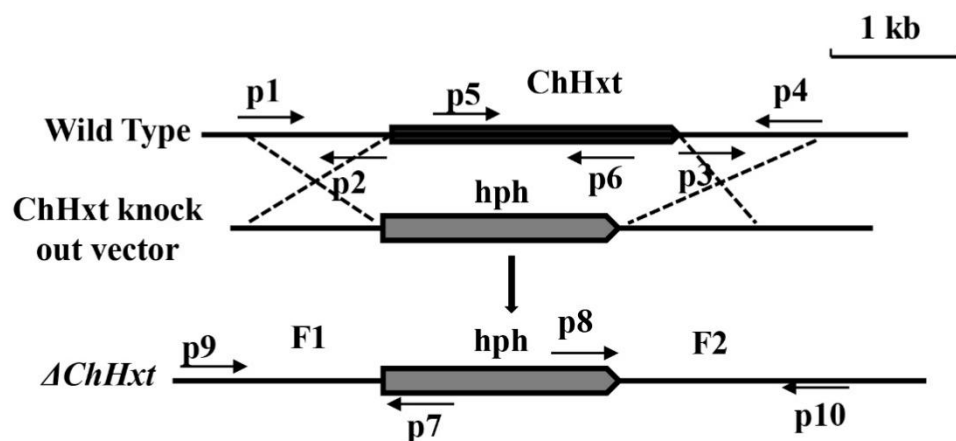


Table S1 Primers used in this study

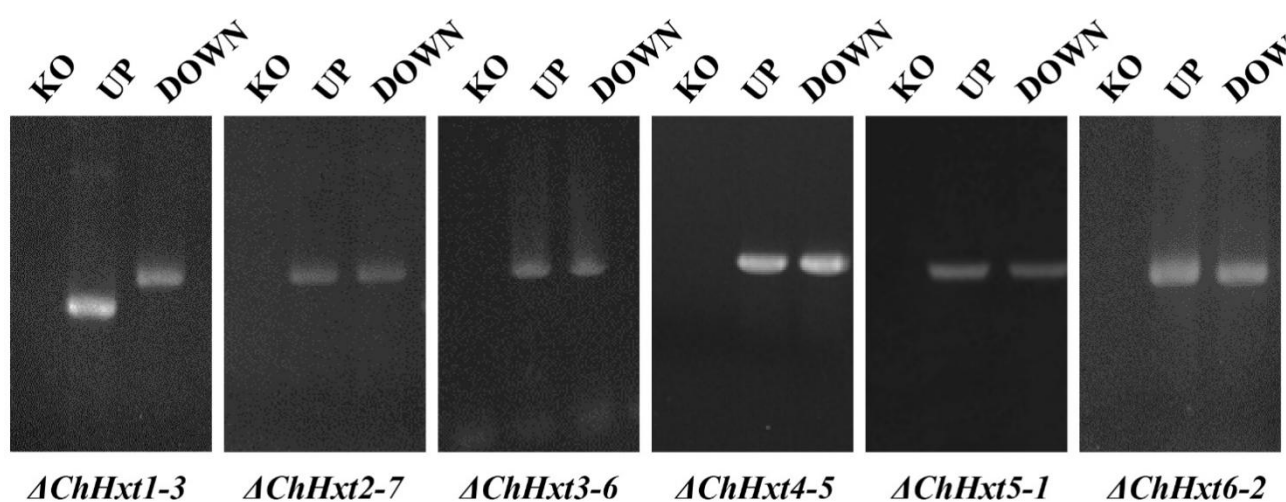
Primer name	Sequence (5'-3')	Remark
ChHxt1F1FP	5'-TGCATTTTCCTCGTAAATTCTGGT-3'	Clone ChHxt1F1
ChHxt1F1RP	5'-CCAAGGAATGCATCTTCAAGCA-3'	
ChHxt1F2FP	5'-TGCACCACTCCTTGATGACC-3'	Clone ChHxt1F2
ChHxt1F2RP	5'-CCTCAAGTCAACACGGAGCA-3'	
ChHxt2F1FP	5'-AAACAGAGCCTCCACGATGC-3'	Clone ChHxt2F1
ChHxt2F1RP	5'-GGTTCCACACAAGGGCCTAA-3'	
ChHxt2F2FP	5'-TCTAACTGGTAAAGCTCCTCAGA-3'	Clone ChHxt2F2
ChHxt2F2RP	5'-ATCCACACCAAACATCCGCA-3'	
ChHxt3F1FP	5'-CTGGCCGTTAATCACGCTCT-3'	Clone ChHxt3F1
ChHxt3F1RP	5'-GAGACGCGGAAAGTCTCAG-3'	
ChHxt3F2FP	5'-ACCCATTTTCTCTTATGGGCCT-3'	Clone ChHxt3F2
ChHxt3F2RP	5'-CACGCCTTTTCTTCCCATGC-3'	
ChHxt4F1FP	5'-AGGCATCCAACACCACCTTATT-3'	Clone ChHxt4F1
ChHxt4F1RP	5'-GATCGTTCCTGCCTGGTTCA-3'	
ChHxt4F2FP	5'-CCTCAAGTCAACACGGAGCA-3'	Clone ChHxt4F2
ChHxt4F2RP	5'-AAACAGAGCCTCCACGATGC-3'	
ChHxt5F1FP	5'-GGTTCCACACAAGGGCCTAA-3'	Clone ChHxt5F1
ChHxt5F1RP	5'-TCTAACTGGTAAAGCTCCTCAGA-3'	
ChHxt5F2FP	5'-ATCCACACCAAACATCCGCA-3'	Clone ChHxt5F2
