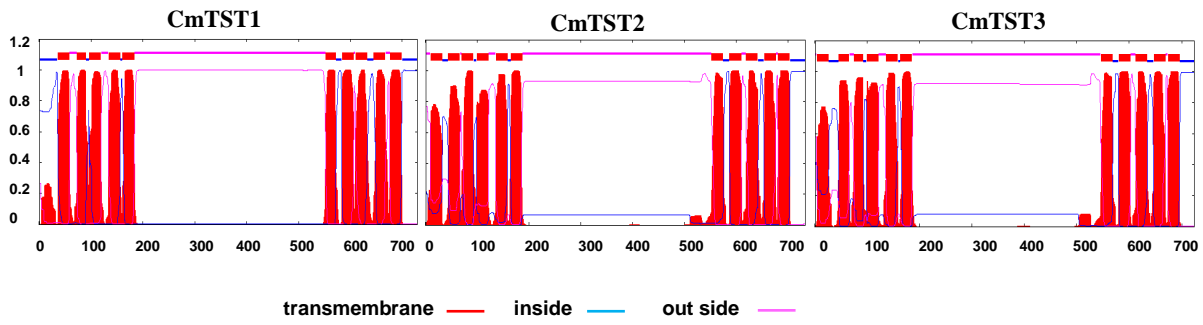
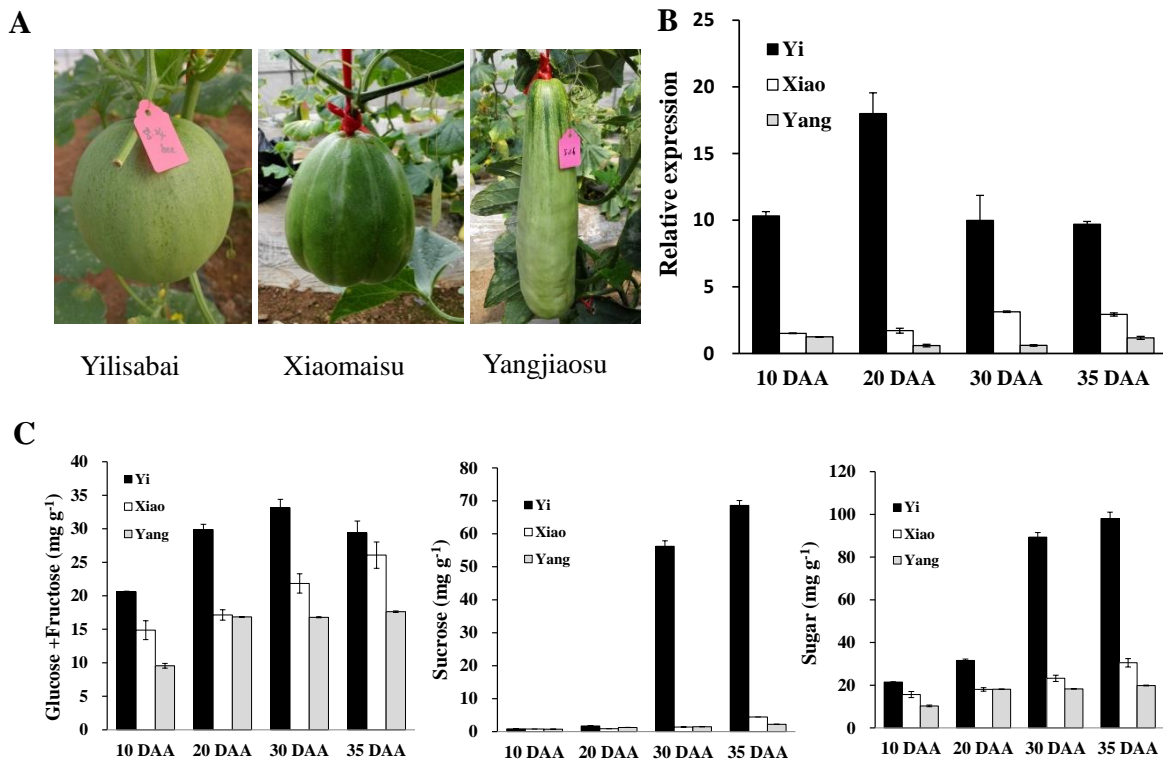


AtTST2	MSGAVLVAAARVGNLIGWDNATIAGAVLYIKKEFNLESNPVEGLIVATSLIGATLITTCSCGVADNLGRRPMLLSSILYFVGSVLWSPNVYVLL	100
BvTST2.1	MSAAVLVAHAATVGDLLYGDWNTIAGAVLYIKKEFNLESSPTEGLIVATSLIGATLITTCSCPTADRLGRRPMMIISVCFVSAIHLWSPNVYVLL	100
CmTST2	MSGSVLVAAARVGNLIGWDNATIAGAVLYIKKEFNLESSPVEGLIVATSLIGATLITTCSCAISDNLGRRLLLFSSVLYFVGGIIMLWSPNVYVLL	100
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AtTST2	IGRLLDGFVGLVITLVEFYISETAPPEIRGLNLTLPQFTGSGMFLSYCMVFGMSLMFSPSWRLMLGLVLFHPSLVFFFLTVFFLPESPRWLVSKGRMLE	200
BvTST2.1	IGRLLDGFVGLVITLVEFYISETAPTDIRGSLNLTLPQFTGSGMFLHAYCMVFGMSLMETPSWRLMLGLVLFVPSLVVYVLLTVFFLPESPRWLVSKGRMNE	200
CmTST2	IGRLLDGFVGLVITLVEFYISETAPPEIRGSLNLTLPQFTGSGMFFSYCMVFGMSLMFSPSWRLMLGLVLFHPSLIYLAITTVFFLPESPRWLVSKGRMLE	200
	<div style="display: flex; justify-content: space-around; width: 100%;"> <span>4</span> <span>5</span> <span>6</span> </div>	
