Current Biology
Supplemental Information

RSL Class I Genes Controlled the Development of Epidermal Structures in the Common Ancestor of Land Plants

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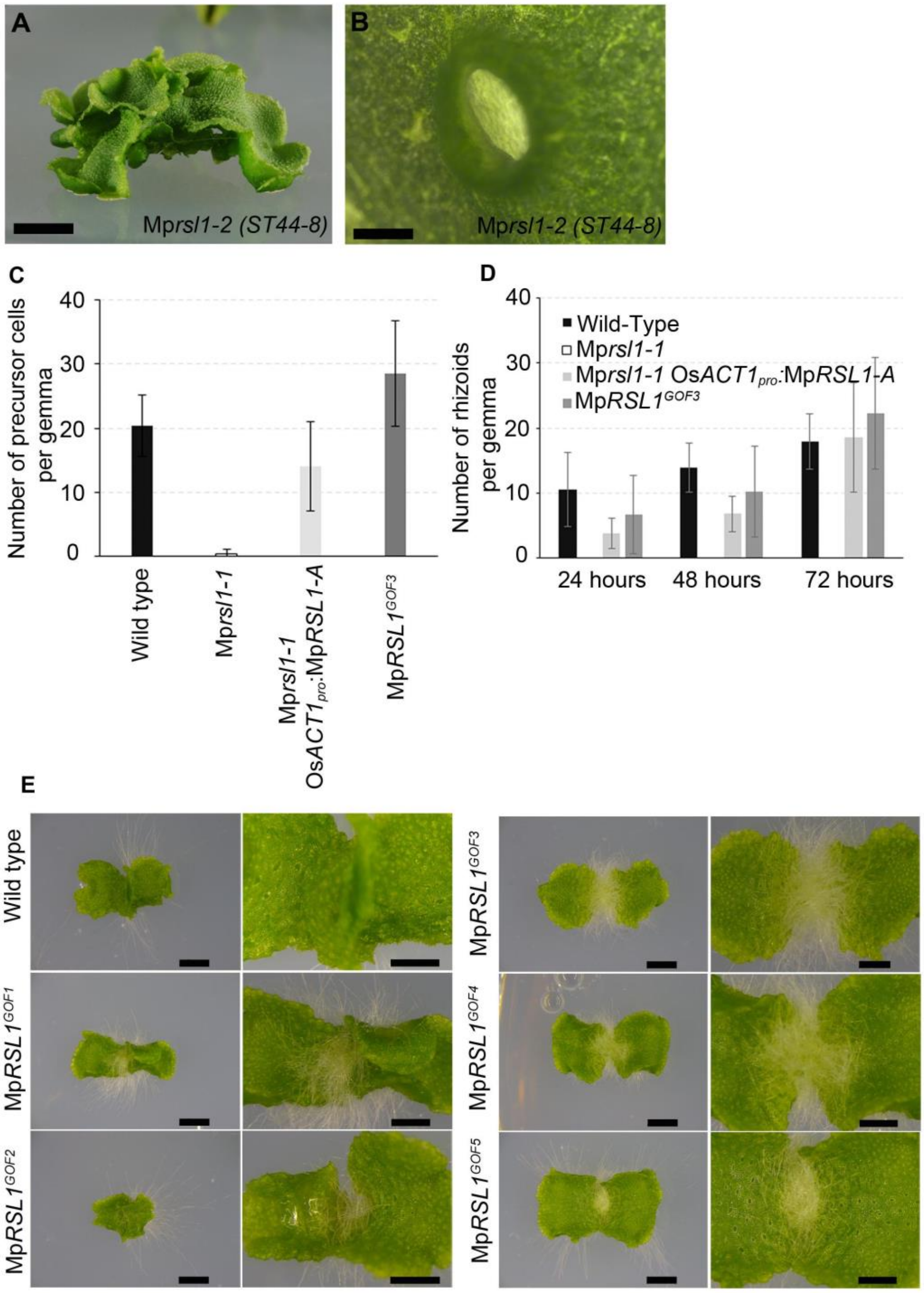
A
Mprsl1-2 (ST44-8)
B
Mprsl1-2 (ST44-8)
C
Number of precursor cells per gemma
40
30
20
10
0
Wild type
Mprsl1-1
Mprsl1-1 OsACT1pro:MpRSL1-A
MpRSL1GOF3
D
Number of rhizoids per gemma
Wild-Type
Mprsl1-1
Mprsl1-1 OsACT1pro:MpRSL1-A
MpRSL1GOF3
24 hours
48 hours
72 hours
E
Wild type
MpRSL1GOF1
MpRSL1GOF2
MpRSL1GOF3
MpRSL1GOF4
MpRSL1GOF5

Figure S1

Figure S1, related to figure 1

Mp*rsl1* and Mp*RSL1*$^{\mathbf{GOF}}$ lines phenotype.

(A) Rhizoids do not develop in 1 month-old Mp*rsl1-2* (ST44-8) plants. Bar, 1 cm.

