Supporting Information

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SI Materials and Methods

Bacterial Strains. SL44 has a deletion of the *nodD1-nodABC* region (1). The *exoA* strain (Rm7031) is described in ref. 2. The *exoX* (MB801), *lpsB* (R1A6), and *ndvB* (B587) mutants are described in refs. 3 and 4.

***A. rhizogenes*–Mediated Hairy Hoot Transformations.** Modifications to the protocol described in ref. 5 are as follows.

We found that transformation efficiency was increased if the plants were first placed on modified Fahraeus medium containing 1 mM α-aminoisobutyric acid (AIB) without selection for 7 days. All emergine roots were removed and plants were transferred to selective media with no ethylene inhibitor and either 25 mg/L kanamycin (remained on selective media for 17–21 days) or 10 mg/L hygromycin (for 10 days). Plants remained on selective media until roots reached ≈2.5 cm (we found that less time was often needed for Jemalong A17 than for Jemalong). Plants were then transferred to 1/2× Gamborg's B5 Basal Salt medium (Sigma) with 1% agar to recover from antibiotic selection. For confocal microscopy studies, 300 mg/mL Augmentin (Research Products International) and 500 mg/L Cefotaxime (Sigma) were added to the B5 medium to reduce the amount of *A. rhizogenes* carryover. Plants remained on B5 for 1 week and were then transferred to buffered nodulation medium (BNM) (6) containing 0.1 μM aminoethoxyvinylglycine (AVG).

Quantitative RT-PCR. To prepare template for qRT-PCR, RNA was DNase-treated by using DNase-free turbo (Ambion). 35 PCR cycles using actin-specific primers (Table S2) were used to check for DNA contamination after DNase treatment. The DNase-treated RNA was used in single-stranded cDNA synthesis with SuperScript III (Invitrogen) and oligo(dT) primer (Invitrogen). 25 PCR cycles were used to check for successful cDNA synthesis. Template concentration per reaction was determined empirically based on relative abundance of the transcript of each *FLOT*; each template was run at two different concentrations. Template quantification was done at the level of total RNA; an internal actin control in each PCR controlled for differences in efficiency of cDNA synthesis. Actin was amplified from cDNA made from 2.5 ng and 7.5 ng of RNA; *FLOT2*, *FLOT3,* and *FLOT4* were amplified from cDNA made from 7.5 and 25 ng of RNA; *FLOT1* was amplified by using cDNA made from 25 and 75 ng of total RNA. qPCR was performed by using the DyNAmo Flash SYBR Green qPCR kit (Finnzymes).

Artificial miRNA Construct Design. The amiRNA web-based designer described in ref. 7 (http://wmd2.weigelworld.org) cross-references with available *Medicago* EST databases. The full-length sequence for the desired *FLOT* target was entered as the "target gene," and available ESTds for that target were entered as "accepted off-targets." The suggested amiRNAs were BLASTed against the available *M. truncatula* genome to ensure that there were no off-target sequences that are absent from the available EST library. The pRS300 plasmid (8) was used as a template to create the amiRNA hairpin with an intron.

Hairy Root Time Course. To determine whether regulation of *FLOT*s was altered in hairy roots, we assayed *FLOT* expression levels in uninoculated and inoculated *M. truncatula* cv. Jemalong seedlings transformed with the amiRNA empty vector construct EX117 (Table S3) at 1, 4, 7, 14, and 21 dpi. Plants were grown as described above and inoculated with 1/2× BNM or Rm1021 in 1/2× BNM. Plants were harvested just below the callus at the appropriate time point and flash-frozen in liquid nitrogen. Three independent replicates of the entire time course were performed; each time point sample was a pool of the 10 plants from a single plate. RNA was isolated with a yield of approximately 50–100 μg per 10 plants.

Protein Localization. pCH010 was constructed in two steps by first inserting eGFP (with added 5′ EcoRI and XmaI sites) into the BamHI/XbaI sites of pJG159; then the NPTII ORF was amplified from the pHELLSGATE8 vector and inserted into the XhoI site in pJG159. pJG159 is a small (7.8 kb) binary vector that was constructed by J. Griffitts (unpublished) by a three-way ligation of inserts A and B from pEGAD (9) (Table S3) and the SphI/XhoI fragment from pCAMBIA1300 (www.cambia.org). To create insert A (Sph-RB-P35S-RI), pEGAD was amplified with primers oJG346/347 and 348/349 followed by overlap extension-PCR with oJG346/349. Insert B (RI-nosT-P35S-XhoI) was amplified from pEGAD with oJG350/351. pQDN03 was constructed by replacing GFP in pDG71 (10) with mCherry (Table S5). FM4-64 was dissolved in 0.1 M phosphate buffer (pH 7.0) to a final concentration of 20 μM and kept on ice until use (11). The GFP/FM4-64 experiment was imaged on a system described in ref. 12 with the same excitation settings listed above for GFP/RFP and 1,000-ms exposures. Typical exposure times were 1,000 ms for GFP, 500 ms for mCherry, and 1,000 ms for FM4-64.