AtTST2	AKRVLCRLRGREDVSGEMALLVEGLGIGGGTTTIEEYITCPADEVTDHDIADVTKDQIKLYGAEGLSWVARPVKGGSTMSVLSRGGSTMSRRQGSLLDPL	300
BvTST2.1	AKRVLCRLRGREDVFAEMALLVEGLRVGGTSTIEEYITCPADLAEDDPMYKDVRLYGSBAGCSWVARPVVGGSMGLIASRQGS.MQSPSVPLMDPL	299
CmTST2	AKRVLCRLRGREDVSGEMALLVEGLVGGGTSLEEFITCPADDLP.DODLLTKDDEIKLYGPEGLSWVARPVVGGSSIGLVSRRGGSIINQSG..LVDPL	297
AtTST2	VTLFGSVHEKLPEDTGSMSRALPFFFGSMFVSGGNQPRHEFDENLVGEGEDVPSLHGD...DSEDDLHSLPLISRQTTSMKDMPHTAHCTLSTFRHG	395
BvTST2.1	VTLFGSVHEKLPEDCGSMLSVIFPFFFGSMFVSGGKPKNEFDWDENTIGDDDDYGHDDDEYAGDAEDDNLRSLSISRQDTGPKRMVAPTSGSMFMSKHS	399
CmTST2	VTLFGSVHEKLPEDTGSMSRSLPFFFGSMFVSGGNQHRNEFDWDESLAREGEDYQSDGAG...NDSDDNLRSLSISRQTTSMKDMVAPAHCSLSSMRQG	393
AtTST2	SQVQGAQEGAGSGMIGGGWQAWKWREREDESQKEGGFKRYYLHQEGFFGSRRSIVSLPGDGTGEADFVQASALVSQPALYSKDLLKEHTIGPAMV	495
BvTST2.1	SWLQSE...ASGIGGGWQAWKWSERGLDQKEGGFKRYYLHQEGDAGSKRGSVLSLAGEVIGDNEYVRAALVSQPALYSRDFMDRDSIGPAMV	494