(B) Gemmae do not develop in Mp*rsl1-2* gemma cups. Bar, 1 mm.

(C) Number of rhizoid precursors in wild type, Mp*rsl1-1*, Mp*rsl1-1* Os*ACT1*$_{pro}$:Mp*RSL1-A*, and Mp*RSL1*^{GOF3} 0 day-old gemmae.

(D) The histograms represent the mean (± SD) number of rhizoid precursor cells on 0 days-old gemmae, n=20. Rhizoid number in wild type, Mp*rsl1-1*, Mp*rsl1-1* Os*ACT1*$_{pro}$:Mp*RSL1-A*, Mp*RSL1*^{GOF3} 4-day-old gemmae. The histograms represent the average (+/- SD) of number of rhizoids per 4-days-old gemmalings in different lines, n=20.

(E) 15-days-old gemmalings of wild type (Tak 1), Mp*RSL1*^{GOF1}, MpRSL1^{GOF2}, Mp*RSL1*^{GOF3}, Mp*RSL1*^{GOF4} and Mp*RSL1*^{GOF5}. Scales bar: right panel, 2 mm; left panel, 1mm.

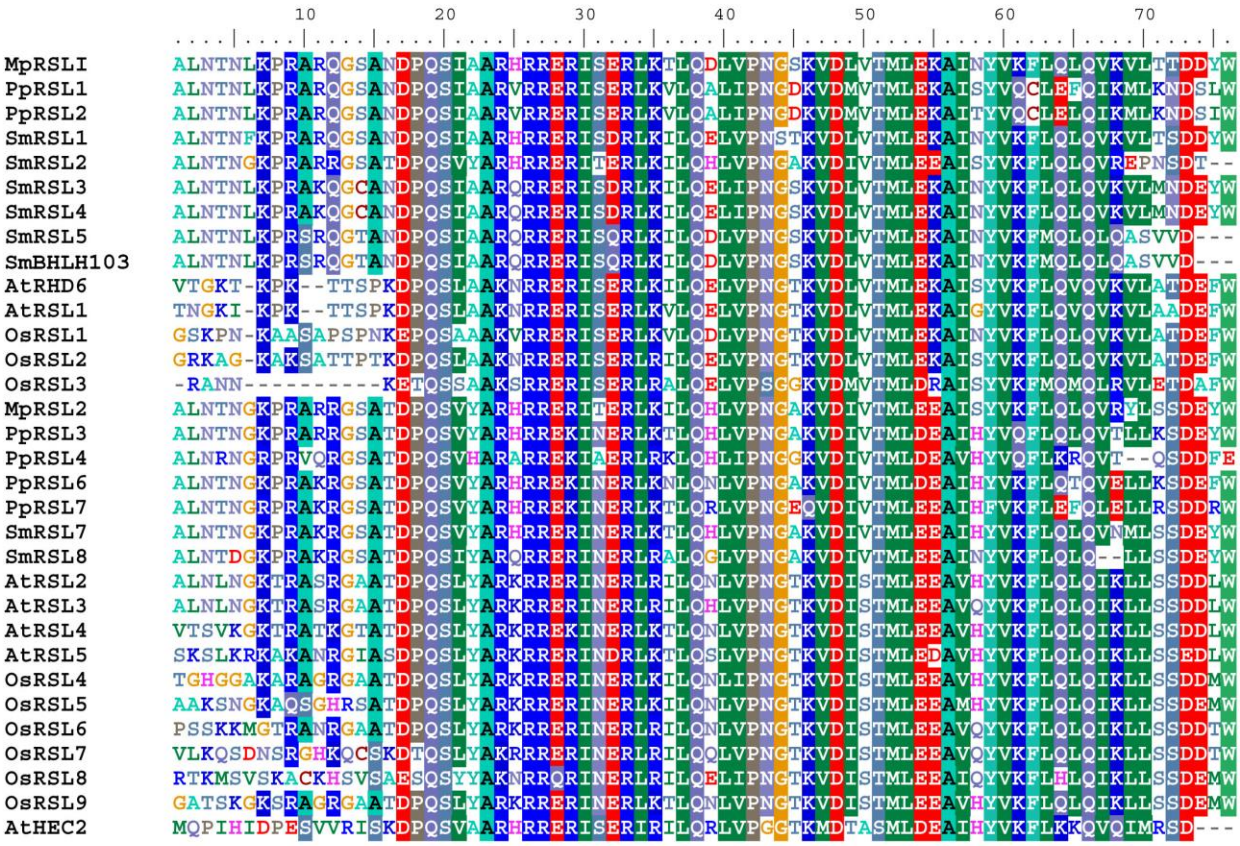
10 20 30 40 50 60 70
MpRSLI ALNTNLKPRARQGSANDPQSIAARHRRERISERLKTLQDLVPNGSKVDLVTMLEKAINYVKFLQLQVKVLTTDDYW
PpRSL1 ALNTNLKPRARQGSANDPQSIAARVRRERISERLKVLQALIPNGDKVDMVTMLEKAISYVQCLEFQIKMLKNDSLW
PpRSL2 ALNTNLKPRARQGSANDPQSIAARVRRERISERLKVLQALIPNGDKVDMVTMLEKAITYVQCLELQIKMLKNDSIW
SmRSL1 ALNTNFKPRARQGSANDPQSIAARHRRERISDRLKILQELVPNSTKVDLVTMLEKAINYVKFLQLQVKVLTSDDYW
SmRSL2 ALNTNGKPRARRGSATDPQSVYARHRRERITERLKILQHLVPNGAKVDIVTMLEEAISYVKFLQLQVREPNSDT--
SmRSL3 ALNTNLKPRAKQGCANDPQSIAARQRRERISDRLKILQELIPNGSKVDLVTMLEKAINYVKFLQLQVKVLMNDEYW
SmRSL4 ALNTNLKPRAKQGCANDPQSIAARQRRERISDRLKILQELIPNGSKVDLVTMLEKAINYVKFLQLQVKVLMNDEYW
SmRSL5 ALNTNLKPRSRQGTANDPQSIAARQRRERISQRLKILQDLVPNGSKVDLVTMLEKAINYVKFMQLQLQASVVD---
SmBHLH103 ALNTNLKPRSRQGTANDPQSIAARQRRERISQRLKILQDLVPNGSKVDLVTMLEKAINYVKFMQLQLQASVVD---
AtRHD6 VTGKT-KPK--TTSPKDPQSLAAKNRRERISERLKILQELVPNGTKVDLVTMLEKAISYVKFLQVQVKVLATDEFW
AtRSL1 TNGKI-KPK--TTSPKDPQSLAAKNRRERISERLKVLQELVPNGTKVDLVTMLEKAIGYVKFLQVQVKVLAADEFW