1. Fisher RF, Egelhoff TT, Mulligan JT, Long SR (1988) Specific binding of proteins from Rhizobium meliloti cell-free extracts containing NodD to DNA sequences upstream of inducible nodulation genes. *Genes Dev* 2:282–293.
2. Leigh JA, Signer ER, Walker GC (1985) Exopolysaccharide-deficient mutants of Rhizobium meliloti that form ineffective nodules. *Proc Natl Acad Sci USA* 82: 6231–6235.
3. Griffitts JS, et al. (2008) A Sinorhizobium meliloti osmosensory two-component system required for cyclic glucan export and symbiosis. *Mol Microbiol* 69: 479–490.
4. Griffitts JS, Long SR (2008) A symbiotic mutant of Sinorhizobium meliloti reveals a novel genetic pathway involving succinoglycan biosynthetic functions. *Mol Microbiol* 67:1292–1306.
5. Boisson-Dernier A, et al. (2001) Agrobacterium rhizogenes-transformed roots of Medicago truncatula for the study of nitrogen-fixing and endomycorrhizal symbiotic associations. *Mol Plant Microbe Interact* 14:695–700.
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7. Schwab R, Ossowski S, Riester M, Warthmann N, Weigel D (2006) Highly specific gene silencing by artificial microRNAs in Arabidopsis. *Plant Cell* 18:1121–1133.
8. Ossowski S, Schwab R, Weigel D (2008) Gene silencing in plants using artificial microRNAs and other small RNAs. *Plant J* 53:674–690.
9. Cutler SR, Ehrhardt DW, Griffitts JS, Somerville CR (2000) Random GFP:cDNA fusions enable visualization of subcellular structures in cells of Arabidopsis at a high frequency. *Proc Natl Acad Sci USA* 97:3718–3723.
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11. Bolte S, et al. (2004) FM-dyes as experimental probes for dissecting vesicle trafficking in living plant cells. *J Microsc* 214:159–173.
12. Paredez AR, Somerville CR, Ehrhardt DW (2006) Visualization of cellulose synthase demonstrates functional association with microtubules. *Science* 312:1491–1495.

