

Figure S1. Exon/intron structure of Arabidopsis secondary cell wall CESA genes. Exons are shown as boxes (white boxes for UTRs and black boxes for CDS regions) while the introns are shown as lines. Positions of the T-DNA insertion in the mutants used in the study are indicated by arrowhead.

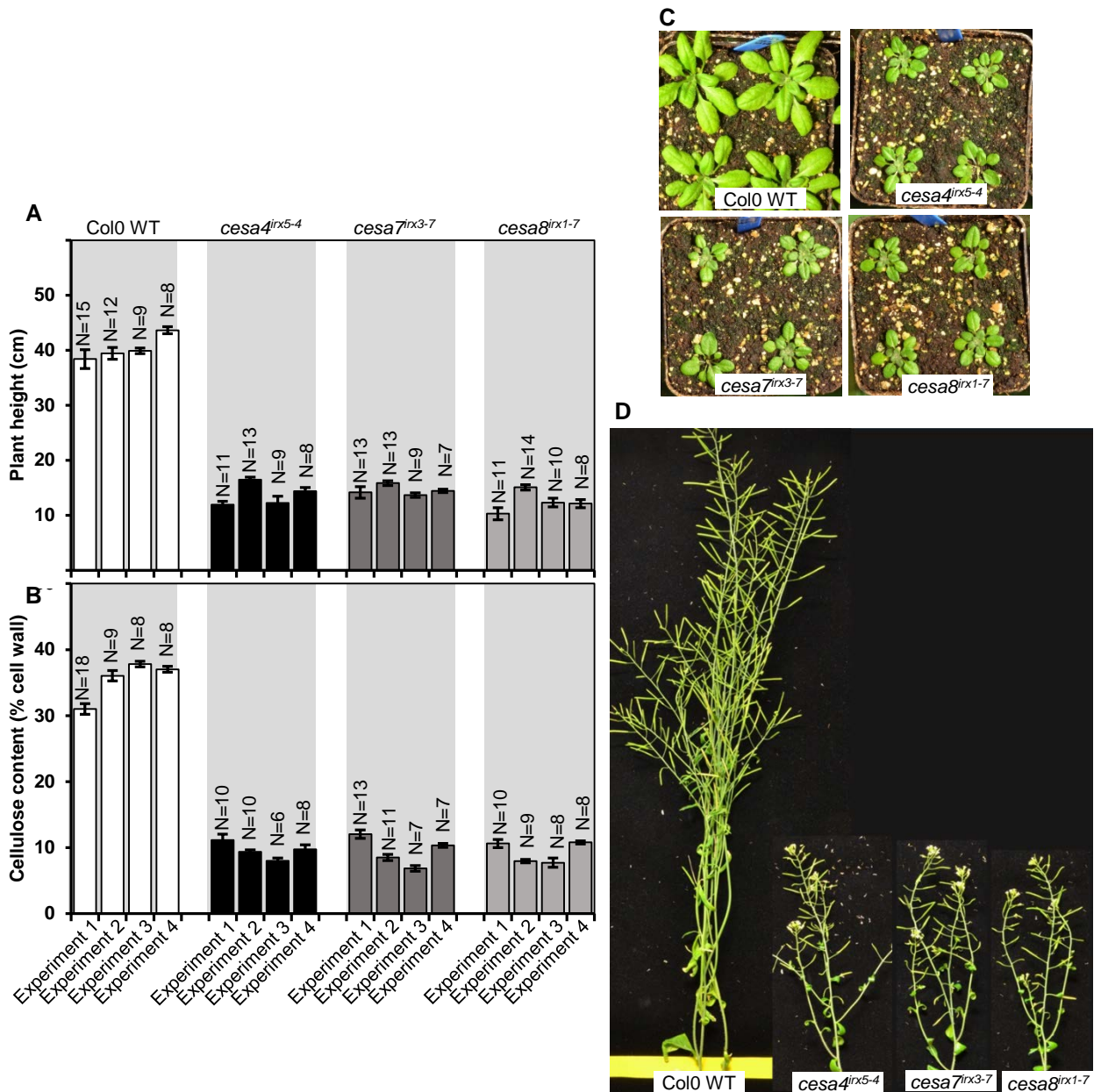


Figure S2. Phenotypes of the *cesa* mutants and wild type control.

- A. Plant height
- B. Cellulose content
- C. Rosette appearance
- D. Inflorescence stem.

Error bars shown in (A) and (B), are SEM. Rosette photos (C) were taken when the plants were 21 days old while the photos of primary inflorescence stem severed at rosette level (D) were taken at 7 week stage.

