

***New Phytologist* Supporting Information**

Article title: HalALMT1 mediates malate efflux in the cortex of mature cluster rootlets of *Hakea laurina*, occurring naturally in severely phosphorus-impooverished soil

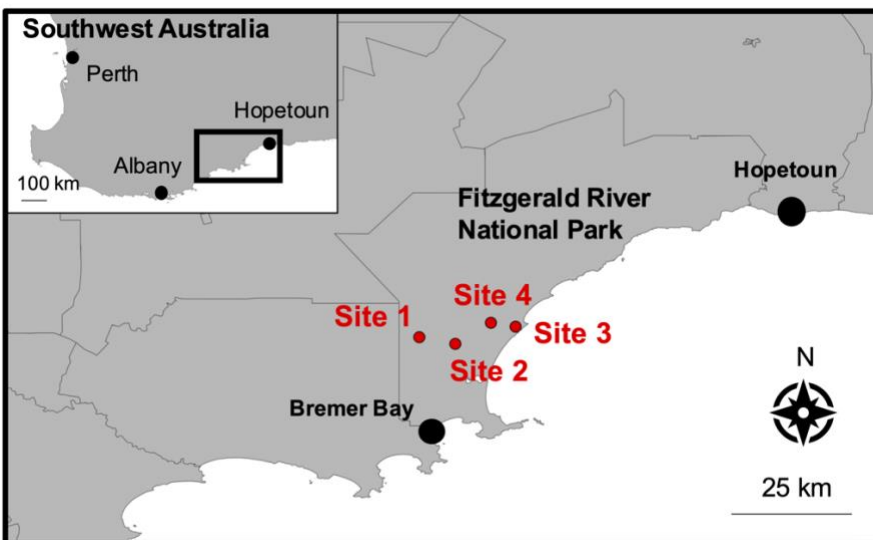
Authors: Hirotsuna Yamada, Lydia Ratna Bunthara, Akira Tanaka, Takuro Kohama, Hayato Maruyama, Wakana Tanaka, Sho Nishida, Tantriani, Akira Oikawa, Keitaro Tawaraya, Toshihiro Watanabe, Shu Tong Liu, Patrick M. Finnegan, Hans Lambers, Takayuki Sasaki, Jun Wasaki

Article acceptance date: 03 February 2025

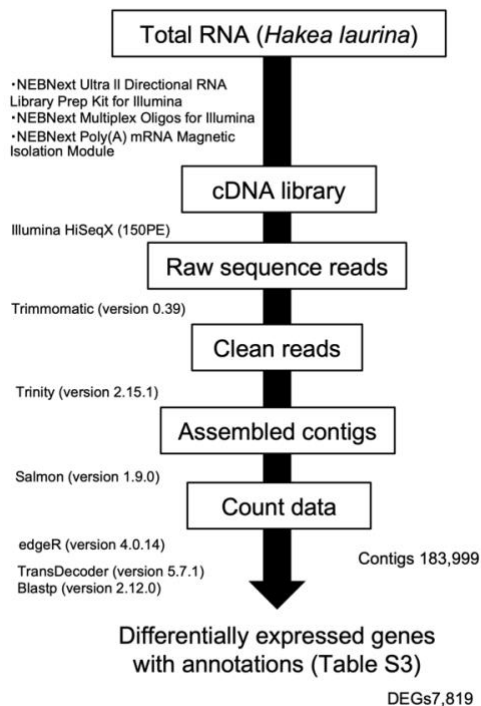
The following Supporting Information is available for this article:

Methods S1 Four sampling sites for *Hakea laurina* in Fitzgerald River National Park in southwest Australia.

The coordinates for site1, site2, site3, and site4 are 34°11'57.0"S 119°21'36.3"E, 34°12'47.4"S 119°26'08.9"E, 34°10'36.0"S 119°33'39.9"E, and 34°10'07.2"S 119°30'35.5"E, respectively. Australian Bureau of Statistics (Jul2021-Jun2026), Digital boundary files, ABS Website, accessed 20 August 2024.



Methods S2 Extraction of genes differentially expressed between tips of cluster-bearing lateral roots (TCR) and mature cluster roots (MCR) formed by *Hakea laurina*. Total RNA extracted from roots of *Hakea laurina* hydroponically cultivated with low phosphorus (P) concentration for approximately three years was sequenced. The cDNA libraries for RNA-Seq were conducted with an NEBNext Ultra II Directional RNA Library Prep Kit for Illumina, NEBNext Multiplex Oligos for Illumina, and NEBNext Poly(A) mRNA Magnetic Isolation Module. After being qualified and quantified by NEBNext Library Quant Kit for Illumina, the libraries were paired-end sequenced each 150 bp. Raw sequence data were cleaned up by removing the adapter sequences and low-quality sequences in Trimmomatic (version 0.39). Cleaned data were analyzed in Trinity (version 2.15.1) to assemble transcript fragments. The counts of assembled contigs were quantified using Salmon (version 1.9.0) and expressed as counts per million (CPM). They were normalized across these samples in edgeR (version 4.0.14).



Methods S3 An alignment of homologous transcripts coding for ALMTs in *Hakea laurina* to design a probe fragment for *in situ* hybridization. The deduced HalALMT1 amino acid sequence was subjected to Blastp (version 2.12.0) against all putative open reading frames of >100 amino acid residues to detect highly homologous proteins. The transcript of *HalALMT1* and the top five homologous transcripts with 3'-UTRs were aligned using ClustalW (Thompson et al., 1994). Arrows indicate parts of the forward and reverse primers used to create the probe fragment (Table S2). The red square box indicates the stop codon of HalALMT1.