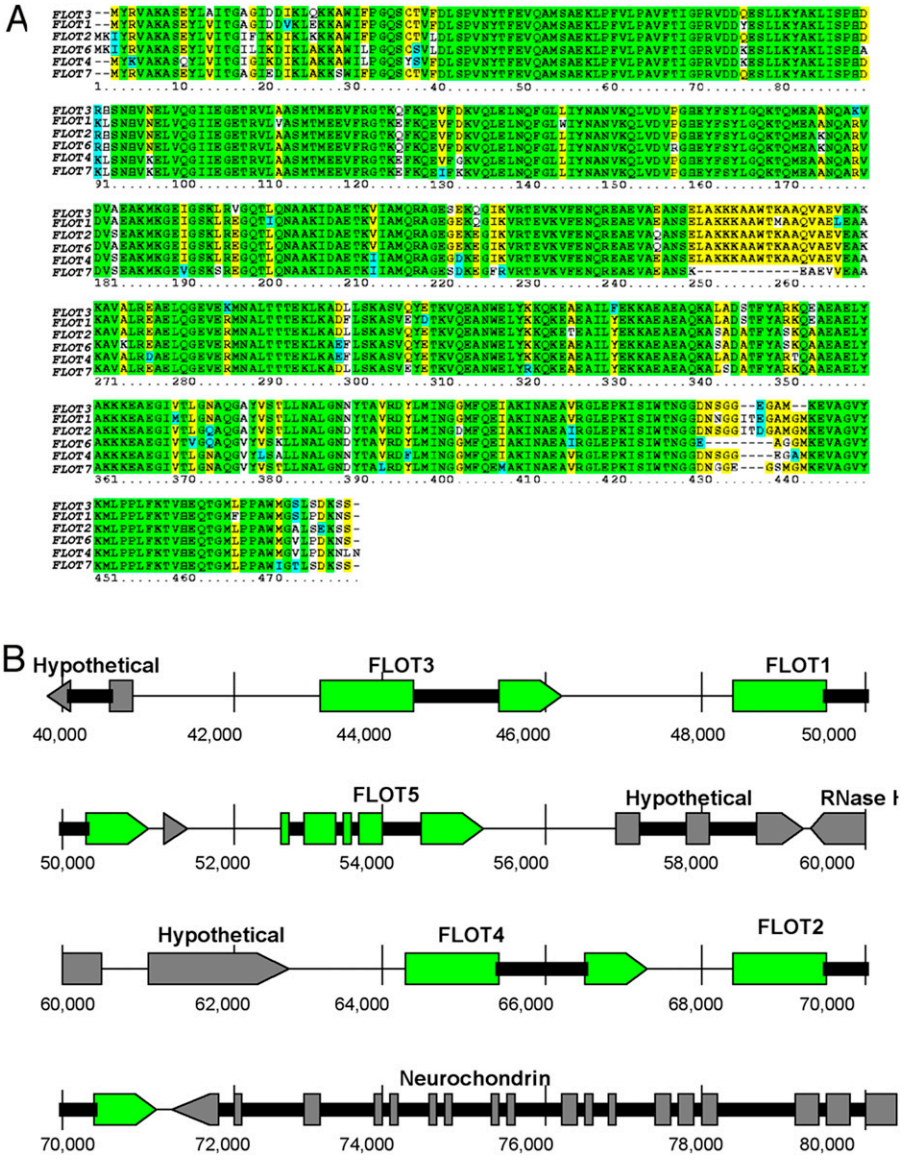

Fig. S1. Alignment of predicted amino acid sequences of FLOTs and arrangement of *FLOT1-5* within a single BAC. (*A*) Sequences were aligned using CLUSTALW available from SDSC Biology Work Bench (http://workbench.sdsc.edu). Conserved residues between all FLOTs are highlighted green, residues conserved between four or more sequences are yellow, and similar residues are blue. Note change in FLOT4 Cys35 to Tyr (residue 37 as numbered). (*B*) BAC CT009553 (mth2-115c19) (IMGAG, http://www.medicago.org/genome/IMGAG/).

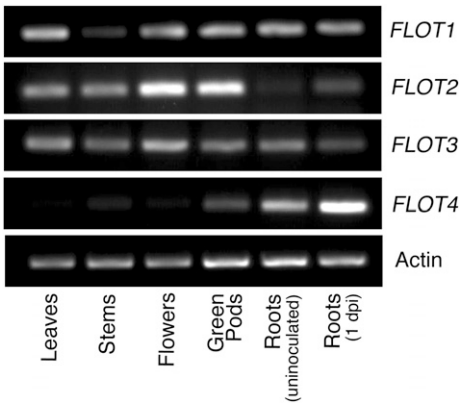

Fig. S2. Expression of *FLOT1-4* in different plant tissues. Semiquantitative RT-PCR was conducted to monitor expression of *FLOT1-7* in leaves, stems, flowers, green pods. Actin primers were used to monitor total input of cDNA. 25 rounds in amplification were used to amplify actin, 30 cycles for *FLOT2,3* and *4* and 35 cycles for *FLOT1*. Expression of *FLOT5,6,* and *7* was not detectible after 35 cycles.

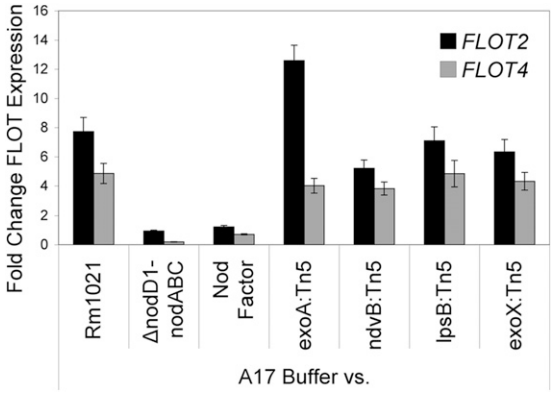

Fig. S3. Expression of *FLOT2* and *FLOT4* in response to polysaccharide mutants. Bacterial mutants in lipopolysaccharide biosynthesis (*lpsB:*Tn5), cyclic β-1,2-glucan synthesis (*ndvB*:Tn5) and exopolysacchride biosynthesis (*exoA*:Tn5 and *exoX*:Tn5) cause up-regulation of *FLOT2* and *FLOT4* at 1 dpi.

