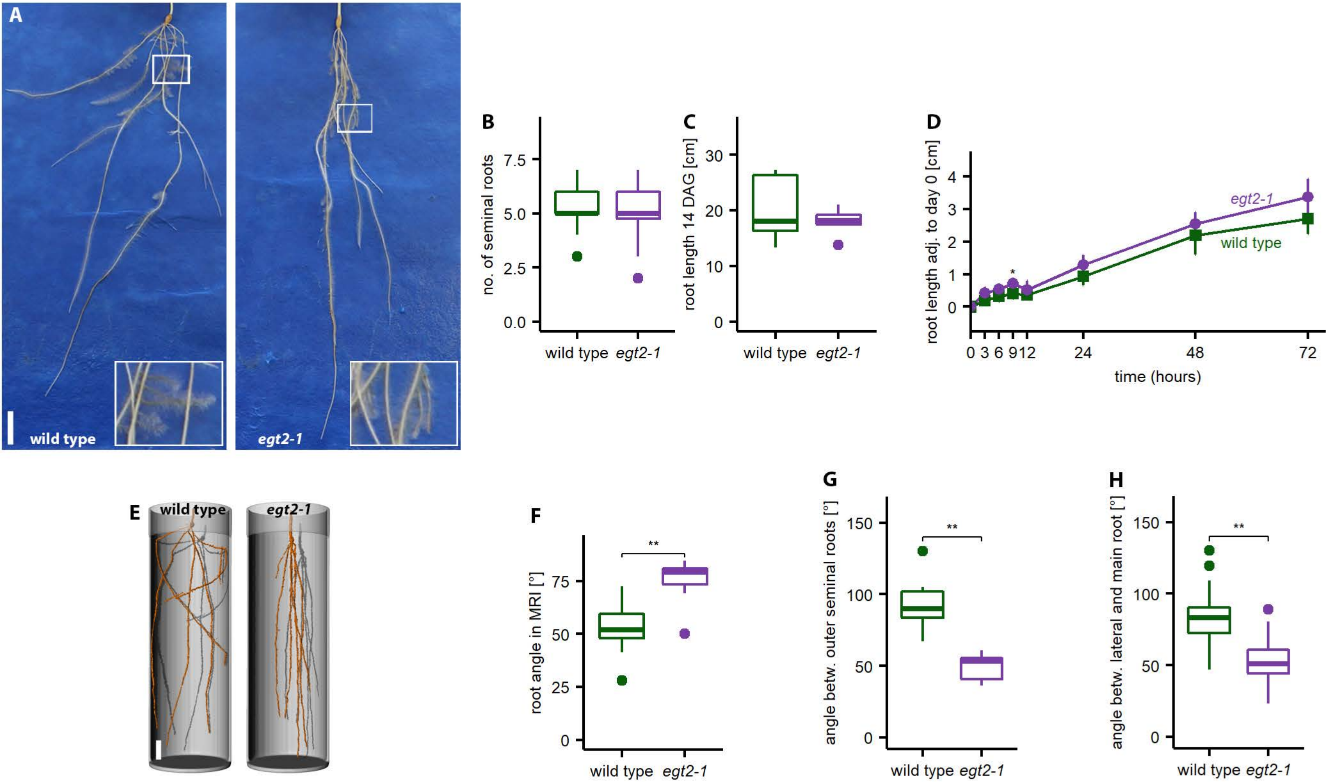

A
wild type
egt2-1
B
no. of seminal roots
0.0
2.5
5.0
7.5
wild type
egt2-1
C
root length 14 DAG [cm]
0
10
20
30
wild type
egt2-1
D
root length adj. to day 0 [cm]
0
1
2
3
4
*
egt2-1
wild type
0 3 6 9 12
24
48
72
time (hours)
E
wild type
egt2-1
F
root angle in MRI [°]
0
25
50
75
**
wild type
egt2-1
G
angle betw. outer seminal roots [°]
0
50
100
150
**
wild type
egt2-1
H
angle betw. lateral and main root [°]
0
50
100
150
**
wild type
egt2-1

Supplementary Figure 1: Root phenotype of *egt2-1*

A Wild type and *egt2-1* roots grown on paper sheets, 14 days after germination (DAG). Scale bar: 2 cm.

B Number of seminal roots 7 DAG; n= 40 per genotype in one experiment.

C Root length 14 DAG; n = 8-9 per genotype in two independent experiments.

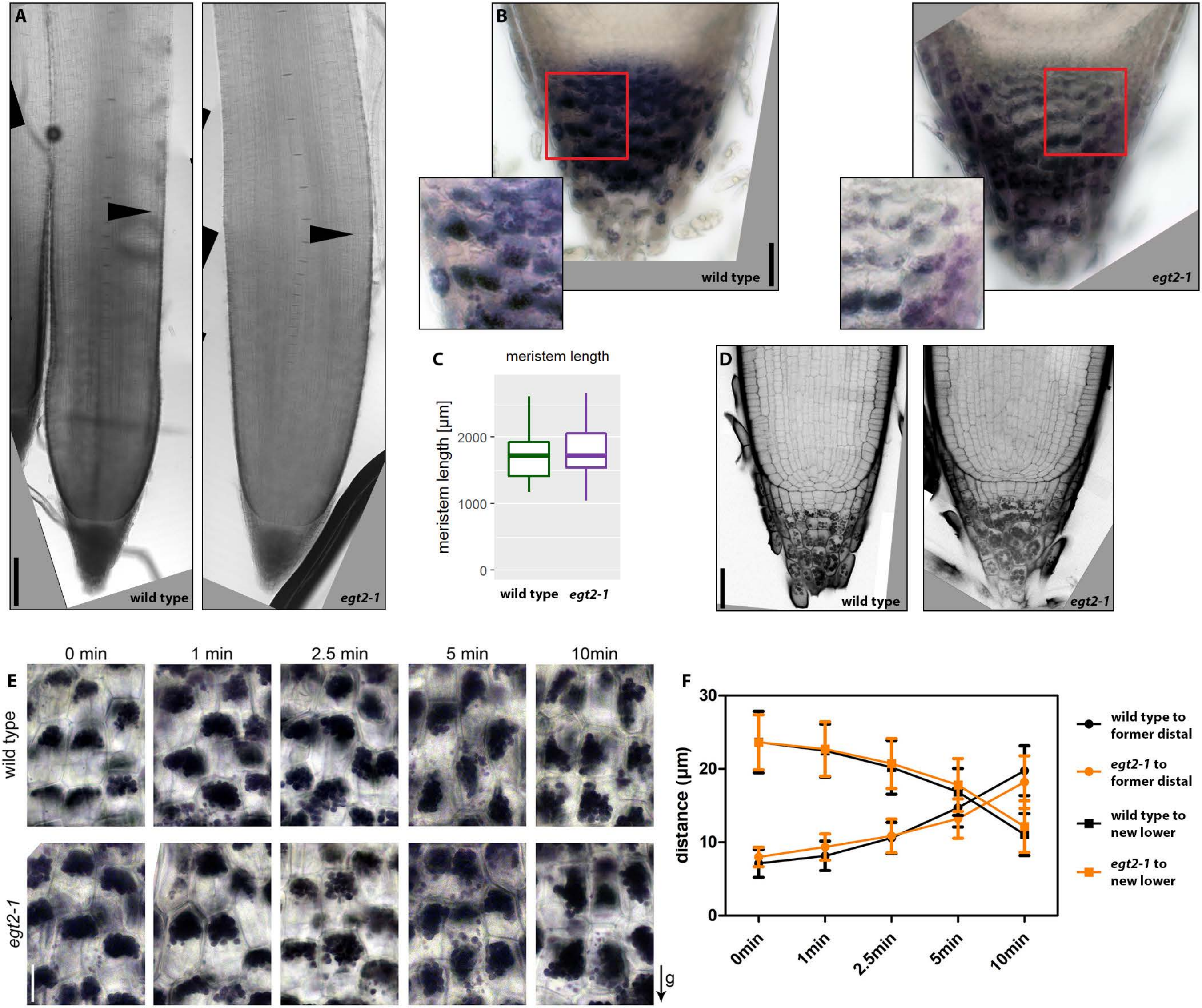
D Root length after rotation; n = 32 in three independent experiments; compare Figure 1G; standard deviation is depicted; two-tailed t-test did not show any significant differences between the genotypes at respective time points; all measurements were normalized to the starting length of the roots at time point 0.

E Magnetic resonance imaging (MRI) pictures of wild type and *egt2-1* roots grown in soil for 7 DAG. Scale bar: 2 cm.

F Root angle of roots grown in soil for 3 DAG and captured by MRI (see D); n = 17-18 per genotype; two-tailed t-test, ** $p < 0.01$.

