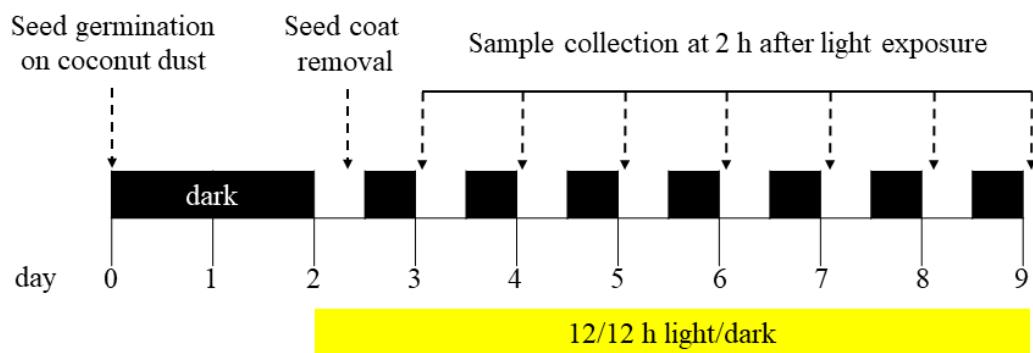


Supplementary Material

A



B



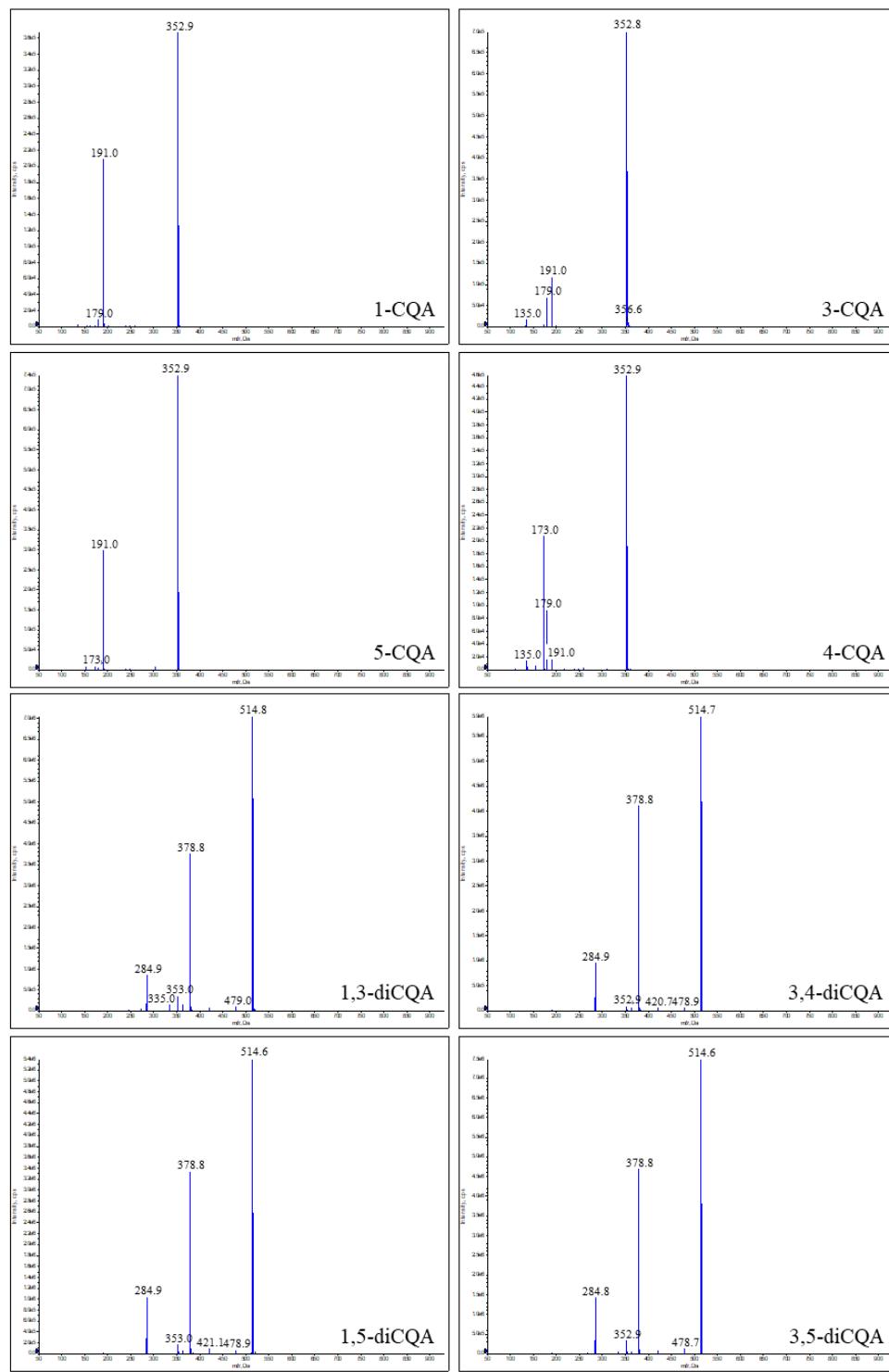
Supplementary FIGURE S1 | Sunflower sprouts used in this study. Schematic diagram of sampling in this study (A) and the representative sunflower sprouts were imaged at different timepoints (day 3 to day 9, from left to right respectively) during their germination period (B).

