```
OsIRX9L         ---------MSRRNAGAMQREGSVK---DWEEFDPSPSPKLAYSQSYVAMRGLLTSVASL
AtIRX9L         -------MASIRRTLSPMYHDRSHENGGSHKGFTIGGSSSKH----------------NS
AoIRX9L         MLYATEKMPSFRRTMSPLQRDGGAVQNGE-TAFDISPPPKLPQNRTYLAIGRLLNSLLNS
SmIRX9L         ------------------------------------------------------------
PpIRX9          ------------------------------------------------------------
OsIRX9          ------------------------------------------------------------
LmIRX9          ------------------------------------------------------------
PtIRX9          ------------------------------------------------------------
AtIRX9          ------------------------------------------------------------
AoIRX9          ------------------------------------------------------------

OsIRX9L         DLVLMSSSLKSAWAAISSHKHARSLERSRSKGMSLKRAMLQLLVCFMVGIFIGFTPPFSV
AtIRX9L         ------SQ-FLSY--LTKL-LGVTSDPKSSRRGPWRRPFYQFLVFFLLGFVLGLTPFGKM
AoIRX9L         ------LD-FPGA--LFSRSYRPAAERSKPKGLHMKRSFFHYCLCFMLGIFVSFTPFFSV
SmIRX9L         ----------MSW--MKMASARQGLDRSKARMQILRKPFFHFVLCFFVGFGIGLTPLSSG
PpIRX9          ------------------------------------------------------------
OsIRX9          ----------MASAGGCKKKTGNSRSRSPRSPVVLRRAMLHSSLCFLVGLLAGLAAPSDW
LmIRX9          ------------MGSAALDHAGGRPKKGSGGSQLWKKALLHSSLCFVMGFFTGFAPSSVS
PtIRX9          ---------------------MGSLERSKKKVQLWKKAIVHFGLCFVMGFFTGFAPGGKA
AtIRX9          ---------------------MGSLERSKKKAQVWKKAVIHFSLCFVMGFFTGFAPAGKA
AoIRX9          ---------------------MGSIERSRKRIQLWKKAAIHFSLCFVMGFFSGFAPTSTA

OsIRX9L         DLPGKIASENGRLPFDGDAIDRR-----QMVE------RQGTKLEPFVAEAESEASSEPQ
AtIRX9L         EDVNG----SDRFSFEIKQPYVE-----E--R------LENRKREE--------------
AoIRX9L         DVSKNLASRQHAFSFEDTAVIDN-----AKQK------ISSVEKEILFIEKSL------S
SmIRX9L         GGGAREQQRPR-ESFTAAK------------R------IA-TEEQVVVLDSSS------P
PpIRX9          ------------------------------------------------------------
OsIRX9          PAAAGAAV----FL----------------------------RTL--RASNV-------
LmIRX9          DWRSAAPV----VGAGGLAV----ESSHL---LRTQHSVGGGNRTL--L-----------
PtIRX9          SIFSSHVV----ASNKSQPV----EMLHQQ--VASTPHASNVNRSL--IA---ES----P
AtIRX9          SFFSNFET----TSYTSTKS----PIPPQPFENATYTQHSLLNRTL--INSQSQA----P
AoIRX9          SIFSGQAN----PYRSTKNLGISAQLAPQPLQ-----NPSENNRSL--MSSEV-------

OsIRX9L         VEEGPPV--PAMLDDEADFVEASPIVHSVNDSGIVVRKHLIIITTTSVR----PHQAYYL
AtIRX9L         ----------AAVDAV--SFVA-ETENGKKEVNFVPKKLLIVVTPTYNR----AMQAYYL
AoIRX9L         IENGSLEMKTEALDASNVTLSAYATPTSQ-HAGLVPQQQLIIVTPTYER----PFQAYYL
SmIRX9L         L-----EIS----------ANSSRHPGEGRMPLQDPKKLLIIVTPTYTR----PFQAMYL
PpIRX9          ------------------------------MPL-EYRKLLIIVTPTYVR----PFGAYYL
OsIRX9          -----------------------IFSRSS-NRPQQP-QLVVVVTTTEQS-DDSERRAAGL
LmIRX9          ------------------------SQGGAAAEAATPRPLLVVVTTTESTPTANGERAAML
PtIRX9          V-----P-TP-LSSKES-EPAKFLEKEEEPKPKLLPRRLAIIVTPISTE---DPYQGVFL
AtIRX9          A-----P-AE-SREAEG-ETRSLSEKEDENQVKVTPRGLVIVVTPIITK---DRYKNVLL
AoIRX9          A-----E-NP-ANSS--------ATHRSTAD-ELSPRNLLIVITTSKPN---DRFSGPFL
                                                        :::*               *

OsIRX9L         NRLAHVLKDVPPPLLWIVAEWPYQSR--E--TAEILRSSGIMYRHLICNRNTTNIRK---
AtIRX9L         NRVAQTLRLVESPVLWIVVEGNVASF--E--TSEILRKTGVMYRHLVCKRNMTSIKD---
AoIRX9L         NRMAHTLKVVPPPLLWIVVEMSSQLT--E--TAKILREAGVMYRHLVCKENVTSIGN---
SmIRX9L         TRLAHTLKLVDPPLLWIVVEMPGQSL--E--TASLLRKTGVMYRHLACEKNLTNVKD---
PpIRX9          TRLAHTLKLVPPPLLWLVVEMHYQSL--E--TAQLLRETGIMYRHLVCDKNLTNVKD---
OsIRX9          TRTAHALRLVSPPLLWLVVEEAPAEKHAAPPTARLLRRTGVVHRHLLMKQGDDDFSMQIS
LmIRX9          TRMAHALLLVPPPLLWVVVEAAPDVP--A--TARLLRDTGLLYRHLTYKDNFTAAEVAAG
PtIRX9          RRLANTIRLVPPPLLWIVVEGQSDSD--E--VSEILRKTGIMYRHLVIKENFTDPEA---
AtIRX9          RRMANTLRLVPPPLLWIVVEKHSDGE--EKSSSTMLRKTGIMYRRIVFKEDFTSLES---
AoIRX9          RRLGNTLKLVSSPLLWIIVEASNEAS--N--NAELLRTTGIMYRHLTYKENFTDAKA---
                 * ...:  *  *:**::.:            : :** :*:::*::  .

OsIRX9L         -IVVCQKNNAIFHIKKHRLDGIVHFADEERAYSADLFEEMRKIRRFGTWPVAIHVGTKYR
AtIRX9L         -RGVHQRNTALEHIELHKLDGIVYFADDDNIYSLELFQSLRQISRFGTWPVAMLAQSKNK
AoIRX9L         -REAHQRNVALSHIEKHQLDGIVHFADDDRMYSVELFGQMRQIRRFGAWPVATLSESKNK
SmIRX9L         -RGTQQRNLALQHIERHQLDGIVYFADDDNFYSLELFDQLREIKRFGTWPVAMLAHSKSK
PpIRX9          -RAASQRNTALAHIEHHELDGIVYFADDGNMYTLELFEQMRNITRFGTWLVGILAPGKSR
OsIRX9          MRREQQRNVALRHIEDHRIAGVVLFGGLADIYDLRLLHHLRDIRTFGAWPVATVSAYERK
LmIRX9          KERHHQRNAALEHIERHRLAGVVHFAGLGDVFQLRFFDQIRQISTFGAWPVARMSRGERK
PtIRX9          -ELDHQRNVALRHIEQHRLSGIVHFAGLSNVYDLGFFDELRQIEVFGTWPVALLSANKNK
AtIRX9          -ELDHQRNLALRHIEHHKLSGIVHFAGLNNIYDLDFFVKIRDIEVFGTWPMALLSANRKR
AoIRX9          -EADHQRNVALNHIEYHRLNGIVHFAEVSNTYNLEFFDEIREIDTFGTWPMAMMSANRRR
                     *:* *: **: *.: *:* *.     :   ::  :*.*  **:* :.     . :

OsIRX9L         VVLEGPVCKGN-----QVTGWHTNQRRGV----------------------SRRFPIGF
AtIRX9L         AILEGPVCNGS-----QVIGWHTNEKSKR----------------------LRRFHVDM
AoIRX9L         VLLEGPVCNGS-----QVIGWHTNQRSKL----------------------SQRFHLDM
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SmIRX9L      TILEGPVCDGH-----KVTGWHTNEKSKR-----------------------LRRFHVDM
PpIRX9       VVFEGPVCEGE-----KVIGWHTSDRSKR-----------------------LRRFHVDM
OsIRX9       VMVQGPLCINTSSSSVITRGWFDMDMDMAAGG-----------ERRAAADRPPPETLMEV
LmIRX9       VLVQGPACSAS-----KVIGWFSRDLSSSTTVSTGQATPTGGTPAGAGAARPSSAPEIDV
PtIRX9       VTIEGPVCDSS-----QVIGWHLKKMNNET----------------------DKRPPIHI
AtIRX9       VVVEGPVCESS-----QVLGWHLRKINNET----------------------ETKPPIHI
AoIRX9       VVVEGPMCRSS-----KVVGWQLKDLSNHTSDTSN---PIPSPDANNGGIAERKHARINI
             . .:** *         . **   .                                : .

OsIRX9L      SGFAFNSTILWDPQRWNS-PTLESIIVHSGGRGGLQESRFIEK-LVEDES--QMEGLG-D
AtIRX9L      SGFAFNSTILWDPKRWRR-PFSHPTRQLDTVKEGFQETSFIEQ-VVADES--EMEGVP-P
AoIRX9L      SGFAFNSTVLWDIKRWHR-PTLEPIRQRHSSSEGFQATTFIEQ-LVEDES--QMEGLL-S
SmIRX9L      SGFGFNSTILWDPRRWKR-PTTQPVRQLDTVKEGFQETTFIEQ-LVEDEN--QMEGLP-S
PpIRX9       SGFAFNSTMLWDPRRWKR-PTLEPIRQLDSIKESSQQTSFIEQ-LVPDES--YMEGRP-P
OsIRX9       GGFAFSSWMLWDPHRWDRFPLSDPDASQESVK-------FVQRVAVEEYNQSTTRGMPDS
LmIRX9       HGFAFNSSVLWDPERWGRYPTSEPDKSQDSIR-------FVQQVVTEDYS--KVKGIP-S
PtIRX9       SSFGFNSSILWDPERWGR-PSSVQQTSQNSIK-------FVKQAALEDET--ELKGIPPE
AtIRX9       SSFAFNSSILWDPERWGR-PSSVEGTKQDSIK-------YVKQVVLEDDT--KLKGLPAQ
AoIRX9       SGFAFNSSILWDPERWGR-PSSVLDTSQDSIK-------FVQEVVLEDES--KLKAIP-S
              .*.*.* :*** .**                       ::..    : .    ..

OsIRX9L      NCTRVMVWNFELEPPQ-VNYPIGWLLQRNLDAVVPIT-------
AtIRX9L      ACSSILNWHLHLDALD-VPYPQGWAIQKNLQALITMK-------
AoIRX9L      NCSKILVWHLHLEAPE-LLNPEGWSIQKNLELVVPLT-------
SmIRX9L      GCSKIMVWHLHLESQI-ASYPSNWKMDKFLEPVIHL--------
PpIRX9       GCLKIMVWHLQLEAPKGFPYPARWTLTTPLEANIPLRKANLVQL
OsIRX9       DCSQIMLWRIQTTL------------------------------
LmIRX9       DCSEIMVWRVEAPSSSRQAPGN--NNRR----------------
PtIRX9       DCSKIMLWRLNLPVSKSPSYHL--STTGSTDASRRKI-------
AtIRX9       DCSKIMLWRLKFPTRTRLST------------------------
AoIRX9       DCSKIMLWHLHIPRTLTASPSS--SQSKNRR-------------
              *  :: *..
```