G Root angle of seminal roots of plants grown in rhizotrons, measured by angle between the outermost seminal roots; 26 DAG; n = 10-11 per genotype; two-tailed t-test, ** $p < 0.01$.

H Root angle of lateral roots of plants grown in rhizotrons, angle measured between main roots and lateral roots; 26 DAG: n = 5 (30 lateral roots per plant) per genotype; two-tailed t-test, ** $p < 0.01$.


A
wild type
egt2-1
B
wild type
egt2-1
C
meristem length
meristem length [µm]
0
1000
2000
wild type
egt2-1
D
wild type
egt2-1
0 min
1 min
2.5 min
5 min
10min
E
wild type
egt2-1
g
F
distance (µm)
0
10
20
30
0min
1min
2.5min
5min
10min
wild type to former distal
egt2-1 to former distal
wild type to new lower
egt2-1 to new lower

Supplementary Figure 2: Meristem phenotype of *egt2-1* resembles the wild type phenotype

A Root meristem of wild type and *egt2-1* 7 DAG; arrow heads mark transition to elongation zone. Scale bar: 200 µm.

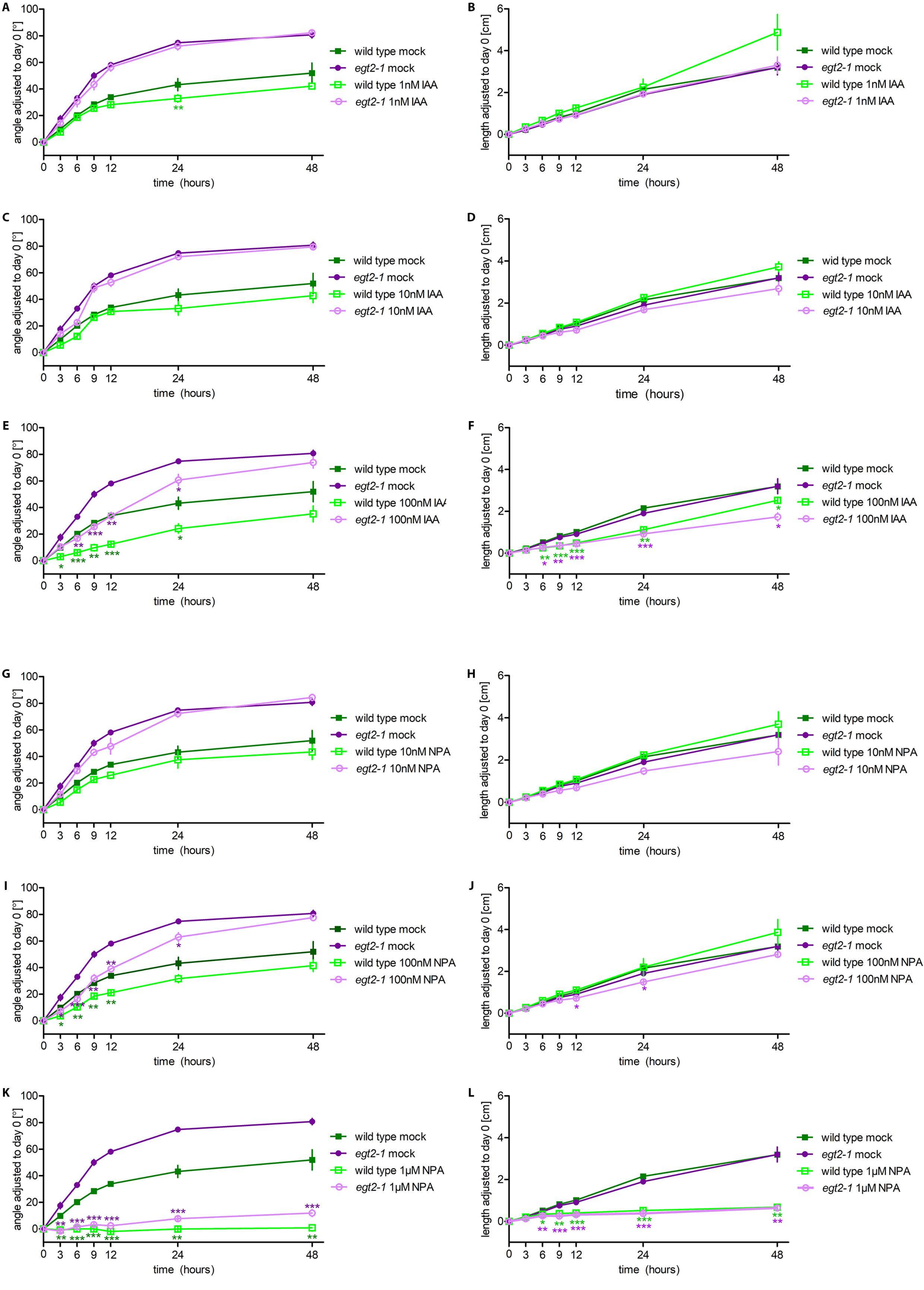
B Wild type and *egt2-1* root cap 7 DAG. Scale bar: 50 µm; brightness adjusted in magnification.

C Quantification of meristem length 7 DAG; n= 15-20 roots per genotype; two-tailed t-test does not show any significant difference.

D Lateral roots of wild type and *egt2-1* plants 14 DAG stained by mPS-PI staining. Scale bar: 50 µm.

E Root caps of wild type and *egt2-1* plants 7 DAG, after 180° rotation at the indicated time points, starch granules stained with Lugol staining. Scale bar: 20 µm.

F Quantification of the starch granule distance to the former distal or new lower cell wall, measured from the center of the starch granules, n = 4-9 plants per time point, 6-10 cells of a size of 25-40 µm were measured per root.


A
angle adjusted to day 0 [°]
time (hours)
wild type mock
egt2-1 mock
wild type 1nM IAA
egt2-1 1nM IAA
B
length adjusted to day 0 [cm]
time (hours)
wild type mock
egt2-1 mock
wild type 1nM IAA
egt2-1 1nM IAA
C
angle adjusted to day 0 [°]
time (hours)
wild type mock
egt2-1 mock
wild type 10nM IAA
egt2-1 10nM IAA
D
length adjusted to day 0 [cm]
time (hours)
wid type mock
egt2-1 mock
wild type 10nM IAA
egt2-1 10nM IAA
E
angle adjusted to day 0 [°]
time (hours)
wild type mock
egt2-1 mock
wild type 100nM IAA
egt2-1 100nM IAA
F
length adjusted to day 0 [cm]
time (hours)
wild type mock
egt2-1 mock
wild type 100nM IAA
egt2-1 100nM IAA
G
angle adjusted to day 0 [°]
time (hours)
wild type mock
egt2-1 mock
wild type 10nM NPA
egt2-1 10nM NPA
H
length adjusted to day 0 [cm]
time (hours)
wild type mock
egt2-1 mock
wild type 10nM NPA
egt2-1 10nM NPA
I
angle adjusted to day 0 [°]
time (hours)
wild type mock
egt2-1 mock
wild type 100nM NPA
egt2-1 100nM NPA
J
length adjusted to day 0 [cm]
time (hours)
wild type mock
egt2-1 mock
wild type 100nM NPA
egt2-1 100nM NPA
K
angle adjusted to day 0 [°]
time (hours)
wild type mock
egt2-1 mock
wild type 1μM NPA
egt2-1 1μM NPA
L
length adjusted to day 0 [cm]
time (hours)
wild type mock
egt2-1 mock
wild type 1μM NPA
egt2-1 1μM NPA

Supplementary Figure 3: *egt2-1* mutant and wild type react similarly to auxin treatment

A, **C**, **E**, **G**, **I, K** Angle of the root tip after indicated treatment and rotation for 90° (at time point 0).

B, **D**, **F**, **H**, **J, L** Length of the roots after indicated treatment and rotation for 90° (at time point 0); phytohormone treated plants were compared to the mock-treated plants of the same genotype at the respective time points by a two-tailed t-test, * $p < 0.05$; **$p<0.01$, and *** $p<0.001$; standard deviation is depicted; to account for the different starting angles of the roots, all measurements were normalized to the starting angle/length of the roots at time point 0.

A, B 1 nM IAA treatment; n = 4 plants per genotype and treatment.

