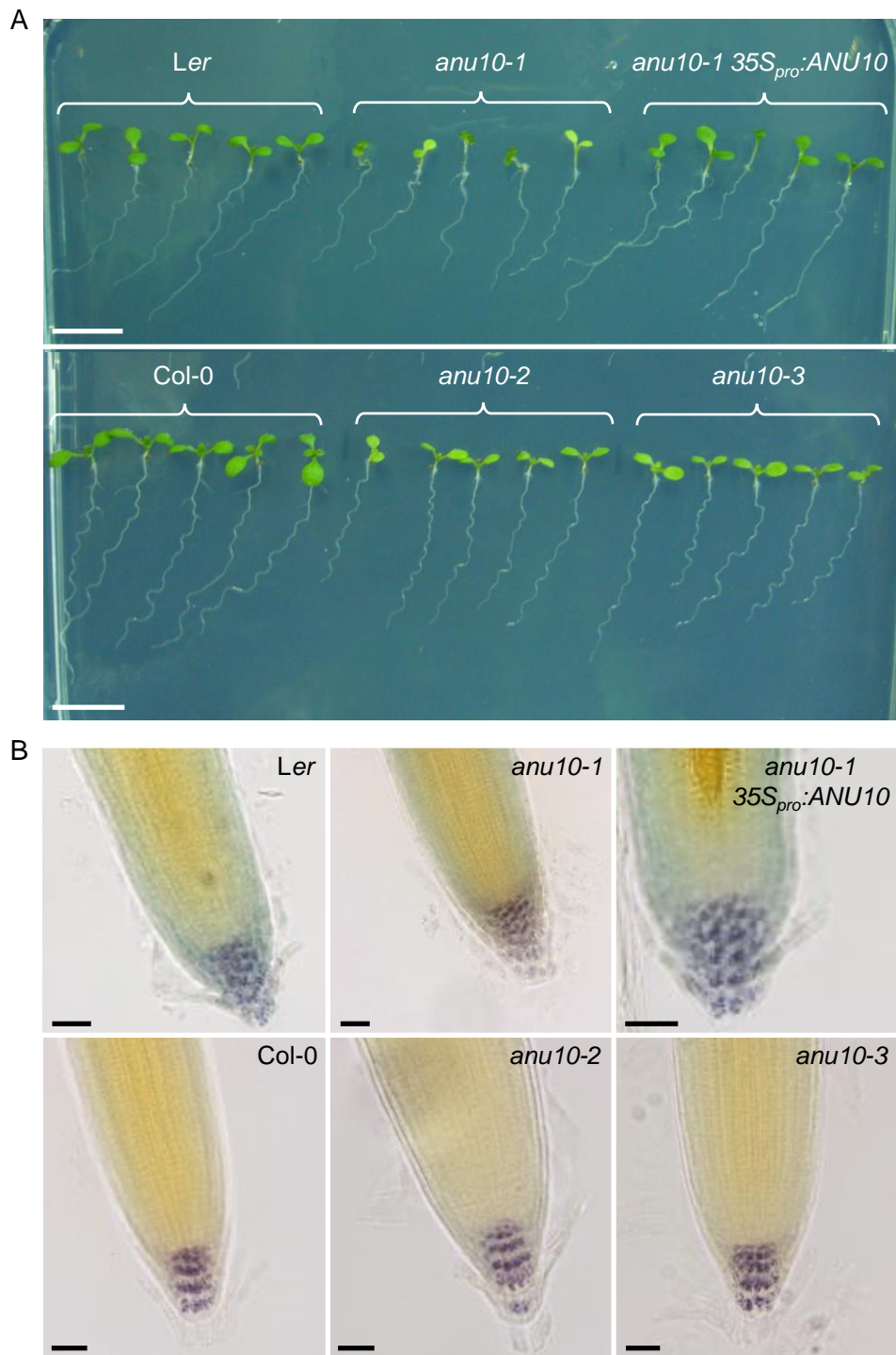
Supplementary Figure S4. Phylogenetic analysis of ANU10 and its putative orthologs. (A) Alignment of the amino acid sequences of the Arabidopsis ANU10 protein and its orthologs in other plant species. Identical and closely related residues that are conserved in >50% of the sequences are shaded black and grey, respectively. Asterisks and dots indicate positions with totally identical or similar residues, respectively, among the orthologs. Numbers indicate residue positions. Amino acids drawn in red and green correspond to the chloroplast transit peptides as predicted by ChloroP1.1 and the transmembrane domains as predicted by SOSUI, respectively. Protein sequences were retrieved from NCBI. The multiple sequence alignment was first made using T-Coffee, subsequently refined using MUSCLE and finally shaded with BOXSHADE 3.21. (B) Unrooted tree showing the evolutionary history of ANU10 and homologous proteins. Analysis of 20 homologs was carried out with MEGA5 following the Neighbor-Joining method. The evolutionary distances were calculated using the Poisson correction method. A bootstrap consensus tree was inferred from 1000 replicates. Numbers correspond to bootstrap percentages. Branches corresponding to partitions reproduced in less than 50% bootstrap replicates were condensed. Plant species and identities (%) between ANU10 and each of its orthologs are: Ppa, *Physcomitrella patens* (18.06%); Sbi, *Sorghum bicolor* (29.46); Osa, *Oryza sativa* (32.00); Ata, *Aegilops tauschii* (28.17); Tur, *Triticum urartu* (28.95); Sly, *Solanum lycopersicum* (45.51); Cru, *Capsella rubella* (85.76); Ath, *Arabidopsis thaliana*; Aly, *Arabidopsis lyrata* (89.98); Csa, *Cucumis sativus* (48.18); Gma, *Glycine max* (52.83 for XP_003541453.1 and 52.97 for XP_003535421.1); Mtr, *Medicago truncatula* (52.15); Car, *Cicer arietinum* (49.85); Rco, *Ricinus communis* (50.94); Tca, *Theobroma cacao* (52.47); Vvi, *Vitis vinifera* (52.55 for CBI30631.3 and 52.39 for XP_002273236.2); Fve, *Fragaria vesca* (54.84) and Ppe, *Prunus persica* (52.58).