Supplemental Figure 1: GT43 IRX9/9L protein sequence alignments from various plant species.
*= amino acid residues conserved across all species, := highly conserved across most species, . = conserved across some species. Grey shading indicates conserved cysteine (C) residues potentially involved in disulphide bonding. Transmembrane helices (TMHs) predicted by ARAMEMNON with high confidence (>0.85) for Arabidopsis and rice sequences are marked with a box. The red D's indicate the aspartic acid residues targeted for site-directed mutagenesis. IRX9/9L sequences from moss (*Physcomitrella patens*, *Pp*; Genbank accession: XP_001751575.1), spikemoss (*Selaginella moellendorfii, Sm*; Genbank accession: XP_002963107.1), black cotton wood (*Populus trichocarpa, Pt*; Genbank accession: XP_006381488.1), Arabidopsis (*Arabidopsis thaliana, At*; Genbank accessions: NP_181246.1, NP_973922.1), rice (*Oryza sativa, Os*; Genbank accessions: NP_001060738.1, NP_001043846.1), asparagus (*Asparagus officinalis, Ao*; Genbank accessions: NP_181246.1, AJF38259.1) and ryegrass (*Lolium multiflorum, Lm*; DDBJ accession: LC127415).

```
SmIRX14         MKSGSAA--AGGVLHPVLRRSLTNAASTPRKSSGSSS------PE----------RGGGG
PpIRX14         ------------------------------------------------------------
AoIRX14B        -----------------MKLSLLPQTYFPRRTNSLPSAE-------------------TT
OsIRX14         -----------------MMKSLLP-QSQLRRSAAAASAARSSGGGAGSGGADGAGSDGGA
LmIRX14         -----------------MMKSLLP-QSQLRRSAAASSAARSSAGAVEG-----AAAASDG
AoIRX14A        MASSITLVASSSTSRLPSKKTHLP-TPLPKPSLSLPKPSPSLPP------AL-TAAAAAV
PtIRX14         MKL--SMLQQ---------------SYMNRRSASFRGSSAPL-------------DSSTD
AtIRX14L        MKL--SVFRL---------------SYWNRRGSSFRSSP-SL-------------DPSFD
AtIRX14         MKL--SALHQ---------------SYLNRRSNSFRSPT-SL-------------DSSVD


SmIRX14         GVLIGPGPLIRLCLHAVACIASLVIGFRFSRETLLVLLIDVGIKF--PYVEPGL------
PpIRX14         ------------------------------------------------------------
AoIRX14B        SPKPSPASISYLLLHFLCCLISLILGFRLSRMVFFLLFSSSTTIITTVT-----------
OsIRX14         GGRAPATSTFWFLLHALCCLVSLFLGFRFSRLLFFLLFSTTALYSSTS--SSSSSAVLRA
LmIRX14         AAKPSSSSTFWFLLHALCCLISLFLGFRFSRLLFFLLFSTSALYAASNTNNPSSSAVLRA
AoIRX14A        AADKSPSSLFWLALHSLCCLASLALGFRFSRLLFFLLFSNPSPNPNPDQNHVAA----PV
PtIRX14         NTIKSPAAIFWLLLHGFCCLISLVLGFRFSRLVFFFLFSTSTTTT--LYIATPL----PH
AtIRX14L        GK--SPSSVFWFVIHGLCCLISLILGFRFSHLVLFFLFSTSVTN---LYTTPFL----FA
AtIRX14         GSGKSLIAVFWLILHCLCCLISLVLGFRFSRLVFFFLFSTSSTN---LYSLPFR----PD


SmIRX14         -GVRD------GV---------------------QDGGGRLPEVRTKSGRVHVGRHEIL
PpIRX14         -------------------------------------------------SSQVYVGRHPIL
AoIRX14B        --------TTTTTLTQTLTLPLL------------------PSPHNKTQSGVAVGRHGIL
OsIRX14         TTTTTTTTTTTTTTTTNTFTLSFQANPNPPPSNLSNHTALDAAGAAGHTQSHVVVGRHGIR
LmIRX14         ITTTTTTTTTTTTTTTNTFTLSFNPSTPPPPTTTNHTPTL-----EDATQSHVVVGRHGIR
AoIRX14A        LP-SRTL---------------------------TLNLPQKLAPNLTASRVVVGRHGIL
PtIRX14         LT-KTN--------NN--INDL-------------PLEIPVINKTLSSSSRVVVGRHGI-
AtIRX14L        GN-GG----------VSQLLRL-------------KPLETATNSTVKKNSRVVVGRHGIR
AtIRX14         LP-VKHL----DVHTIGRTLDP-------------GANGTTVVATATKSSRVVVGRHGIR
                                                                 . * **** *

SmIRX14         IRPWPHPDSAEIYRAHALLERVQVEQQS-LYGPKERKTVIAITPTFARTFQAIHLLGVMH
PpIRX14         KRPWPHPDPIEMAQAYNMLARVQLEQQR-LYGIENWKPIIAITPTYFRTFQSLHLSGLMH
AoIRX14B        IRPWPHPDPDQTMRAHRLIERVQKEQRTVYYGIKKPKPLIVITPTYVRTFQAMYLTGLMH
OsIRX14         IRPWPHPDPVEVMRAHRIMERVQEEQRR-WYGVKEPRHVLVVTPTYSRAFQALHLTGLLH
LmIRX14         IRPWPHPDPVEVMRAHRIMDRVQEEQRR-WYGVREPRHVLAVTPTYHRAFQALHLTGLLH
AoIRX14A        IRPFPHPDPTETMKAHRIMERVQQEQRA-QYGVKNPRPVVVITPTYVRTFQTLHLTGLMH
PtIRX14         RRPWPHPNPSEVMKAHQIIETVQREQRT-QFGVKSPRTLIVVTPTYVRTFQTLHLTGVMH
AtIRX14L        IRPWPHPNPIEVLRAHQLLVRVQKEQKS-MYGVRSPRTVIVVTPTYVRTFQALHLTGVMH
AtIRX14         IRPWPHPNPVEVMKAHQIIGRVQKEQKM-IFGMKSSKMVIAVTPTYVRTFQALHLTGVMH
                 **:***:  :  :*: ::  ** **:   :* .. : ::.:***: *:**:::* *::*

SmIRX14         TLRAAPGPVIWIVVEAGGRSNETASILASSRL-EFVHLGVKD--AMPVAWEQRRRMETRL
PpIRX14         TLSLVRRPVTWIVIEASGISAETAELLRQVRVHKLVHLGASE--HLPRTLQDRIILEARL
AoIRX14B        SLMLVPFDFTWLVVEAGGVSNETASILDRSHL-NYVHVPFDEGKKMPMEWGERRKMEARM
OsIRX14         SLRNVPYPLTWIVVEAGGTTNATASLLARSDL-TIVHIPFPD--RMPHDWADRHATENRM
LmIRX14         SLRNVPYPLTWLVVEAGGVTNATAELLARSGL-TVVHVPFPD--RMPLDWPERHATENRM
AoIRX14A        TLMLVPYDLTWIVVEAGGISNETADLIERSKI-KTIHLGFER--GMPLKWPDRHKMEALM
PtIRX14         SLMLVPYDVVWIVVEAGGATNETASIIAKSSI-KTFHIGFTQ--KMPNSWEGRHKLETKM
AtIRX14L        SLMLVPYDLVWIVVEAGGITNETASFIAKSGL-KTIHLGFDQ--KMPNTWEDRHKLETKM
AtIRX14         SLMLVPYDLVWIVVEAGGATNETGLIIAKSGL-RTIHVGIDQ--RMPNTWEDRSKLEVFM
                :*  .   . *:*:**.* :  *. ::    :   .*:       :*     *   *  :

SmIRX14         RIEGLSHVRREKLDGLILFADDSNVHSLQLFDEIQKVKWIGALSVGLLETGSGATETASS
PpIRX14         RTEGLRYVREQNLEGVIVFADESNVYSMQFFDEVQKVKWVGALPVGTLGYAGFEDPALL-
AoIRX14B        RLHALRVVREKKMDGIVVFADDSNIHSTEMFDEIQKVKHIGAISVGFLTHSDI-------
OsIRX14         RLHALRVIRERKMDGVIVFADDSNVHSLELFDEVQKVQWMGAVSVGILAHTGTADQPRL-
LmIRX14         RLHALRVIRERKMDGVVVFADDSNVHSMELFDEAQKVQWMAAVSVGILAHTGAAEQPRL-
AoIRX14A        RIQALRVVKERKLDGIVVFADDSNMHSLELFDEIQKVKWIGALSVGILAHSGNSEPLVQN
PtIRX14         RLRA-RVVREEMMDGIVMFADDSNMHSMELFDEIQNVKWFGAVSVGILAHSGGGGESSSA
AtIRX14L        RLHALRVVREKKLDGIVMFADDSNMHSMELFDEIQTVKWFGALSVGILAHSGNADELSSI
AtIRX14         RLQALRVVREEKLDGIVMFADDSNMHSMELFDEIQNVKWFGTVSVGILAHSGNAEEMVLS
                * ..   ::.. ::*:::***:**::* ::*** *.*: ..:: ** *

SmIRX14         MV------AAASSAKPRLPVQGPACNETCHVVGWHVLRPSPVDGEDDSSSSSFTDVAGGL
PpIRX14         --------RDKVSRNTVLQVQGPTCDSSENITGWRAFRPLSLDDVLINE------YRD--
AoIRX14B        -----------EEENHSIPIQGPECTPSGKFVGMQTLDPS--------------------
OsIRX14         --------SEEDKQNMPLPVQGPACNSSGHLAGWHTFNSLPFAGKTATV------VGEAA
LmIRX14         --------TEDDRKSMPLPVQGPACNASGQLAGWHTFNTLPFSGKTAAV------VGEAA
AoIRX14A        EKK-----SEEDKENMPLPIQGPACNSSNQLVGWHTFNSLPFMENIATT------VGDVG
PtIRX14         VAEK----VKPNLSNPAMPVQGPACNASNKLVGWHTFNSLPYEGKSAVY------IDDRA
AtIRX14L        LKNE----QGKNKEKPSMPIQGPSCNSSEKLVGWHIFNTQPYAKKTAVY------IDEKA
AtIRX14         MEKRKEMEKEEEEESSSLPVQGPACNSTDQLIGWHIFNTLPYAGKSAVY------IDDVA
                              .  : :*** *  : .. * : :
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SmIRX14       TDVATHLEWSGFVINSRAVWDEAESENEEEEASSSTMEDEQQEEDATRRKPEWINEWKEW
PpIRX14       --EQTNLEWSGFVLNARTVWASA------------------------PDRPKWIREWVEW
AoIRX14B      ---SNKLEWAGFVLSSRLLWENV------------------------EERPTWVQNIDD-
OsIRX14       PVLPRGLEWAGFVLNSRILWKEA------------------------EGKPDWVKDLDAV
LmIRX14       PVLPRGLEWAGFVINSRMLWKEA------------------------EGKPEWVKDLDAV
AoIRX14A      TVLPTKIEWAGFVLNSKLVWGED------------------------EGKPEWVRDLDSV
PtIRX14       TVLPRKLEWAGFVLNSRLLLKEA------------------------QDKPEWVKDLDLV
AtIRX14L      PVMPSKMEWSGFVLNSRLLWKESL-----------------------DDKPAWVKDLSLL
AtIRX14       AVLPQKLEWSGFVLNSRLLWEEA------------------------ENKPEWVKDFGSL
                    :**:***:.:: :  .                           :* *:.:

SmIRX14       IS---EDSGSPLRLAKDQGFIEALGNCG--REVMLWWIRVEARADSRYPPRQVALCPTRG
PpIRX14       ARPEQRRYIDPRSLLSDETKVETLGSCGNGKAVLVWWARIEARSDSKYPP----------
AoIRX14B      -----LEIESPLALLKDASYVEPLGNCG--KKILVWWIRSEARADSKFPA----------
OsIRX14       GE-NGEEIENPLILLNDPSSVEPLGNCG--KKILLWWLRVEARADSKFPQ----------
LmIRX14       GE-NGEEVENPLTLLNDASFVEPLGNCG--KKILLWWLRVEARADSKFPA----------
AoIRX14A      GL-NGEEIESPLALLKEASGVEPLGSCG--KKVMLWWLRVEARADSKFPP----------
PtIRX14       ----DENIESPLALLKDPSMVEPLGSCG--RQVLLWWLRVEARADSKFPP----------
AtIRX14L      DD-GYAEIESPLSLVKDPSMVEPLGSCG--RRVLLWWLRVEARADSKFPP----------
AtIRX14       NE-N-EGVESPLSLLKDPSMVEPLGSCG--RQVLLWWLRVEARADSKFPP----------
                       .*  * .:   :* **.**  : :::** * ***:**::*

SmIRX14       YHIYQLTRLPHRWSLTSPLEITVPAQRTPWPE----------------------------
PpIRX14       -----------RWNLDLPLEVVVPARKTPWPE----------------------------
AoIRX14B      -----------GWVIDSPSKSPLPSGHSSSTSDYLTRLRHDKMNNT--------------
OsIRX14       -----------GWVIEPPLDIVVPAKRTPWPE-TTAELSAELVDSKQDQEGRRLS-----
LmIRX14       -----------GWIIEPPLEVVIPAKRTPWPE-TTTEVSSEQLEGKQEQEDRRLS-----
AoIRX14A      -----------GWIINPPLDITVPAKRTPWPD-APPQLPSEKLPIDQDP---AEK-----
PtIRX14       -----------GWIIDPPLEITVPSKRTPWPD-APPELPSNKKLTINQE--QTIK-----
AtIRX14L      -----------GWIIKSPLEITVPSKRTPWPD-SSSELPAAAIKEAKSN-----------
AtIRX14       -----------GWIIDPPLEITVAAKRTPWPD-VPPEPPTKKKDQMPLSQGNTVVVIPKQ
                          * :  * .  : : ::   .

SmIRX14       ---------------------------------------------
PpIRX14       ---------------------------------------------
AoIRX14B      ------------------NKEHMERHLDGKIYLRRD---------
OsIRX14       -----RTDRSSRSRSTTKRKEN-----------------------
LmIRX14       -----RTNRSARPRSTTTKRKG-----DVQN--------------
AoIRX14A      --HPSKSGRSRSRHSSRSKRKHEQRAGNAQITGATP---------
PtIRX14       --RSPKTR--SPRSKRRSKRKHEAKLVETQVS-----TRHSEQN-
AtIRX14L      ----SKPR--VSKSKSY-KEKQEPKAFDGVKVSATS---------
AtIRX14       QQHPTKIR--KPKRKSK-KSKHEPRPTDTTTQVYSSSSKHQERN-
```