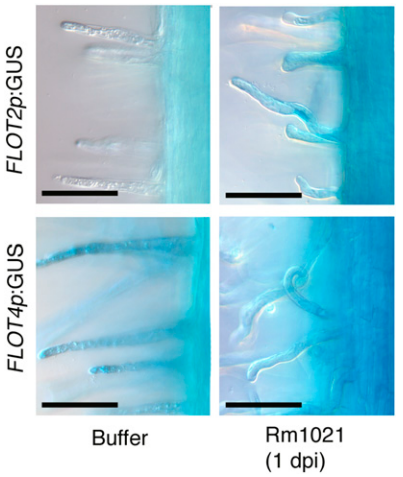

Fig. S4. *FLOT2* and *FLOT4* are expressed in inoculated root hairs. *M. truncatula* cv Jemalong A17 plants were transformed to generate hairy roots expressing *FLOT2* and *4* promoter-GUS fusions; GUS activity is shown for buffer- and Rm1021-inoculated roots at 1 dpi. Ten transgenic lines were observed for each construct at each time point. (Scale bars: 30 nm.) A representative sample at the indicated time points is shown.

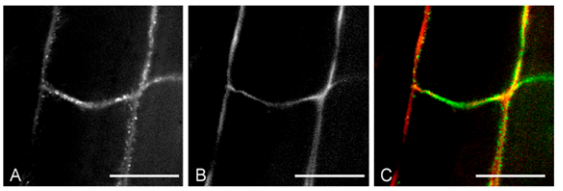

Fig. S5. FLOT2 and FLOT4 localize to membrane-associated puncta. We generated A17 hairy roots expressing 35S:FLOT2::GFP or FLOT4p:FLOT4:GFP. Transgenic roots were visualized using a spinning disk confocal microscope (scale bars: 15 μm). At least six transgenic lines were observed for each treatment. Representative images are shown. (*A*) 35S:FLOT2:GFP in root cells is punctate. (*B*) FM4-64 membrane-associated dye. (*C*) Colocalization of FLOT2:GFP puncta (green) and FM4-64 (red).