TRINITY_DN4236_c0.g1
TRINITY_DN54045_c0.g1
TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

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ACGATTCTTTCACTAAAGGATGCTCTTCAGTCTCATAGACAGCCGACCC
CTGATTGAAGCTCTTAATGGCTGCATCACTCCGAATTCAG---GTGCC
TGCTGGAAGCTCTTAATGGCTGCATCACTGCAGAGGTTTCAAG---GCGCC
ATGGTCATGGCTTTGCATGGATGTATCTTCAGAGATACAG---GCACC
ATGGTCCTCA-TGTACGCGGTCTGTCGTC---AACCTCAAGCTGCTCC
* * * * *

TRINITY_DN4236_c0.g1
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TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

ACACACTGTTGAGGCATCTTTAGAGATCCATGTATCCGAGTGGCAGGAA
AGTGCCTTATGAGGCCTCATTATGAAGCCATGTGAAGCAGTGGCTCTC
ACCTGAATTCGAAAGAAAATTCAGAAGGCTGCGACAGCATGAGCTTGG
TGACTTTTGAAGAAGCTTTTCAGTGATGCTGCATGATATTAAGCTCCA
CCGAGAACGAAGAAAAGCTTTTCATAGGAGCTTCTGAAGTGGGTACTG
AGGGGCTCTCGTGGCGCTGTCACCA---CCGTGATGATCAGGGCTACG
* * * * *

TRINITY_DN4236_c0.g1
TRINITY_DN54045_c0.g1
TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

AACATCAGAGATACCTGATGGAACCTGCTGACAGCATTAACCAACCA
AGCTTGCTTACACTGAGGAACTAGGGATAGCATCATCA---AATGA
GATGTGGCAAGCCTTGAAGGAATGGCTCAGCAATCACAGA---GATGA
ATTCTCCGTAGCTTGAAGGAATTAGCCATTACATGACTAC---CATGC
AAGCTGCAAAATGTTGCTGGAATAGGCCAAAGTCAAAA---AATGG
AGCTCAGGTTAATGTTCTAGGCCAGCTTTCATGCCACCGC---CCTGC
* * * * *

TRINITY_DN4236_c0.g1
TRINITY_DN54045_c0.g1
TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

CGCTGCTTACAGGGA-AACCTCTAATCAACTCTATGAGGCACTGCAA-
AGAGATTGGCCAGC-AGT-----GTTAATAGTAGCAA-GTTGCA-
GGCAACCTTGCTTGGCTGATCTCCGATGAAAGTGCCAAA-AACGCT--
GAAGATCTGTAACACAGAATTAGCAGTTGGAGATATGAA-GATGCA--
AGAAGTTGATGATGGAGATATCT-CTCAGGTGATCATGAAGCAGAGA
AGGAGTA---CCAGCCCATCTACGAGCTG---GCGCCGTA---CCGCTC
* * * * *

TRINITY_DN4236_c0.g1
TRINITY_DN54045_c0.g1
TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

GTTCTCGATTATCAGTCA-AATCACAACCTCTT-----TT---ATTGG
GTTATCAAGAATAGAGCTT-AGTGA-AGCTGTAC-----TT---CCTTG
GCCGATGACCTTAAGGCCACACTCAAAACCGCAA-----T---ACTAGA
GTAGAAGAGCTCAAGATTGCTTGAAGTCTTACCAGCTT---ACTCAG
AGAATTGCAGAAGAAGATAGATCAGAAGTCTATCTCTTGTGGACTCGG
GCCACGTGGCGGGCGCTGCTGCTGGCTTCTGCTGGCTGCTGCTGCTTTC
* * * * *

TRINITY_DN4236_c0.g1
TRINITY_DN54045_c0.g1
TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

TGGC---TCAAAAGATCATGAAGGACCA--AGATGCTGATCTTGCTGC
ATGA---TCAGGACACTACAGACGATGA--AGATT-----
GAAAC---TCAGACCTCT-----TAGCA--TAATGC-----
GTGCCCATACCAATTGCGACAGACCTG--AGGGGAAGAAAGAGCT
AGAG---TTGGAAATCGGAAACGCCAGTTCAGAGATTGGAATATCCC
CTAC-----CCGCTACGGAGCGCAGATCG-TGAGGCTC-----
* * * * *

TRINITY_DN4236_c0.g1
TRINITY_DN54045_c0.g1
TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

TGCAACTCAGATCAAAAGTCCGACATGGAGGAAT-----TGGGGATAGG
TGCAATGGCAGGCTTTGATTTCTGCTGATTGAGA-----TAGTGAAA--
--CAGGGGCGACAGTTGCTTCACTGCTAGTTGAGA-----TTGTTAAC--
CATTTCACACCCACTCTTTCACTCTGATGGAGG-----TTGTTCTCT
GAGGATGCTCTCAACCTCTAAATGATGAGAGAAGTTTCTGGGACCAA
-GAGTGGCGCGCGCTGCACTGCTCACCAGT--CGTTGGCATCG-
* * * * *

TRINITY_DN4236_c0.g1
TRINITY_DN54045_c0.g1
TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

TTCAAC--TTCTTTGAAGACAG---AC-AACCTGGCGACAGTTGGTTG
-----ATGTTGG---AA-AAATTAGCAAAAGAGTGC---
-----TGACCTG---AC-AGATTGCAAAATCTGTCC---
TGTCACAGTTGCTTCACTACTA---ATTGAATAGTGGCAAGATG---
GTCCCTCAGTGAAGCTGTCTGGATATTAGGTCAAGTCCATGCTCAAAA
-----CCCTGAGCGG---GCTGACCTGGCGCACCGGCTGGTG
* * * * *