ChHxt5F2RP	5'-CTGGCCGTTAATCACGCTCT-3'	
ChHxt6F1FP	5'-CCTCAAGTCAACACGGAGCA-3'	Clone ChHxt6F1
ChHxt6F1RP	5'-AAACAGAGCCTCCACGATGC-3'	
ChHxt6F2FP	5'-GGTTCCACACAAGGGCCTAA-3'	Clone ChHxt6F2
ChHxt6F2RP	5'-TCTAACTGGTAAAGCTCCTCAGA-3'	
HPH-UP	5'-CACGCCTTTTCTTCCCATGC-3'	Validation of the ChHxt1 knockout mutants
HPH-DOWN	5'-ATCCACACCAAACATCCGCA-3'	
ChHxt1-KFP	5'-ATCCACACCAAACATCCGCA-3'	
ChHxt1-KRP	5'-CTGGCCGTTAATCACGCTCT-3'	
ChHxt1-UP	5'-TGCAGTGACAGATGAGCACAA-3'	Validation of the ChHxt2 knockout mutants
ChHxt1-DOWN	5'-ACTCTCCTGAGCTCCACAGT-3'	
ChHxt2-KFP	5'-CTGGCCGTTAATCACGCTCT-3'	
ChHxt2-KRP	5'-GAGACGCGGAAAGTCTCAG-3'	
ChHxt2-UP	5'-TGCACCACTCCTTGATGACC-3'	Validation of the ChHxt3 knockout mutants
ChHxt2-DOWN	5'-CCTCAAGTCAACACGGAGCA-3'	
ChHxt3-KFP	5'-ACCCATTTTCTCTTATGGGCCT-3'	
ChHxt3-KRP	5'-CACGCCTTTTCTTCCCATGC-3'	
ChHxt3-UP	5'-CTGGCCGTTAATCACGCTCT-3'	Validation of the ChHxt4 knockout mutants
ChHxt3-DOWN	5'-GAGACGCGGAAAGTCTCAG-3'	
ChHxt4-KFP	5'-AGGCATCCAACACCACCTTATT-3'	
ChHxt4-KRP	5'-GAGACGCGGAAAGTCTCAG-3'	