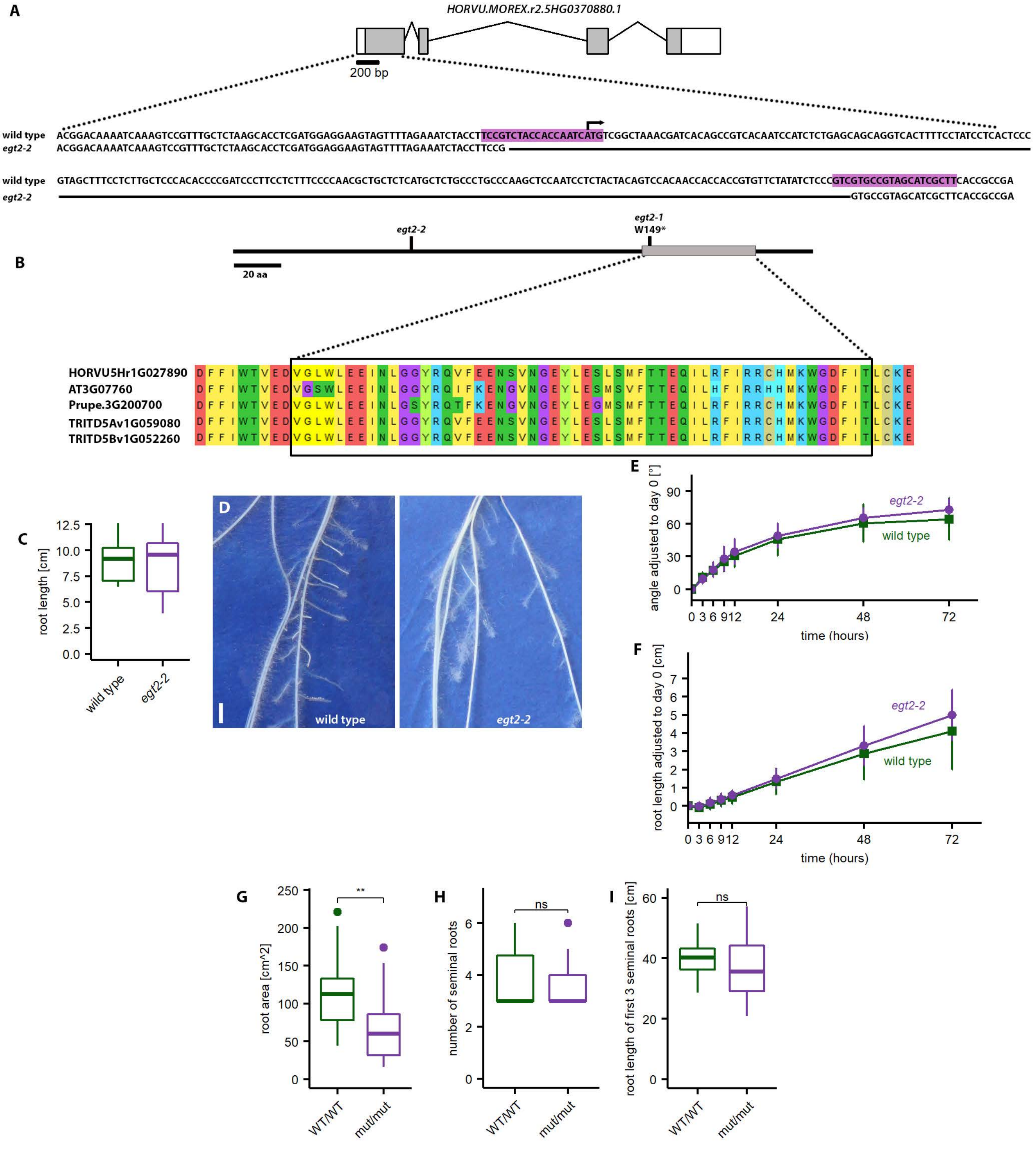
C, D 10 nM IAA treatment; n = 4 plants per genotype and treatment in two independent experiments.

E, F 100 nM IAA treatment; n = 4 plants per genotype and treatment in two independent experiments.

G, H 10 nM NPA treatment; n = 4 plants per genotype and treatment in two independent experiments.

I, J 100 nM NPA treatment; n = 4 plants per genotype and treatment in two independent experiments.

K, L 1 µM NPA treatment; n = 4 plants per genotype and treatment in two independent experiments.


A
HORVU.MOREX.r2.5HG0370880.1
200 bp
wild type ACGGACAAAATCAAAGTCCGTTTGCTCTAAGCACCTCGATGGAGGAAGTAGTTTTAGAAATCTACCTTCCGTCTACCACCAATCATGTCGGCTAAACGATCACAGCCGTCACAATCCATCTCTGAGCAGCAGGTCACTTTTCCTATCCTCACTCCC
egt2-2 ACGGACAAAATCAAAGTCCGTTTGCTCTAAGCACCTCGATGGAGGAAGTAGTTTTAGAAATCTACCTTCCG
wild type GTAGCTTTCCTCTTGCTCCCACACCCCGATCCCTTCCTCTTTCCCCAACGCTGCTCTCATGCTCTGCCCTGCCCAAGCTCCAATCCTCTACTACAGTCCACAACCACCACCGTGTTCTATATCTCCCGTCGTGCCGTAGCATCGCTTCACCGCCGA
egt2-2 GTGCCGTAGCATCGCTTCACCGCCGA
B
egt2-2
egt2-1
W149*
20 aa
HORVU5Hr1G027890 D F F I W T V E D V G L W L E E I N L G G Y R Q V F E E N S V N G E Y L E S L S M F T T E Q I L R F I R R C H M K W G D F I T L C K E
AT3G07760 D F F I W T V E D V G S W L E E I N L G G Y R Q I F K E N G V N G E Y L E S M S V F T T E Q I L H F I R R H H M K W G D F I T L C K E
Prupe.3G200700 D F F I W T V E D V G L W L E E I N L G S Y R Q T F K E N G V N G E Y L E G M S M F T T E Q I L R F I R R C H M K W G D F I T L C K E
TRITD5Av1G059080 D F F I W T V E D V G L W L E E I N L G G Y R Q V F E E N S V N G E Y L E S L S M F T T E Q I L R F I R R C H M K W G D F I T L C K E
TRITD5Bv1G052260 D F F I W T V E D V G L W L E E I N L G G Y R Q V F E E N S V N G E Y L E S L S M F T T E Q I L R F I R R C H M K W G D F I T L C K E
C
root length [cm]
12.5
10.0
7.5
5.0
2.5
0.0
wild type
egt2-2
D
wild type
egt2-2
E
angle adjusted to day 0 [°]
90
60
30
0
egt2-2
wild type
0 3 6 9 12 24 48 72
time (hours)
F
root length adjusted to day 0 [cm]
7
6
5
4
3
2
1
0
egt2-2
wild type
0 3 6 9 12 24 48 72
time (hours)
G
root area [cm^2]
250
200
150
100
50
0
**
WT/WT
mut/mut
H
number of seminal roots
6
4
2
0
ns
WT/WT
mut/mut
I
root length of first 3 seminal roots [cm]
60
40
20
0
ns
WT/WT
mut/mut

Supplementary Figure 4: CRISPR/Cas9 induced mutation in *EGT2* and conserved function in wheat

A Gene model of *EGT2* and partial DNA sequence; arrow marks translation start site; purple boxes mark the CRISPR target sites; in the *egt2-2* mutant line, CRISPR/Cas induced a deletion between the target sites as depicted.

B Protein structure of EGT2 with the SAM domain from amino acid 172 - 225; protein alignment of EGT2, AtSAM5 (At3g07760) and peach WEEP (Prupe. 3G200700) in the SAM region.

C Root length 7 DAG, two-tailed t-test does not show a significant difference (p <0.05); n = 15-17 in two independent experiments.

D Wild type GP and *egt2-2* lateral roots 14 DAG. Scale bar: 1 cm.

E Root tip angle after rotation; plants 5 DAG were rotated by 90° (time 0) and the root tip angle was measured over time; n = 20 per genotype in two independent experiments; the two genotypes were compared between each other at the respective time points by a two-tailed t-test and no significant difference was detected; standard deviation is depicted; to account for the different starting angles of the roots, all measurements were normalized to the starting angle of the roots at time 0.