Supplementary FIGURE S2 | Multiple sequence alignment of HQTs and HCTs from sunflower (HaHQTs and HaHCTs) and other representative species. Amino acid sequences of HaHQTs and HaHCTs are shown and aligned with those from other representative species including tomato (SlHQT; NP_001234850.2), globe artichoke (CcsHQT1; ABK79689.1, CcsHQT2; ADL62854.1, CcsHQT3; ADL62855.1, CcsHCT1; AAZ80046.1, and CcsHCT2; KVH99042.1), and chicory (CiHQT1; ANN12610.1, CiHQT2; ANN12611.1, CiHQT3; ANN12612.1, CiHCT1; ANN12608.1, and CiHCT2; ANN12609.1).

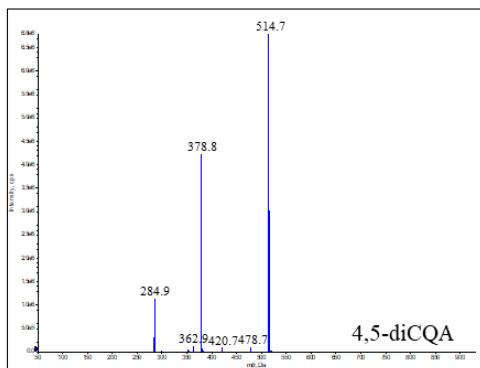
Supplementary FIGURE S2 | Multiple sequence alignment of HQTs and HCTs from sunflower (HaHQTs and HaHCTs) and other representative species. Amino acid sequences of HaHQTs and HaHCTs are shown and aligned with those from other representative species including tomato (SHQT; NP_001234850.2), globe artichoke (CcsHQT1; ABK79689.1, CcsHQT2; ADL62854.1, CcsHQT3; ADL62855.1, CcsHCT1; AAZ80046.1, and CcsHCT2; KVH99042.1), and chicory (CiHQT1; ANN12610.1, CiHQT2; ANN12611.1, CiHQT3; ANN12612.1, CiHCT1; ANN12608.1, and CiHCT2; ANN12609.1). (continue)