ChHxt4-UP	5'-CAAACATGGCTAAGCAGGGC-3'	Validation of the ChHxt4 knockout mutants
ChHxt4-DOWN	5'-GTCACAAGGCCTCTGCTTTTG-3'	
ChHxt5-KFP	5'-ACCCATTTTCTCTTATGGGCCT-3'	Validation of the ChHxt5 knockout mutants
ChHxt5-KRP	5'-CACGCCTTTTCTTCCCATGC-3'	
ChHxt5-UP	5'-GTCACAAGGCCTCTGCTTTTG-3'	Validation of the ChHxt6 knockout mutants
ChHxt5-DOWN	5'-AGTTACACAAAGCCGTGACAA-3'	
ChHxt6-KFP	5'-AGGCATCCAACACCACCTTATT-3'	Validation of the ChHxt6 knockout mutants
ChHxt6-KRP	5'-GATCGTTCCTGCCTGGTTCA-3'	
ChHxt6-UP	5'-CCTCAAGTCAACACGGAGCA-3'	Complementation of ChHxt6
ChHxt6-DOWN	5'-AAACAGAGCCTCCACGATGC-3'	
ChHxt6C-FP	5'-GGTTCCACACAAGGGCCTAA-3'	Create mutant ChHxt6T169S
ChHxt6C-RF	5'-GGTCTTCCACGATGTAACGAAGCA-3'	
ChHxt6T169S-FP	5'-CTGACTATTATGGTCCTACTATTTTCA-3'	Create mutant ChHxt6P221L
ChHxt6T169S-RP	5'-TGAAAATAGTAGGACCATAATAGTCACAG-3'	
ChHxt6P221L-FP	5'-TTTTTCTACTACGGTGCCGTTATTTTCAAGTCA-3'	Clone ChHxt1
ChHxt6P221L-RP	5'-TGACTTGAAAATAACGGCACCGTAGTAGAAAAA-3'	
ChHxt1FP	5'-ACACTGACATTGATCGCAGC-3'	Clone ChHxt2
ChHxt1RP	5'-GACAAGCTATGCAATGCCACA-3'	
ChHxt2FP	5'-CAAACATGGCTAAGCAGGGC-3'	Clone ChHxt3
ChHxt2RP	5'-GTCACAAGGCCTCTGCTTTTG-3'	
ChHxt3FP	5'-AGTTACACAAAGCCGTGACAA-3'	Clone ChHxt4
ChHxt3RP	5'-CCATATTAAGCACTGGACGAGC-3'	
ChHxt4FP	5'-ACTGACATTGATCGCAGCGG-3'	Clone ChHxt5
ChHxt4RP	5'-GTCACAAGGCCTCTGCTTTTG-3'	
ChHxt5FP	5'-TGTAACAAGGTTAGAGTTCAGGT-3'	Clone ChHxt6 and ChHxt6 point mutants
ChHxt5RP	5'-CCAGGGGAGTAACCATTTTTCG-3'	
ChHxt6FP	5'-AAACGAGGCAGACAAACATGC-3'	
ChHxt6RP	5'-TCTTCCACGATGTAACGAAGCA-3'	

A



B



C

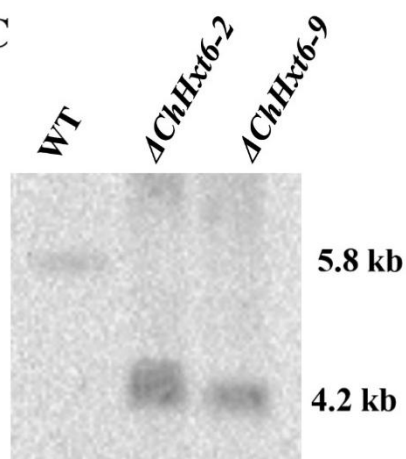


Figure S1 Gene knockout of ChHxts. A: Deletion strategy for ChHxt1~ChHxt6 genes in *C. higginsianum*. The p1 to p10 represents primers used for construction of ChHxt deletion vectors and validation of ChHxt mutants. p1: ChHxtF1FP, p2: ChHxtF1RP, p3: ChHxtF2FP, p4: ChHxtF2RP, p5: ChHxt-KFP, p6: ChHxt-KRP, p7: HPH-UP, p8: HPH-DOWN, p9: ChHxt-UP, p10: ChHxt-DOWN. B: Validation of ChHxt deletion mutants by PCR with primer pairs of p5/p6 (KO), p7/p9 (UP) and p8/p10 (DOWN). C: Validation of ChHxt6 knockout mutants by Southern blotting.