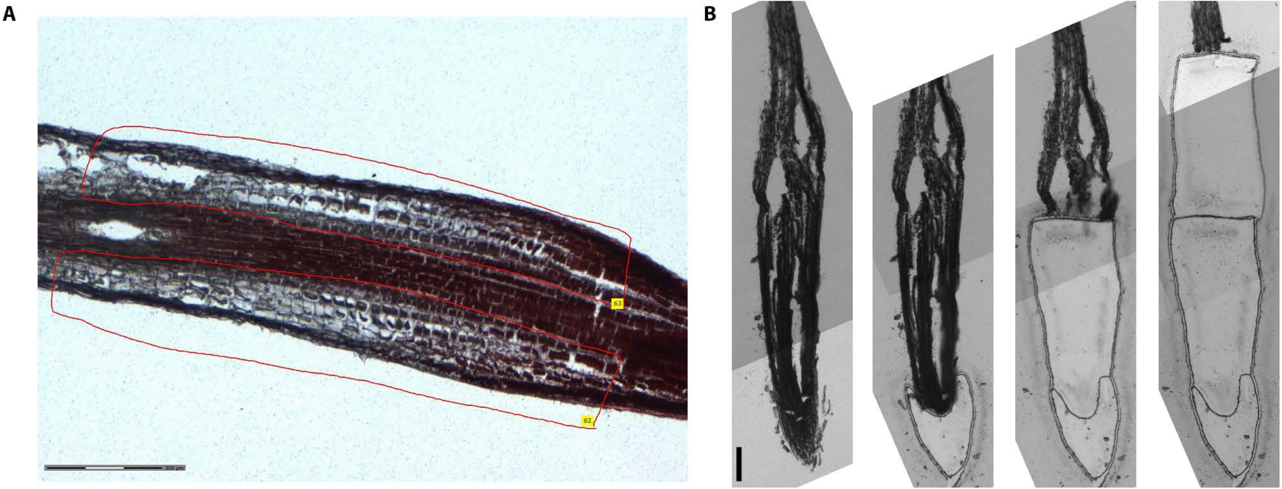
F Root length after rotation as described in D; n = 20 per genotype in two independent experiments.

G Area enclosed by the first three seminal root of wild type (WT/WT) and *egt2* (mut/mut) wheat seedling at 7 DAG; n = 16 and 35 for wild type and mutant, respectively.

H Number of seminal roots of wild type (WT/WT) and *egt2* (mut/mut) wheat seedling at 7 DAG; n = 18 and 39 for wild type and mutant, respectively.

I Length of the first three seminal root in wild type (WT/WT) and *egt2* (mut/mut) wheat seedling at 7DAG; n = 16 and 35 for wild type and mutant, respectively.

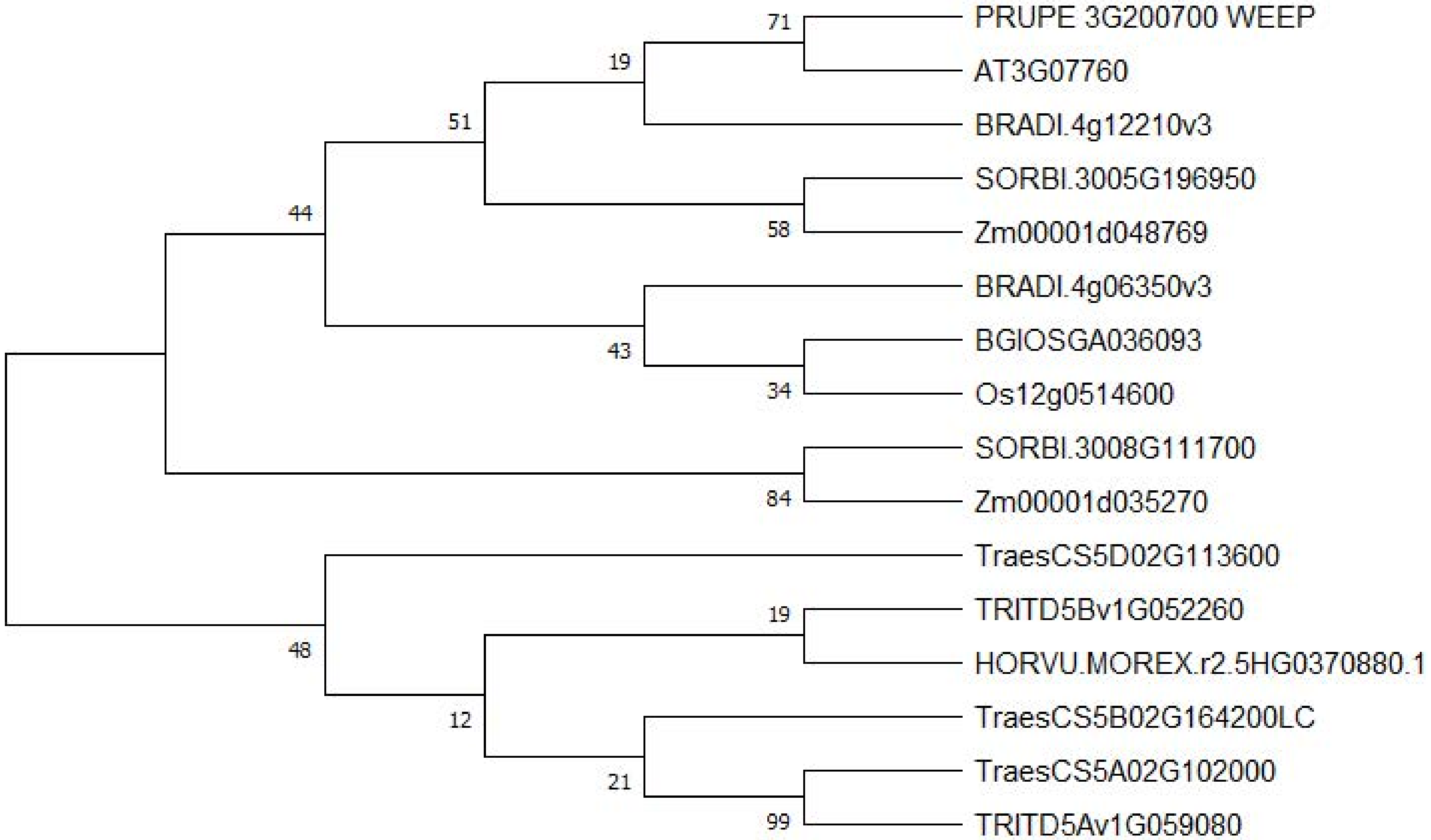
Wheat plants analyzed in G, H and I were derived from two independent segregating populations; the two genotypes were compared by a two-tailed t-test (* p <0.05, ns = no significant difference).


A
63
62
300 µm
B

Supplementary Figure 5: LCM samples

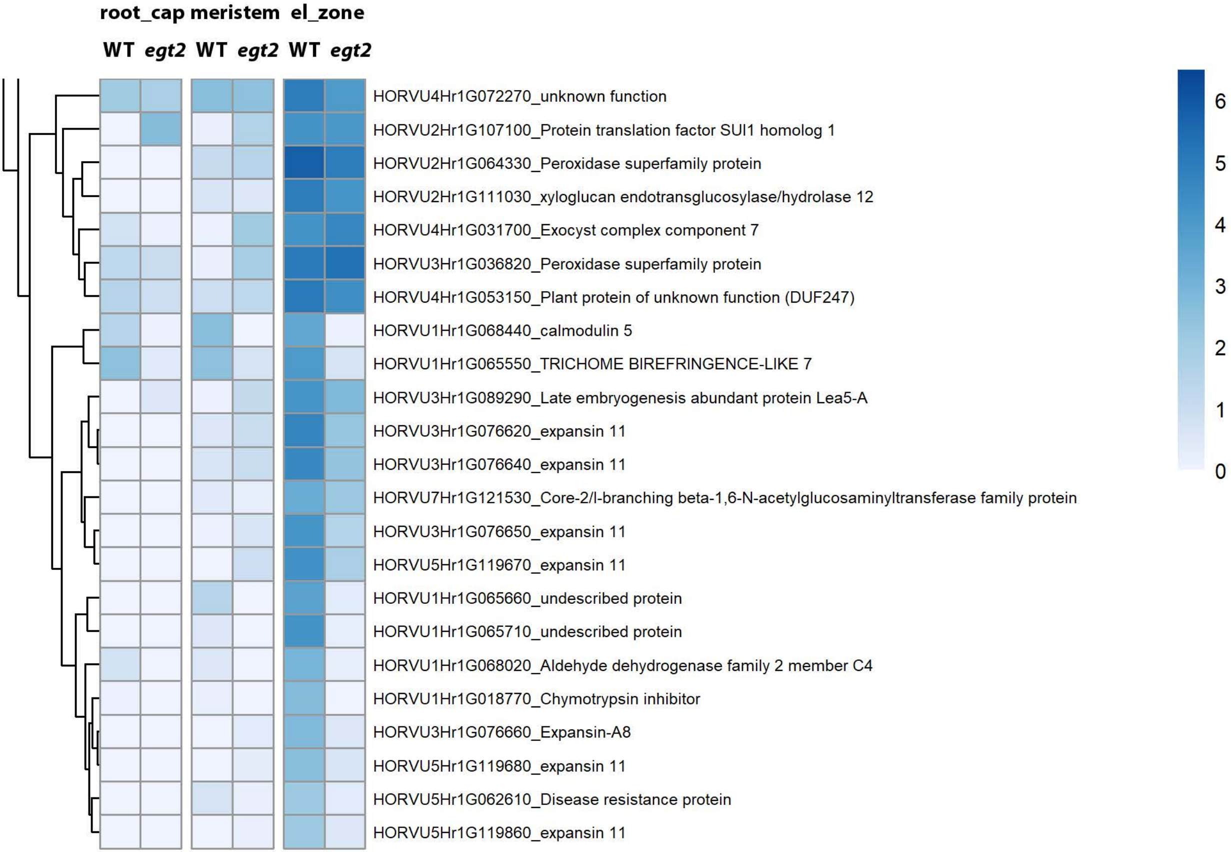
A Representative picture of rotated root before tissue dissection at the LCM. Scale bar: 300 µm.