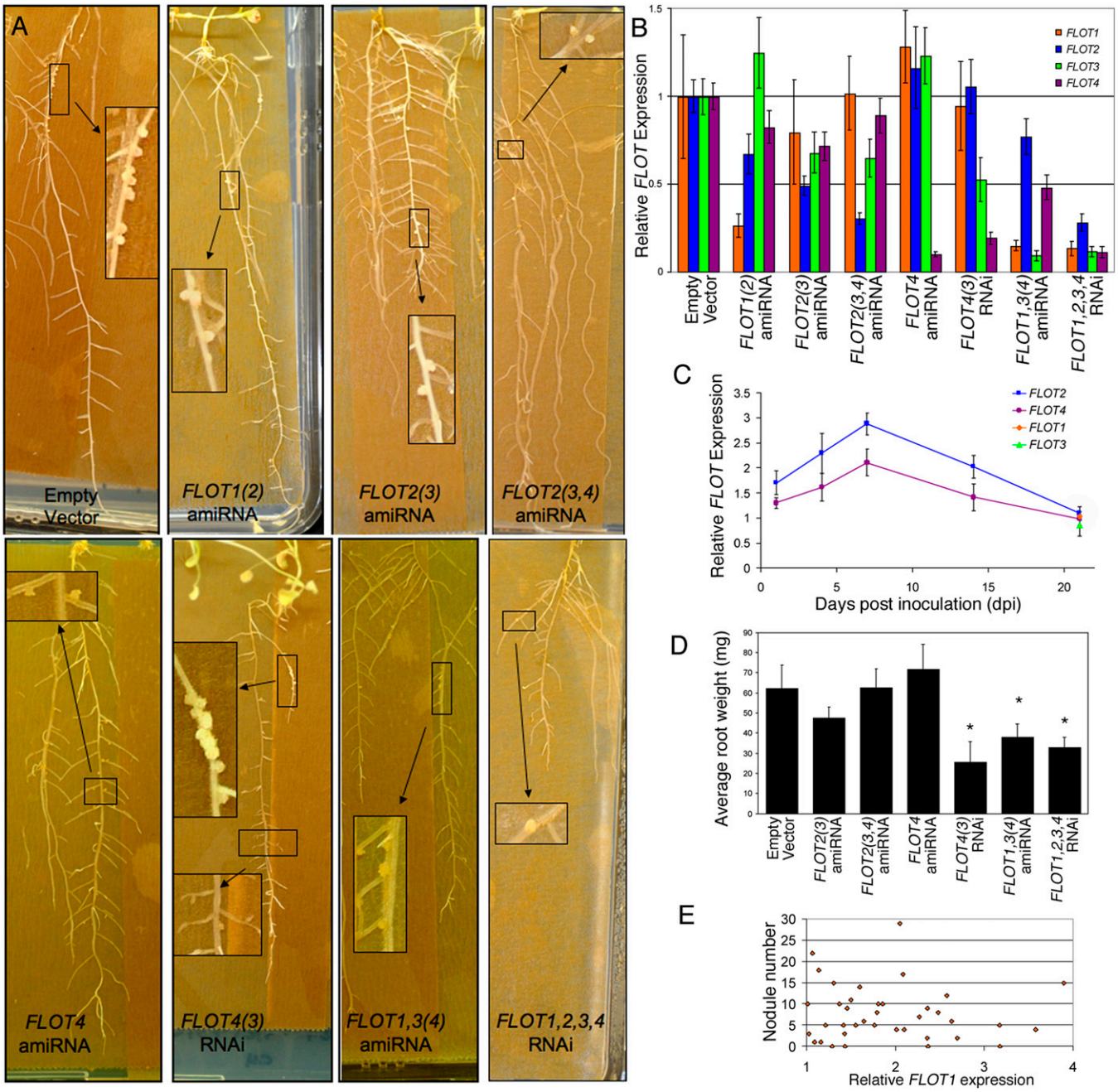

Fig. S6. Root and nodule phenotypes of roots transformed with amiRNA and RNAi constructs, expression of *FLOT1-4* in hairy root time course, and silencing data for *FLOT1*. (*A*) A representative plant for amiRNA and RNAi constructs described in this study is shown. Nodules that formed in silenced lines were small and white (with the exception of *FLOT1+3(4)* amiRNA line). Note smaller overall roots in *FLOT3*-silenced lines, shorter primary roots in *FLOT2*-silenced lines and increase in short secondary lateral roots in *FLOT4*-silenced lines. (*B*) Silencing data including the data for *FLOT1* and one additional construct that primarily targets *FLOT1* (*FLOT1(2)* amiRNA). Gene expression of *FLOTs* in individual hairy roots expressing the indicated RNAi or amiRNA construct was assessed using qRT-PCR, normalized to an internal actin control and then to expression in control plants (average of at least 10 roots). Constructs are designated by their primary target gene(s); numbers in parentheses show genes that have partial but significant ($P < 0.05$) reduction in expression due to cross silencing. (*C*) Hairy root timecourse. Jemalong seedlings were transformed using *A. rhizogenes* with the amiRNA empty vector to generate hairy roots. Plants were inoculated with *S. meliloti* Rm1021 or 1/2× BNM and harvested at the indicated time. qRT-PCR was performed to analyze expression of *FLOT2* and *FLOT4* at all time points; *FLOT1* and *FLOT3* expression were monitored at 21 dpi only. Expression of each gene is normalized to an actin internal control; the ratio of inoculated to uninoculated plants is shown. Error bars are standard error of the ratio. (*D*) Average root weight of silenced lines. The ten plants per construct used to count infection events (Fig. 4 *A*–*G*) were weighed. Error bars represent standard error; pair-wise *t* tests were done to determine significance (*, $P < 0.05$). (*E*) Linear regressions were conducted on plants described in Fig. S4*B* to determine whether a correlation exists between expression of *FLOTs* and nodule number. *FLOT1* expression does not correlate with nodule number (*P* slope = 0.9).

Table S1. Summary of the *FLOT* gene family

Gene name	EST (TIGR)	Affymetrix probe set (expressed?)	BAC ID (IMGAG, GenBank)	Genomic locationchromosome (position, cM)	Number of exons	Estimated spliced mRNA size
FLOT1	BF644444	Mtr.5691.1 (No)	Mth2-115c19, CT009553	3 (71.8)	2	1,434 bp
FLOT2	EX527915		Mth2-115c19, CT009553	3 (71.8)	2	1,440 bp
FLOT3	TC139669	Mtr.45231.1 (Yes)	Mth2-115c19, CT009553	3 (71.8)	2	1,422 bp
FLOT4	TC133140, TC127236	Mtr.11786.1 (Yes), Mtr.42072.1 (Yes)	Mth2-115c19, CT009553	3 (71.8)	2	1,425 bp
FLOT5	TC126348		Mth2-115c19, CT009553	3 (71.8)	5	?
FLOT6	AW574030	Mtr.3447.1 (No)	Mth2-193c3, AC161241	1 (49.4)	2	1,416 bp
FLOT7	TC117648	Mtr.10214.1 (No)	Mth2-135j6, AC151528	1 (43.9)	2–3	?
FLOT7			Mth2-58k14, AC174291	Unanchored	2–3	?

Location, size, gene structure, and available ESTs for putative *M. truncatula* flotillin-like genes are shown. Data were compiled from the Noble foundation (http://bioinfo.noble.org/gene-atlas/), GenBank (http://www.ncbi.nlm.nih.gov/Genbank/), the International *Medicago* Genome Annotation Group (IMGAG, http://www.medicago.org/genome/IMGAG/), and the *M.t.* Gene Index (http://compbio.dfci.harvard.edu/tgi/cgi-bin/tgi/gimain.pl?gudb=medicago).