OsRSL1 GSKPN-KAASAPSPNKEPQSAAAKVRRERISERLKVLQDLVPNGTKVDLVTMLEKAINYVKFLQLQVKVLATDEFW
OsRSL2 GRKAG-KAKSATTPTKDPQSLAAKNRRERISERLRILQELVPNGTKVDLVTMLEKAISYVKFLQLQVKVLATDEFW
OsRSL3 -RANN----------KETQSSAAKSRRERISERLRALQELVPSGGKVDMVTMLDRAISYVKFMQMQLRVLETDAFW
MpRSL2 ALNTNGKPRARRGSATDPQSVYARHRRERITERLKILQHLVPNGAKVDIVTMLEEAISYVKFLQLQVRYLSSDEYW
PpRSL3 ALNTNGKPRARRGSATDPQSVYARHRREKINERLKTLQHLVPNGAKVDIVTMLDEAIHYVQFLQLQVTLLKSDEYW
PpRSL4 ALNRNGRPRVQRGSATDPQSVHARARREKIAERLRKLQHLIPNGGKVDIVTMLDEAVHYVQFLKRQVT--QSDDFE
PpRSL6 ALNTNGKPRAKRGSATDPQSVYARHRREKINERLKNLQNLVPNGAKVDIVTMLDEAIHYVKFLQTQVELLKSDEFW
PpRSL7 ALNTNGRPRAKRGSATDPQSVYARHRREKINERLKTLQRLVPNGEQVDIVTMLEEAIHFVKFLEFQLELLRSDDRW
SmRSL7 ALNTNGKPRAKRGSATDPQSVYARHRRERINERLKTLQHLVPNGAKVDIVTMLEEAIHYVKFLQLQVNMLSSDEYW
SmRSL8 ALNTDGKPRAKRGSATDPQSIYARQRRERINERLRALQGLVPNGAKVDIVTMLEEAINYVKFLQLQ--LLSSDEYW
AtRSL2 ALNLNGKTRASRGAATDPQSLYARKRRERINERLRILQNLVPNGTKVDISTMLEEAVHYVKFLQLQIKLLSSDDLW
AtRSL3 ALNLNGKTRASRGAATDPQSLYARKRRERINERLRILQHLVPNGTKVDISTMLEEAVQYVKFLQLQIKLLSSDDLW
AtRSL4 VTSVKGKTRATKGTATDPQSLYARKRREKINERLKTLQNLVPNGTKVDISTMLEEAVHYVKFLQLQIKLLSSDDLW
AtRSL5 SKSLKRKAKANRGIASDPQSLYARKRRERINDRLKTLQSLVPNGTKVDISTMLEDAVHYVKFLQLQIKLLSSEDLW
OsRSL4 TGHGGAKARAGRGAATDPQSLYARKRRERINERLKILQNLIPNGTKVDISTMLEEAVHYVKFLQLQIKLLSSDDMW
OsRSL5 AAKSNGKAQSGHRSATDPQSLYARKRRERINERLKILQNLVPNGTKVDISTMLEEAMHYVKFLQLQIKLLSSDEMW
OsRSL6 PSSKKMGTRANRGAATDPQSLYARKRRERINERLRILQNLVPNGTKVDISTMLEEAVQYVKFLQLQIKLLSSDDTW
OsRSL7 VLKQSDNSRGHKQCSKDTQSLYAKRRRERINERLRILQQLVPNGTKVDISTMLEEAVQYVKFLQLQIKLLSSDDTW
OsRSL8 RTKMSVSKACKHSVSAESQSYYAKNRRQRINERLRILQELIPNGTKVDISTMLEEAIQYVKFLHLQIKLLSSDEMW
OsRSL9 GATSKGKSRAGRGAATDPQSLYARKRRERINERLKTLQNLVPNGTKVDISTMLEEAVHYVKFLQLQIKLLSSDEMW
AtHEC2 MQPIHIDPESVVRISKDPQSVAARHRRERISERIRILQRLVPGGTKMDTASMLDEAIHYVKFLKKQVQIMRSD---