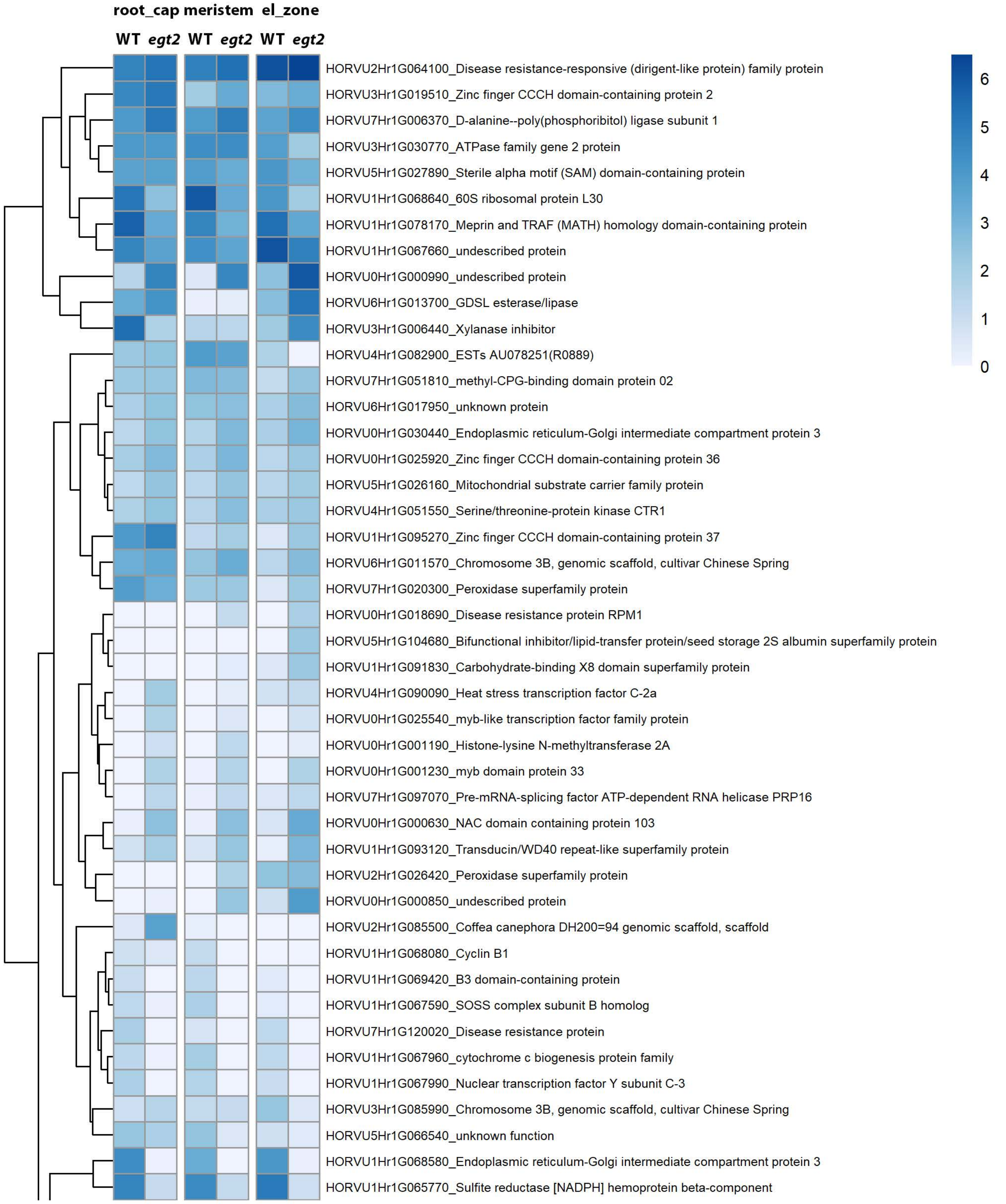
B Representative picture of root before and after tissue dissection at the LCM for RNAseq. Scale bar: 300 µm.


71
19
51
44
58
43
34
84
48
19
12
21
99
PRUPE 3G200700 WEEP
AT3G07760
BRADI.4g12210v3
SORBI.3005G196950
Zm00001d048769
BRADI.4g06350v3
BGIOSGA036093
Os12g0514600
SORBI.3008G111700
Zm00001d035270
TraesCS5D02G113600
TRITD5Bv1G052260
HORVU.MOREX.r2.5HG0370880.1
TraesCS5B02G164200LC
TraesCS5A02G102000
TRITD5Av1G059080

Supplementary Figure 6: Phylogenetic tree of EGT2 and related proteins

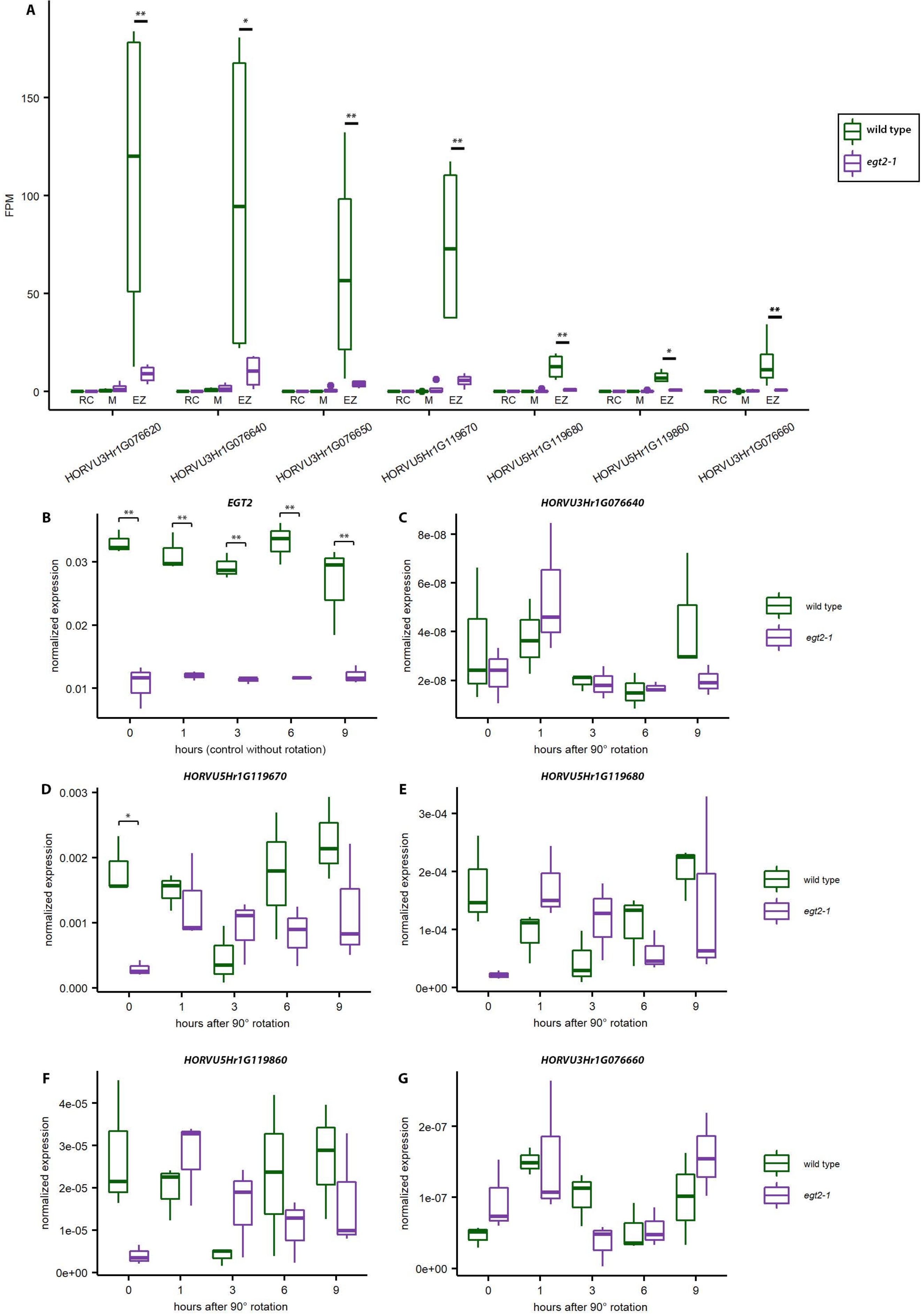
Species abbreviations: AT: *Arabidopsis thaliana*; BGIOS: *Oryza sativa Indica Group*; BRADI: *Brachypodium dystachyon*; Os: *Oryza sativa Japonica Group;* PRUPE: *Prunus persica*; SORBI: *Sorghum bicolor;* Traes: *Triticum aestivum;* TRITD: *Triticum durum;* Zm: *Zea mays.*