ChHxt1p	MG..IKPAGVPGKAWFAIAI..FFVAFGLLEGYDTGTNGILLESYWRLESTGYVNPAGNPDVSPSQESAIVSIL	74
ChHxt2p	MGLGKLSVRINGAECGAELMLL..AITSIGGLFGYDTGQISGMLLFSDFKKRATGSLGPDGLPQWVPTTQSLMVSIM	78
ChHxt3p	MEKETGVTRPVPQGLAVTD.DVNATEAPVTWKAYLIC.AFASFGGTFFGVDSGYINGVLGSSIFIHAVE.....GPGATSIGESDTSLIVSIL	86
ChHxt4p	MAIAMGW.QKPDNVAGSSAPAIMV..LFVASGGLLEGYDTGTNGILSMTAFRRDFTTGYT.ENGLPAISPAESSIIIVAIL	78
ChHxt5p	MQFPKLGKGVHKKDRTDADAPTDSNSVDLVAEQKVTFACFLG.LVASIGGFMFGYVSGQISGFFELMDFMERFGE...PQNGTYIFSAARQCTIVGLL	96
ChHxt6p	...MGWNLKLNKSDDRHALESTTPSTTKTVSWLKDPGLRPLNLFLELLFGDVAFSGDSTLNNLQQINKWQEDFD.....HPRKSLLGALSASY	87
ChHxt1p	SAGTFFSALASPLLDSTGRRLGTAASCWFNIGVVLQIA...TDIPVELAGRFFAGFGVGLISALVPLYQSETAPKWIRGAIYGAYQWATTIGLLAA	171
ChHxt2p	LINFGVRNFQDSDASWRIVIGLGIGFSLPLGLGILFVPRSPRWLAGQDWEGRMALARLRLGLKDDPHCDLVEKDLQEMFKVIEFESKTGY....GTWA	176
ChHxt3p	SCGTFFSALLIAGDLADMGRKWTIVILGCLIMIGVLIQMITNVNTALGPIVAGRIIAGLGVGFEASVILYMBEISPKKVRGALSGYQFCITIGLLAA	186
ChHxt4p	SAGTILGALLAAPIDSWGRRIISLISVGVSPFGGIFQVCA...HDIPMLLVGRFFAGVGVSISVLPVYQSEMAPKWIRGTLCAVQLSITIGLLAS	175
ChHxt5p	CTGCLVSAITAGKLADTTGRRLTISLFAFFSCIGIVIEISS..TNKWYQFAIGRLVNGVIGIGALSVPVPMYQSESTAIIRGVISTYQLFITLGIWLAE	194
ChHxt6p	WIGNILGVVLIPLVADRIGRIAAAGSLLCIGGAACAAT...TSNGAFIAGRILLMCGVMCSVSTVLMTELAYPAHRETATALSSTTYSVGCIALAA	184
ChHxt1p	IVNNATHNRQDT.GSYRIPIAIOFAWSIILFVSMIILPETPRFLIKSCHVDRATKALAKLRRL..PENDPYVAEEIAEIKANHDETSIGT....ASYL	263
ChHxt2p	LINFGVRNFQDSDASWRIVIGLGIGFSLPLGLGILFVPRSPRWLAGQDWEGRMALARLRLGLKDDPHCDLVEKDLQEMFKVIEFESKTGY....GTWA	271
ChHxt3p	CVVYATKDRDIT.DGSYRIPIAIOFPWAILLGGELMLFPPSPRYFVKKRLADARSLSRLRGQ..PENSEYIQVELAEIVANEYERQLVPNTTWFGTWA	283
ChHxt4p	FVNILTEKQLTA.AAYRVPLGLQLVWAVVLAICLLVLPEPRFLVVKQCKPEAAGLSRLRL..DITHALLEELQELIANHEVELTLGP....DSYK	267
ChHxt5p	IVNFATHNKVGS.ASWRIENGLGEAWALILGASTILLPESPRYAFROGREDEARRNIAGV..EPNAMSVNAQIDDIRAKVAESAGAD....TSIW	286
ChHxt6p	WVAFGSFRIEDS.WAWRTTTLIQAFPSAMVLCLEFLPESPRWLCSSRKEQTLDTIAKYHGAG.NVDVAVVQHEYAEIRDITIEAFITQKNKP...FHLK	279
ChHxt1p	DCFR.....EPVLKQFTGMALQALQQLTGINFIFYYCTQYFQNS.GFSNGFVIGMTS...SINVVSTIPGMYAVDRWGRPMMLWGAVGMCVSQFLV	353