S1HQT	SALDYLELQPDLSLIRGPAYFASPNLNINSWTRLPVHECDFGWGRPIHMGPACILYEGT	395
CcsHQT1	SAIDYLELQPDLTALVRGPTYFASPNLNINSWTRLPIYESDFGWGRPIFMGPASILYEGT	400
CcsHQT2	SAIDYLESQPDLSALIRGPSYFASPNLNINAWTRLPVYDADLGWGRPIFMGPACILYEGT	409
CcsHQT3	SAIDYPEVQPDLSALIRGPSYFASPNLNINTWTRLPVHDADFGWGRPVFMGPACILYEGT	398
CcsHCT1	SALDYLELQPDLKALVRGAHTFKCPNLGITSWARLPIHADAFGWGRPIFMGPGGIAYEGL	402
CcsHCT2	SALDYLELQPDLKALVRGAHTFKCPNLGITSWARLPIHADAFGWGRPIFMGPGGIAYEGL	396
CiHQT1	SAIDYLELQLDL SALIRGPTYFASPNLNVNSWTRLPLYESDFGWGRPIFMGPANILYEGT	404
CiHQT2	SAIDYLESQKDL SALIRGPTYFASPNLNINAWTRLPVHDADFGYGRPIFMGPQILYEGT	405
CiHQT3	SAIDYLEVQPDLSALIRGPTYFASPNLNINTWTRLPVHDADFGWGRPVFMGPAVILYEGT	407
CiHCT1	SALDYLELQPDLKALVRGAHSFRCPNLGITSWARLPIHADAFGWGRPIFMGPGGIAYEGL	400
CiHCT2	SALDYLELQPDLKALVRGAHTFKCPNLGITSWARLPIHADAFGWGRPIFMGPGGIAYEGL	396
HaHQT1	SAIDYLESITDL SLIRGPTYFASPNLNVNSWTRLPIYDSDFGWGRPIFMGPASILYEGT	401
HaHQT2	SAIDYLETQPDISALIRGPTYFASPNLNINAWTRLPVHDADFGWGRPIFMGPATILYEGT	406
HaHQT3	SAIDYLEVQPDLSALIRGPTYFASPNLNINTWTRLPVHDADFGWGRPVFMGPACILYEGT	413
HaHCT1	SALDYLELQPDLKALVRGAHTFKCPNLGITSWARLPIHADAFGWGRPIFMGPGGIAYEGL	400
HaHCT2	SALDYLELQSDLKALVRGAHTFKCPNLGITSWARLPIHADAFGWGRPIFMGPGGIAYEGL	396
	:* * * :*: * :* * . * . ***. :	
S1HQT	IYIIPSPNSKDRNLRLAVCLDAHMSLFEKYLYEL	430
CcsHQT1	IYIIPSPSG-DRSVSLAVCLDPDHMSLFRKLYDF	434