Table S2. Primers and constructs described in this study: PCR and qPCR primers to monitor *FLOT* expression

Experiment	Intended target	Fig.	Primer name	Primer sequence
qPCR	Actin	1, 4*A*, S4	chh256	CACGAGACCACCTACAACTCT
			chh257	GGACTTAGAAGCACTTCCTGT
qPCR	FLOT1	1, 4*A*, S4	chh250	CTGAGCTTGAGGCTGCTAAAG
			chh251	TTAGCCTCTTGTACCTTAGTGTC
qPCR	FLOT2	1, 4*A*, S4	chh234	CAAGAGCTTTTCTCAGATAAGGC
			chh237	ACTGATGGTGCCATGGGTATG
qPCR	FLOT3	1, 4*A*, S4	chh244	AGTTGGCCAAAAAGAAGGCTG
			chh247	TTAGCCTCTTGTACCTTAGTTTC
qPCR	FLOT4	1, 4*A*, S4	chh241	TGCGTCTGCTAATGCTTTCTGTG
			chh243	CCGAAGTTGAGGCTGCCAAAG
PCR/expression	FLOT1		chh073	TATGACATAGTAGTAGTTTG
			chh068	ATGTACCGGGTAGCAAAAGCA
PCR/expression	FLOT2		chh258	AGTCGAGGCGAAGAAGGCTGT T
			chh259	AGTCTCTTTCTGTTTCTTGTACAGC
PCR/expression	FLOT2		chh067	ATGAAAATTTACCGGGTCGCG
			chh074	TCAGGCATGTATGATCACGTA
PCR/expression	FLOT3		chh068	ATGTACCGGGTAGCAAAAGCA
			chh075	TGCATCTCCTAATTAAGACTT
PCR/expression	FLOT4		chh068	ATGTACCGGGTAGCAAAAGCA
			chh076	GCCAAAATAAAATTCCACAAT
PCR/expression	FLOT5		chh090	GTGGGACTTCATCGTGTAGC
			chh091	CCTAATACTTGCATTGCATCAT
PCR/expression	FLOT6		chh067	ATGAAAATTTACCGGGTCGCG
			chh077	CTACTATAAACCTCTAAACCC
PCR/expression	FLOT6		chh252	GGGTGAAGCAGGTGGTATG
			chh253	CTACTACTATAAACCTCTAAACCC
PCR/expression	FLOT6		chh254	AGGCGAAGAAGGCTGTGAAAC
			chh255	ACCCCTTGAGCTTGTCCAACT
PCR/expression	FLOT7/8		chh230	GTAGTTTAGTAATTTAGTAGTTTAAG
			chh231	TCAAAGAGAGGCTGAAGTGGCTGAGG
PCR/expression	FLOT7/8		chh231	TCAAAGAGAGGCTGAAGTGGCTGAGG
			chh107	AGTCTCTTTCTGTTTCTTGT

Table S3. Primers and constructs described in this study: RNAi construct primers (Figs. 4 and 5 and Fig. S5)

Construct (original)	Construct (this study)	Intended target	Primer name	Primer sequence
E	FLOT1-4 RNAi	FLOT1-4	chh112	CCACAATTCTGTATGAGAAGAA
			chh113	GTTTCCAAGTGCATTGAGAAG
J			chh224	CACCACCTTTTATTGTGTGATATGTGTT
		FLOT1 UTR	chh225	TGTAAACTAAATCATCATATGACATAG
K			chh226	CACCGGTTTTGGTAATTTAGTAGCAGT
		FLOT2 UTR	chh227	ATTAAATTGGCATTTAATATAAGAG
L	FLOT4(3) RNAi		chh228	CACCAATTAGGATGATGCAACTATTATG
		FLOT3 UTR	chh229	ACCTGAAAATCTGAAAGACTAGTGA
M			chh277	CCACATCTTTACCTGACAAAAACTC
		FLOT1 UTR	chh278	GACTGGTGAATAAAACACATATCAC
N			chh279	CCACAGCCTTATCTGAGAAAAGCTC
		FLOT2 UTR	chh280	TCACGTAATAAAAAGTACTGCTAC
O		FLOT2	chh281	CCACATGAAAATTTACCGGGTCGC
			chh282	GAGTTTGATGTCTTTGATGAAAATAC
Q		FLOT1-4	Chh285	CACCGTAAGGGATTACTTGATGATAAA
			Chh286	TATTATCACCACCATTAGTCCAAAT