TRINITY_DN4236_c0.g1
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TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

AAAGGAGAGTAAAAAATTCAGA-----GACCACCTCAAGGAGTCTGAG
-AAGAACTTGCAGATTATGAG-----GTTTTCACAAAC-AGCTTAAAG
-ATGAACCTGCCAA-----CTTG-----GCTCATTCAAGAAATGTCAC
-AAGGAGTGGTTGAA---GCAGT-----GAACGACTTGAAGAACTGG-
CTGGGATGCTCCGAAATAGTATACAGTTGTGAATCCCGACCTCGTGT
AAGGAGGCTCTGACGACGGGCC-----GGACGAGCAAGGCGGAAGAA
* * * * *

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TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

CGT-CAAAATTAAGACCACCA-----TTGAGTAAGATTGCAATGCC
TAT-CCAGATTGTTGGCAACA-----T-ATCAA-----CGAAAGCC
C---CAACAGTTACACCTGAAA-----A-ACCA-----CAATTACT
---CAGAGTTCAAGCTGAGA-----TTGAGGAACCAATGAACAAGC
CGTGACAGAGTATGTTCAAAACATTTCTTCACTGTTCTCCCTCAATCC
GCTGACGAGGTGCGCGCCAGC-----TGTTCCGCAAGATGGCGCTGC
* * * * *

TRINITY_DN4236_c0.g1
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TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

A-GTTTGAATTCCTGAAGCACTCCCTTCGCTGCTTGTCTCATTGC
A-GTTTGGTATCACAGGGTTACCAACAACATAAAGAGGAAGCA--GA
ACACAGGGGATCCATAAAACCACTTCAGGTGATGAAGT-CCTCAT--G
CAACTTCAGATTACCAAGAAC--GCAAACTGAAAGTCTTCAACAA
TAATGGTGTATTGAAGAGAAATCAAAACATATGAAGTGAAGTGCCT
TGCTGCCGATCTGCGGAGCAC-GTCGAGTGGCAGAGTGGGAGCC---
* * * * *

TRINITY_DN4236_c0.g1
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TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

TATTGGAGACAGTTGCTAGGATTGATCTGTTATTATGAGGTTGACAAA
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GTTCAGCTCTAAATATTACTTGAGACAGAACACAT---CTCTCGTC
TGCTTTTGGCAACATTGCTTCACTCTTGATTGAATTT---GTTGCGAGG
TGACTGGGCGGCTGCTGGCCGCTGATAGTACAACCGGATCATCTTGC
* * * * *

TRINITY_DN4236_c0.g1
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TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

TTAGGGAAGTTGGCATGCTTCAAGAGTATGAATCTAAGAAGAAATCAC
TTTGATCA--CCACATAC--ACAAGTGCGAGTCTGATGGGCGCTGATA
CCTGAATG-----TGT-----AAATA-AACCTTCTGACATTCAAC
CCCGGGCA-----AAGAGAAAAAATGAGGTTCTTCAAGATTATATA
CTTCAAAATATTGTTGACTCATATGAAGAACTCTGGAAGAACAAAT
GTGGCACA-----CGCATCTCCAGTACTCAGCTGGCTGCTAGCT
* * * * *

TRINITY_DN4236_c0.g1
TRINITY_DN54045_c0.g1
TRINITY_DN28314_c0.g1
TRINITY_DN845_c1.g1-Ho1ALMT1
TRINITY_DN1394_c0.g1
TRINITY_DN293420_c0.g1

AATTGTTCTGA-----GAGAACACCT-AGAAAAACACCGAAGATTGC
TAGTGATAGAG-----GTAGACACAA-ATTTCCCTGCTTACAATTTGC
AAACAT-----A-----AGACACCCAG-AACTGTAATCTCCAGA---AC
TATGACACAGGCACTGATCTACAA-ATTTGTAATGTTGTGCTCATG
TAAAGAACCTCACTGAG-GAACCTGCAGCAGAGGTTGGTGGTTTGG
GAGCAT-----ACACGCGCGGAGCCCTCGGAAGCCGAGCGCG-
* * * * *

Fig. S1 The relative amount of organic anions in root exudates from *Hakea laurina*. Carboxylates were detected by CE-TOF/MS. The relative carboxylate concentration in root exudates was calculated relative to the concentration of malate in mature cluster root (MCR) (= 100). Relative amounts of root exudates were compared using the Tukey-Kramer test. Different letters indicate statistically-significant differences ($p < 0.05$; Tukey-Kramer, $n = 3$). Error bars indicate standard errors.