CcsHQT2	IYVLPPSPNN-DRSVSLAVCLDANEQPLFEKFLYEF	443
CcsHQT3	IYVLPPSPNN-DRSMSLAVCLDADEQPLFEKFLYDF	432
CcsHCT1	SFVLPPSPIN-DGSLSIVISLQAEHMKLFSKFLYDI	436
CcsHCT2	SFVLPPSPIN-DGSLSIAISLQSEHMKLFSKFLYDI	430
CiHQT1	IYIIPSPTD-DRSLKLA VCLDSDHMSLFQKYLYDF	438
CiHQT2	VYVLPPSPNN-DRSVSLAVCLDANEQPLFEKFLYEM	439
CiHQT3	IYVLPPSPNN-DRSMSLAVCLDADEQPLFEKFLYEF	441
CiHCT1	SFVLPPSPIN-DGSLSIAISLQAEHMKVFSKFLYDI	434
CiHCT2	SFVLPPSPVN-DGSIWIAISLGEHMKLFSKFLYDI	430
HaHQT1	IYILPGPGG-DRSVALAVCLDPDHMALFKERLYDF	435
HaHQT2	VYVLPPSPNN-DRSVSLAVCLDAKEQPLFEKYLYEL	440
HaHQT3	IYVLPPSPNN-DRSMSLAVCLDADEQPLFEKFLYDF	447
HaHCT1	SFVLPPSPDN-DGSLSIAISLQAEHMKLFSNLLYDI	434
HaHCT2	SFVLPPSPIN-DGSLSIAISLQAOQHMKLFSNYLYDI	430
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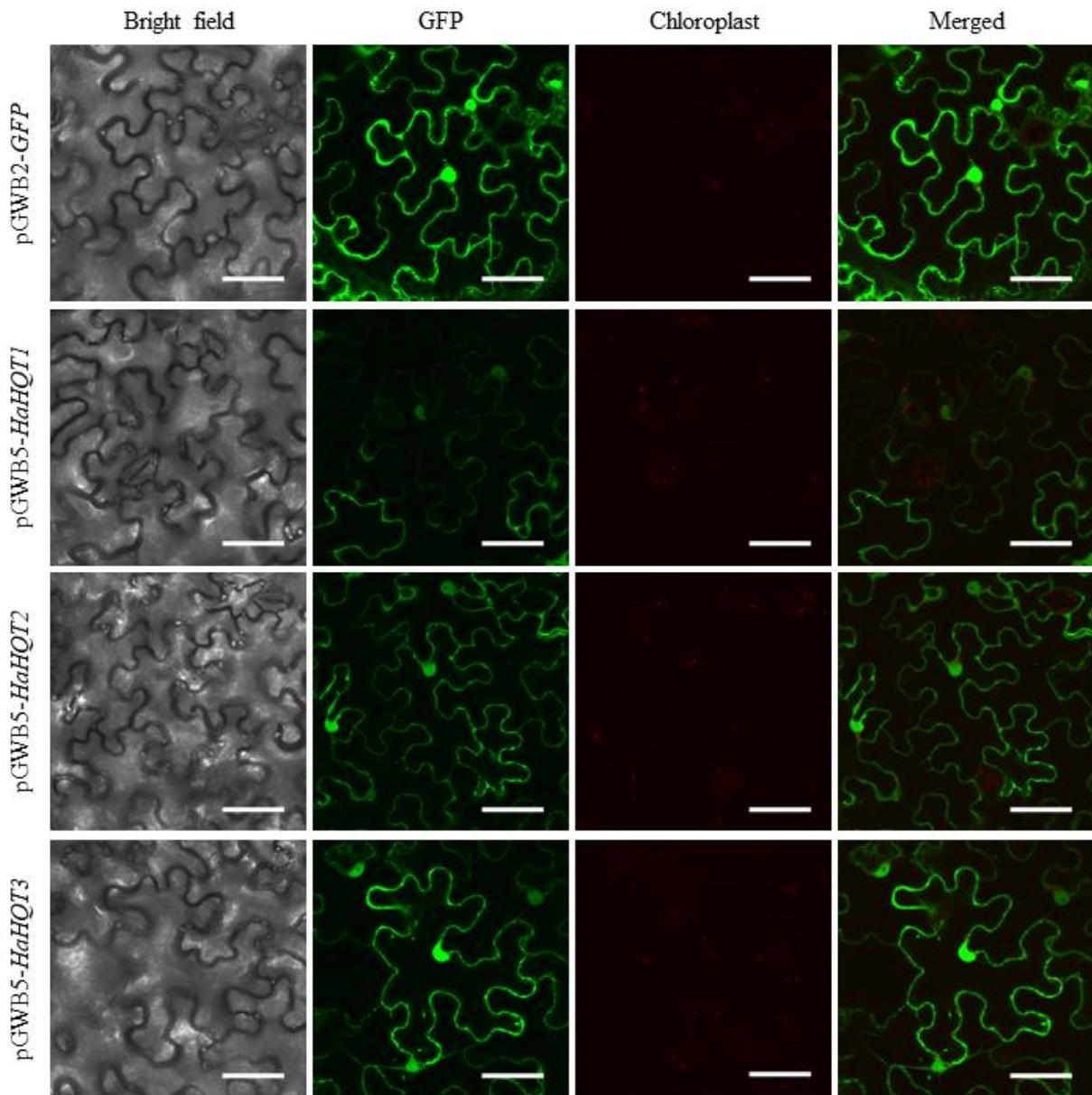
Supplementary FIGURE S2 | Multiple sequence alignment of HQTs and HCTs from sunflower (HaHQTs and HaHCTs) and other representative species. Amino acid sequences of HaHQTs and HaHCTs are shown and aligned with those from other representative species including tomato (SIHQT; NP_001234850.2), globe artichoke (CcsHQT1; ABK79689.1, CcsHQT2; ADL62854.1, CcsHQT3; ADL62855.1, CcsHCT1; AAZ80046.1, and CcsHCT2; KVH99042.1), and chicory (CiHQT1; ANN12610.1, CiHQT2; ANN12611.1, CiHQT3; ANN12612.1, CiHCT1; ANN12608.1, and CiHCT2; ANN12609.1). (continue)