Figure S2

Figure S2, related to figure 2

Alignment of RSL Class I (pink box), RSL Class II (blue box) and AtHEC2 protein used for the construction of the maximum likelihood tree.

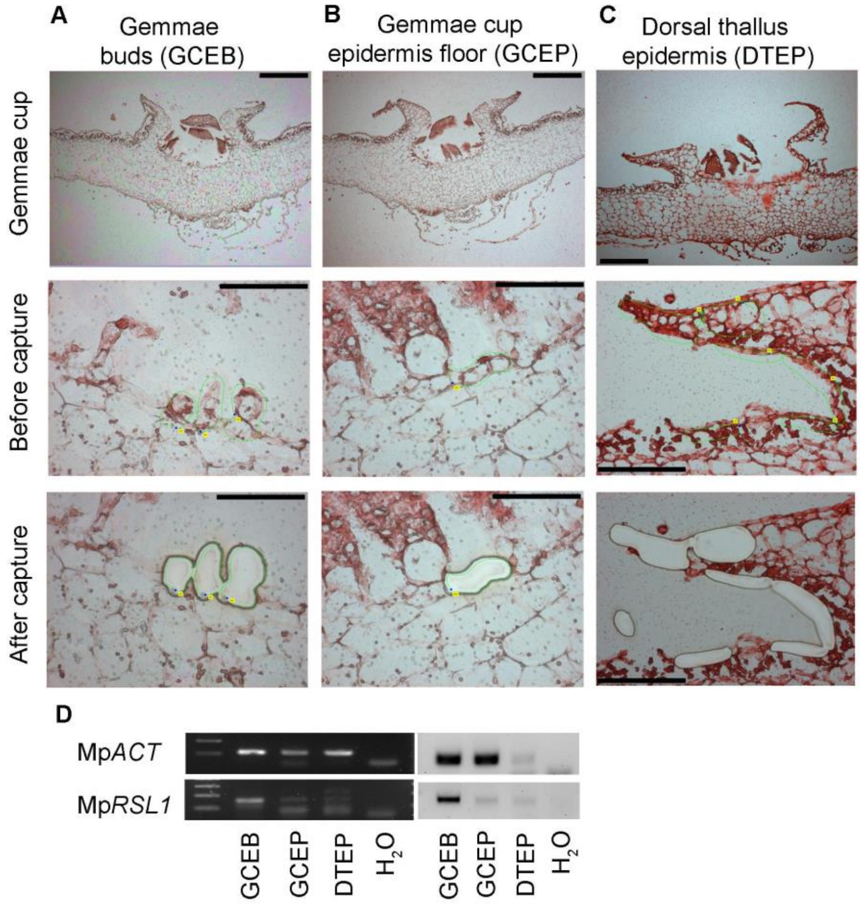
A
Gemmae buds (GCEB)
B
Gemmae cup epidermis floor (GCEP)
C
Dorsal thallus epidermis (DTEP)
Gemmae cup
Before capture
After capture
D
MpACT
MpRSL1
GCEB
GCEP
DTEP
H2O
GCEB
GCEP
DTEP
H2O

Figure S3

Figure S3, related to figure 3

Mp*RSL1* transcript levels in cells captured by laser microdissection.

(A) gemma cup epidermal buds, (B) gemma cup epidermal cells and (C) dorsal thallus epidermal cells before dissection (top and middle panel) and after capture (bottom panel). Bar: top panel, 300 µm; middle and bottom panel, 75 µm.

(D) RT-PCR analysis of Mp*RSL1* transcript level in gemma cup epidermal buds (GCEB), gemmae cup epidermal cells (GCEP) and thallus epidermal cells (DTEP). Mp*ACT* was used as reference gene.