Table S4. Primers and constructs described in this study: amiRNA construct primers (Figs. 4 and 5 and Fig. S5)

Construct (original)	Construct (this study)	Intended target	Primer name	Primer sequence
		pRS300	amiRNA-A	CACCCTGCAAGGCGATTAAGTTGGGTAAC
			amiRNA-B	GCGGATAACAATTTCACACAGGAAACAG
EX101		FLOT 1–4	chh287	gaTCAAGTGTCACAATCCCCTATtctctcttttgtattcc
			chh288	gaATAGGGGATTGTGACACTTGAtcaaagagaatcaatga
			chh289	gaATCGGGGATTGTGTCACTTGTtcacaggtcgtgatatg
			chh290	gaACAAGTGACACAATCCCCGATtctacatatatattcct
EX102	FLOT1,3 (4) amiRNA	FLOT 1–4	chh291	gaTTTCACTTCAGTTCTCACTTAtctctcttttgtattcc
			chh292	gaTAAGTGAGAACTGAAGTGAAAtcaaagagaatcaatga
			chh293	gaTACGTGAGAACTGTAGTGAATtcacaggtcgtgatatg
			chh294	gaATTCACTACAGTTCTCACGTAtctacatatatattcct
EX103		FLOT2	chh295	gaTTTAACGTGATTGGAGTCCCGtctctcttttgtattcc
			chh296	gaCGGGACTCCAATCACGTTAAAtcaaagagaatcaatga
			chh297	gaCGAGACTCCAATCTCGTTAATtcacaggtcgtgatatg
			chh298	gaATTAACGAGATTGGAGTCTCGtctacatatatattcct
EX104		FLOT2	chh299	gaTGGTCCAAGACAGTGCACGGTtctctcttttgtattcc
			chh300	gaACCGTGCACTGTCTTGGACCAtcaaagagaatcaatga
			chh301	gaACAGTGCACTGTCATGGACCTtcacaggtcgtgatatg
			chh302	gaAGGTCCATGACAGTGCACTGTtctacatatatattcct
EX105	FLOT2(3) amiRNA	FLOT2	chh303	gaTTTCTCGGCACTCATAGCTCGtctctcttttgtattcc
			chh304	gaCGAGCTATGAGTGCCGAGAAAtcaaagagaatcaatga
			chh305	gaCGCGCTATGAGTGGCGAGAATtcacaggtcgtgatatg
			chh306	gaATTCTCGCCACTCATAGCGCGtctacatatatattcct
EX106		FLOT3	chh307	gaTACTAGTGAATCTCAGACGTGtctctcttttgtattcc
			chh308	gaCACGTCTGAGATTCACTAGTAtcaaagagaatcaatga
			chh309	gaCAAGTCTGAGATTGACTAGTTtcacaggtcgtgatatg
			chh310	gaAACTAGTCAATCTCAGACTTGtctacatatatattcct
EX107	FLOT2(3,4) amiRNA	FLOT3	chh311	gaTACTAGTGAATCTCAGACACGtctctcttttgtattcc
			chh312	gaCGTGTCTGAGATTCACTAGTAtcaaagagaatcaatga
			chh313	gaCGCGTCTGAGATTGACTAGTTtcacaggtcgtgatatg
			chh314	gaAACTAGTCAATCTCAGACGCGtctacatatatattcct
EX108		FLOT1	chh315	gaTAAATATTCATGACCCGCGACtctctcttttgtattcc
			chh316	gaGTCGCGGGTCATGAATATTTAtcaaagagaatcaatga
			chh317	gaGTAGCGGGTCATGTATATTTTtcacaggtcgtgatatg
			chh318	gaAAAATATACATGACCCGCTACtctacatatatattcct
EX109		FLOT1	chh319	gaTATTGAAGCAACGAGGACGCGtctctcttttgtattcc
			chh320	gaCGCGTCCTCGTTGCTTCAATAtcaaagagaatcaatga
			chh321	gaCGAGTCCTCGTTGGTTCAATTtcacaggtcgtgatatg
			chh322	gaAATTGAACCAACGAGGACTCGtctacatatatattcct
EX110	FLOT4 (amiRNA)	FLOT4	chh323	gaTAAAGGTGTAATTTACAGGCGtctctcttttgtattcc
			chh324	gaCGCCTGTAAATTACACCTTTAtcaaagagaatcaatga
			chh325	gaCGACTGTAAATTAGACCTTTTtcacaggtcgtgatatg
			chh326	gaAAAAGGTCTAATTTACAGTCGtctacatatatattcct
EX111		FLOT4	chh327	gaTGCACTTAGATACACCCGTTCtctctcttttgtattcc
			chh328	gaGAACGGGTGTATCTAAGTGCAtcaaagagaatcaatga
			chh329	gaGACCGGGTGTATCAAAGTGCTtcacaggtcgtgatatg
			chh330	gaAGCACTTTGATACACCCGGTCtctacatatatattcct
EX112	FLOT1(2) amiRNA)	FLOT1+2	chh331	gaTATAATCCGAATTGGTTCAGCtctctcttttgtattcc
			chh332	gaGCTGAACCAATTCGGATTATAtcaaagagaatcaatga
			chh333	gaGCCGAACCAATTCCGATTATTtcacaggtcgtgatatg
			chh334	gaAATAATCGGAATTGGTTCGGCtctacatatatattcct
EX116		None	chh349	gaTAGCCATAGCTAACTACTTCCtctctcttttgtattcc
			chh350	gaGGAAGTAGTTAGCTATGGCTAtcaaagagaatcaatga
			chh351	gaGGCAGTAGTTAGCAATGGCTTtcacaggtcgtgatatg
			chh352	gaAAGCCATTGCTAACTACTGCCtctacatatatattcct
EX117	empty vector	None	chh353	gaTATCAATCTTCTGTCACTCTTtctctcttttgtattcc
			chh354	gaAAGAGTGACAGAAGATTGATAtcaaagagaatcaatga
			chh355	gaAAAAGTGACAGAACATTGATTtcacaggtcgtgatatg
			chh356	gaAATCAATGTTCTGTCACTTTTtctacatatatattcct