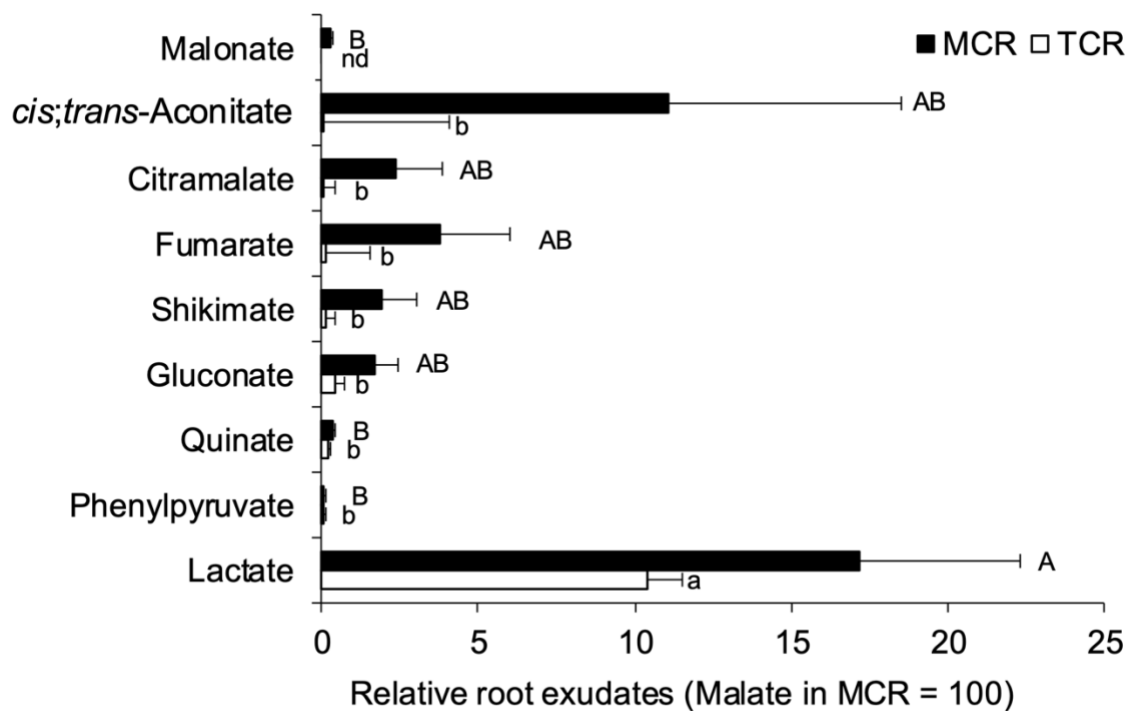
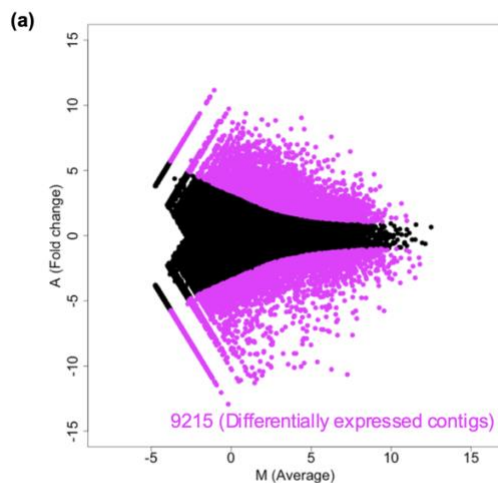


Fig. S2 Results of differentially expressed genes between tips of cluster-bearing lateral roots (TCR) and mature cluster roots (MCR) formed by *Hakea laurina*. (a) Differentially-expressed contigs with $q < 0.05$ and $|\log_2 \text{fold-change of (MCR/TCR)}| \geq 1$ were detected as differentially-expressed contigs. The x-axis (M) shows the average $\log_2\text{CPM}$, and the y-axis (A) shows the \log_2 fold-change gene expression. Magenta represents 9,215 significantly differently-expressed contigs. (b) Contigs with CPM below 0.4 were removed. In total, 77,014 contigs were detected. Putative open reading frames of >100 amino acid residues were searched in TransDecoder (version 5.7.1) and annotated homologs of *Arabidopsis thaliana* by Blastp (version 2.12.0). The *A. thaliana* reference proteome data were retrieved from TAIR website (https://www.arabidopsis.org/download_files/Proteins/TAIR10_protein_lists/TAIR10_pep_20110103_representative_gene_model). Finally, 7,819 annotated differentially-expressed genes (DEGs) were detected and shared in Table S3.



(b)

Analysis	the number of genes
CPM>0.4	77,014
Differentially expressed contigs	9,219
Annotated DEGs	7,819
DEGs upregulated in MCR	4,210
DEGs upregulated in TCR	3,609

Fig. S3 qPCR of *HaALMT1* and phylogenetic tree of ALMT family.

(a) Relative *HaALMT1* expression in tips of cluster-bearing lateral roots (TCR) and mature cluster roots (MCR) of *Hakea laurina* grown without phosphorus (–P) and when exposed to AlCl_3 (–P+Al treatment). The expression in TCR is used as standard (= 1.0). As housekeeping gene, we selected a gene encoding an elongation factor. Relative expression was compared using the Tukey-Kramer test between TCR and MCR under –P treatment, as well as between –P and –P+Al treatments in MCR. ** indicates a statistically-significant difference ($p < 0.01$; Student's *t*-test, $n = 3$). Error bar indicates standard error. (b) Phylogenetic relationships among *HaALMT1* found in the *Hakea laurina* and several homologs from *Arabidopsis thaliana* (At), *Brassica napus* (Bn), *Glycine max* (Gm), *Lupinus albus* (La), *Secale cereale* (Sc), *Triticum aestivum* (Ta) based on amino acid sequences. A red-circle highlighted ALMT is a target *HaALMT1*. Blue square-highlighted ALMTs have been characterized as Al^{3+} -activated or Al^{3+} -induced ALMTs, and green triangle-highlighted ALMTs have been reported to be involved in P uptake.