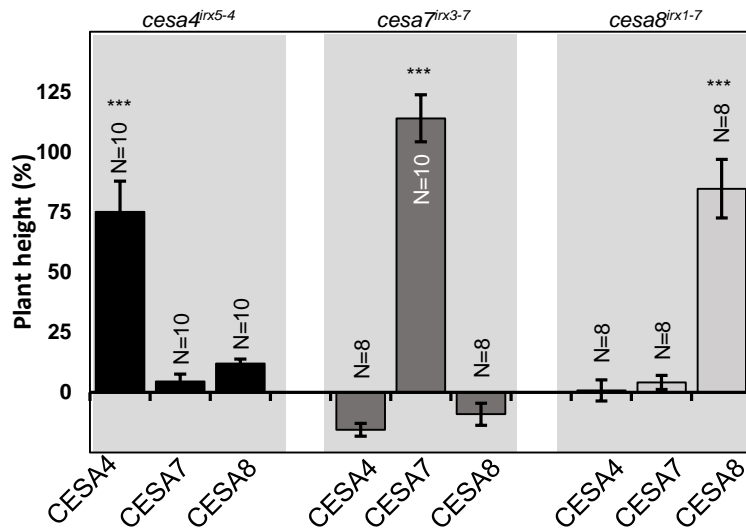


Figure S3. Height of different *cesa* mutant plants transformed with WT CESA genes. Error bars are SEM. Significance levels from univariate ANOVA between the genotype and the mutant background are shown; *** Significant at 0.001.

Promoter	Gene	Mutant	Plant height (%)										Cellulose content (%)									
pCESA7	CESA4	<i>cesa4^{ix5-4}</i>	179	121	112	108	94	90	72	68	45	0	108	35	27	16	7	NA	91	11	2	61
	CESA7	<i>cesa7^{ix3-7}</i>	208	178	149	149	139	139	129	104	94	89	NA	118	110	89	145	95	82	97	84	108
	CESA8	<i>cesa8^{ix1-7}</i>	182	174	103	99	95	95	66	61	61	57	82	128	81	57	50	78	17	57	61	52
pCESA4	CESA4	<i>cesa4^{ix5-4}</i>	106	96	96	72	69	54	19				98	55	85	92	57	84	78			
	CESA8	<i>cesa8^{ix1-7}</i>	104	82	75	71	71	64	53	28			81	64	57	54	54	57	49	58		
pCESA8	CESA4	<i>cesa4^{ix5-4}</i>	93	93	43	21							51	65	59	28						
	CESA8	<i>cesa8^{ix1-7}</i>	88	81	70	70	63	60	35				96	82	81	89	54	72	53			

Figure S4. Complementation of secondary cell wall *cesa* mutants using different promoters. CESA4 and 8 were expressed under control of all three SCW CESA promoters. Plant height (%) and cellulose content (%) data is shown for up to 10 independent lines. Plant height (%) = (Plant height of the line – plant height of the background mutant)/(Plant height of Col0 WT– plant height of the background mutant) *100. Data is colour coded and signifies the % complementation: dark green: >60%, green: 40-60%, light green: 12.5-40%, white: <12.5%.