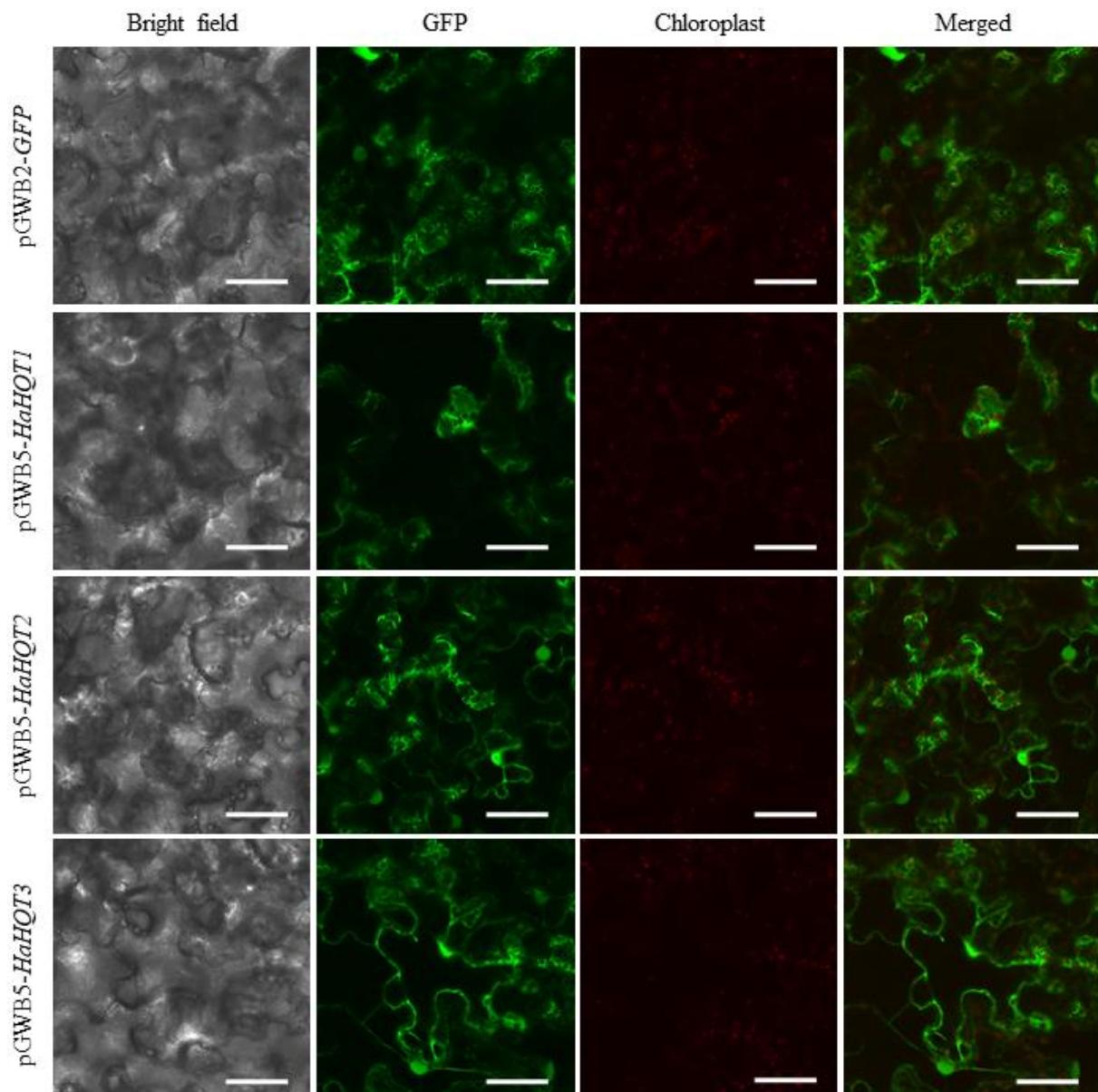
Supplementary FIGURE S3 | Negative ion MS^2 spectra of monocaffeoylquinic acids and dicaffeoylquinic acids. Enhanced Product Ion (EPI) profile of caffeoylquinic acid derivatives from authentic standards, including monnocoaffeoylequinic acids such as 1-, 3-, 4- and 5-CQA, and diaffeoylequinic acids such as 1,3-, 1,5-, 3,4-, 3,5- and 4,5-diCQA.