CmTST2	SLAGEPVG...ASGIGGGWQAWKWREREPDGNKEGGFKRYYLHQEGTSGPQQGSIVSLPGDALTGGYIQAAALVSQPALYSKELMNQHPVGPAMV	489
AtTST2	HPSET.TKGSINHHDDHPGVKRALVGVGLQTLQQFSGINGVLYYTPQILECAGVGLLSNMGISGSSASLLISALTFVMLPATAVAMRLMDLSCRRTL	594
BvTST2.1	HPSEASAKRPSWRDFLEPGVRRALVGVGLQTLQQFSGINGVLYYTPQILECAGVGLLSHMGIGASSALLISALTTLLMLPATAVAMRLMDLSCRRTL	594
CmTST2	HPESI.TKGPSVDFDFPFGVKHALVGVGLQTLQQFSGINGVLYYTPQILEKAGVGLLSNLGIGSSASLLISGLTLLMLPSTAVAMRLMDLSCRRTL	588
	<div style="display: flex; justify-content: space-around; width: 100%;"> <span>7</span> <span>8</span> <span>9</span> </div>	
AtTST2	LLTFIPHLIASLLVLSNVLVMSNIVHAWLSTVSVVLYFCFFVMFGFPPNILCAEIFFPTRVRGICIAICALTFWICDIIVTYSLFVLLKSLGLAGVFG	694
BvTST2.1	LLTFIPVLELSLVVLIILANVIRKNTIYAVVSTVAVVLYFCFFVMFGFPPNILCAEIFFPTRVRGICIAICALTFWICDIIVTYLFPMMKAVGLAGVFG	694
CmTST2	LLTFIPALIASLIHLVIGSLVQMSIWNASISTVSVVLYFCFFVMFGFPPNILCAEIFFPTRVRGICIAICALTFWICDIIVTYLFLVLLNSIQLGGVFG	688
	<div style="display: flex; justify-content: space-around; width: 100%;"> <span>9</span> <span>10</span> <span>11</span> <span>12</span> </div>	