Supplemental Figure 2. GT43 IRX14 protein sequence alignment from various plant species.

See supplemental Figure 1. legend for annotations. IRX14/14L sequences from moss (*Physcomitrella patens*, *Pp*; Genbank accession: XP_001778969.1), spikemoss (*Selaginella moellendorfii, Sm*; Genbank accession: XP_002980520.1), black cotton wood (*Populus trichocarpa, Pt*; Genbank accession: AEG25426.1), Arabidopsis (*Arabidopsis thaliana, At*; Genbank accession: NP_195407.2), NP_201524.1, rice (*Oryza sativa, Os*; Genbank accession: Q653F4.1), *asparagus (Asparagus officinalis, Ao*; Genbank accessions: *AJF38261.1, AJF38262) and ryegrass (Lolium multiflorum, Lm*; DDBJ accession: *LC127417).*

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PpIRX10       MEHPLECADSCSLAMSWFCNKKCRGWGLMKRTVVASGLRSVVLLLLFIYFVQDVTAEMGH
LmIRX10       ----------------------------MEKPRVALAL--AAALALLLSCSDVVVRAQDT
SmIRX10       ------------------------------------MGAAWIALALGLALCASAEQQQQH
OsIRX10       ----------------------------MRRWVLAIAILAAAVCFFLGAQAQEVRQGHQT
AtIRX10       ----------------------------MKIHSCLSAILLFLF-----FSASSAKQNVRT
AtIRX10L      ----------------------------MKLSSCVL--IFLLCNTFSSISAFRLSRSQPT
AoIRX10       ----------------------------MRLGIWVFSILLF-SALIAEIEAQEIERRHRT
PtIRX10       ----------------------------MEIWKWVFVGVLWYAGFVLSTGAVELGRSQHT


PpIRX10       QRISGSAGDVLEDNPVGRLKVFIYDIPSKYNTDWLKKDPRCLTHMFAVEEYLHDFLTESP
LmIRX10       ERIEGSAGDVLEDDPVGRLKVYIYELPTKYNKKMVAKDSRCLSHMFAAEIFMHRFLLSSA
SmIRX10       ERISGSAGDVLEDDPVGRLKVFVYELPVKYNRKVLEKDPRCLTHMFATEIFVHEFLLGSA
OsIRX10       ERISGSAGDVLEDDPVGRLKVYVYDLPSKYNKKLLKKDPRCLNHMFAAEIFMHRFLLSSA
AtIRX10       ERISGSAGDVLEDDPVGKLKVYVYELPSKYNKKLLQKDPRCLTHMFAAEIFMHRFLLSSP
AtIRX10L      ERISGSAGDVLEDDPVGRLKVFVYELPSKYNKKILQKDPRCLNHMFAAEIYMQRFLLSSP
AoIRX10       ERISGSAGDVLEDDPVGRLKVFVYELPSKYNKKILQKDQRCLNHMFAAEIFMHRFLLSSP
PtIRX10       ERISGSAGDVLEDDPVGRLKVFVYELPRKYNKKILQKDPRCLTHMFAAEIFMHRFLLSSP
              :**.*********:***:***::*::* *** . : ** ***.****.* ::: **  *

PpIRX10       VRTLNPEEADWFYTPVYTTCDLTPNGLPLPFKSPRVMRSAISYISSHWPYWNRTDGADHF
LmIRX10       TRTMNPEEADWFYTPVYTTCDLTPWGHPLPFKSPRIMRSAVQFISSHWPYWNRTEGADHF
SmIRX10       VRTLNPEEADWFYTPVYTTCDLTPNGLPLPFKSPRMMRSVIQYISNQWPYWNRTEGADHF
OsIRX10       VRTFNPEEADWFYTPVYTTCDLTPSGLPLPFKSPRMMRSAIELIATNWPYWNRSEGADHF
AtIRX10       VRTRNPDEADWFYTPIYPTCDLTPTGLPLPFKSPRMMRSSIQLISSNWPYWNRTEGADHF
AtIRX10L      VRTLNPEEADWFYVPVYTTCDLTPNGLPLPFKSPRMMRSAIQLIASNWPYWNRTEGADHF
AoIRX10       VRTLNPEEADWFYTPVYTTCDLTPNGLPLPFKSPRMMRSAIQLISSNWPYWNRTEGADHF
VvIRX10       VRTLNPEEADWFYTPVYTTCDLTPNGLPLPFKSPRMMRSAIQLISSNWPYWNRTEGADHF
PtIRX10       VRTLNPDEADWFYTPVYTTCDLTTNGLPLPFKSPRMMRSAIQLISSNWPYWNRTEGADHF
              .** **:******.*:* *****  * ********:*** :. *:..******::*****