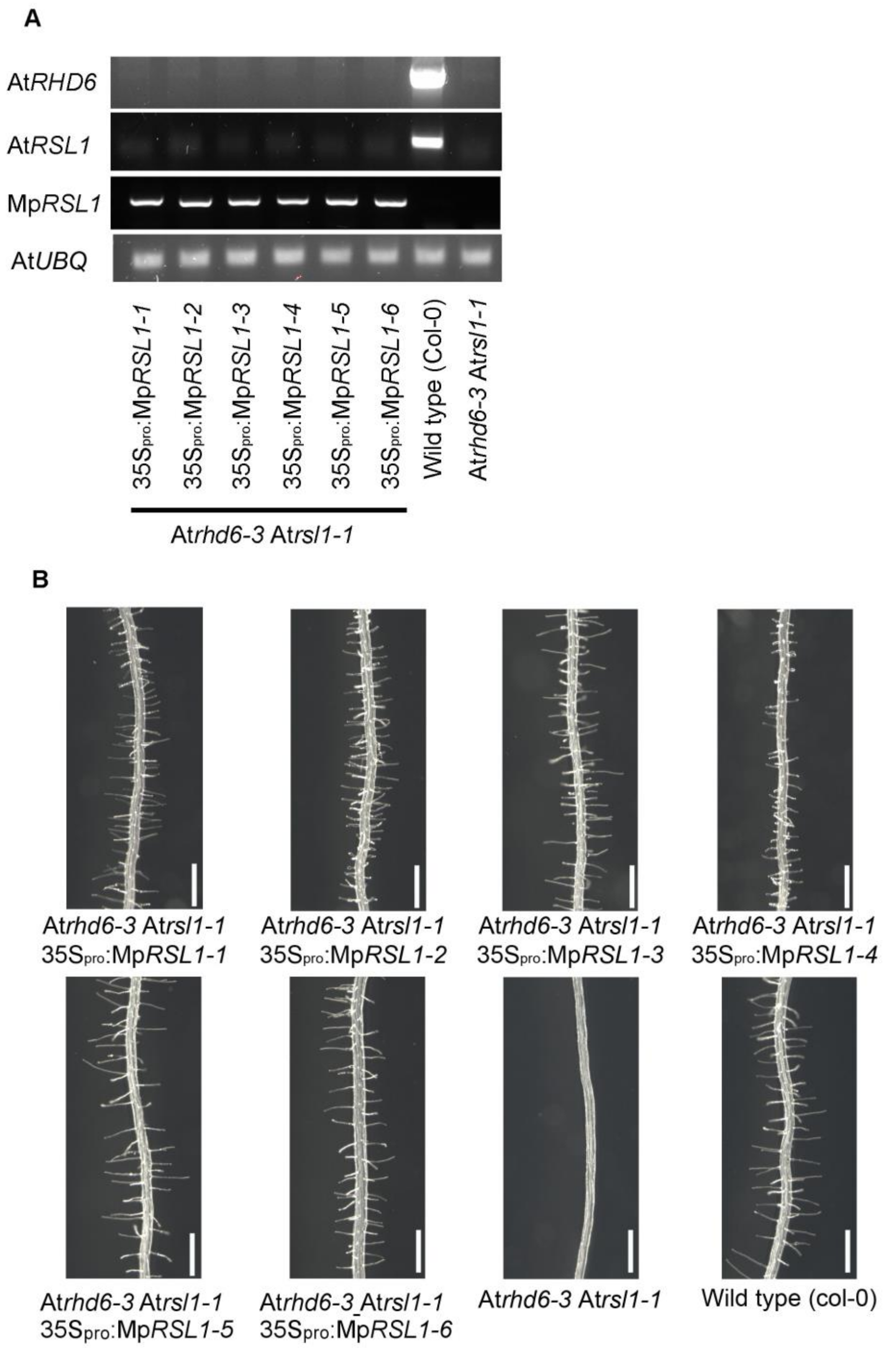
A
AtRHD6
AtRSL1
MpRSL1
AtUBQ
35Spro:MpRSL1-1
35Spro:MpRSL1-2
35Spro:MpRSL1-3
35Spro:MpRSL1-4
35Spro:MpRSL1-5
35Spro:MpRSL1-6
Wild type (Col-0)
Atrhd6-3 Atrsl1-1
Atrhd6-3 Atrsl1-1
B
Atrhd6-3 Atrsl1-1 35Spro:MpRSL1-1
Atrhd6-3 Atrsl1-1 35Spro:MpRSL1-2
Atrhd6-3 Atrsl1-1 35Spro:MpRSL1-3
Atrhd6-3 Atrsl1-1 35Spro:MpRSL1-4
Atrhd6-3 Atrsl1-1 35Spro:MpRSL1-5
Atrhd6-3 Atrsl1-1 35Spro:MpRSL1-6
Atrhd6-3 Atrsl1-1
Wild type (col-0)

Figure S4

Figure S4, related to figure 4

Expression of Mp*RSL1* restores root hair development in the root hairless At*rhd6-3* At*rsl1-1* double mutant.

(A) Semi-quantitative RT-PCR analysis of At*RHD6*, At*RSL1* and Mp*RSL1* expression in Wild type (Col-0), At*rhd6-3* At*rsl1-1* double mutant and At*rhd6-3* At*rsl1-1* double mutants transformed with *$35S_{pro}$:*Mp*RSL1*.