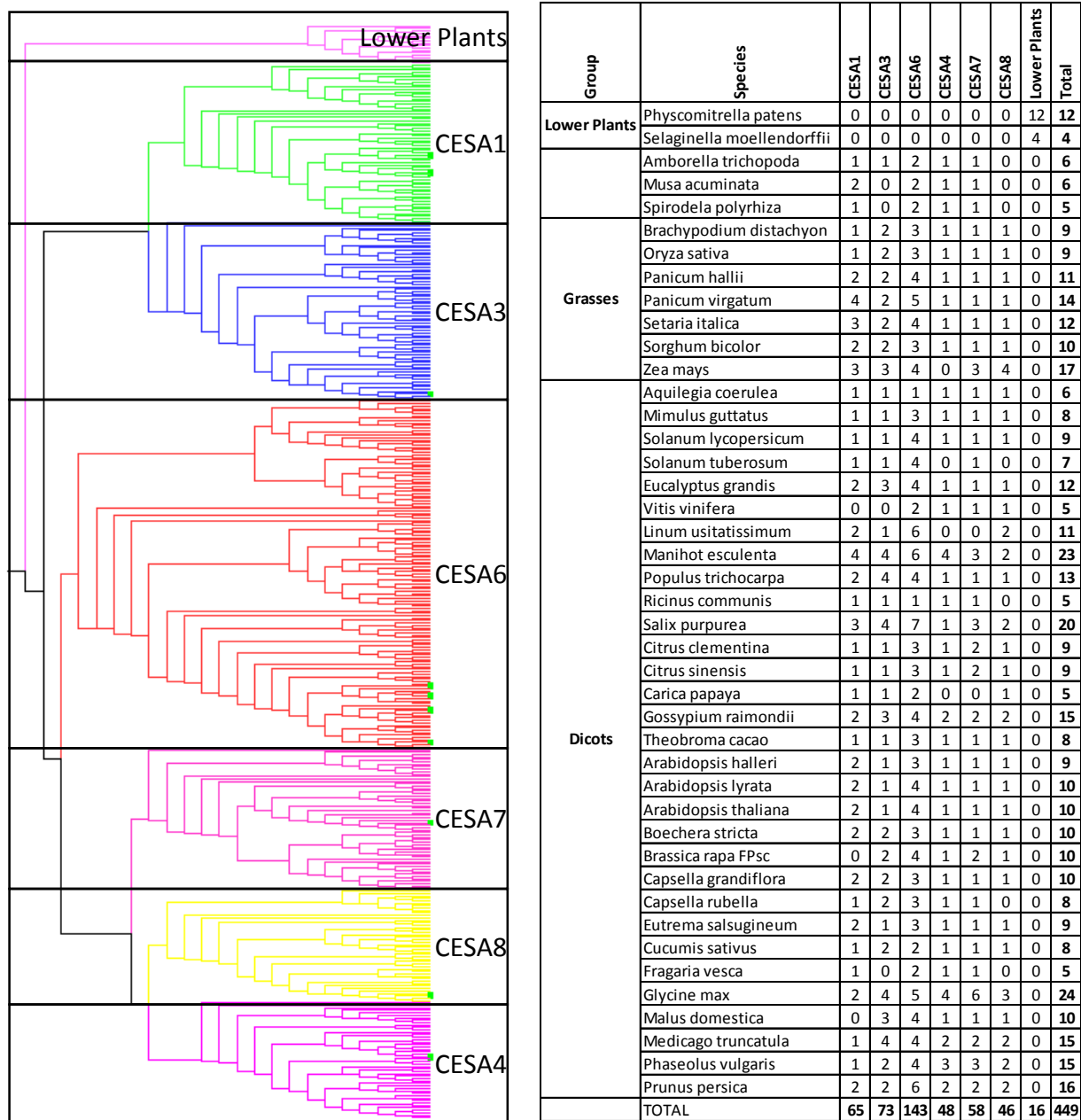


Figure S5. Classification of CESA proteins into 6 classes.

The trees is based upon 449 sequences from Phytozome that have been manually curated. The clades were named based on the *Arabidopsis* members which are indicated with green dots in the tree above. The table on the right lists the number of proteins in each CESA class for all the species used in the analysis.