PpIRX10       FVVPHDFAACFHYQEEKAIERGILPLLKRATLIQTFGQNHHVCLKEDSIVIPPYAPPERM
LmIRX10       FVVPHDFGACFHYQEEKAIERGILPLLRRATLVQTFGQKDHVCLKEGSINIPPYAPPQKM
SmIRX10       FVVPHDFGACFHYQEEKAIERGILHLLQRATLVQTFGQRYHVCLKEGSIVVPPYCPPQKM
OsIRX10       FVTPHDFGACFHYQEEKAIGRGILPLLQRATLVQTFGQKNHVCLKDGSITIPPYAPPQKM
AtIRX10       FVVPHDFGACFHYQEEKAIERGILPLLQRATLVQTFGQRNHVCLDEGSITIPPFAPPQKM
AtIRX10L      FVVPHDFGACFHYQEEKAIGRGILPLLQRATLVQTFGQRNHVCLKEGSITVPPYAPPQKM
AoIRX10       FVVPHDFGACFHYQEEKAIERGILHLLSRATLVQTFGQRNHVCLKDGSITIPPYAPPQKM
PtIRX10       FVVPHDFGACFHYQEEKAIERGILPLLQRATLVQTFGQRNHVCLKDGSITVPPYAPPQKM
              **.****.*********** **** ** ****:*****. ****.: ** :**:.**::*

PpIRX10       QTRLNPPSTPRSIFAYFRGLFYDPGNDPEGGYYARGARAAIWENFKDNPLFDISTEHPAT
LmIRX10       KTHLVPPETPRSIFVYFRGLFYDTANDPEGGYYARGARASVWENFKNNPLFDISTDHPPT
SmIRX10       QAHLIPPSIPRSIFVYFRGLFYDYGNDPEGGYYARGARAAVWENFKDNPLFDISTEHPIT
OsIRX10       QAHLIPPDTPRSIFVYFRGLFYDTSNDPEGGYYARGARASVWENFKNNPLFDISTDHPPT
AtIRX10       QAHFIPPDIPRSIFVYFRGLFYDVNNDPEGGYYARGARAAVWENFKNNPLFDISTDHPTT
AtIRX10L      QSHLIPEKTPRSIFVYFRGLFYDVGNDPEGGYYARGARAAVWENFKDNPLFDISTEHPTT
AoIRX10       QAHLIPPDTPRSIFVYFRGLFYDVGNDPEGGYYARGARASVWENFKDNPLFDISTEHPTT
PtIRX10       QTHLIPEKTPRSIFVYFRGLFYDVGNDPEGGYYARGARAAVWENFKDNPLFDISTEHPAT
              :::: * . *****.********  **************::*****:********:** *

PpIRX10       YYEDMQRAIFCLCPLGWAPWSPRLVEGVIFGCIPVIIADDIVLPFADAIPWEKIGVFVEE
LmIRX10       YYEDMQRAIFCLCPLGWAPWSPRLVEAVVFGCIPVIIADDIVLPFADAIPWDEIGVFIAE
SmIRX10       YYEDMQRAIFCLCPLGWAPWSPRLVEGVIFGCIPVIIADDIVLPFADAIPWEEIGVFVAE
OsIRX10       YYEDMQRSVFCLCPLGWAPWSPRLVEAVVFGCIPVIIADDIVLPFADAIPWEEIGVFVAE
AtIRX10       YYEDMQRAIFCLCPLGWAPWSPRLVEAVVFGCIPVIIADDIVLPFADAIPWEEIGVFVAE
AtIRX10L      YYEDMQRAIFCLCPLGWAPWSPRLVEAVIFGCIPVIIADDIVLPFADAIPWEDIGVFVDE
AoIRX10       YYEDMQRAIFCLCPLGWAPWSPRLVEAVVFGCIPVIIADDIVLPFADAIPWEDIGVFVAE
PtIRX10       YYEDMQRAVFCLCPLGWAPWSPRLVEAVIFGCIPVIIADDIVLPFADAIPWEEIGVYVDE
              *******::*****************.*:**********************:.***:: *

PpIRX10       KDVPILDKILCTINHEEVLEKQRLLANPAMKQAMLFPRPAKPGDAFHQILNGLARKLPHD
LmIRX10       DDVPKLDTILTSIPMEVILRKQRLLANPSMKQAMLFPQPAQPGDAFHQILNGLARKLPHD
SmIRX10       KDVPKLDTILTSIPPEVILKKQRLLATPAMKQAMLFPQPAQPGDAFHQILNGLARKLPHD
OsIRX10       EDVPKLDSILTSIPTDVILRKQRLLANPSMKQAMLFPQPAQAGDAFHQILNGLARKLPHG
AtIRX10       KDVPELDTILTSIPTEVILRKQRLLANPSMKRAMLFPQPAQPGDAFHQILNGLARKLPHD
AtIRX10L      KDVPYLDTILTSIPPEVILRKQRLLANPSMKQAMLFPQPAQPGDAFHQVLNGLARKLPHE
AoIRX10       KDVPNLDTILTSISPDVILRKQRLLANPSMKQAMLFPQPAQSGDAFHQVLNGLARKLPHD
PtIRX10       EDVPNLDTILTSIPPEVILRKQRLLANPSMKQAMLFPQPAQPGDAFHQVLNGLARKLPHD
              .*** **.** :*  : :*.******.*:**:*****:**: ******:**********

PpIRX10       PSIYLQPGQSFLNWTEGPPGDLYPWGNDL------------------------
LmIRX10       KGVYLTPGQKALNWTQGPKGDLKPW----------------------------
SmIRX10       RRIYLNPGEKALNWSSGPPGDLKPCKRNTLDRESRNWHQLLQLLRHWCRVLLF
OsIRX10       ENVFLKPGERALNWTAGPVGDLKPW----------------------------
AtIRX10       KSIYLKTGEKALNWTAGPVADLKPW----------------------------
AtIRX10L      RSVYLRPGEKLLNWTAGPVADLKPW----------------------------
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AoIRX10       ESLYLKPGEKLLNWTAGPVGDLKPW----------------------------
PtIRX10       RSVYLRPSEKILNWTAGPVGDLKPW----------------------------
                ::*  .:  ***: ** .** *
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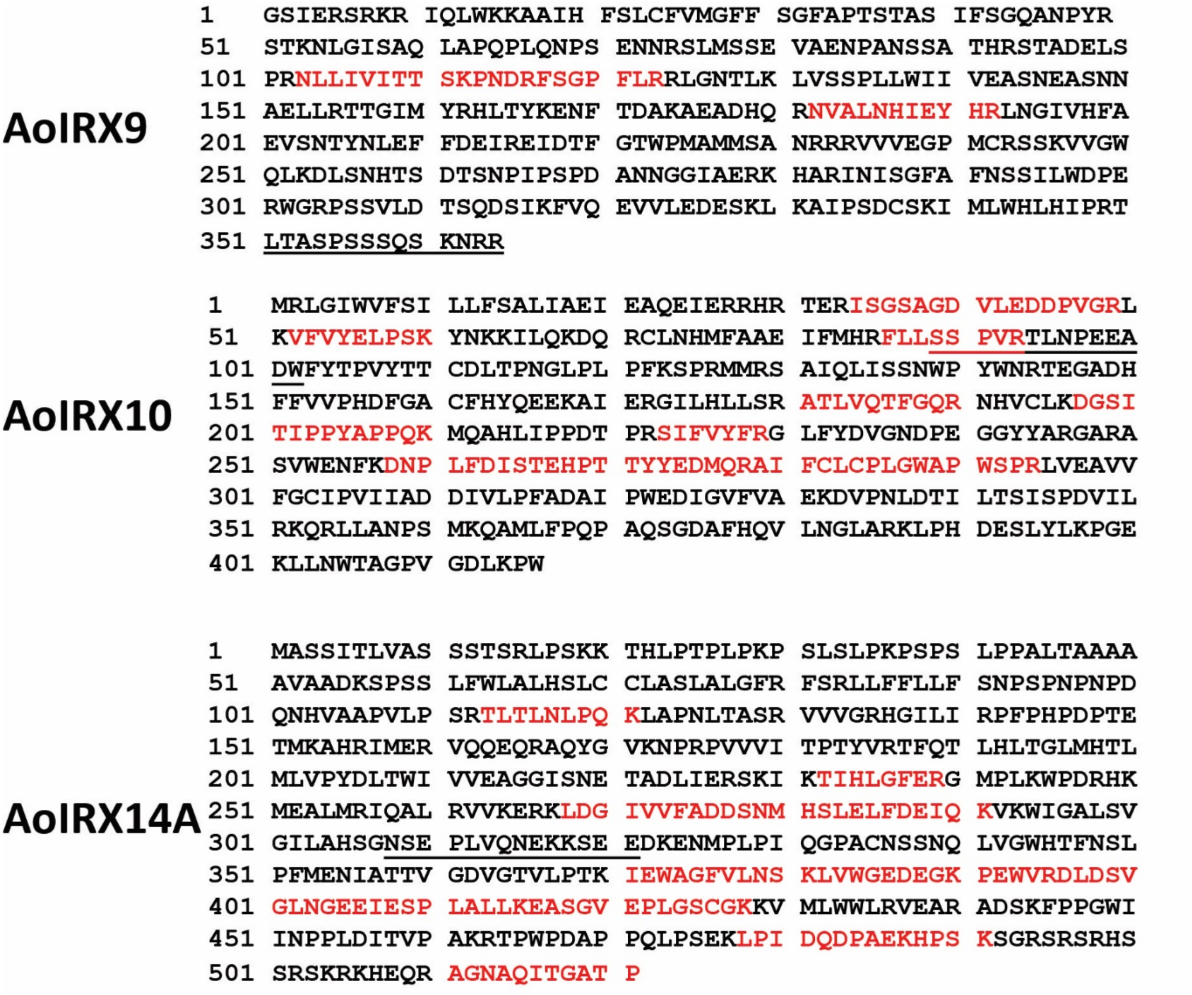
Supplemental Figure 3. GT47 IRX10 protein sequences alignment from various plant species.