(B) Root hair phenotype of At*rhd6-3* At*rsl1-1* double mutants, At*rhd6-3* At*rsl1-1* 3*$5S_{pro}$:*Mp*RSL1-1*, At*rhd6-3* At*rsl1-1 $35S_{pro}$:*Mp*RSL1-2,* At*rhd6-3* At*rsl1-1 $35S_{pro}$:*Mp*RSL1-3,* At*rhd6-3* At*rsl1-1 $35S_{pro}$:*Mp*RSL1-4,* At*rhd6-3* At*rsl1-1 $35S_{pro}$:*Mp*RSL1-5,* At*rhd6-3* At*rsl1-1 $35S_{pro}$:*Mp*RSL1-6.* Bar: 500 μm.

Table S1, related to figure S2

Mp*RSL1*^{GOF} lines possess a single T-DNA insertion.

	Mutant phenotype		Wild-Type phenotype		
	HygR	HygS	HygR	HygS	T-DNA insertion
Mp*RSL1*^{GOF1}	106	0	0	123	1
Mp*RSL1*^{GOF2}	108	0	0	122	1
Mp*RSL1*^{GOF3}	117	0	0	151	1
Mp*RSL1*^{GOF4}	76	0	0	97	1
Mp*RSL1*^{GOF5}	116	0	0	102	1

Table S2, related to figure S2

Mp*RSL1*^{GOF} lines allelic test.

Crosses	Plants F1 exhibiting a hairy phenotype (%)
Mp*RSL1*^{GOF3} x Mp*RSL1*^{GOF1}	100 %
Mp*RSL1*^{GOF3} x Mp*RSL1*^{GOF2}	100 %
Mp*RSL1*^{GOF3} x Mp*RSL1*^{GOF4}	100 %
Mp*RSL1*^{GOF3} x Mp*RSL1*^{GOF5}	100 %

Supplemental experimental procedures

Plant material and growth

Marchantia polymorpha growth and transformation

Takaragaike-1 (Tak-1) male and Takaragaike-2 (Tak-2) female *M. polymorpha* accessions were used in this study [S1]. Mutants grew from spores resulting from a cross between Tak-1 and Tak-2. Gemmae or meristematic segments were propagated on ½ Johnson's medium [S2] supplemented with 1% agar in Petri dishes under a 16 h light: 8 h dark photoperiod at 23°C and a light intensity of 56 $\mu E.m^{-2}.s^{-1}$. Sexual organ development was stimulated with far red light [S3, S4].

M. polymorpha sporelings and regenerating thalli were transformed following the protocols previously described [S1, S5]. After 2 days of co-cultivation with agrobacterium GV3301 carrying the pCambia1300, or Os*ACT1*$_{pro}$:Mp*RSL1* binary vector, transformed sporelings or regenerating thalli were selected on Johnson's medium supplemented with cefotaxime 100 $\mu g.ml^{-1}$ and hygromycin 10 $\mu g.ml^{-1}$ or gentamycin 100 $\mu g.ml^{-1}$.

Physcomitrella patens

Gransden wild type and Pp*rsl1* Pp*rsl2* [S6] double mutant plants were propagated in Petri dishes on KNOPS medium supplemented with 0.7% Agar under a 16 h light: 8 h dark photoperiod at 23°C and a light intensity of 56 $\mu E.m^{-2}.s^{-1}$.

Arabidopsis thaliana growth and transformation

Col-0 and At*rhd6-3* At*rsl1-1* double mutant [S6] plants were grown on soil: 2/3 of peat-based compost (Levington M2), 1/3 of vermiculite medium (Sinclair). For phenotypic analysis, seeds were surface sterilized with a 70% Ethanol 0.1% Triton solution for 5 minutes followed by 99% ethanol solution for 5 minutes and grown as previously described [S6].

At*rhd6-3* At*rsl1-1 Arabidopsis* plants were transformed with agrobacterium GV3301 carrying *35S*$_{pro}$:Mp*RSL1* binary vector using the floral-dip method. Transformants were then selected on MS medium supplemented with 1% agar and 50 μg.ml^{-1} of hygromycin.

Phenotypic analysis

Images were captured with a Leica DFC310 FX camera mounted on a Leica M165 FC microscope.

In *Physcomitrella patens*, number of axillary hairs was measured in 5 nodes of 15 gametophores from 1-month-old wild type colonies and 5 nodes of 15 gametophores from 1-month-old Pp*rsl1* Pp*rsl2* double mutant colonies.