Supplementary Figure S5. Subcellular localization of the ANU10:GFP fusion protein in the *anu10-1 ANU10_{pro}:ANU10:GFP* transgenic line. Confocal micrographs of the subepidermal layer of mesophyll cells showing (A) chlorophyll autofluorescence of the chloroplasts, (B) GFP fluorescence and (C) an overlay of the chlorophyll and GFP signals. Pictures were taken from first-node leaves collected 16 das. Scale bars indicate 20 μm .



Supplementary Figure S6. Phenotypic rescue of *anu10-1* by the ANU10:GFP fusion protein. Pictures were taken (A-C) 16 and (D-F) 21 das. Scale bars indicate 2 mm.



Supplementary Figure S7. Growth and starch content of *anu10* roots. (A) Plants grown on vertically oriented agar plates. (B) Lugol staining of root apices. Pictures were taken (A) 7 and (B) 9 das. Scale bars indicate (A) 1 cm and (B) 100 μ m.

Supplementary Table S1. Primer sets used in this work

Purpose	Oligonucleotide name(s)	Oligonucleotide sequences (5'→3')	
		Forward primer (F)	Reverse primer (R)
Linkage analysis	AthZFPG_F/R	TTGCGTTTCCACATTTGTTTAAAC	AGAAATGGGTCAATTCACATGTA
	SNP10026_F/R	CAAGCCCTCTTATGCTTTAGAAA	ACTATTGACTTCGTGTTAACAAAG
	nga392_F/R	TTGAATAATTTGTAGCCATGTTAAT	GGGGTGTTAAATGCGGTGTTG
	F3M18_F/R	GCTGTCAATTTAGGGCATTGA	AAAAGTTGGTTTGAAAAAGTTGAGA
	F1K23_F/R	CCTGTCAAGGGGTCGTAGTC	TTGTGCCTCATCTTCTCGTG
	cer479911_F/R	CAAATAGAACCAACACCGTAAA	ATTCACTTTCCAATAGATCACTC
	SO392_F/R	GACGATATCTTCCTGCGAAAGA	TGTAAAGAGAAGCAGAGTGTCG
<i>ANU10</i> sequencing	ANU10_F1/R1	TGGGGAAATACGGTTTTGAG	CGAGATGCTGATATGTTGTGTG
	ANU10_F2/R2	AGGACTGGCAATTGGTTCTG	TTTCGCCGACGAAGAAGTAT
	ANU10_F3/R3	AGAGCGGAGATGCATATAACT	TTATTTTGGAGACATGTTATGAG
	ANU10_R4		GGAATTGATACTGATAGGTTG
Topo and Gateway cloning	ANU10cds_F/R	CACCATGGCTTCCATTTCTACTGCT	TTGTGGCAATAATGGTCTGAC
	ANU10cds_F2/R2	GCAGCTGTGGAACCTCCAGC	ACAGTACCAACGAGACTGAGC
	ANU10cds_F3	ATCCTTTAGCTAGAGAACTTTATG	
	ANU10pro_F/R	GGGGACAAGTTTGTACAAAAAAGCAGGCT TTGCTGCTCTGCATAGACACA	GGGGACCACTTTGTACAAGAAAGCTGGGT GAATGGAAGCCATTGACGAAC
	ANU10pro_F2	CGCAGATACGGAATCATTTTTCT	
	ANU10cds-pro	TCTGTTTCGTCAATGGCTTCCATTTCTACTGCTT	
	ANU10pro-cds	ATGGAAGCCATTGACGAACAGATAAAAATCACAGA	
qRT-PCR	M13_F/R	GTAAAACGACGGCCAG	CAGGAAACAGCTATGACCATGATT
	18S RRNA_F/R	GGTGGTGGTGCATGGCCGTTCTT	GACGGGCGGTGTGTACAAAGGGC
	LHCB1_F/R ¹	GCCTTCGCTACCAACTTCGTC	AACCGGATACACAACTCGATC
	LHCB2_F/R	CTCAAAGCATCTGGTACGGA	CGGTTAGGTAGGACGGTGT
	LHCB3_F/R	AGTACACTATGGGAAATGATCT	GTGAGGTAAGACGGAGTTTG
	LHCB5_F/R	GGTATGGTCCTGACAGGAG	CTTCTTTCCAAGACCAAATGG