See supplemental Figure 1. legend for annotations. IRX10/10Lsequences from moss (*Physcomitrella patens*, *Pp*; Genbank accession: XP_001753186.1), spikemoss (*Selaginella moellendorfii, Sm*; Genbank accession: XP_002993593.1), black cotton wood (*Populus trichocarpa, Pt*; Genbank accession: XP_002318180.1), Arabidopsis (*Arabidopsis thaliana, At*; Genbank accession: NP_174064.1, NP_568941.1), rice (*Oryza sativa, Os*; Genbank accession:Q8S1X7.1), asparagus (*Asparagus officinalis, Ao*; Genbank accession: AJF38260.1) and ryegrass (*Lolium multiflorum, Lm*; Genbank accession: LC127416)



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1   GSIERSRKR IQLWKKAAIH FSLCFVMGFF SGFAPTSTAS IFSGQANPYR
51  STKNLGISAQ LAPQPLQNPS ENNRSLMSSE VAENPANSSA THRSTADELS
101 PRNLLIVITT SKPNDRFSGP FLRRLGNTLK LVSSPLLWII VEASNEASNN
151 AELLRTTGIM YRHLTYKENF TDAKAEADHQ RNVALNHIEY HRLNGIVHFA
201 EVSNTYNLEF FDEIREIDTF GTWPMAMMSA NRRRVVVEGP MCRSSKVVGW
251 QLKDLSNHTS DTSNPIPSPD ANNGGIAERK HARINISGFA FNSSILWDPE
301 RWGRPSSVLD TSQDSIKFVQ EVVLEDESKL KAIPSDCSKI MLWHLHIPRT
351 LTASPSSSQS KNRR

AoIRX10
1   MRLGIWVFSI LLFSALIAEI EAQEIERRHR TERISGSAGD VLEDDPVGRL
51  KVFVYELPSK YNKKILQKDQ RCLNHMFAAE IFMHRFLLSS PVRTLNPEEA
101 DWFYTPVYTT CDLTPNGLPL PFKSPRMMRS AIQLISSNWP YWNRTEGADH
151 FFVVPHDFGA CFHYQEEKAI ERGILHLLSR ATLVQTFGQR NHVCLKDGSI
201 TIPPYAPPQK MQAHLIPPDT PRSIFVYFRG LFYDVGNDPE GGYYARGARA
251 SVWENFKDNP LFDISTEHPT TYYEDMQRAI FCLCPLGWAP WSPRLVEAVV
301 FGCIPVIIAD DIVLPFADAI PWEDIGVFVA EKDVPNLDTI LTSISPDVIL
351 RKQRLLANPS MKQAMLFPQP AQSGDAFHQV LNGLARKLPH DESLYLKPGE
401 KLLNWTAGPV GDLKPW

AoIRX14A
1   MASSITLVAS SSTSRLPSKK THLPTPLPKP SLSLPKPSPS LPPALTAAAA
51  AVAADKSPSS LFWLALHSLC CLASLALGFR FSRLLFFLLF SNPSPNPNPD
101 QNHVAAPVLP SRTLTLNLPQ KLAPNLTASR VVVGRHGILI RPFPHPDPTE
151 TMKAHRIMER VQQEQRAQYG VKNPRPVVVI TPTYVRTFQT LHLTGLMHTL
201 MLVPYDLTWI VVEAGGISNE TADLIERSKI KTIHLGFERG MPLKWPDRHK
251 MEALMRIQAL RVVKERKLDG IVVFADDSNM HSLELFDEIQ KVKWIGALSV
301 GILAHSGNSE PLVQNEKKSE EDKENMPLPI QGPACNSSNQ LVGWHTFNSL
351 PFMENIATTV GDVGTVLPTK IEWAGFVLNS KLVWGEDEGK PEWVRDLDSV
401 GLNGEEIESP LALLKEASGV EPLGSCGKKV MLWWLRVEAR ADSKFPPGWI
451 INPPLDITVP AKRTPWPDAP PQLPSEKLPI DQDPAEKHPS KSGRSRSRHS
501 SRSKRKHEQR AGNAQITGAT P
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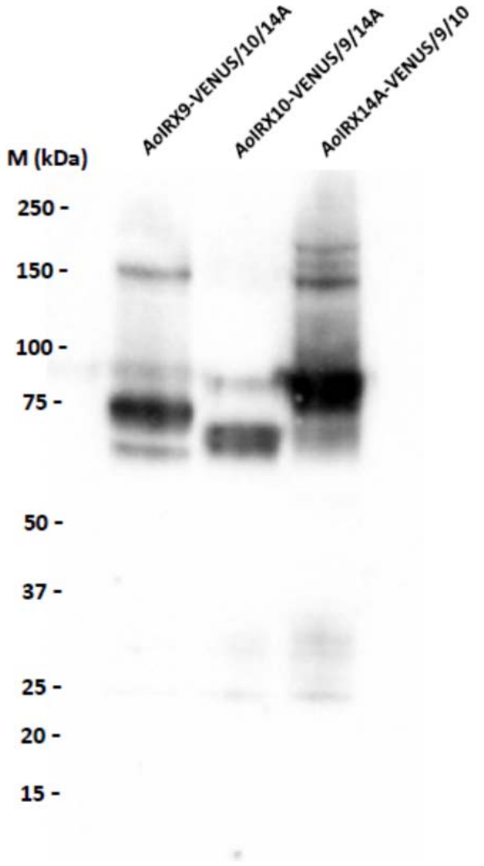
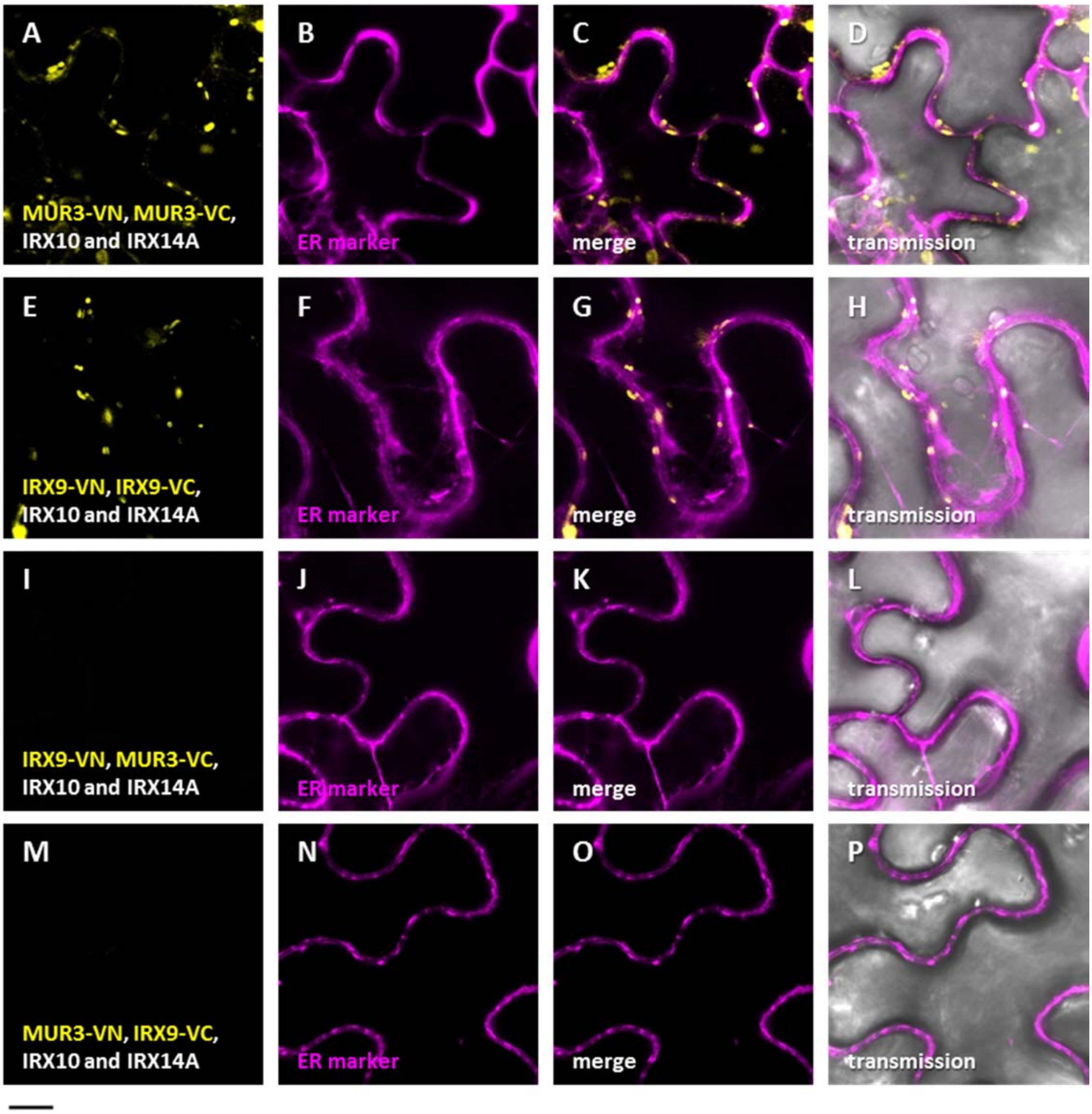
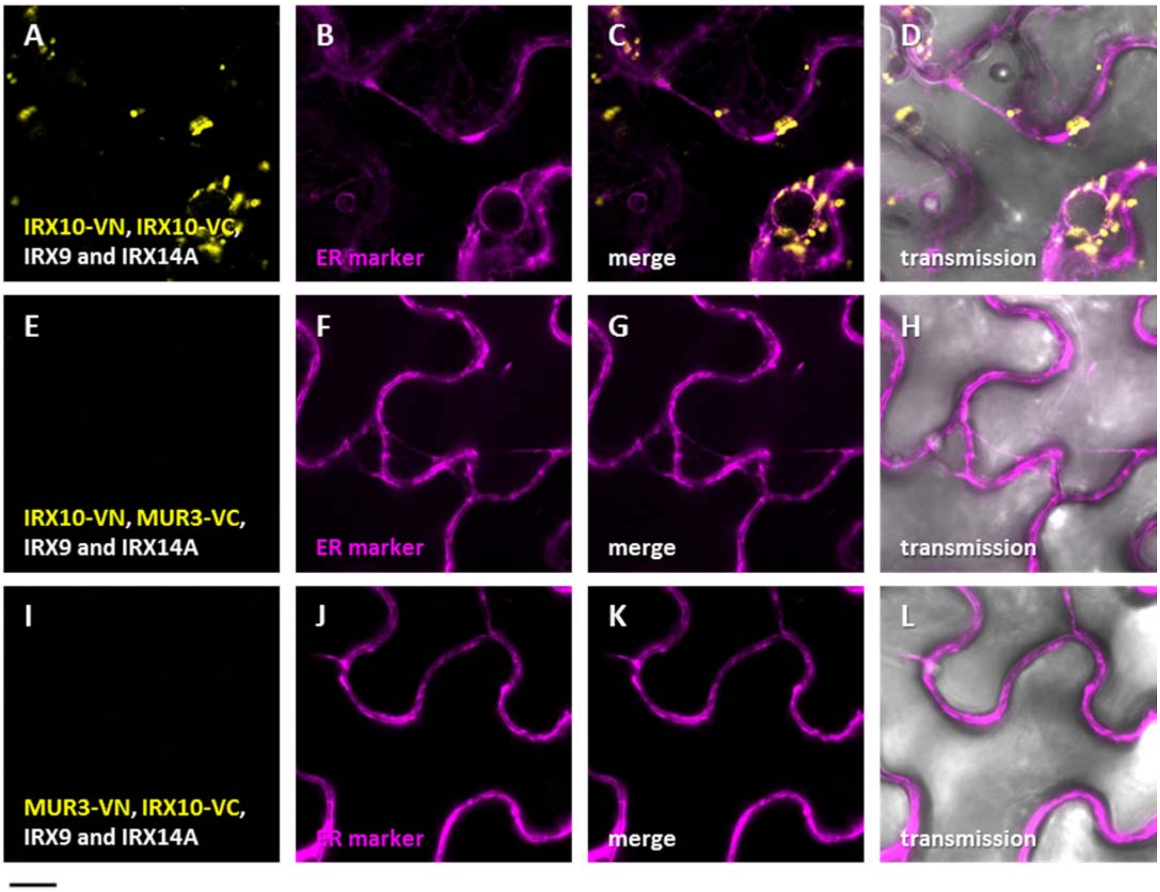
Supplemental Figure 4: Proteomic analysis of proteins co-immunoprecipitated with the *Ao*IRX9 antibody from solubilised microsomal membranes (MMs) of *N. benthamiana* leaves co-expressing *Ao*IRX9, *Ao*IRX10 and *Ao*IRX14A. The underlined regions are the peptide antigens targeted to generate antibodies. The red regions are the tryptic peptides detected via proteomic analysis.