Supplementary FIGURE S3 | Negative ion MS² spectra of monocaffeoylquinic acids and dicaffeoylquinic acids. Enhanced Product Ion (EPI) profile of caffeoylquinic acid derivatives from authentic standards, including monnocoaffeoylequinic acids such as 1-, 3-, 4- and 5-CQA, and diaffeoylequinic acids such as 1,3-, 1,5-, 3,4-, 3,5- and 4,5-diCQA. (continued)



Supplementary FIGURE S4 | Subcellular localization of GFP-tagged HaHQTs in the epidermis of *Nicotiana benthamiana* leaves. Confocal microscopy image of epidermal cells from *N. benthamiana* leaves infiltrated with pGWB2-GFP (control), pGWB5-HaHQT1, pGWB5-HaHQT2, or pGWB5-HaHQT3. Bars=50 μm



Supplementary FIGURE S5 | Subcellular localization of GFP-tagged HaHQTs in the mesophyll of *Nicotiana benthamiana* leaves. Confocal microscopy image of epidermal cells from *N. benthamiana* leaves infiltrated with pGWB2-GFP (control), pGWB5-HaHQT1, pGWB5-HaHQT2, or pGWB5-HaHQT3. Bars=50 μm

Supplementary Table S1 | All primers used in this study.

Experiment	Gene	Accession number	Primer Sequence (5'→3')		Product size (bp)
			Forward	Reverse	
Cloning	<i>HaHQT1</i>	MK598073	ATGAACCTAACAGTAAACAAATCACTAA	CTAGAAATCATACAAACGTTCCCTAAA	1,305
	<i>HaHQT2</i>	MK598074	ATGAAAACCAGATCAAACCAAA	TTACAATTCATACAAGTACTTCTCAAAAA	1,323
	<i>HaHQT3</i>	MK598075	ATGGGGACTGTTCAAAAGACA	TTAGAAGTCGTATAAGAACCTTCTCAAAACA	1,326
Subcellular localization	<i>HaHQT1</i>	MK598073	ATGAACCTAACAGTAAACAAATCACTAA	GAAATCATACAAACGTTCCCTAAA	1,302
	<i>HaHQT2</i>	MK598074	ATGAAAACCAGATCAAACCAAA	CAATTCATACAAGTACTTCTCAAAAA	1,320
	<i>HaHQT3</i>	MK598075	ATGGGGACTGTTCAAAAGACA	GAAGTCGTATAAGAACCTTCTCAAAACA	1,323
qRT-PCR /ddPCR	<i>ETIF5</i>	XM_022156448.1	TTGTGCCCTCGTCCCATAAC	AAATCTGCTCCCTCCCCATC	231
	<i>EF12</i>	XM_022137686.1	TTGACCGCTAACGCCACGTT	AGGCACCAAGAAAACCCGAAT	199
	<i>ACT7</i>	XM_022154554.1	TTGGAATGGAAAGCTGCTGGT	CTGGAGGAGCAACCACCTTG	201
<i>HaHQT1</i>	<i>HaHQT1</i>	MK598073	ACCGCAAGGGAGAACATCCACAA	CGTCCCCATCCAAAGTCAGA	194
	<i>HaHQT2</i>	MK598074	TGGTAAACGCTGCGAACACTG	GTCGGGGTCCCATAAAAAT	223
	<i>HaHQT3</i>	MK598075	CCCGATGACCAACTAACCA	GCTGATAGATGGGCTGCAC	246
<i>HaHCT1</i>	<i>HaHCT1</i>	MK598076	GAGCTGCAGCCCGATCTAAA	CCGGCTTGGTAACACAAAAA	184
	<i>HaHCT2</i>	MK598077	CGGGCTTGGATTACTTGGAG	CACCAGGCCATGAATATG	159