AtTST2	MYAVVCIISWVEVFLKVPETKGMPLVITTEFFSVGARKQAAAKNE	739
BvTST2.1	FYAVVILISWVEVFLKVPETKGMPLVITTEFFALGARQASH	735
CmTST2	MYAVVCIISWVEVFLKVPETKGMPLVITTEFFSVGARKLLAARKNG	733

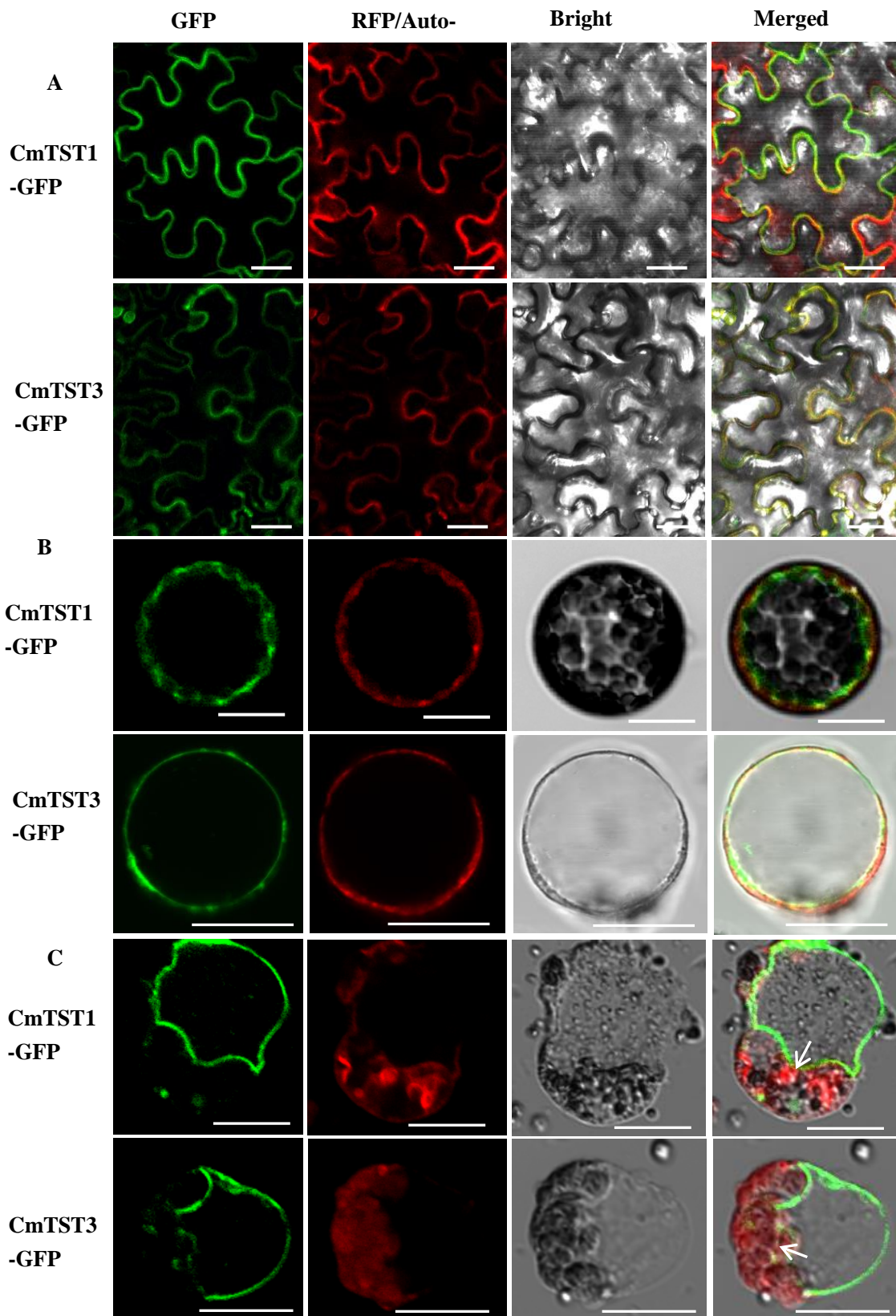
**Supplementary Fig. S1.** Comparison of amino acid sequence of CmTST proteins and previously identified tonoplast localized sugar transporter AtTST2 (Arabidopsis) and BvTST2.1 (sugar beet). Black boxes indicate identical amino acid residues. Dark gray boxes represent amino acid residues with an identity of more than 50%. The 12 putative trans-membrane domains of CmTST2 are underlined. The sequence alignment was performed using the DNAMAN software package.



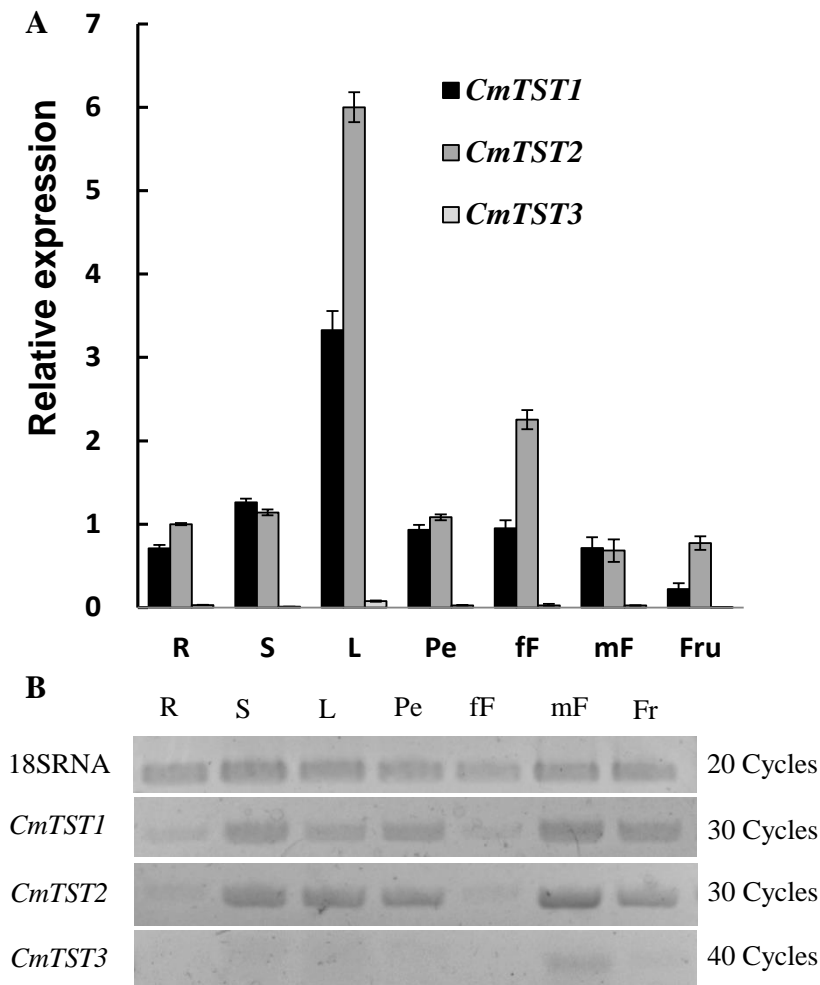
**Supplementary Fig. S2.** Membrane-spanning model of CmTSTs. The predicted website was TMHMM <http://genome.cbs.dtu.dk/services/TMHMM-2.0>.