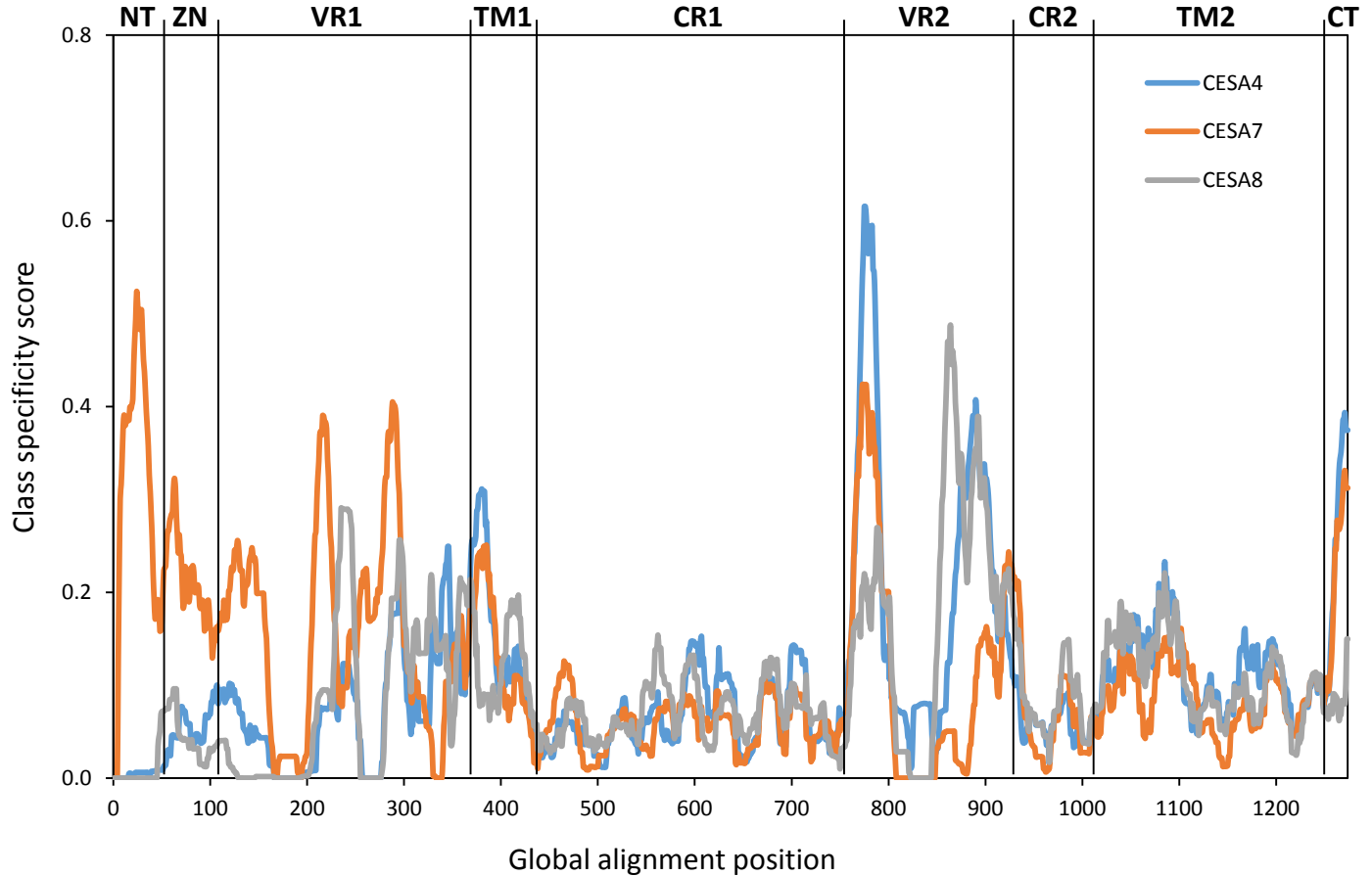


Figure S6. Analysis of CESA protein class specificity.

A global alignment of 449 CESA proteins from 43 species was produced as shown in Figure S5. Class specificity scores were calculated at each amino acid position in the global alignment exactly as described in Carroll and Specht (2011). The scores for the 3 SCW CESA classes, CESA4, 7 and 8 were plotted. The curves were smoothed by taking average scores for a sliding window of 10 amino acids.

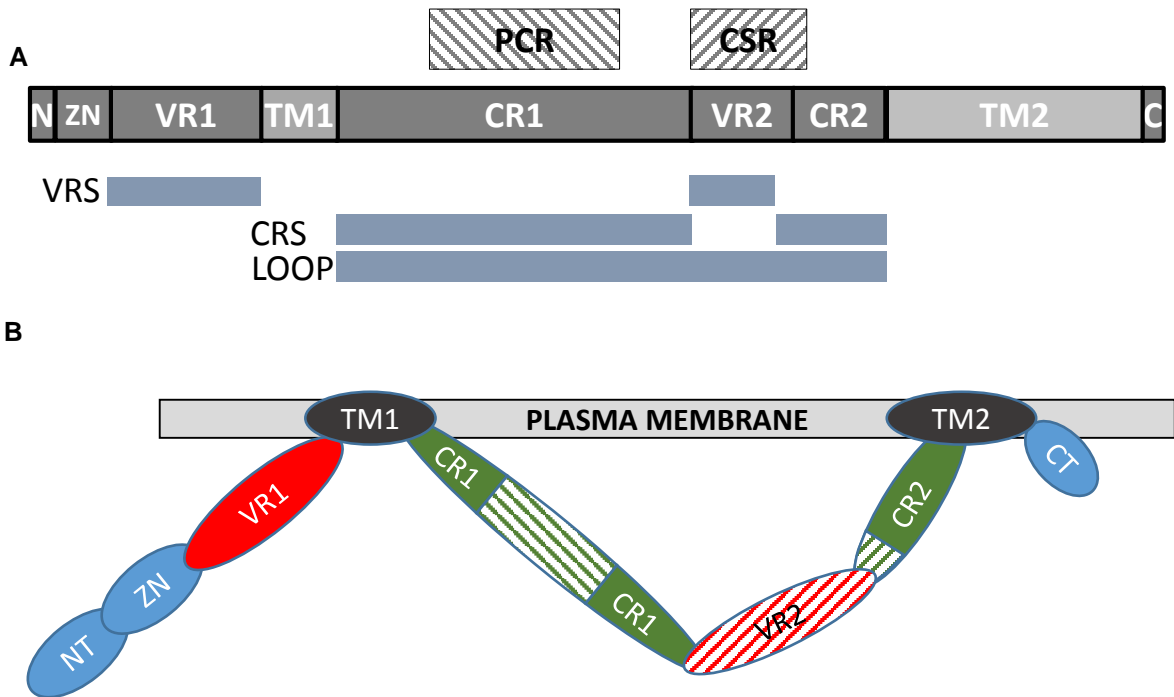


Figure S7. Schematic representation of CESA proteins showing the location of the regions used in this study.