ChHxt2p	ECFPGSS.GIKTVYRTLLGLSLHFIQWTCVNFPPFYGATIFESA.GIEDPIMMOLLG...AVNVFCIFFGLYAVEKYGRRWPLIGAVWQTAWLTVF	366
ChHxt3p	NCFKGSVFKANSNLRTILGTSLOMMQWTVGNFIHYSTPFLQSTGAIDNTFLISLVT...LVNVCSPLSFWTVERFGRRTILLWGATGMLICQFLV	380
ChHxt4p	EIFYG...SEHLGRKRTLTGCLQMLQQLTGINFIFYYCTQYFQNS.GFSNGFVIGMTS...SINVVSTIPGMYAVDRWGRPMMLWGAVGMCVSQFLV	359
ChHxt5p	EIFYG...EPMLYNTILGVLQAGQQLIGANFFFYGTTFVSAT.GLSDSYVTQLLG...SVNVACTFGGLLVVKKSGRPMALIIGALWMMCMFFVY	377
ChHxt6p	ALPAT.....PGRNRWSPFIWCGICKQWSENGLVSYLGSMLKSA.GITKQIETTLTATSQMFSAFCEAFALPARVGRRLMLTSMAGMWIVFAMI	373
ChHxt1p	AMLG.....TLTTGQDDAGKIIIVNLPQKAAIAIVCIYIEFFASTWGFANVVNGELFGLKTRAKSLSLSTATNWLNLWAIAYATPYLVNYGDSYANL	447
ChHxt2p	AAVG.....VAMP.....PETNSTTGIVMIVSACMFASFSTWGMCMCVIGETFLRTRAKQASLATAGNWLGNFLIAELTPYATA.....GI	446
ChHxt3p	AIIIGVTGPNHHTPDPADATKSIANNIPVNAQIAIAIFTEWFASTWGFANVNLGELFELPLPMSRGVALSTASNWLWNTIIAVITPYMVGENRG..NL	478
ChHxt4p	ASF.....TTAAGEN.....LQQAQRTILIVCAVNIFFPAASWGFVCHVITSELYPLKVRKANSISTASNWLNLFGIAYGTBFMVQGTGSADI	445
ChHxt5p	AFVGH.....FALDQND.....PQNTPTAGTVLVAESCLFIAGPATWGLVAVVAELYPARYAPAMALATASNWLNLFLMSFFTRYITD.....AI	461
ChHxt6p	TATS.....GAYVETG.....NRHASYYTTVAEIIYLSGVHNLGWTGAQMLYVEILLYTIRAKGMAMFSLVAGTCGAFNTYVNLGIA.....AM	453
ChHxt1p	QSKIFFVWFACFLCIAFYFFIYETKGLTLEVEEELYAEVSVASKS.ANWRPATFRERQAAEGDKAVPHNSDGDTAHHEKTVDIA.....	532
ChHxt2p	GYSYGFVEAACNLAGAVVWFFLYETKMLSLNVDNRMYSDDPSVKPYSSSKWVPPGYITRKQKDDSAFQASVHSENTAVGGDQHKKEKTSDDERPFSEHHT	546
ChHxt3p	KSSVFFVWGGCLSCALVITYFLVPETKGLSLPQVDKMEET.TPRTS.AKWVPHETFASQVARNG..VLDKVTVDNVERRSSNV.....	558
ChHxt4p	GPKIFFLWGAFCILAVLFVWCMVBETSKISLEQIDEMYERVDYAWN.TREEPSWSEFQILDEGWSPSAQPPLEHELQQTQTNSTTQTQTSHTNSNAT	544
ChHxt5p	DYLYGVFAGCCAAALAVIYFFEVIESKDRTLEIDITMYVQR.VNPITSAKWR..GEVRKNHHNESSEAEKAEATV.....	534
ChHxt6p	SWKPYEFYVGIWIVQFVVYLVFPETKGPSLEQIALLEDGK.DAQVRVNAVADMLDEKEGGVAVETRERQ.....	524
ChHxt1p		532
ChHxt2p	KQEPLANNV.....	555
ChHxt3p		558
ChHxt4p	STNTGATGNSTSAEKMLSQMGNVDFS	571
ChHxt5p		534
ChHxt6p		524

Figure S2 Multi-alignment of ChHxt1p to ChHxt6p. The amino acid sequences of ChHxt1p to ChHxt6p were aligned with DNAMAN (v9). Homology parts were highlighted with dark shading.