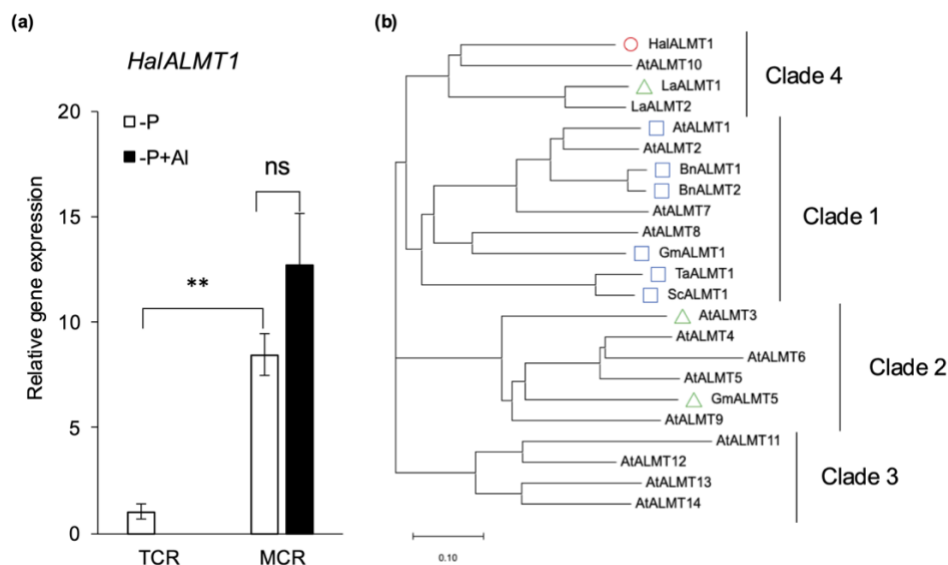


Fig. S4 The intensity of an electric current (I) recorded from oocytes expressing or not expressing HalATLM1, with or without malate injection, at an applied voltage of -100 mV.

These values are derived from Fig. 4b. The intensity of an electric current between -AI and +AI under HalALMT1-expressing oocytes, and between oocytes expressing or not expressing HalATLM1 with malate induction under both -AI and +AI conditions at -100 mV was compared using Student's *t*-test (*, $p < 0.05$; **, $p < 0.01$; ns, not significant, i.e., $p > 0.05$). Data are presented as means \pm standard error ($n = 12-14$).

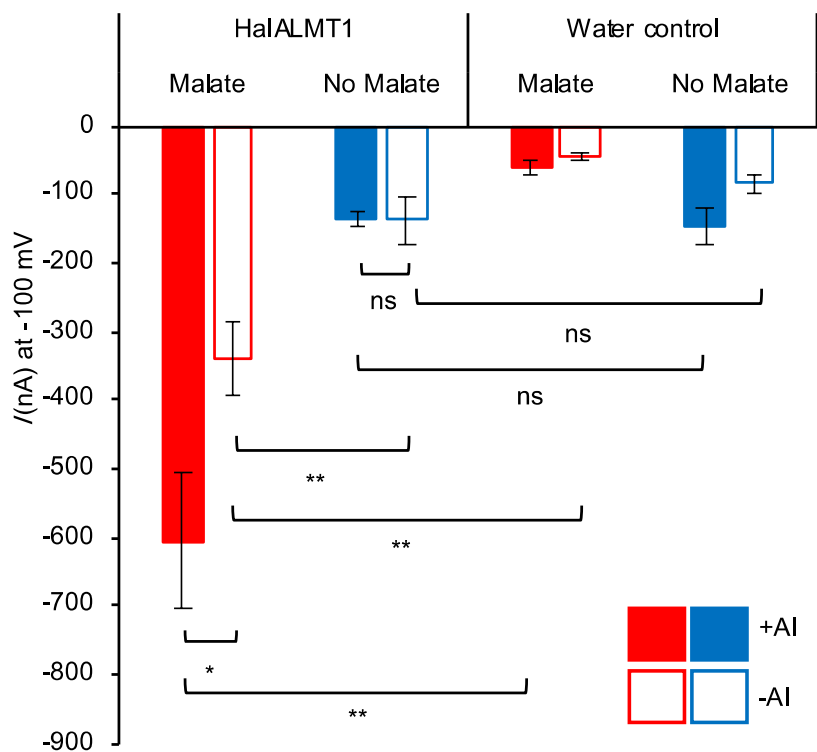


Fig. S5 Mean current-voltage curves recorded from oocytes expressing HalALMT1 and oocytes injected with water, malate, and citrate. The oocytes were used directly (no injection) for the two-electrode voltage-clamp measurements. The oocyte membrane was clamped to the holding potentials (-20 mV), and voltage pulses were applied in 20 mV increments from -140 to +40 mV. The bath solutions were ND 96 pH 4.5 [96 mM NaCl, 1.8 mM CaCl₂, 1 mM KCl, 0.1 mM LaCl₃, 210 mOsm kg⁻¹ with sorbitol]. All measurements were performed with AlCl₃ (100 μ M) in the bath solution. Data are presented as means \pm standard error (n = 6-7).