**Supplementary Fig. S3.** The expression of *CmTST2* in different melon genotypes with different sugar accumulation. (A) The fruit appearances of the three melon varieties, Yilisabai (Yi), Xiaomaisu (Xiao), and Yangjiaosu (Yang). (B) Relative expression of *CmTST2* in the melon varieties Yi, Xiao and Yang during different developmental stages were determined by qRT-PCR. *TUA* was used as reference gene. (C) Different types of sugar content in the three melon varieties' fruits during different developmental stages were determined by GC-MS. 'Sugar' means the total of glucose, fructose and sucrose. Error bars represent the SE for three technical replicates of three biological replicates.



**Supplementary Fig. S4.** Subcellular localization of CmTST1/3-GFP fusion protein in tobacco. (A, B) Colocalization of CmTST1/3-GFP and AtRtip-RFP (tonoplast localization marker) in tobacco epidermal cell (A) or protoplast (B). (C) Tonoplast localization of CmTST1/3-GFP in tobacco protoplast. The laser-scanning confocal microscopy images show fluorescence and merged images. The chlorophyll auto fluorescence (Auto-) in (C) and the bright field images are also presented. CmTST1/3-GFP fusion protein is clearly inserted into the tonoplast, which separates the vacuolar volume from the cytoplasm containing chloroplasts (white arrows). Bars=20  $\mu$ m.



**Supplementary Fig. S5.** Spatial expression analysis of *CmTST* genes in various melon organs by qRT-PCR (A) and RT-PCR (B). R, root; S, stem; L, leaf; Pe, petiole; fF, female flower; mF, male flower; Fr, fruit (20 DAA). Error bars represent the SE for the three technical replicates of the three biological replicates.

**Supplementary Table S1.** Primers used in the paper (Lowercases indicate the restriction enzyme sites or recombination connector sequence).

<i>primers</i>	<i>Description</i>	<i>Sequence (5' - 3')</i>
TST1FLF	ORF clone	ATGAAGGGAGCTTTGCTAGTGGC
TST1FLR	ORF clone	TCAATTAATACTACCTTTTCGCAGCT
TST2FLF	ORF clone	ATGAGTGGTTCTGTTTTGGTGGCTG
TST2FLR	ORF clone	TCAACCATTTTTAGCAGCTAAAAGCTGTTTGG
TST3FLF	ORF clone	ATGAGGGGAGCTGTTCTCGTTGCAATCGC
TST3FLR	ORF clone	TTAATCAGCATTATCAGCCTGTTTCGC
TubulinF	Quantitative RT-PCR	GCGGTGCTTCTAGACAATGA
TubulinR	Quantitative RT-PCR	CCTGAGATACAAGACGGTTGAG
TST1qF	Quantitative RT-PCR	ATAATAGGTCCTGCTGAAGA
TST1qR	Quantitative RT-PCR	AAAGGGTGACAAGTGGGT
TST2qF	Quantitative RT-PCR	GGATGGGATAATGCGACTA
TST2qR	Quantitative RT-PCR	GCTCCGATAAGGGATGTG
TST3qF	Quantitative RT-PCR	GGTCGTCGTCCTCTAATG
TST3qR	Quantitative RT-PCR	ACCAAGGTAACCGCAAGT
Cm18SF	Semi RT-PCR	CGAGTCTGGTAATTGGAATGAGTA
Cm18SR	Semi RT-PCR	CTACGAGCTTTTTAACTGCAACAA
TST2F	Semi RT-PCR	AAGGATGGGATAATGCGACTA
TST2R	Semi RT-PCR	TGACAAGCCAGCGAGGTGA
TST1F	Semi RT-PCR	GCATCAGAGGCTGGTGGAG
TST1R	Semi RT-PCR	TAGCACAGGGATGGTAGCG
TST3F	Semi RT-PCR	GTCAGCCAATCAGCACTCC
TST3R	Semi RT-PCR	AGGCACCAAAGACTCCACC
TST1GFPF	Fusion to GFP	attacgccgaggtcATGAAGGGAGCTTTGCTAGTGGC