root_cap
meristem
el_zone
WT
egt2
WT
egt2
WT
egt2
HORVU4Hr1G072270_unknown function
HORVU2Hr1G107100_Protein translation factor SUI1 homolog 1
HORVU2Hr1G064330_Peroxidase superfamily protein
HORVU2Hr1G111030_xyloglucan endotransglucosylase/hydrolase 12
HORVU4Hr1G031700_Exocyst complex component 7
HORVU3Hr1G036820_Peroxidase superfamily protein
HORVU4Hr1G053150_Plant protein of unknown function (DUF247)
HORVU1Hr1G068440_calmodulin 5
HORVU1Hr1G065550_TRICHOME BIREFRINGENCE-LIKE 7
HORVU3Hr1G089290_Late embryogenesis abundant protein Lea5-A
HORVU3Hr1G076620_expansin 11
HORVU3Hr1G076640_expansin 11
HORVU7Hr1G121530_Core-2/I-branching beta-1,6-N-acetylglucosaminyltransferase family protein
HORVU3Hr1G076650_expansin 11
HORVU5Hr1G119670_expansin 11
HORVU1Hr1G065660_undescribed protein
HORVU1Hr1G065710_undescribed protein
HORVU1Hr1G068020_Aldehyde dehydrogenase family 2 member C4
HORVU1Hr1G018770_Chymotrypsin inhibitor
HORVU3Hr1G076660_Expansin-A8
HORVU5Hr1G119680_expansin 11
HORVU5Hr1G062610_Disease resistance protein
HORVU5Hr1G119860_expansin 11
6
5
4
3
2
1
0


root_cap meristem el_zone
WT egt2 WT egt2 WT egt2
HORVU2Hr1G064100_Disease resistance-responsive (dirigent-like protein) family protein
HORVU3Hr1G019510_Zinc finger CCCH domain-containing protein 2
HORVU7Hr1G006370_D-alanine--poly(phosphoribitol) ligase subunit 1
HORVU3Hr1G030770_ATPase family gene 2 protein
HORVU5Hr1G027890_Sterile alpha motif (SAM) domain-containing protein
HORVU1Hr1G068640_60S ribosomal protein L30
HORVU1Hr1G078170_Meprin and TRAF (MATH) homology domain-containing protein
HORVU1Hr1G067660_undescribed protein
HORVU0Hr1G000990_undescribed protein
HORVU6Hr1G013700_GDSL esterase/lipase
HORVU3Hr1G006440_Xylanase inhibitor
HORVU4Hr1G082900_ESTs AU078251(R0889)
HORVU7Hr1G051810_methyl-CPG-binding domain protein 02
HORVU6Hr1G017950_unknown protein
HORVU0Hr1G030440_Endoplasmic reticulum-Golgi intermediate compartment protein 3
HORVU0Hr1G025920_Zinc finger CCCH domain-containing protein 36
HORVU5Hr1G026160_Mitochondrial substrate carrier family protein
HORVU4Hr1G051550_Serine/threonine-protein kinase CTR1
HORVU1Hr1G095270_Zinc finger CCCH domain-containing protein 37
HORVU6Hr1G011570_Chromosome 3B, genomic scaffold, cultivar Chinese Spring
HORVU7Hr1G020300_Peroxidase superfamily protein
HORVU0Hr1G018690_Disease resistance protein RPM1
HORVU5Hr1G104680_Bifunctional inhibitor/lipid-transfer protein/seed storage 2S albumin superfamily protein
HORVU1Hr1G091830_Carbohydrate-binding X8 domain superfamily protein
HORVU4Hr1G090090_Heat stress transcription factor C-2a
HORVU0Hr1G025540_myb-like transcription factor family protein
HORVU0Hr1G001190_Histone-lysine N-methyltransferase 2A
HORVU0Hr1G001230_myb domain protein 33
HORVU7Hr1G097070_Pre-mRNA-splicing factor ATP-dependent RNA helicase PRP16
HORVU0Hr1G000630_NAC domain containing protein 103
HORVU1Hr1G093120_Transducin/WD40 repeat-like superfamily protein
HORVU2Hr1G026420_Peroxidase superfamily protein
HORVU0Hr1G000850_undescribed protein
HORVU2Hr1G085500_Coffea canephora DH200=94 genomic scaffold, scaffold
HORVU1Hr1G068080_Cyclin B1
HORVU1Hr1G069420_B3 domain-containing protein
HORVU1Hr1G067590_SOSS complex subunit B homolog
HORVU7Hr1G120020_Disease resistance protein
HORVU1Hr1G067960_cytochrome c biogenesis protein family
HORVU1Hr1G067990_Nuclear transcription factor Y subunit C-3
HORVU3Hr1G085990_Chromosome 3B, genomic scaffold, cultivar Chinese Spring
HORVU5Hr1G066540_unknown function
HORVU1Hr1G068580_Endoplasmic reticulum-Golgi intermediate compartment protein 3
HORVU1Hr1G065770_Sulfite reductase [NADPH] hemoprotein beta-component
6
5
4
3
2
1
0

Supplementary Figure 7: Heat map of differentially expressed genes (FDR <5% and log2FC >|2|) between wild-type and egt2-1 in root cap, root meristem and elongation zone

Average of log1p of fpm normalized counts of four biological replicates of the respective genotype and tissue is shown in blue scale.


A
FPM
0
50
100
150
**
*
**
**
**
*
**
RC
M
EZ
HORVU3Hr1G076620
HORVU3Hr1G076640
HORVU3Hr1G076650
HORVU5Hr1G119670
HORVU5Hr1G119680
HORVU5Hr1G119860
HORVU3Hr1G076660
wild type
egt2-1
B
EGT2
normalized expression
0.01
0.02
0.03
0
1
3
6
9
hours (control without rotation)
C
HORVU3Hr1G076640
2e-08
4e-08
6e-08
8e-08
hours after 90° rotation
D
HORVU5Hr1G119670
0.000
0.001
0.002
0.003
E
HORVU5Hr1G119680
0e+00
1e-04
2e-04
3e-04
F
HORVU5Hr1G119860
0e+00
1e-05
2e-05
3e-05
4e-05
G
HORVU3Hr1G076660
0e+00
1e-07
2e-07

Supplementary Figure 8: Expression of expansins

A Fpm normalized expression values of the expansins differentially expressed in the gene set; padj is depicted with * $p < 0.05$, ** $p < 0.01$; RC: root cap, M: meristem, EZ: elongation zone.

B qRT-PCR of *EGT2* expression in meristem and elongation zone without rotation as control to Figure 3B; normalized to *tubulin;* two-tailed t-test, * $p < 0.05$, ** $p < 0.01$.

C qRT-PCR of *HORVU3Hr1G076640* expression in meristem and elongation zone after rotation of 90°; normalized to *tubulin*.

D qRT-PCR of *HORVU5Hr1G119670* expression in meristem and elongation zone after rotation of 90°; normalized to *tubulin*; two-tailed t-test, ** $p < 0.01$.

E qRT-PCR of *HORVU5Hr1G119680* expression in meristem and elongation zone after rotation of 90°; normalized to *tubulin*.

F qRT-PCR of *HORVU5Hr1G119860* expression in meristem and elongation zone after rotation of 90°; normalized to *tubulin*.

G qRT-PCR of *HORVU3Hr1G076660* expression in meristem and elongation zone after rotation of 90°; normalized to *tubulin*.