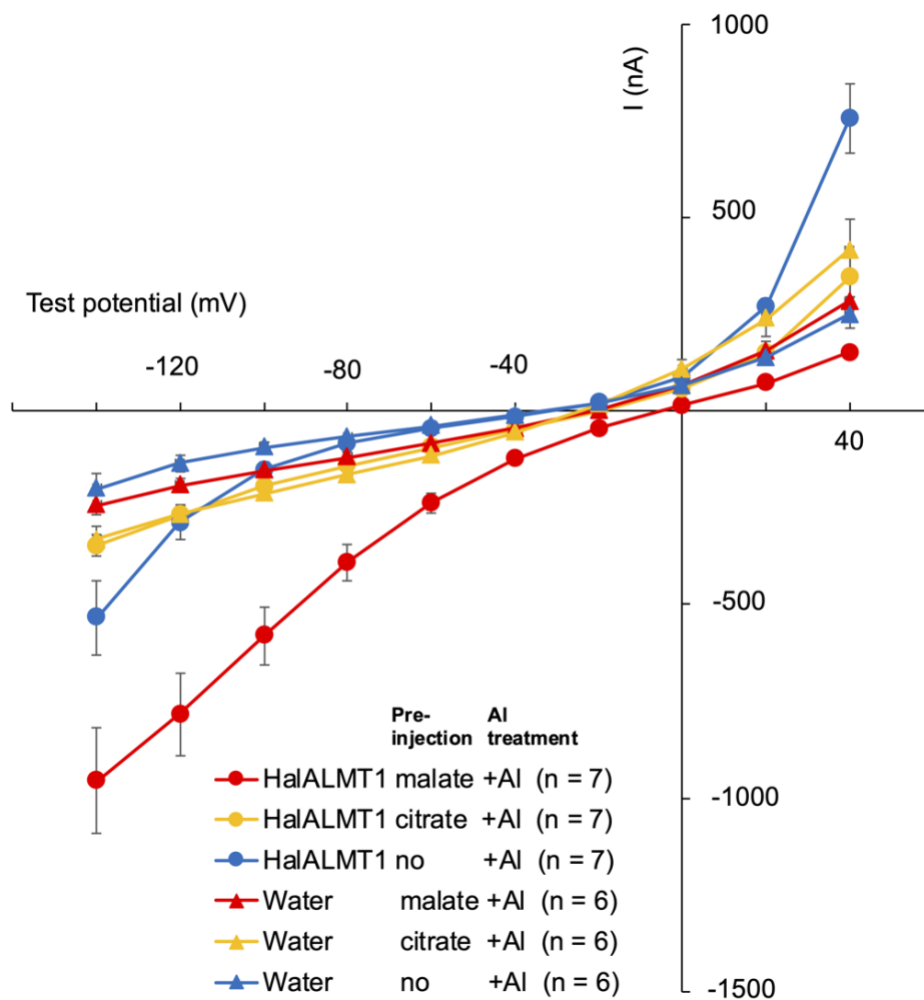


Fig. S6 qPCR of *HaIALMT1* in roots of Col-0 (wildtype; WT) and two transgenics 35Spro-*HaIALMT1* in Col-0 (OX1 and OX2) of *Arabidopsis thaliana*. Relative gene expression for *HaIALMT1* in roots. The expression of WT is standard (=1.0). Relative expression was compared using the Student's *t*-test between transgenic lines. Different letters show a statistically-significant difference ($p < 0.05$; Student's *t*-test). Error bars indicate standard error.

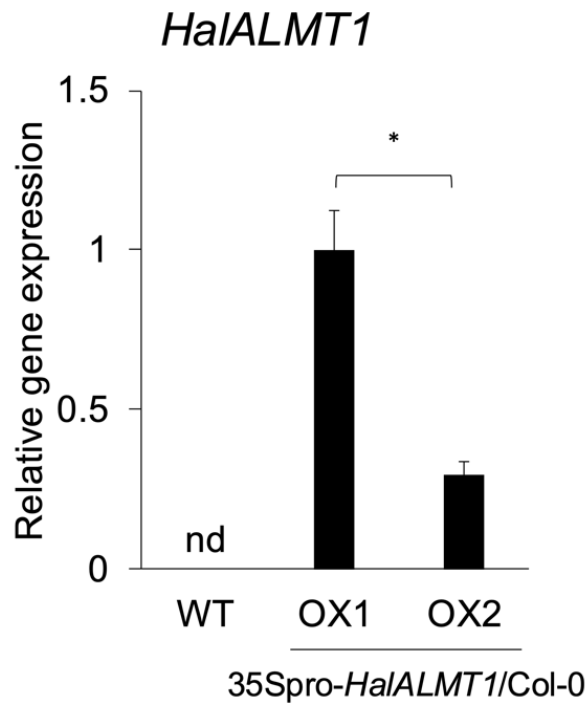


Fig. S7 The effect of long-term Al³⁺ treatment on root growth of wildtype (WT), *atalmt1*, and two transgenics 35Spro-*HaALMT1* in *atalmt1*(OX1 and OX2) of *Arabidopsis thaliana*.

Relative root length of 35Spro-*HaALMT1*/*atalmt1* under Al condition. WT, *atalmt1*, OX1, and OX2 cultivated under ½ MS medium for 10 days were transferred to ½ MS plates with or without 300 µM AlCl₃ (pH 4.8). The length of roots after seven days of cultivation with or without aluminum (+Al or -Al) was measured. (a) Relative gene expression for *AtALMT1* and *HaALMT1* in roots. The expression of wildtype and OX1 is standard (=1.0) for *AtALMT1* and *HaALMT1*, respectively. Relative gene expression of *AtALMT1* and *HaALMT1* was compared using the Tukey-Kramer test among wildtype under -Al and +Al treatments, an *atalmt1* mutant, and two transgenic lines, as applicable. (b) Photograph of representatives for *atalmt1* under -Al/+Al and 35S:*HaALMT1* in *atalmt1* under -Al/+Al. (c) Relative root length under Al condition (%) was calculated as (root length under +Al)/(root length under -Al)*100. Relative root length under Al conditions was tested using the Tukey-Kramer test among wildtype, an *atalmt1* mutant, and two transgenic lines. Different letters show a statistically-significant difference ($p < 0.05$; Tukey-Kramer). Error bars indicate standard error.