- A. Linear representation of a CESA proteins with the 9 regions used in this study shown as boxes with grey fill and black lines. The name of each region is indicated inside each box. The length of each box is proportional to actual region size in amino acids. Position of the PCR and CSR regions described in Veragara and Carpita (2001) is shown above it. A linear representation of the regions involved in the multi-region swap experiments- VRS, CRS and LOOP, are indicated below.
- B. A cartoon representation of the 9 regions and their predicted topology.

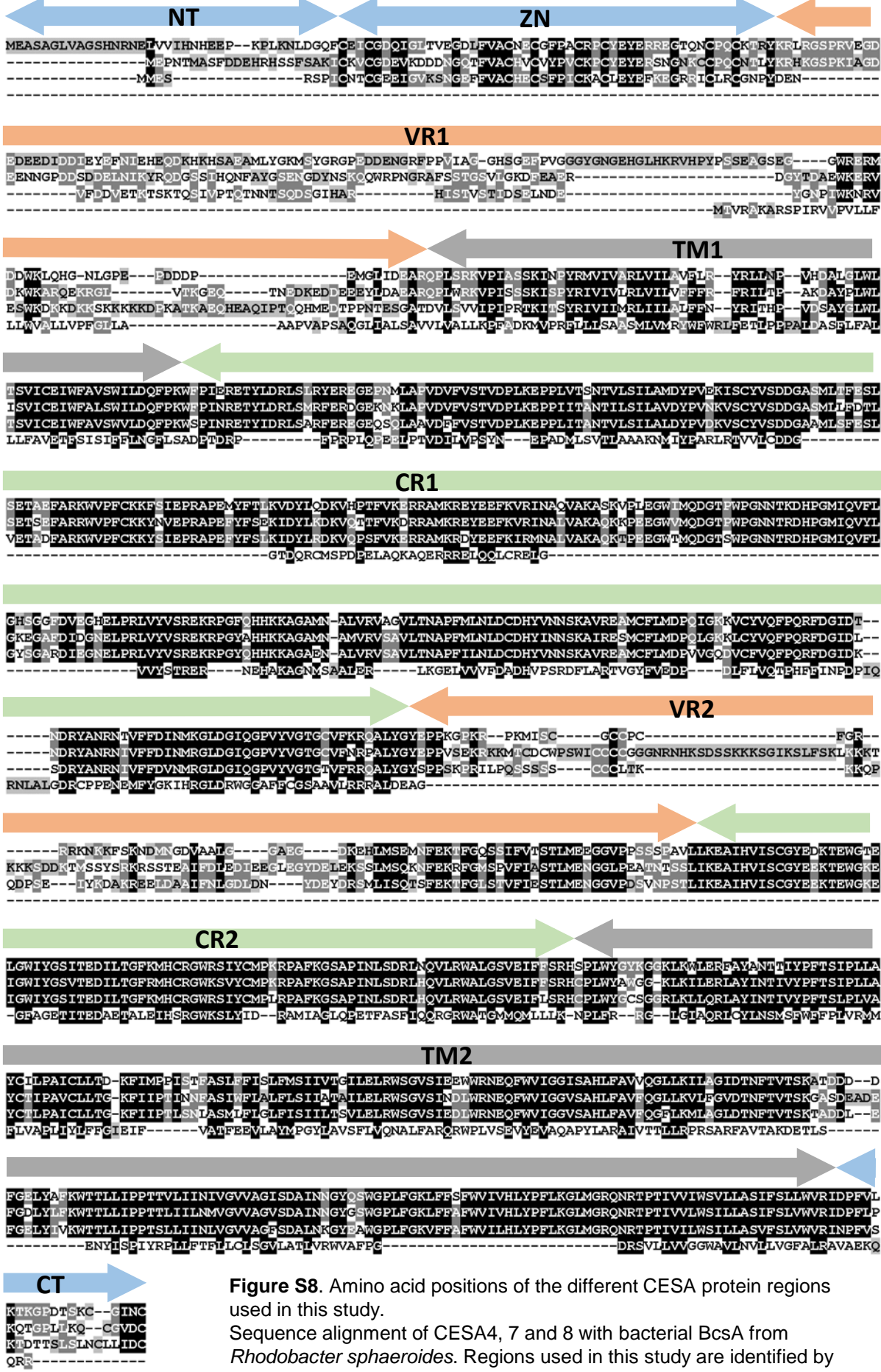
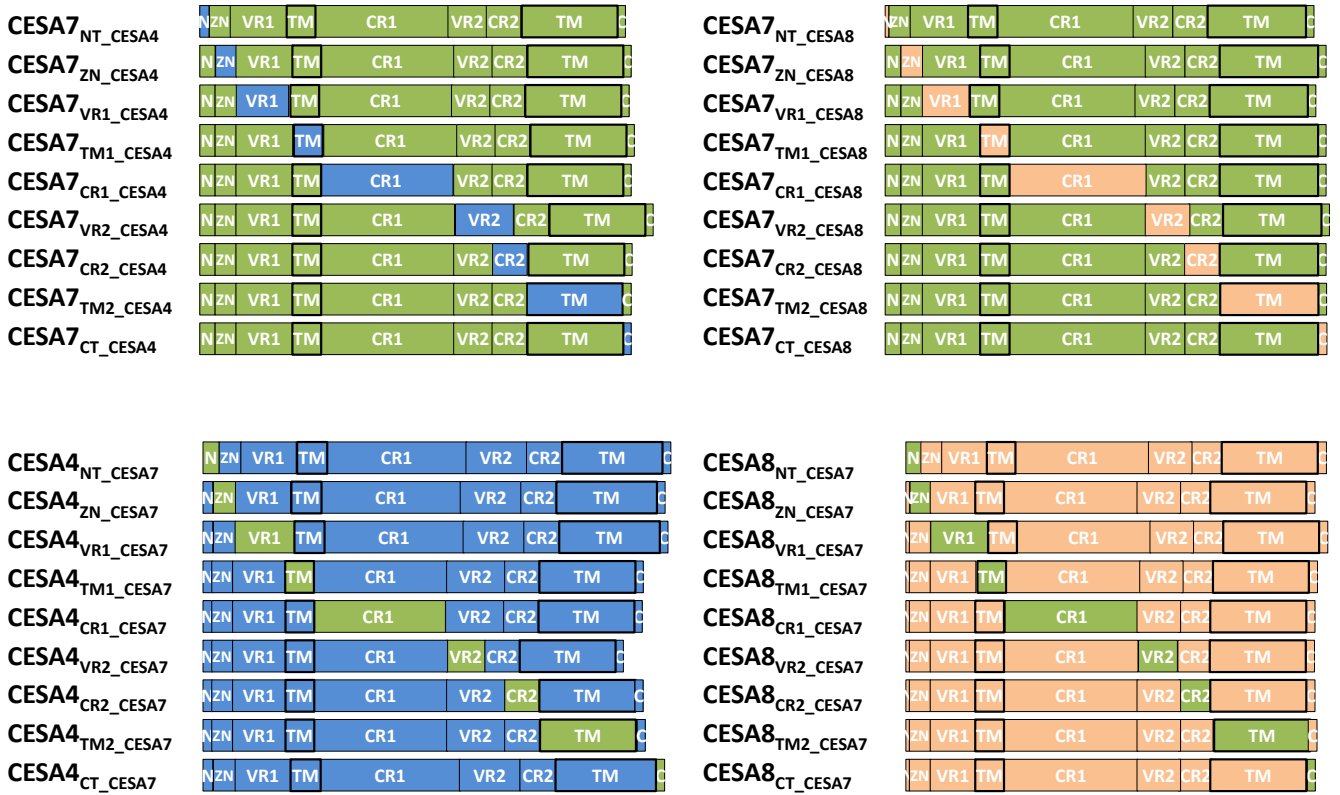
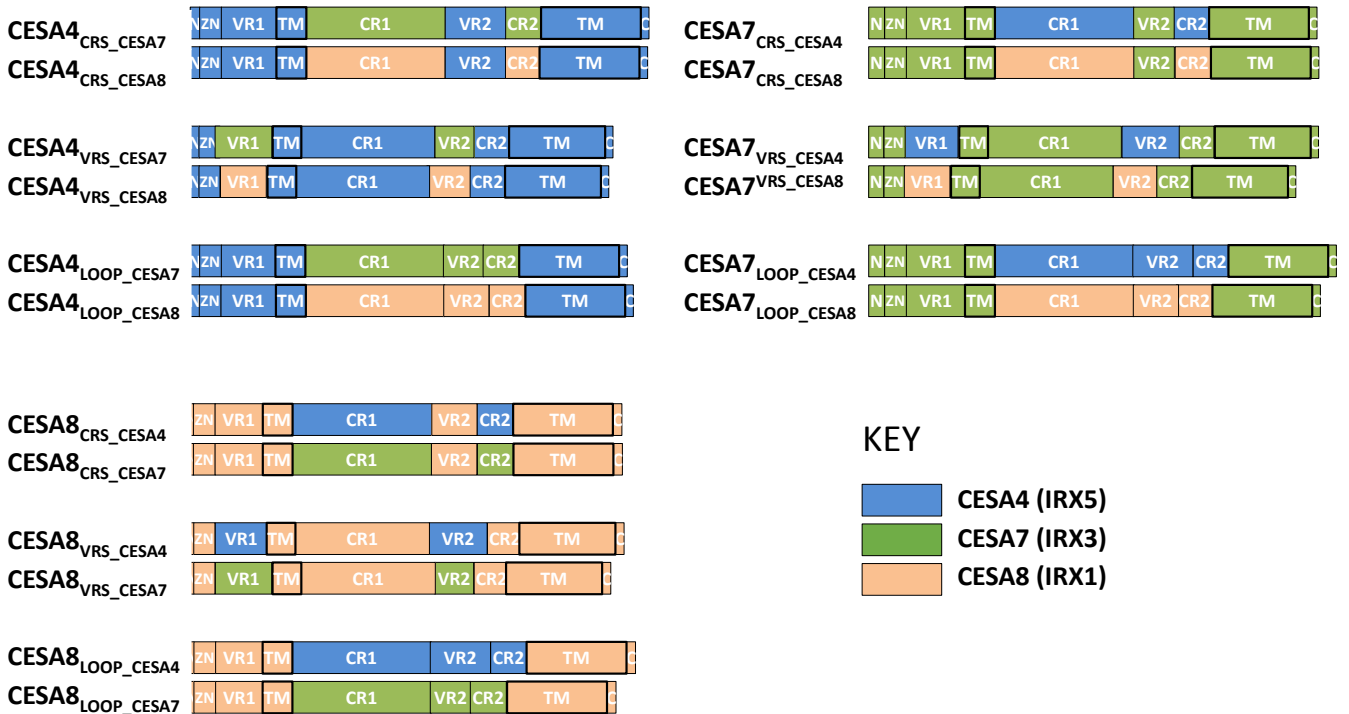