TST1GFPR	Fusion to GFP	tagggaagaggTCAATTAATACTACCTTTTCGCAGCT
TST2GFPF	Fusion to GFP	attacgccgaggtcATGAGTGGTTCTGTTTTGGTGGCTG
TST2GFPR	Fusion to GFP	tagggaagaggTCAACCATTTTTAGCAGCTAAAAGCTGTTTGG
TST3GFPF	Fusion to GFP	attacgccgaggtcATGAGGGGAGCTGTTCTCGTTGCAATCGC
TST3GFPR	Fusion to GFP	tagggaagaggTTAATCAGCATTATCAGCCTGTTTCGC
TST1PF	Promoter clone	gaccatgattacccaagcttGTCATATACTTACTTTAGCTCAACCTC
TST1PR	Promoter clone	ggactgaccaccgggatccTTATTCAATTCAAGCTCCAATGTGGTCAG
TST2PF	Promoter clone	gaccatgattacccaagcttCAAAGTCGTTTAGATTTAGTTACCC
TST2PR	Promoter clone	ggactgaccaccgggatccTTTGCTTCTTCAAATCGACAGGAAA
TST3PF	Promoter clone	gaccatgattacccaagcttCTTATTGTGCATCACAGCATAGAGG
TST3PR	Promoter clone	ggactgaccaccgggatccCTTACCAATGGGGTGAATTAGAATATC
TST2-121F	Clone in to PBI121	tgetctagaATGAGTGGTTCTGTTTTGGTGGCTG
TST2-121R	Clone in to PBI121	tccccggGTCAACCATTTTTAGCAGCTAAAAGCTGTTTGG

**Supplementary Table S2.** Potential cis-acting regulatory elements identified in promoter regions of *CmTST* genes.

Gene name	Tissue/organ specific relative elements	Sugar / hormone responsive elements	Abiotic stress-responsive elements
<i>CmTST1</i>	<b>CAATBOX1</b> (CAAT, tissue-specific), <b>ROOTMOTIFTAPOX1</b> (ATATT, organ-specific), <b>RAV1AAT</b> (CAACA, high express in rosette leaves and roots), <b>OSE1ROOTNODULE</b> (AAAGAT, organ-specific) <b>OSE2ROOTNODULE</b> (CTCTT, organ-specific)	<b>WBOXHVISO1</b> (TGACT, sugar-responsive), <b>SREATMSD</b> (TTATCC, sugar-responsive), <b>PYRIMIDINEBOXOSRAMY1A</b> (CCTTTT, gibberellin-responsive), <b>P-box</b> (CCTTTTG, gibberellin-responsive) <b>ARFAT</b> (ACTTTA, auxin-responsive), <b>ERE</b> (ATTTCAA, ethylene-responsive)	<b>ELRECOREPCR1</b> (TTGACC, pathogen- and wound-induced), <b>TC-rich</b> (ATTTTCTTCA, defense and stress responsiveness)
<i>CmTST2</i>	<b>CAATBOX1</b> (CAAT, tissue-specific), <b>ROOTMOTIFTAPOX1</b> (ATATT, organ-specific), <b>RAV1AAT</b> (CAACA, high express in rosette leaves and roots), <b>RAV1BAT</b> (CACCTG, high express in rosette leaves and roots), <b>OSE2ROOTNODULE</b> (CTCTT, organ-specific), <b>RHERPATEXPA7</b> (KCACGW, root hair-specific)	<b>WBOXHVISO1</b> (TGACT, sugar-responsive), <b>PYRIMIDINEBOXOSRAMY1A</b> (CCTTTT, gibberellin-responsive), <b>GARE</b> (AAACAGA, gibberellin-responsive) <b>ARFAT</b> (ACTTTA, auxin-responsive), <b>DPBFCOREDCDC3</b> (ACACNNG, ABA-responsive), <b>TCA-element</b> (CAGAAAAGGA, salicylic acid-responsive),	<b>TC-rich</b> (ATTTTCTTCA, defense and stress responsiveness)
<i>CmTST3</i>	<b>POLLENILELAT52</b> (AGAAA, pollen-specific motif) <b>CAATBOX1</b> (CAAT, tissue-specific), <b>ROOTMOTIFTAPOX1</b> (ATATT, organ-specific)	<b>SREATMSD</b> (TTATCC, sugar-responsive), <b>PYRIMIDINEBOXOSRAMY1A</b> (CCTTTT, gibberellin-responsive), <b>GARE</b> (AAACAGA, gibberellin-responsive) <b>ERE</b> (ATTTCAA, ethylene-responsive),	<b>TC-rich</b> (ATTTTCTTCA, defense and stress responsiveness)