Table S5. Primers and constructs described in this study: Vector construction

Construct	Insert	Template	Backbone vector	Fig.	Primer name	Primer sequence
pJG159	A (Sph-RB-P35S-RI)	pEGAD	pCAMBIA 1300		oJG346	CGAGGCGCATGCCAAATGGACGAACGGATAAAC
					oJG347	GGTACCGGAAGCTTCGAGCTCTTAATTAAGGCCT
					oJG348	CGAAGCTTCCGGTACCCCGTCAACATGGTGGAGC
					oJG349	TCTAGAGAATTCGGATCCGTCCTCTCCAAATGAAATG
pJG159	B (RI-osT-P35S-XhoI)		pCAMBIA 1300		oJG350	GGATCCGAATTCTCTAGACCCGATCGTTCAAACATTTG
					oJG351	CTCGCGCTCGAGCTGCAGTCCTCTCCAAATGAAATGAA
pCH010	eGFP	pDG71	pJG159		chh050	AAAAGGATCCAAGAATTCAACCCGGGATGGTGAGCAAGGGCGAGGAG
					chh051	AAAATCTAGATTACTTGTACAGCTCGTCCATG
pCH010	NPTII	pHELLS-GATE8	pJG159		chh039	TTTTCTCGAGCACTCTCAATCCAAATAATCTGCA
					chh040	TTTTCTCGAGTCAGAAGAACTCGTCAAGAAGGCG
pCH035	FLOT2 ORF	BAC Mth2-115c19	pCH010	3	chh156	TTTTGAATTCATGAAAATTTACCGGGTCGCG
					chh172	TTTTCCCGGGAGAGCTTTTCTCAGATAAGGC
pCH118	FLOT4 genomic region	BAC Mth2-115c19	pCH010	3,5	chh180	TTTCCCGGGATTCAAGTTTTTGTCAGGCAAGA
					chh181	TTTGGTACCTTCCCATGAACTTAACTCAATTG
pCH037	FLOT2 promoter	BAC Mth2-115c19	pMDC163	2	chh209	CACCTTGGCGGATATATGTGTGTGAT
					chh210	TCTTGATGATTTTGAGAAGAG
pCH038	FLOT3 promoter	BAC Mth2-115c19	pMDC163	2	chh211	CCACCCTCTAAAATCAGAATTGTCTG
					chh212	GTTGATTTTGGTTTGAATTTAG
pCH041	FLOT1 promoter	BAC Mth2-115c19	pMDC163	2	chh233	CACCGTCTTAATTAGGAGATGCAAGTA
					chh218	GTTGATAATGGTTTGAATTCGAGAAG
pCH042	FLOT4 promoter	BAC Mth2-115c19	pMDC163	2	chh232	CACCTTCCCATGAACTTAACTCAATTG
					chh220	CGTTGATTTTGATTTAATTTTTAAAAA
pQDN03	mCherry	pRSET-B mCherry	pDG71	5	QDN01	TTTGGATCCACCATGGTGAGCAAGGGC
					QDN02	TTTTCTAGATTACTTGTACAGCTCGTCCATGC