qRT-PCR	HEMA1_F/R ²	GTGAGCTCTCTGCTTCTTCTGATTCTG	CTGCTTCTTTTCTTTGTATATCGATCAGCT
	ORE1_F/R ³	CTTACCATGGAAGGCTAAGATGGG	TCGGGTATTTCCGGTCTCTCAC
	accD_F/R ⁴	GCTAAGTAAAGCAATGGATAGTTT	CGAATGTCCTTGGAGCTAACTAA
	rbcL_F/R ⁵	GTGTTGGGTTCAAAGCTGGT	CATCGGTCCACACAGTTGTC
	psbA_F/R ⁵	GAGCAGCAATGAATGCGATA	CCTATGGGGTCGCTTCTGTA
	rrn16_F/R ⁵	CGGTATCTGGGAATAAGCA	GATTTGACGGCGGACTTAAA
	rrn23_F/R ⁵	GGGCGACTGTTTACCAAAAA	TTACCCGACAAGGAATTTTCG
	atpB_F/R ⁵	GAGCTCGTATGAGAGTTGGT	ACCCAATAAGGCGGATACCT
Genotyping of mutant and transgenic plants	LB1_F ^a	GCCTTTTTCAGAAATGGATAAATAGCCTTGCTTCC	
	N831342_F/R	TGTGTCAACCAGATTGGTCAG	TTGCTAGGTTTCATTCCCACAC
	N828696_F/R	TTTACTGTATTGCCGAATCGC	AGCGAAGTTAACCTCTCGAGG
	35S_F ^b	GACAGTGGTCCCAAAGATG	
	GFP_R ^c		TATGTTGCATCACCTTCACCCT
	GUS_R ^d		CACAAACGGTGATACGTACACT

^{a-d}Each of these oligonucleotides was used together with: ^aN831342_R and N828696_R to genotype the *anu10-2* and *anu10-3* mutants; ^bANU10cds_R2, ^cANU10cds_F2 or ANU10cds-pro, and ^dANU10pro_F2 to genotype transgenic plants putatively carrying the ^b35S_{pro}:ANU10, ^c35S_{pro}:ANU10:GFP or ANU10_{pro}:ANU10:GFP, and ^dANU10_{pro}:GUS transgenes.¹Adhikari *et al.* (2011), ²Mochizuki *et al.* (2008), ³Rauf *et al.* (2013), ⁴Hricová *et al.* (2006), ⁵Gao *et al.* (2011).

Supplementary Table S2. Transmembrane domains and chloroplast transit peptides in the ANU10 protein and its putative orthologs

Organism	Protein ^a	Protein length (aa)	Transmembrane domains ^b		Chloroplast transit peptides ^c	
			Number	Coordinates	Presence	Length (aa) ^c
<i>Physcomitrella patens</i>	XP_001764411.1	579	1	417-439	No	-
<i>Sorghum bicolor</i>	XP_002451363.1	642	0	-	Yes	59
<i>Oryza sativa</i>	BAD07831.1	630	1	400-422	Yes	45
<i>Aegilops tauschii</i>	EMT06249.1	514	2	2-24, 284-306	No	-
<i>Triticum urartu</i>	EMS59110.1	561	1	331-353	No	-
<i>Solanum lycopersicum</i>	XP_004234099.1	631	1	416-438	Yes	46
<i>Capsella rubella</i>	EOA37571.1	609	1	396-418	Yes	34
<i>Arabidopsis thaliana</i>	NP_174172.2	614	1	421-443	Yes	37
<i>Arabidopsis lyrata</i>	XP_002893534	616	1	404-426	Yes	55
<i>Cucumis sativus</i>	XP_004134942.1	627	0	-	Yes	50
<i>Glycine max</i>	XP_003535421.1	607	1	408-430	Yes	38
<i>Glycine max</i>	XP_003541453.1	603	1	404-426	Yes	34
<i>Medicago truncatula</i>	XP_003593674.1	614	0	-	Yes	63
<i>Cicer arietinum</i>	XP_004485849.1	637	1	436-458	Yes	83
<i>Ricinus communis</i>	XP_002522507.1	628	1	403-425	Yes	54
<i>Theobroma cacao</i>	EOY28124.1	624	0	-	Yes	46
<i>Vitis vinifera</i>	CBI30631.3	666	1	415-437	Yes	45
<i>Vitis vinifera</i>	XP_002273236.2	624	1	414-436	Yes	45
<i>Fragaria vesca</i>	XP_004295562.1	614	1	403-425	Yes	56
<i>Prunus persica</i>	EMJ12593.1	588	0	-	No	-

^aGeneBank accession number. ^{b,c}Predictions shown in this table were obtained from ^bSOSUI and ^cChloroP 1.1.

Supplementary Data References

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