Supplemental Figure 5: Western blot of *N. benthamiana* MMs expressing either *Asparagus* IRX9, IRX10 or IRX14A proteins conjugated with VENUS. MMs extracted from *N. benthamiana* leaves co-expressing Asparagus IRX9, IRX10 and IRX14A. *Ao*IRX9, *Ao*IRX10 and *Ao*IRX14A, respectively, were fused at the COOH-terminus with VENUS. MMs were separated on an SDS-PAGE gel and a GFP antibody used as probe to demonstrate the relative expression level of the three tagged proteins. The bands ~75 kDa in size demonstrate the asparagus IRX proteins are conjugated with VENUS and do not undergo substantial cleavage.

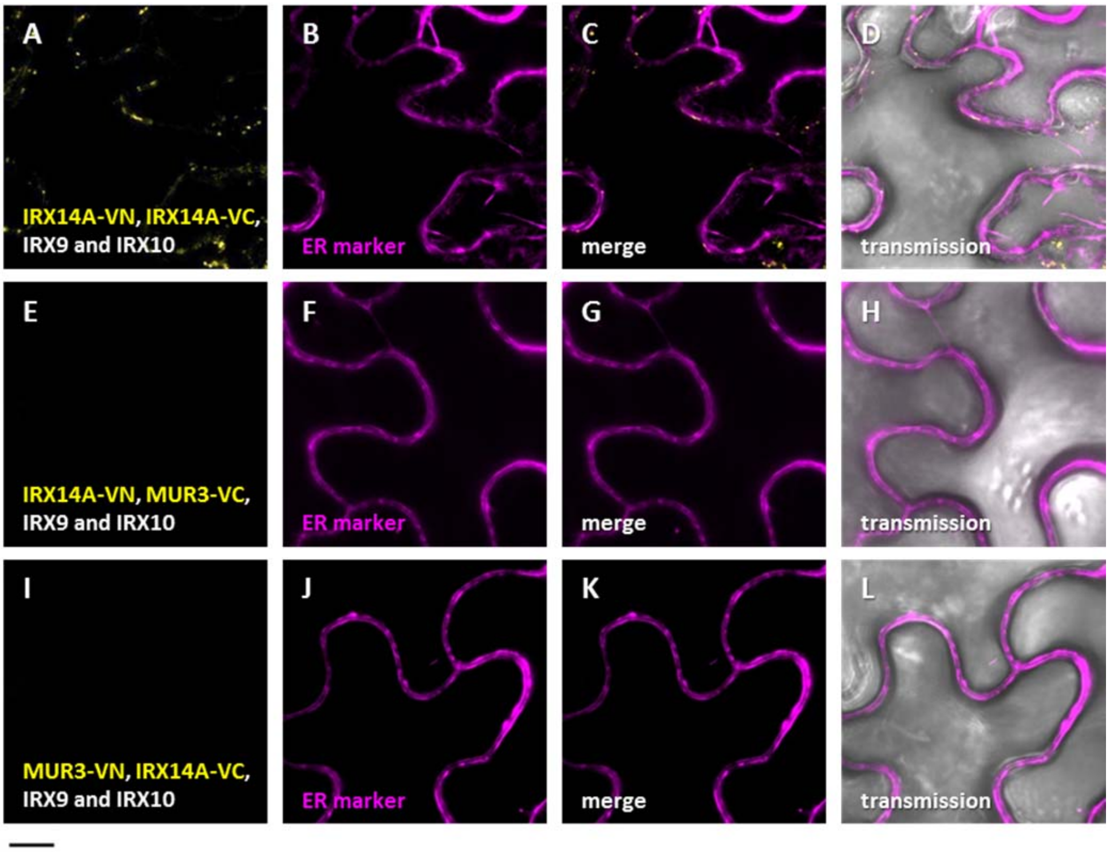


Supplemental Figure 6: MUR3 positive control and expanded images from Figures 9A-C.
MUR3 is known to form homo-dimers *in planta* (Søgaard *et al.*, 2012) and so it was used as a positive control for BiFC experiments. Strong fluorescent signal indicates this system is working (A-D). E-H shows the expanded figures from Figure 9A-C where the IRX9 homo-dimerisation is independent of the ER marker (MANI(49)-RFP). The ER marker was expressed as a transformation control, to be sure that infiltration had occurred and that the fluorescently tagged protein is expressed, even if fluorescence is not observed in the interaction test. Also, MUR3 is not expected to be involved in the xylan synthesis pathway, it was used as a negative control of random interactions with the IRX proteins. IRX9 did not interact with MUR3 as shown by lack of signal in I-P.

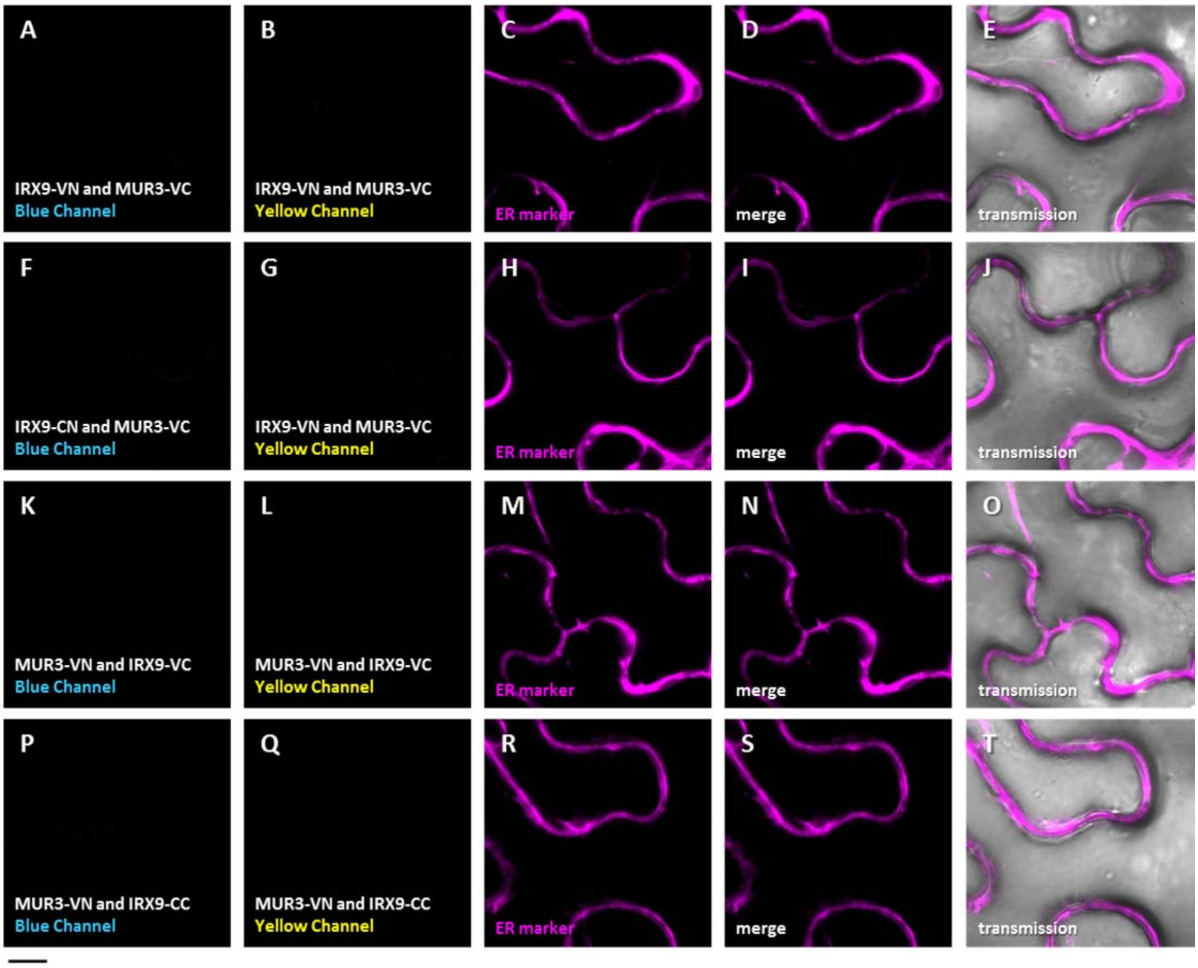


Supplemental Figure 7: Expanded images from Figure 9D-F.