Phylogenetic analysis

The RSL Class I and RSL class II proteins—AtRHD6 (At1g66470), AtRSL1 (At5g37800), AtRSL2 (At4g33880), AtRSL3 (At2g14760), AtRSL4 (At1g27740), AtRSL5 (At5g43175), OsRSL1 (Os01g02110), OsRSL2 (Os02g48060), OsRSL3 (Os06g30090), OsRSL4 (Os03g10770), OsRSL5 (Os03g42100), OsRSL6 (Os07g39940), OsRSL7 (Os11g41640), OsRSL8 (Os12g32400), OsRSL9 (Os12g39850), SmRSL1 (EFJ25918.1), SmRSL2 (EFJ10890.1), SmRSL3 (EFJ29938.1), SmRSL4 (EFJ25105.1), SmRSL5 (EFJ20125.1), SmbHLH103 (EFJ14254.1), SmRSL7 (EFJ36606.1), SmRSL8 (EFJ19083.1), PpRSL1 (EF156393), PpRSL2 (EF156394), PpRSL3 (EF156395), PpRSL4 (EF156396), PpRSL5 (EF156397), PpRSL6 (EF156398) and PpRSL7 (EF156399)—from *Arabidopsis thaliana* (At), *Oryza sativa* (Os), *Selaginella moellendorffii* (Sm) and *Physcomitrella patens* (Pp) were retrieved from public databases following a published classification [S7]. MpRSL1

(KT633827), MpRSL2 (KT633828), were isolated then cloned. AtHEC2 (AT3G50330) was retrieved from NCBI database and used to root the tree. Alignment of bHLH domains from RSL Class I, RSL Class II and AtHEC2 proteins was performed with MAFFT (http://mafft.cbrc.jp/alignment/software/) and manually edited with bioedit (https://www.bioedit.com/). A maximum likelihood phylogenetic analysis was carried out on the sequence alignment using the LG model of amino acid [S8] and a Shimodaira-Hasegawa-like approximate ratio test [S9] with the program PhyML 3.0 [S10] (http://atgc.lirmm.fr/phyml/). The resulting tree was edited with the program figtree (http://tree.bio.ed.ac.uk/software/figtree/).

Generation of Os*ACT1pro:*Mp*RSL1* and *35Spro:MpRSL1*.

35S$_{pro}$-GW has been generated by inserting a Gateway cassette into the *Cla*I restriction site of pCambia1300a vector containing a 35S promoter and a Nos terminator. The vector pOsAct-108-hygro [S11] has been digested with *Sma*I and *Pst*I to remove the promoter Os*ACT1*$_{pro}$. The promoter Os*ACT1*$_{pro}$ was then subcloned into pCambia1300a previously digested with *Eco*RI and *Pst*I. Then, the resulting vector was digested with *Xba*I and *Hin*dIII to remove the cassette Os*ACT1*$_{pro}$:*Pst*I:Term. This cassette was then placed into the vector pMpGW207 containing gentamycin resistance [S12] previously digested with *Sac*I and *Hin*dIII. Os*ACT1*$_{pro}$-GW-Term has been obtained by insertion of a gateway cassette into the *Pst*I restriction site. Full length cDNA of Mp*RSL1* was amplified with specific primers (See the list of primers sequences below) and cloned into pCR8-GW pTOPO (Invitrogen). A subsequent LR reaction allowed the insertion of full length cDNA into pOs*ACT1*$_{pro}$-GW or *35S*$_{pro}$-GW vector to generate Os*ACT1*$_{pro}$:Mp*RSL1* and *35S*$_{pro}$:Mp*RSL1*.

Molecular analysis of mutants

Genomic DNA extraction

Genomic DNA was isolated from 1-month-old plants grown in sterile conditions. Tissues were ground in liquid nitrogen and the DNA was extracted with 2% cetyltrimethyl ammonium bromide (CTAB) buffer as previously described [S13].

Identification of DNA sequences flanking T-DNA insertions

Tail PCR was performed in Mp*RSL1*^{GOF} and Mp*rsl1* lines to identify sequences flanking the T-DNA insertion. Specific primers to the T-DNA sequence (TR1 to TR3 and TL1 to TL3 for right border T-DNA and left border T-DNA respectively) and universal adaptor primers (AD1 to AD6) were used as described previously (See the list of primers sequences below) [S1, S14].

Measurement of steady state mRNA levels using quantitative RT-PCR

Total RNA was extracted from frozen tissues using a plant RNeasy plant mini kit (Qiagen) according to the manufacturer's protocol. cDNA was synthesized from 1 µg of total RNA at 42 °C for 1 h with 200 U the protoscript 2 reverse transcriptase (NEB) and 2 µM of dT17 oligonucleotides. Quantitative RT-PCR was carried on a 7300 Applied Biosystem thermocycler. Amplification reactions were performed into 10 µl volume medium containing 5 µl of 2X SYBR green mastermix (Applied Biosystem), 500 nM of each primer (See the list of primers sequences below), and 4 µl of 1:10 diluted cDNA (corresponding to 20 ng of reverse transcribed total RNA). A two-step program composed of a denaturation step at 94°C for 15 seconds and a hybridization-elongation step at 60°C for 1 minute was repeated 40 times, then, a dissociation stage was performed. The transcript levels of genes were calculated with Linreg v2012.0 [S15, S16].