Figure S8. Amino acid positions of the different CESA protein regions used in this study. Sequence alignment of CESA4, 7 and 8 with bacterial BcsA from *Rhodobacter sphaeroides*. Regions used in this study are identified by the double headed arrows above the alignment.

Single region swaps



Multi-region swaps



KEY

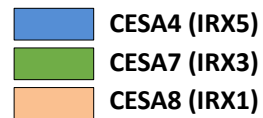


Figure S9. Schematic representation of the swap constructs used in this study. All region lengths are proportional to their respective amino acid size.

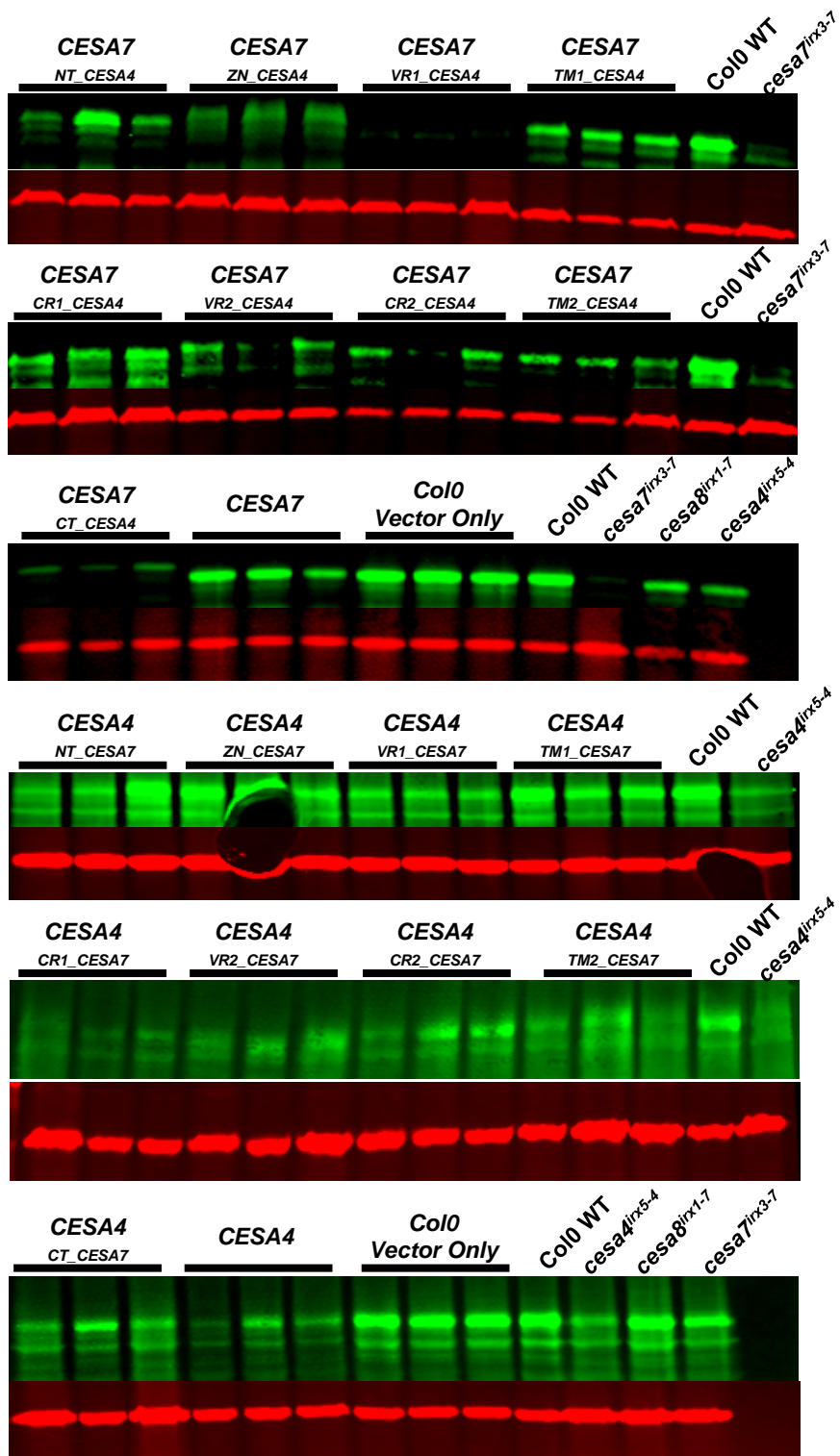


Figure S10. Analysis of protein expression in the swap constructs used in this study. Level of expression was measured for CESA7 (A) or CESA4 (B) using quantitative western blot analysis. Crudes extracts prepared from 3 independent lines for each CESA7_{CESA4} construct and probed with anti-CESA7 antibody while the crude extracts prepared from three independent lines for each CESA4_{CESA7} construct were probed with anti-CESA4 antibody (green bands). All extracts were also simultaneously probed with anti-HSP73 antibody to normalise for loading variations (red bands). HSP73 normalised signals were then expressed as percentage of Col0 WT. Protein expression (%WT) was plotted against cellulose content (%WT) to produce Figure 7.

Region	AtCesA04		AtCesA07		AtCesA08	