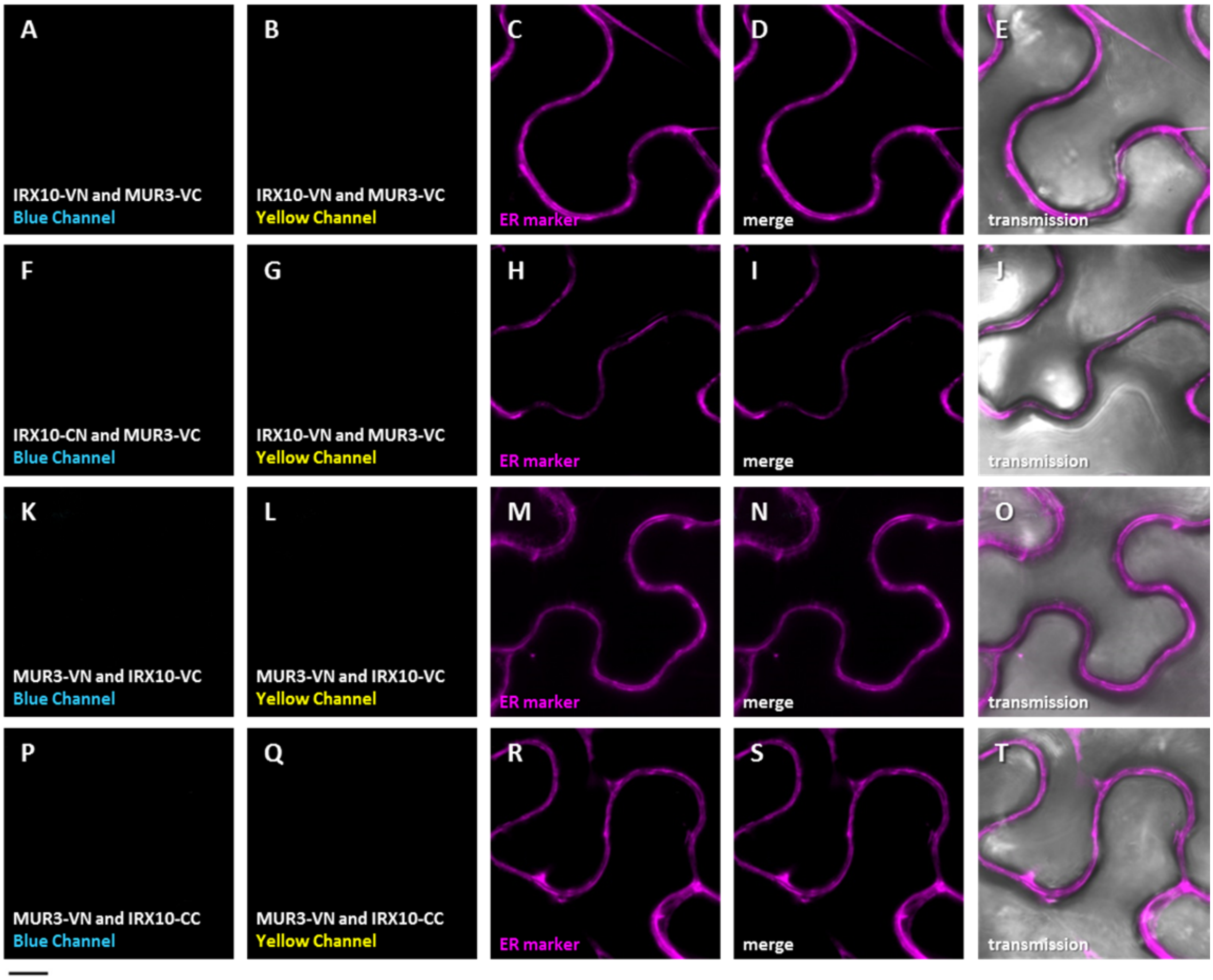
A-D shows the expanded figures from Figure 9-F where the IRX10 homo-dimerisation is independent of the ER marker (MANI(49)-RFP). The ER marker was expressed as a transformation control. MUR3 is not expected to be involved in the xylan synthesis pathway and, as expected, did not interact with IRX10 as shown by lack of signal in E-L.



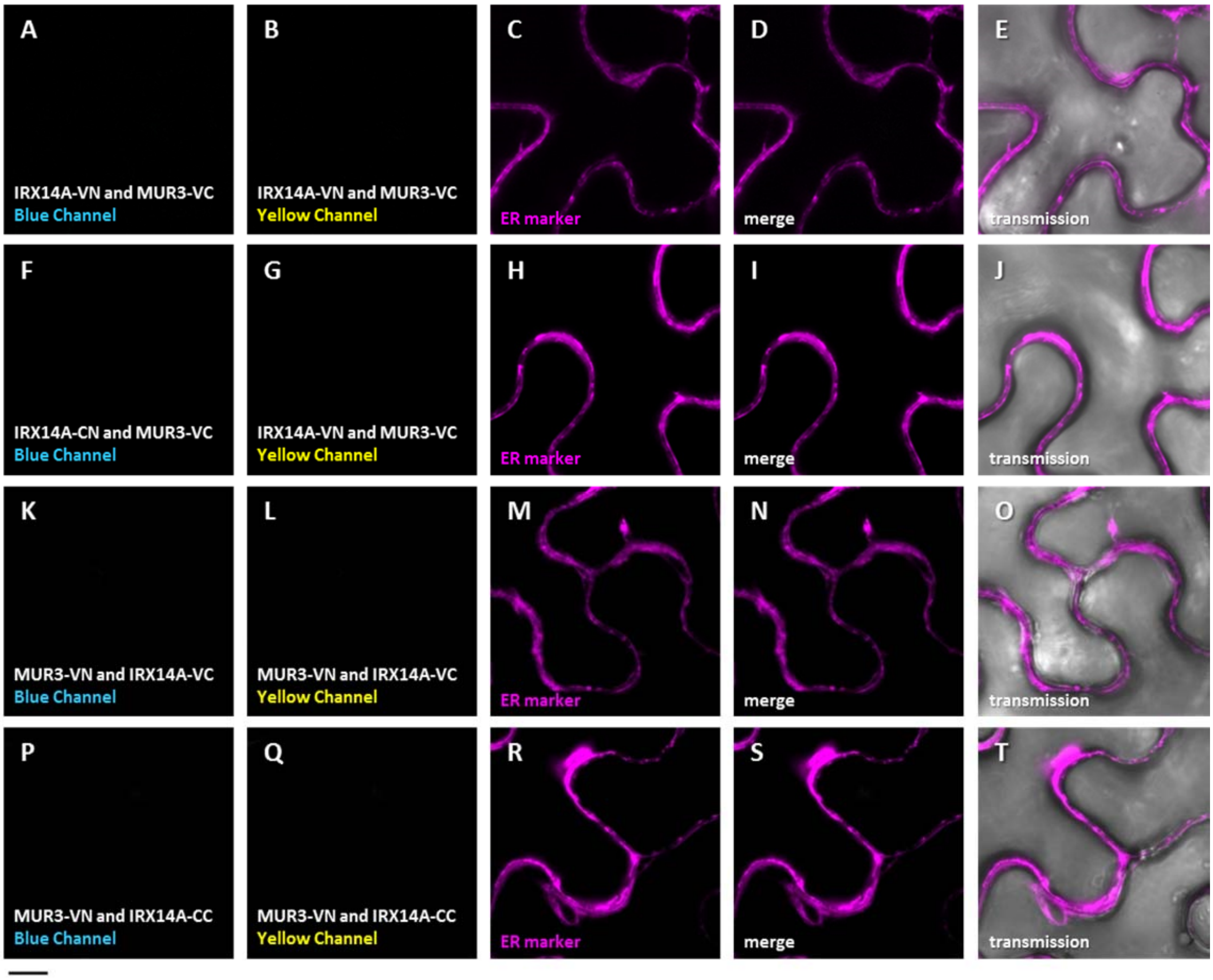
Supplemental Figure 8: Expanded images from Figure 9G-I.
A-D shows the expanded figures from Figure 9G-I where the IRX14A homo-dimerisation is independent of the ER marker (MANI(49)-RFP). The ER marker was expressed as a transformation control. MUR3 is not expected to be involved in the xylan synthesis pathway, and as expected, did not interact with IRX14 as shown by lack of signal in E-L.



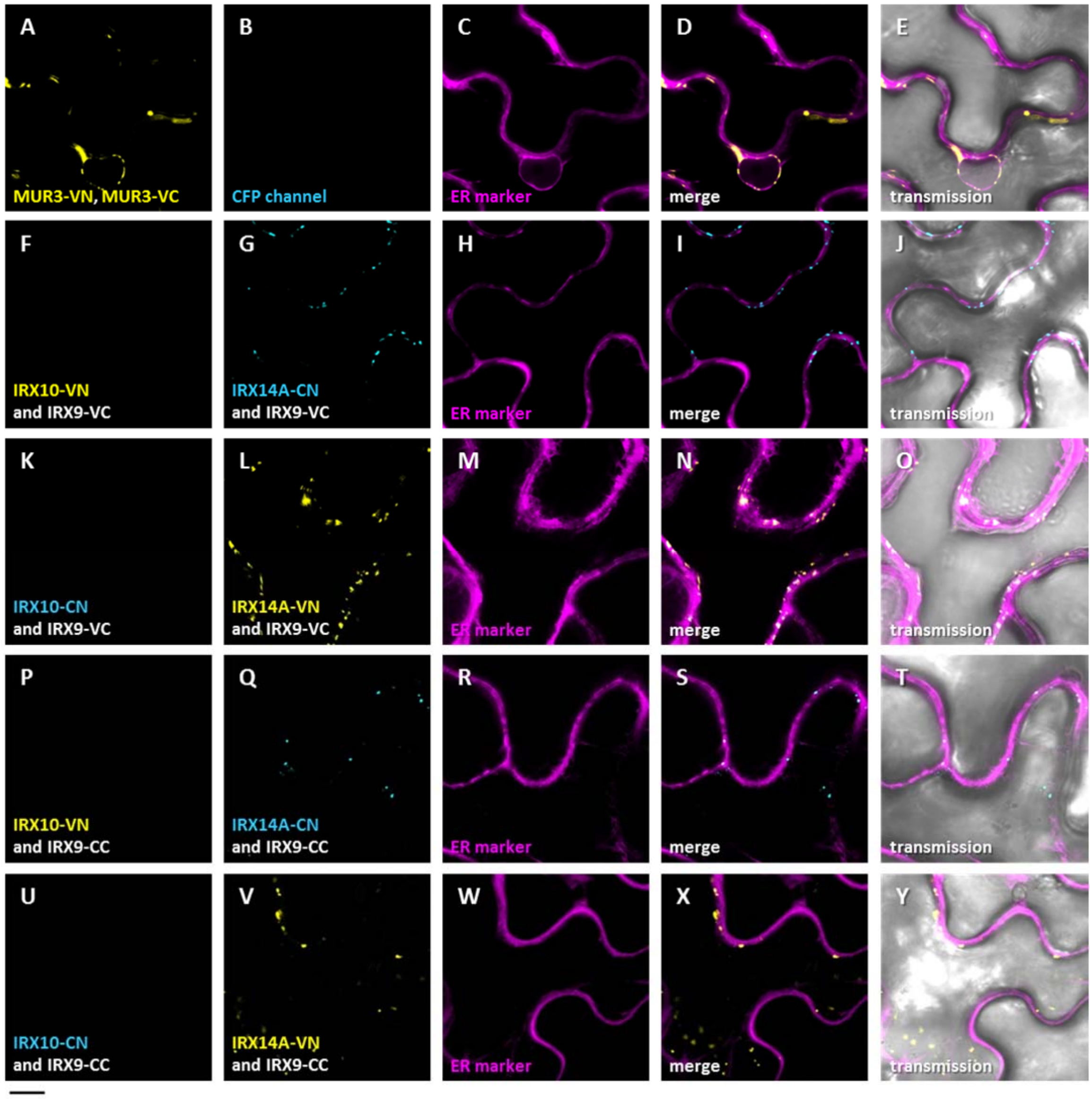
Supplemental Figure 9: Negative controls for IRX9 in Figure 10.
MUR3 is also not expected to be involved in the xylan synthesis pathway, and so it was used as a negative control of random interactions with the IRX proteins. In all cases, MUR3 did not interact with IRX9. The ER marker (MANI(49)-RFP) was used as a transformation control.