Table S1. Mutated high confidence protein coding genes in the egt2 mapping intervall on chromosome 5H

Gene ID	Start	End	Gene length	Gene function	5´Position of mutation	Type of mutation	ID of mutation
HORVU5Hr1G027890	160633192	160636469	3278	Sterile alpha motif (SAM) domain-containing protein	160635280	G to A	chr5H_part1_160635280_A
HORVU5Hr1G029670	178370028	178374066	4039	F-box family protein	178376238	C to T	chr5H_part1_178376238_T
HORVU5Hr1G029910	182793217	182822426	29210	ATPase-like fidgetin	182789159	C to T	chr5H_part1_182789159_T
HORVU5Hr1G030940	192355205	192356197	993	50S ribosomal protein L14	192351249	C to T	chr5H_part1_192351249_T
HORVU5Hr1G031470	199378390	199523314	144925	Chaperone protein ClpB	199516725	G to A	chr5H_part1_199516725_A
HORVU5Hr1G031630	202056154	202057757	1604	40S ribosomal protein S19	202062593	G to C	chr5H_part1_202062593_C
HORVU5Hr1G032430	211283193	211283420	228	Non-specific serine/threonine protein kinase	211283531	G to A	chr5H_part1_211283531_A

Table S1. cont.

Gene ID	Mutation effect on transcript	Internal annotation of the mutant	Affected isoform	Position of mutation in cDNA	Position of Mutation in protein
HORVU5Hr1G027890	premature stop codon	EXON_5H_part1_160633192_160633623	HORVU5Hr1G027890.1	c.525	p.175
HORVU5Hr1G029670	downstream coding region	EXON_5H_part1_178370028_178370491	HORVU5Hr1G029670.1	c.*2517	
HORVU5Hr1G029910	upstream coding region	EXON_5H_part1_182793217_182793720	HORVU5Hr1G029910.8	c.-4159	
HORVU5Hr1G030940	downstream coding region	CDS_5H_part1_192355205_192355579	HORVU5Hr1G030940.1	c.*3956	
HORVU5Hr1G031470	upstream coding region	EXON_5H_part1_199378390_199378652	HORVU5Hr1G031470.8	c.-4996	
HORVU5Hr1G031630	upstream coding region	EXON_5H_part1_202056154_202056231	HORVU5Hr1G031630.1	c.-4838	
HORVU5Hr1G032430	upstream coding region	CDS_5H_part1_211283193_211283420	HORVU5Hr1G032430.1	c.-111	

Table S2. Overview of RNAseq reads and mapping results

Lane	Sample	PF Clusters	Yield (Mbases)	% >= Q30 bases	Mean Quality Score
1	107229	22,376,577	4,520	95.56	36.22
1	107231	21,932,224	4,430	95.24	36.1
1	107233	19,561,632	3,951	95.25	36.14
1	107235	20,630,560	4,167	95.71	36.24
1	107237	19,386,123	3,916	95.37	36.17
1	107239	20,053,643	4,051	95.72	36.23
1	107241	20,626,439	4,167	95.6	36.21
1	107243	20,166,570	4,074	95.61	36.2
1	107245	21,604,056	4,364	96.06	36.31
1	107247	18,975,817	3,833	95.71	36.22
1	107249	19,326,567	3,904	95.38	36.17
1	107251	19,635,769	3,966	94.26	35.98
1	107253	20,791,711	4,200	95.23	36.14
1	107255	21,350,631	4,313	95.93	36.26
1	107257	22,062,775	4,457	95.79	36.26
1	107259	21,507,018	4,344	95.87	36.25
1	107261	20,644,170	4,170	95.52	36.19
1	107263	22,681,047	4,582	95.2	36.13
1	107265	18,683,628	3,774	95.72	36.22
1	107267	24,278,896	4,904	96.05	36.3
1	107269	19,355,318	3,910	95.8	36.25
1	107271	21,218,515	4,286	95.53	36.2
1	107273	20,290,927	4,099	93.83	35.9
1	107275	18,430,934	3,723	95.64	36.22
2	107229	22,562,294	4,558	95.69	36.25
2	107231	22,189,773	4,482	95.36	36.12
2	107233	19,752,586	3,990	95.37	36.17
2	107235	20,904,791	4,223	95.83	36.26
2	107237	19,529,397	3,945	95.49	36.2
2	107239	20,310,327	4,103	95.84	36.25
2	107241	20,856,789	4,213	95.72	36.23
2	107243	20,436,490	4,128	95.73	36.23
2	107245	21,795,430	4,403	96.19	36.33
2	107247	19,111,224	3,860	95.81	36.24
2	107249	19,473,688	3,934	95.51	36.19
2	107251	19,846,346	4,009	94.36	36
2	107253	20,955,605	4,233	95.34	36.16
2	107255	21,594,671	4,362	96.03	36.28
2	107257	22,234,718	4,491	95.92	36.28
2	107259	21,711,249	4,386	95.98	36.27
2	107261	20,793,488	4,200	95.66	36.22
2	107263	22,956,790	4,637	95.33	36.16
2	107265	18,835,184	3,805	95.83	36.24
2	107267	24,545,072	4,958	96.18	36.32
2	107269	19,540,410	3,947	95.92	36.27
2	107271	21,405,829	4,324	95.65	36.22

Table S2. cont.

Lane	Sample	PF Clusters	Yield (Mbases)	% >= Q30 bases	Mean Quality Score
2	107273	20,532,358	4,148	93.92	35.92
2	107275	18,578,126	3,753	95.77	36.25

Table S3: Differentially expressed genes

	Elongation zone		**Meristem**		**Root_cap**		**Gene function**
Gene ID	**Log2FC**	**padj**	**Log2FC**	**padj**	**Log2FC**	**padj**	
HORVU0Hr1G000630	4.9	<0.001	6.2	<0.001	5.9	<0.001	NAC domain containing protein 103
HORVU0Hr1G000850	5.0	<0.001	8.2	<0.001	no DE	no DE	undescribed protein
HORVU0Hr1G000990	5.1	<0.001	7.1	<0.001	5.0	<0.001	undescribed protein
HORVU0Hr1G001190	no DE	no DE	6.3	0.005	no DE	no DE	Histone-lysine N-methyltransferase 2A
HORVU0Hr1G001230	6.7	0.002	5.5	0.027	6.9	0.002	myb domain protein 33
HORVU0Hr1G018690	7.0	<0.001	6.1	0.016	no DE	no DE	Disease resistance protein RPM1
HORVU0Hr1G025540	no DE	no DE	no DE	no DE	6.9	0.003	myb-like transcription factor family protein
HORVU0Hr1G025920	no DE	no DE	1.8	0.006	no DE	no DE	Zinc finger CCCH domain-containing protein 36
HORVU0Hr1G030440	1.7	0.013	2.0	<0.001	no DE	no DE	Endoplasmic reticulum-Golgi intermediate compartment protein 3
HORVU1Hr1G018770	-7.6	<0.001	no DE	no DE	no DE	no DE	Chymotrypsin inhibitor
HORVU1Hr1G065550	-5.8	<0.001	-3.5	<0.001	-4.6	0.014	TRICHOME BIREFRINGENCE-LIKE 7
HORVU1Hr1G065660	-6.4	<0.001	-6.7	0.003	no DE	no DE	undescribed protein
HORVU1Hr1G065710	-7.9	<0.001	no DE	no DE	no DE	no DE	undescribed protein
HORVU1Hr1G065770	-6.7	<0.001	-5.3	0.001	-5.7	<0.001	Sulfite reductase [NADPH] hemoprotein beta-component
HORVU1Hr1G067590	no DE	no DE	-6.4	0.001	no DE	no DE	SOSS complex subunit B homolog
HORVU1Hr1G067660	-1.9	<0.001	no DE	no DE	no DE	no DE	undescribed protein
HORVU1Hr1G067960	no DE	no DE	-6.8	<0.001	no DE	no DE	cytochrome c biogenesis protein family
HORVU1Hr1G067990	no DE	no DE	-6.0	0.001	-7.3	0.001	Nuclear transcription factor Y subunit C-3
HORVU1Hr1G068020	-6.4	<0.001	no DE	no DE	no DE	no DE	Aldehyde dehydrogenase family 2 member C4
HORVU1Hr1G068080	no DE	no DE	-5.2	0.046	no DE	no DE	Cyclin B1
HORVU1Hr1G068440	-8.7	<0.001	-7.8	<0.001	no DE	no DE	calmodulin 5
HORVU1Hr1G068580	-8.5	<0.001	-9.5	<0.001	no DE	no DE	Endoplasmic reticulum-Golgi intermediate compartment protein 3
HORVU1Hr1G068640	-3.2	<0.001	-3.6	<0.001	-3.9	<0.001	60S ribosomal protein L30
HORVU1Hr1G069420	no DE	no DE	-6.3	0.002	no DE	no DE	B3 domain-containing protein
HORVU1Hr1G078170	-2.8	<0.001	-2.4	<0.001	-3.5	<0.001	Meprin and TRAF (MATH) homology domain-containing protein
HORVU1Hr1G091830	3.5	0.043	no DE	no DE	no DE	no DE	Carbohydrate-binding X8 domain superfamily protein
HORVU1Hr1G093120	6.2	<0.001	3.4	0.005	no DE	no DE	Transducin/WD40 repeat-like superfamily protein
HORVU1Hr1G095270	3.5	0.012	no DE	no DE	no DE	no DE	Zinc finger CCCH domain-containing protein 37
HORVU2Hr1G026420	no DE	no DE	6.4	0.042	no DE	no DE	Peroxidase superfamily protein
HORVU2Hr1G064100	no DE	no DE	0.8	0.024	no DE	no DE	Disease resistance-responsive (dirigent-like protein) family protein
HORVU2Hr1G064330	-1.3	0.044	no DE	no DE	no DE	no DE	Peroxidase superfamily protein
HORVU2Hr1G085500	no DE	no DE	no DE	no DE	6.0	<0.001	Coffea canephora DH200=94 genomic scaffold, scaffold_890
HORVU2Hr1G107100	no DE	no DE	no DE	no DE	8.5	<0.001	Protein translation factor SUI1 homolog 1
HORVU2Hr1G111030	-1.1	0.026	no DE	no DE	no DE	no DE	xyloglucan endotransglucosylase/hydrolase 12
HORVU3Hr1G006440	no DE	no DE	no DE	no DE	-5.5	0.002	Xylanase inhibitor
HORVU3Hr1G019510	no DE	no DE	2.0	0.034	no DE	no DE	Zinc finger CCCH domain-containing protein 2
HORVU3Hr1G030770	-2.6	0.044	no DE	no DE	no DE	no DE	ATPase family gene 2 protein
HORVU3Hr1G036820	no DE	no DE	5.1	0.031	no DE	no DE	Peroxidase superfamily protein
HORVU3Hr1G076620	-3.7	<0.001	no DE	no DE	no DE	no DE	expansin 11
HORVU3Hr1G076640	-3.3	0.043	no DE	no DE	no DE	no DE	expansin 11
HORVU3Hr1G076650	-4.1	<0.001	no DE	no DE	no DE	no DE	expansin 11