Supplementary Table S2 | List of regulatory elements found in the promoter regions of *HaHQT2* and *HaHQT3*.

IUPAC family	Number of elements found		Function
	<i>HaHQT2</i> promoter	<i>HaHQT3</i> promoter	
P\$FAM002	3	12	light responsiveness, auxin/ salicylic acid/ abscisic acid/ jasmonate responsiveness, stress regulation, dehydration responsiveness
P\$FAM003	5	3	phenylpropanoid biosynthesis, phytochrome regulation, MYB
P\$FAM008	1	1	water stress responsiveness, MYB
P\$FAM010	8	4	elicitor responsiveness, defense response regulation, salicylic acid responsiveness, sucrose responsiveness
P\$FAM012	3	7	light responsiveness, light regulation, MYB
P\$FAM013	1	3	abscisic acid responsiveness, dehydration responsiveness, low temperature responsiveness
P\$FAM014	4	8	phytochrome regulation, sugar repression, MYB
P\$FAM015	0	2	sugar repression
P\$FAM022	3	1	storage protein
P\$FAM024	3	4	storage protein
P\$FAM025	1	0	sugar repression
P\$FAM026	1	0	light responsiveness
P\$FAM027	2	1	light regulation, circadian rhythms
P\$FAM061	1	1	pathogen, ethylene, and jasmonate responsiveness
P\$FAM069	1	2	auxin responsiveness, sulfur responsiveness
P\$FAM085	0	1	light responsiveness
P\$FAM098	0	2	auxin responsiveness
P\$FAM099	1	0	phytochrome responsiveness
P\$FAM107	1	1	sugar starvation
P\$FAM116	1	0	abscisic acid and drought responsiveness
P\$FAM124	1	0	ethylene responsiveness
P\$FAM162	2	1	low temperature responsive element
P\$FAM170	1	1	gibberellin responsiveness, MYB
P\$FAM171	1	5	MYB
P\$FAM172	4	2	dehydration responsiveness
P\$FAM205	0	2	gibberellin responsiveness and sugar repression, Dof
P\$FAM227	2	0	storage protein
P\$FAM234	0	1	storage protein
P\$FAM243	2	6	storage protein
P\$FAM244	2	0	storage protein
P\$FAM245	0	2	storage protein
P\$FAM251	1	0	light responsiveness
P\$FAM261	2	2	low temperature responsiveness

Supplementary Table S2 | List of regulatory elements found in the promoter regions of *HaHQT2* and *HaHQT3*. (continue)

IUPAC family	Number of elements found		Function
	<i>HaHQT2</i> promoter	<i>HaHQT3</i> promoter	
P\$FAM262	1	0	light responsiveness and circadian expression
P\$FAM263	5	3	abscisic acid responsiveness
P\$FAM266	6	2	dehydration responsiveness, MYB
P\$FAM267	10	17	auxin responsiveness, Dof
P\$FAM273	1	1	gibberellin responsiveness, sugar starvation
P\$FAM290	7	4	pathogen- and salt-induced
P\$FAM302	2	3	light responsiveness, axillary bud outgrowth
P\$FAM310	2	1	cytokinin
P\$FAM311	1	1	low CO ₂ , MYB
P\$FAM315	0	1	light responsiveness
P\$FAM324	0	2	calmodulin
P\$FAM325	3	0	cell cycle, MYB
P\$FAM329	0	1	wood formation