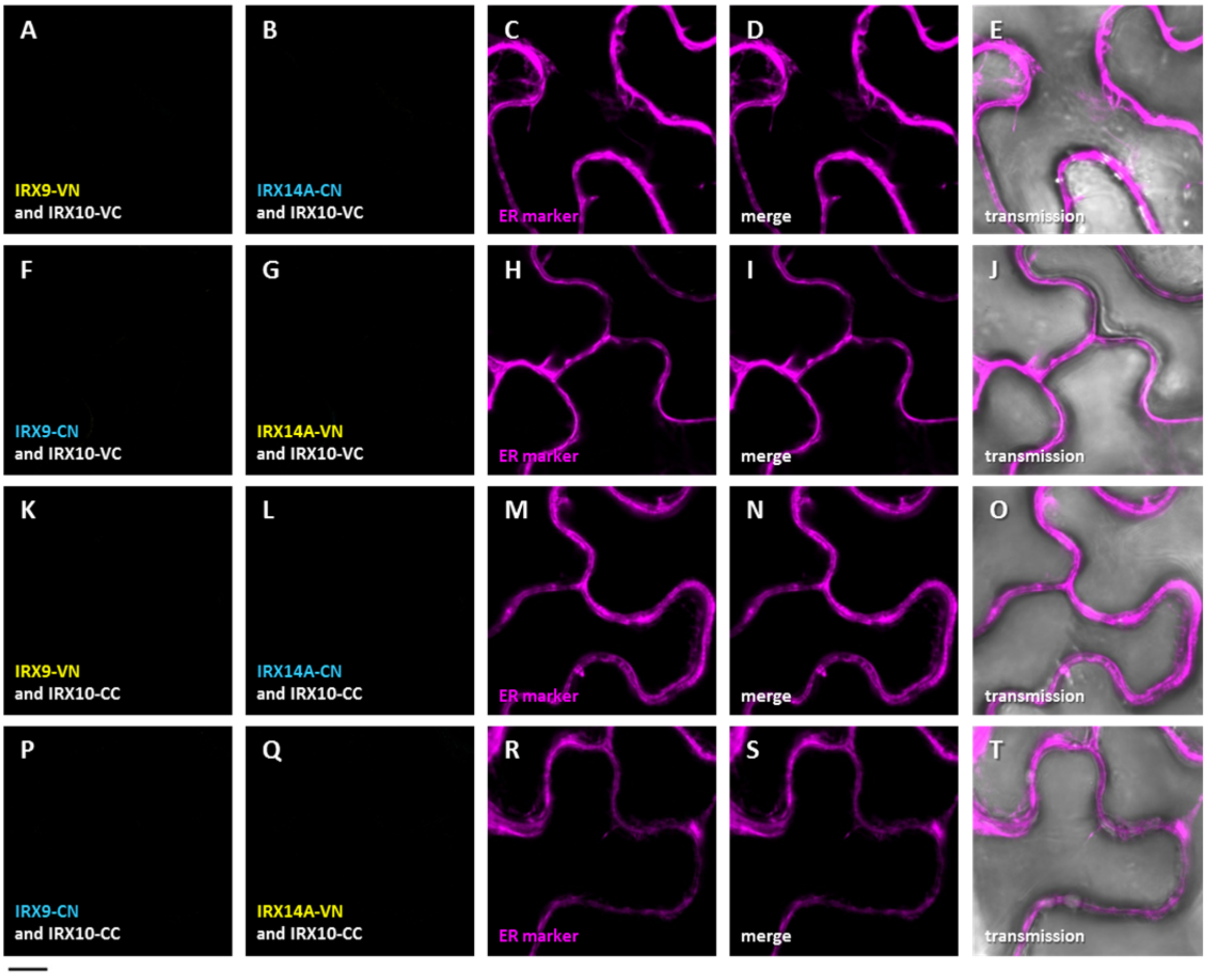
Supplemental Figure 10: Negative controls for IRX10 in Figure 10.
MUR3 is also not expected to be involved in the xylan synthesis pathway, and so it was used as a negative control of random interactions with the IRX proteins. In all cases, MUR3 did not interact with IRX10. The ER marker (MANI(49)-RFP) was used as a transformation control.



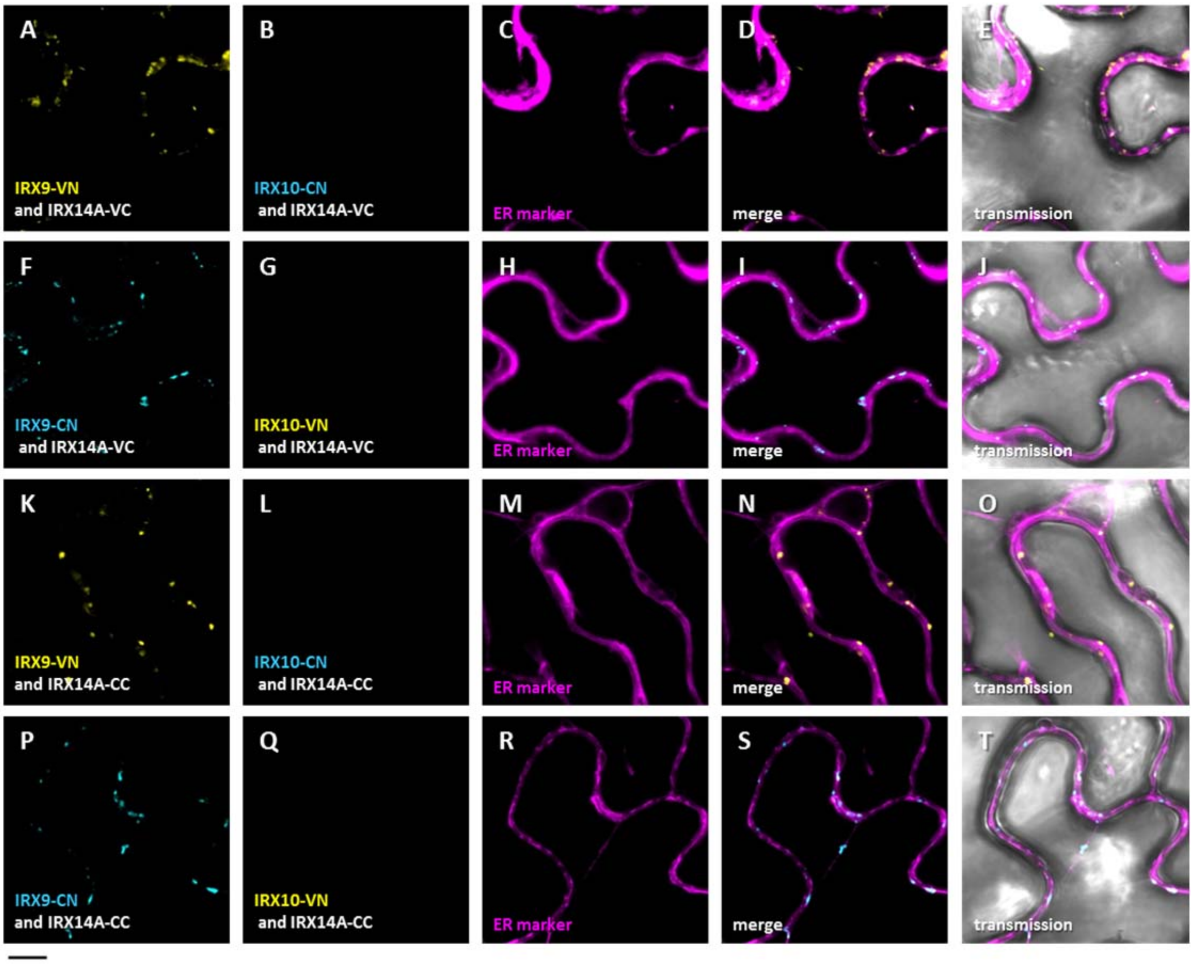
Supplemental Figure 11: Negative controls for IRX14A in Figure 10.
MUR3 is also not expected to be involved in the xylan synthesis pathway, and so it was used as a negative control of random interactions with the IRX proteins. In all cases, MUR3 did not interact with IRX14A. The ER marker (MANI(49)-RFP) was used as a transformation control.



Supplemental Figure 12: MUR3 positive control and expanded images from Figure 10A-D.
MUR3 is known to form homo-dimers *in planta* (Søgaard *et al.*, 2012) and so it was used as a positive control for BiFC experiments. Strong fluorescence signal indicates this system is working (A-E). B shows the CFP channel, which is not able to pick up any signal emitted by VENUS fluorescent proteins. F-Y show the expanded figures from Figure 10A-D where IRX9-CC or IRX9-VC is the bait protein. Fluorescent signal indicates when another protein is able to interact with IRX9. In all cases, IRX14A interacts with IRX9 producing either yellow or blue fluorescence depending on the IRX14A fusion. In all cases IRX10 does not interact with IRX9 as no fluorescent signal is observed. The ER marker (MANI(49)-RFP) was used as a transformation control.



Supplemental Figure 13: Expanded images from Figure 10E-H.
A-T show the expanded images from Figure 10E-H where IRX10-CC (K-T) or IRX10-VC (A-J) is the bait protein. Fluorescent signal indicates when another protein is able to interact with IRX10. In all cases, neither IRX9 nor IRX14A interacted with IRX10. The ER marker (MANI(49)-RFP) was used as a transformation control.



Supplemental Figure 14: Expanded images from Figure 10I-L
A-T show the expanded images from Figure 10 I-L where IRX14A-CC or IRX14A-VC is the bait protein. Fluorescent signal indicates when another protein is able to interact with IRX14A. In all cases, IRX14A interacts with IRX9 producing either yellow or blue fluorescence depending on the IRX9 fusion. In all cases IRX10 does not interact with IRX14A as no fluorescent signal is observed. The ER marker (MANI(49)-RFP) was used as a transformation control.