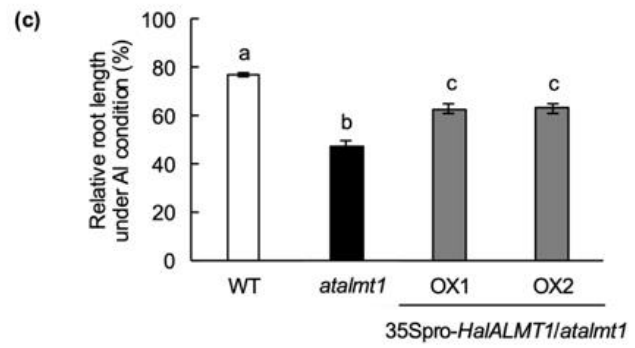
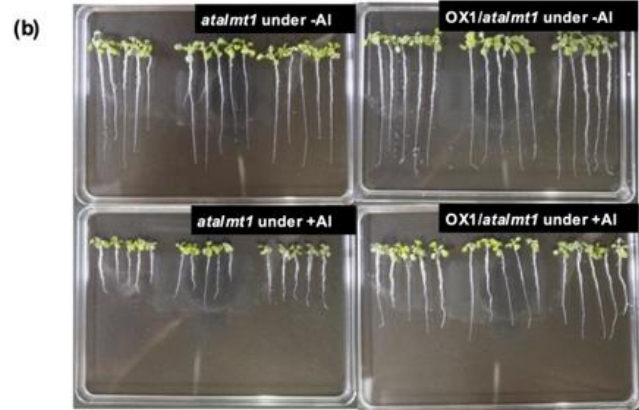
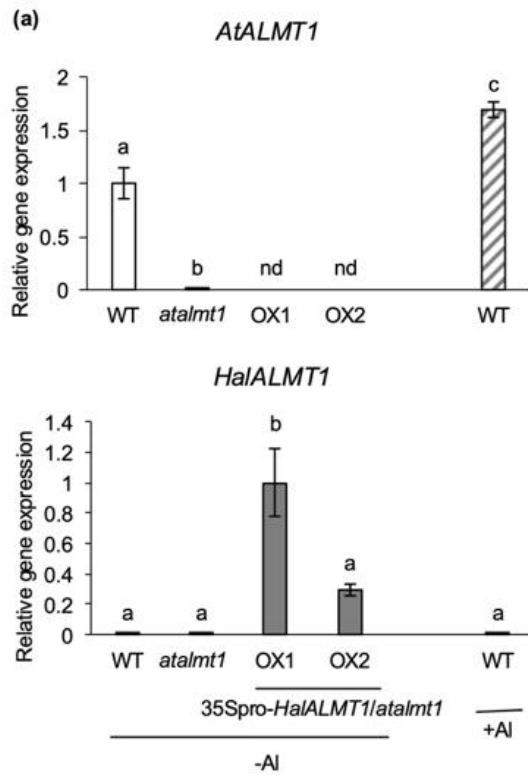


Fig. S8 Other observed images of *HalALMT1* mRNA in cluster rootlets of *Hakea laurina* using the *in situ* hybridization presented in Fig. 5. Cluster roots were harvested from hydroponically-cultivated *Hakea laurina* under low-phosphorus conditions. (a), (b), (c), and (d) are longitudinal sections and (e) is a cross-section of a rootlet. All scale bars: 200 μ m.