Fixation and wax inclusion for laser microdissection experiments

Plants were fixed in 100% acetone for 24 h at 4°C. The samples were then incubated in 1/1 acetone with Histoclear (v/v) and a 100% histoclear solution, for 5 minutes each, then embedded in wax with Tissue Tek® VIPTM. 8 µm sections were then placed on Membrane Slide 1.0 PEN (D) (#415190-9041-000, Zeiss) and dried overnight at 37°C.

Laser capture microdissection

Laser capture micro-dissection was performed on 8 µm thin sections of wax embedded specimens (see supplemental information) using a Carl Zeiss PALM microbeam microscope and PALMRobo 4.5V software driving the laser (laser power: 43, laser focus: 35, LPC delta +27, 1 cycle of cutting). Captured cells were collected in AdhesiveCap 500 clear (D) tubes (#415190-9211-000, Zeiss). RNA was extracted with Arcturus PicoPureTM RNA isolation kit (#KIT0204, Lifetechnologie) then cDNA were synthesized and amplified with Ovation RNA-Seq System v2 (#7102, NuGen) following the manufacturer's protocol. 1 µl of non-diluted amplified cDNA was used for expression analysis by PCR.

Plastic sectioning

Isolated gemmae cups from 1-month-old plants were fixed in fresh 4% paraformaldehyde in phosphate buffered saline (PBS) buffer (8 $g.L^{-1}$ of NaCl, 0.2 $g.L^{-1}$ of KCl, 1.44 $g.L^{-1}$ of NaH_2PO_4 and 0.24 $g.L^{-1}$ of KH_2PO_4) for 24 hours at 4°C. Chlorophyll was removed by incubation for 1 h in 30%, 70% and 100% ethanol. After rehydration in an ethanol series (100, 70 and 30%) for 20 minutes each, samples were placed in 1% molten agarose. Once the agarose solidified, the embedded samples were washed twice in sterile water then dehydrated in an ethanol series (30, 70 and 100%). Dehydrated samples were then embedded in Technovit® 7100 cold-polymerising resin (#14653, Kulzer) [S17]. µm) 4 µm sections were made with an Ultracut E (Reichert-Jung). Images were taken captured with a Micropublisher 5.0 RTV (Q-Imaging) camera mounted on a Leica DMRB microscope.

Sequences of primers used in this study

Cloning

MpRSL1-ATG	ATGGCGAATTATGATAGCAGC
MpRSL1-Stop	GCAGACAACAACTCGTCCTGA

Genotyping

Primer A	ATGGGGCAAAGTCAGGGTAT
Primer B	GTGAATTCGACTTGGTGTAAG
Primer 1	CCGTAAGTCAATTAAGGAG
Primer 2	CTGTTTCCACGAACTCCTC
Primer 3	GTGGTGTGGAGGAGTCGTTG
Primer 4	CAGTATCGTATCAAGCCGAAG
Primer 5	GACCCTGATACACAATTTCGC
Primer 6	TCCGTCCACACACATTCTAGG
Primer R	GCTGGCGTAATAGCGAAGAGG
Primer L	GGTTTCGCTCATGTGTTGAGC

Tail PCR:

Primer AD1	NGTCGASWGANAWGAA
Primer AD2	TGWGNAGSANCASAGA
Primer AD3	AGWGNAGWANCAWAGG
Primer AD4	GTNCGASWCANAWGTT
Primer AD5	NTCGASTWTSGWGTT
Primer AD6	WGTGNAGWANCANAGA
Primer TR1	CCTGCAGGCATGCAAGCTTGG

Primer TR2	GCTGGCGTAATAGCGAAGAGG
Primer TR3	CCTGAATGGCGAATGCTAGAG
Primer TL1	CAGATAAGGGAATTAGGGTTCCTATAGG
Primer TL2	TATAGGGTTTCGCTCATGTGTTGAGC
Primer TL3	AGTACATTAAAAACGTCCGCAATGTG
Gene expression:	
MpRSL1-F	AGATGAGTCTGGGGCAACC
MpRSL1-R	GGATGAGCGCTTTAGAGTG
MpEF1-F	CCGAGATCCTGACCAAGG
MpEF1-R	GAGGTGGGTACTCAGCGAAG
MpCUL1-F	AGGATGTGGACAAGGATAGACG
MpCUL1-R	GTTGATGTGGCAACACCTTG
MpACT-F	AGGCATCTGGTATCCACGAG
MpACT-R	ACATGGTCGTTCCTCCAGAC
AtRHD6-F	CCTAAATCCGCTGGAAACAA
AtRHD6-R	CTCTTGGATTCTTGGCTGCT
AtRSL1-F	CCCTAAACTGGCTGGCAATA
AtRSL1-R	TCTTGGCTGCTAGGCTTTGT
AtUBQ-F	GGCCTTGTATAATCCCTGATGAATAAG
AtUBQ-R	AAAGAGATAACAGGAACGGAAACATAGT

Supplemental references

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