Table S3: cont.

	Elongation zone		**Meristem**		**Root_cap**		**Gene function**
Gene ID	**Log2FC**	**padj**	**Log2FC**	**padj**	**Log2FC**	**padj**	
HORVU3Hr1G076660	-4.4	0.003	no DE	no DE	no DE	no DE	Expansin-A8
HORVU3Hr1G085990	-4.0	0.044	no DE	no DE	no DE	no DE	Chromosome 3B, genomic scaffold, cultivar Chinese Spring
HORVU3Hr1G089290	-2.1	0.003	no DE	no DE	no DE	no DE	Late embryogenesis abundant protein Lea5-A
HORVU4Hr1G031700	no DE	no DE	6.4	0.009	no DE	no DE	Exocyst complex component 7
HORVU4Hr1G051550	no DE	no DE	1.9	<0.001	no DE	no DE	Serine/threonine-protein kinase CTR1
HORVU4Hr1G053150	-1.0	0.002	no DE	no DE	no DE	no DE	Plant protein of unknown function (DUF247)
HORVU4Hr1G072270	-1.5	<0.001	no DE	no DE	no DE	no DE	unknown function
HORVU4Hr1G082900	-6.8	0.004	no DE	no DE	no DE	no DE	ESTs AU078251(R0889)
HORVU4Hr1G090090	no DE	no DE	no DE	no DE	7.4	0.009	Heat stress transcription factor C-2a
HORVU5Hr1G026160	no DE	no DE	1.6	0.029	no DE	no DE	Mitochondrial substrate carrier family protein
HORVU5Hr1G027890	-1.5	0.016	no DE	no DE	no DE	no DE	Sterile alpha motif (SAM) domain-containing protein
HORVU5Hr1G062610	-4.3	0.002	no DE	no DE	no DE	no DE	Disease resistance protein
HORVU5Hr1G066540	no DE	no DE	-3.8	0.028	no DE	no DE	unknown function
HORVU5Hr1G104680	7.6	0.036	no DE	no DE	no DE	no DE	Bifunctional inhibitor/lipid-transf. prot./seed storage 2S albumin prot.
HORVU5Hr1G119670	-3.8	<0.001	no DE	no DE	no DE	no DE	expansin 11
HORVU5Hr1G119680	-3.9	<0.001	no DE	no DE	no DE	no DE	expansin 11
HORVU5Hr1G119860	-3.3	0.019	no DE	no DE	no DE	no DE	expansin 11
HORVU6Hr1G011570	no DE	no DE	1.4	0.005	no DE	no DE	Chromosome 3B, genomic scaffold, cultivar Chinese Spring
HORVU6Hr1G013700	3.8	0.006	no DE	no DE	no DE	no DE	GDSL esterase/lipase
HORVU6Hr1G017950	1.5	0.036	no DE	no DE	no DE	no DE	unknown protein
HORVU7Hr1G006370	no DE	no DE	1.5	0.003	no DE	no DE	D-alanine--poly(phosphoribitol) ligase subunit 1
HORVU7Hr1G020300	3.3	0.026	no DE	no DE	no DE	no DE	Peroxidase superfamily protein
HORVU7Hr1G051810	2.2	0.013	no DE	no DE	no DE	no DE	methyl-CPG-binding domain protein 02
HORVU7Hr1G097070	no DE	no DE	no DE	no DE	6.3	0.047	Pre-mRNA-splicing factor ATP-dependent RNA helicase PRP16
HORVU7Hr1G120020	no DE	no DE	no DE	no DE	-7.5	0.005	Disease resistance protein
HORVU7Hr1G121530	-1.6	<0.001	no DE	no DE	no DE	no DE	Core-2/I-branching beta-1,6-N-acetylglucosaminyltransferase prot.

Table S4: Oligonucleotide primer sequences

Name	**Purpose**	**Barley_identifier**	**Sequence 5´ - 3´**
αTub-F	qRT_PCR	HORVU1Hr1G081280	GAGGTTTGACGGTGCTCTGA
αTub-R	qRT_PCR	HORVU1Hr1G081280	GGTGATCTCGGCAACAGACA
GK-SAM-qRT-fw4	qRT_PCR	HORVU5Hr1G027890	ATTCTGCGGTTTATCAGGCG
GK-SAM-qRT-rv4	qRT_PCR	HORVU5Hr1G027890	AAACACCACCGATAGGCACG
HORVU3Hr1G076640-q-fw	qRT_PCR	HORVU3Hr1G076640	GGGATCGCCTATTTGTGC
HORVU3Hr1G076640-q-rv	qRT_PCR	HORVU3Hr1G076640	GTTGGAGGAGGAAGATGTCC
HORVU5Hr1G119670-q-fw	qRT_PCR	HORVU5Hr1G119670	TGGCAACAGTTCGACTACTAGC
HORVU5Hr1G119670-q-rv	qRT_PCR	HORVU5Hr1G119670	CGGGCAAGGTGATCATAATT
HORVU5Hr1G119680-q-fw	qRT_PCR	HORVU5Hr1G119680	GGCTGCCGGTCAGCTATACC
HORVU5Hr1G119680-q-rv	qRT_PCR	HORVU5Hr1G119680	AAATTCCGATGCTTGTCCG
HORVU5Hr1G119860-q-fw	qRT_PCR	HORVU5Hr1G119860	GGAAGTGCCCAGATGCATAC
HORVU5Hr1G119860-q-rv	qRT_PCR	HORVU5Hr1G119860	CCCGCTTCCAGCATATATTG
HORVU3Hr1G076660-q-fw	qRT_PCR	HORVU3Hr1G076660	CACGAACCGGGACAAATAAG
HORVU3Hr1G076660-q-rv	qRT_PCR	HORVU3Hr1G076660	GTTGAAAATGGAGCTCGGAG
GK-Hyg-fw	CRISPR validation (insertion)		ACTCACCGCGACGTCTGTCG
GK-Hyg-rv	CRISPR validation (insertion)		GCGCGTCTGCTGCTCCATA
GK_HvSAM_CRISPR_seq_fw	CRISPR validation (mutation)	HORVU5Hr1G027890	AGACGTGTTGGATACTATGTCG
GK-HvSAM_gDNA_rv1	CRISPR validation (mutation)	HORVU5Hr1G027890	GCA GAA ACC ACG ATC AAG CAT CAC C
GL-HvSAM-pGGC-fw-V2	EGT2 cloning (greengate plasmids)	HORVU5Hr1G027890	AAAGGTCTCAGGCTTAATGTCGGCTAAACGATCAC
GK-HvSAM-pGGC-rv	EGT2 cloning (greengate plasmids)	HORVU5Hr1G027890	AAAGGTCTCTCTGAAGGTTCCAGCTTTAGGGAGAC