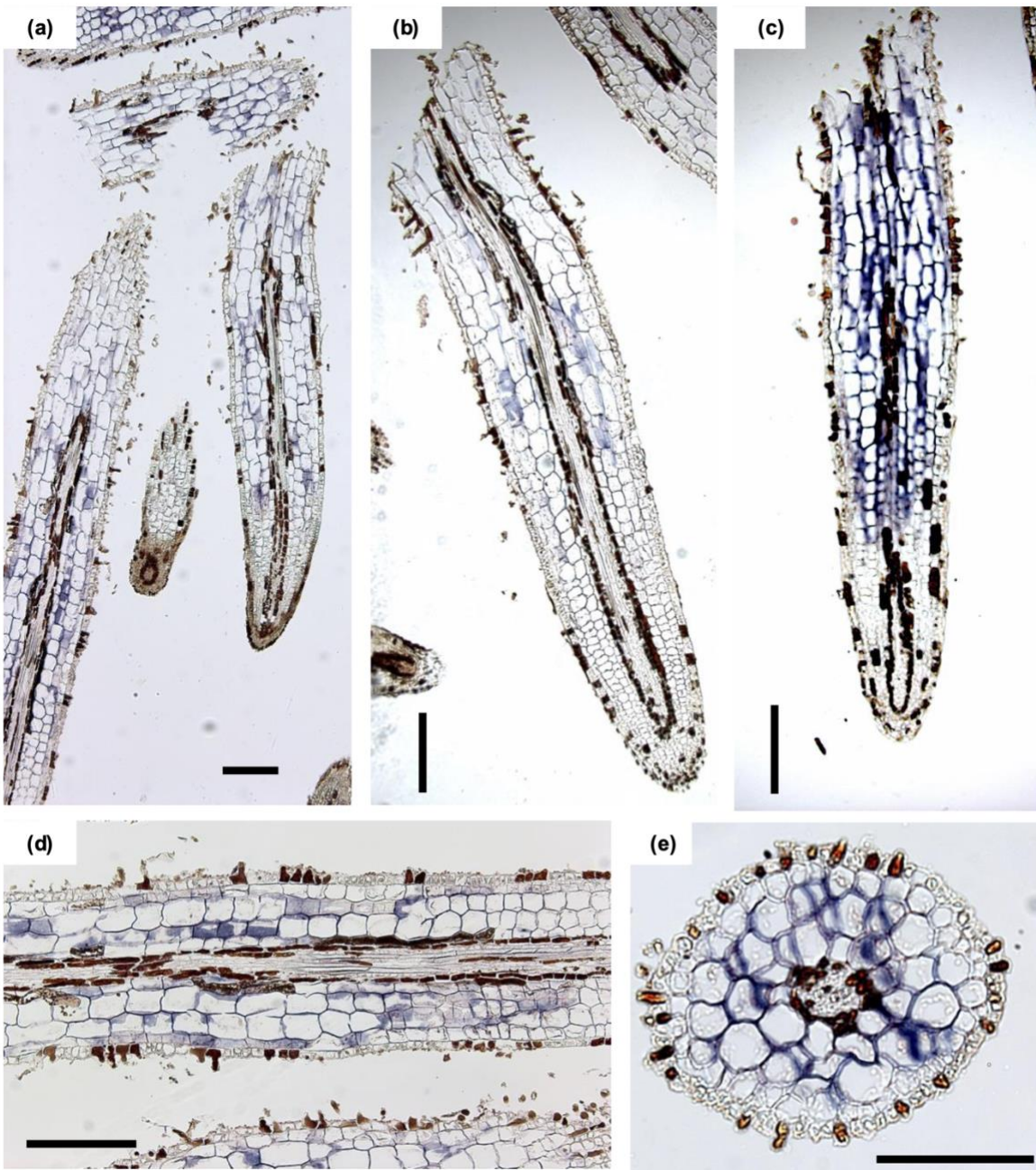


Table S1 Primer sequences for qPCR

gene	primer	Sequence (5'→3')
<i>HalALMT1</i> (in Arabidopsis)	Forward	TATTGCGTTCCACCGTACGT
	Reverse	TGAATCTCTTGCATCAGAGGA
<i>HalEF1a</i>	Forward	GTCGATTCTGGAAAGTCGACC
	Reverse	AATGTCAATGGTGATACCACGC
<i>AtALMT1</i>	Forward	GGCCGACCGTGCTATACGAG
	Reverse	CATGAGTCCTGTGAACTCCC
<i>HalALMT1</i> (in Arabidopsis)	Forward	ACGAACACTGATGGCAAGGT
	Reverse	ACACCCAACTGCAATACCCT

Table S2 Primer sequences for cloning

Name of construct	primer	Sequence (5'→3')
pXBG2 (HalALMT1)	Forward	CAACTTTGGCAGATCATGGTTCATGCCAAGGAACTT
	Reverse	AGTGGTAACCAGATCTCAAACCTTGTGGAAGAACTTTCATGG
pCR TM 8/GW/TOPO (HalALMT1-full) (HalALMT1 without stop codon)	Forward	ATGGTTCATGCCAAGGAACTTC
	Reverse	TCAAACCTTGTGGAAGAACTTTCATGG
	Reverse	AACTTGTGGAAGAACTTTCATGGTTTG
HalALMT1 inner	Forward	CACTGGGTAGCTAACCAATC
	Reverse	ATCATGCAGGCATCACTGAG
pGWB5 -> pXBG2 (HalALMT1::GFP)	Forward	CAACTTTGGCAGATCATGGTTCATGCCAAGGAACTT
	Reverse	AGTGGTAACCAGATCTTACTTGTACAGCTCGTCCATGCCG
pGWB6 -> pXBG2 (GFP::HalALMT1)	Forward	CAACTTTGGCAGATCATGGTGAGCAAGGGCGA
	Reverse	AGTGGTAACCAGATCTCAAACCTTGTGGAAGAACTTTCATGG
Nested 1 st PCR	Forward	CAGAATCTGGGTCTTCCACA
	Reverse	CATTGCTTCTCTGTAGCCTCC
Nested 2 nd PCR	Forward	GCCTGGCTAGGGTTCTTAGAT
	Reverse	GAAGTTTCCTTGGCATGAACC

Primer walking 1	Forward	CAGCTTGATTTCCGGTATCGGG
Primer walking 2	Forward	GCTAAACCTTCTCCCACTGC
Primer walking 3	Reverse	GTGACTATCGATCTCAGCCACA
Primer walking 4	Reverse	GAGGAATGATAGCTAAGTTTGGC
Primer walking 5	Reverse	CTTCCACTCAGAGCCACTTC
Primer walking 6	Reverse	CTTCAAAGCACTCACTCCTCAC
Promoter cloning	Forward	Same as Primer walking 2
(pHalALMT1)	Reverse	Same as Primer walking 5
Probe	Forward	CGGGCCCCCCTCGAATGGCTCTGTGCATTATATATAATCTTG
	Reverse	TACCGTCGACCTCGATCAGTGATGCCTGCATGATATTAAG

See separate Excel file for:

Table S3 The counts per million (CPM) of differentially expressed genes (DEGs) up-regulated in mature cluster roots (MCR) and tips of cluster-bearing lateral roots (TCR) .

Table S4 The results of gene ontology (GO) analysis and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway analysis using differentially expressed genes (DEGs) up-regulated in mature cluster roots (MCR) and tips of cluster-bearing lateral roots (TCR).