	coordinates	length	coordinates	length	coordinates	length
NT	aa 1-22	22	aa 1-36	36	aa 1-8	8
Zn	aa 23-72	50	aa 37-86	50	aa 9-58	50
VR1	aa 73-197	125	aa 87-220	134	aa 59-166	108
TM1-2	aa 198-265	68	aa 221-288	68	aa 167-234	68
CR1	aa 266-583	318	aa 289-606	318	aa 235-552	318
VR2	aa 584-720	137	aa 607-698	92	aa 553-655	103
CR2	aa 721-802	82	aa 699-780	82	aa 656-737	82
TM3-8	aa 803-1030	228	aa 781-1007	227	aa 738-964	227
CT	aa 1031-1049	19	aa 1008-1026	19	aa 965-985	21
CRS	CR1+CR2	400	CR1+CR2	400	CR1+CR2	400
VRS	VR1+VR2	262	VR1+VR2	226	VR1+VR2	211
LOOP	CR1+VR2+CR2	537	CR1+VR2+CR2	492	CR1+VR2+CR2	503
TOTAL	1049		1026		985	

Table S1. Protein coordinates for regions of AtCESA4, 7 and 8 used in the swap constructs.

Swap Name	Primer BF	Primer CF
CESA7 ^{NT} _CESA4	gtttccagtcacgacgttgtaaacgacggccag	tcattctcggctaagatttgtgagatatgtggagat
CESA7 ^{ZN} _CESA4	aatctagatggacaattctgcaaagtctgtggcgat	caatgcaacactctttacaagcgtctcagaggaagc
CESA7 ^{VR1} _CESA4	cagtgaagactcgttacaaacgccacaaaggctct	ctagatgctgaagcaagacagccactctcgcggaaa
CESA7 ^{TM1} _CESA4	ctgatcgacgaggcacggcaacctctctggagaaaa	cttgatcagttcccgaatggttccctattgaacgt
CESA7 ^{CR1} _CESA4	cttgatcagttcccgaatggttccgattaaccga	agaccagcattatacGGAtatgaaccaccaaagggt
CESA7 ^{VR2} _CESA4	cgacaagctctgtatGGAtacgaacctcggatcgc	tcggtgatcaaagaagctatccatgtcataagctgc
CESA7 ^{CR2} _CESA4	gtgctcctaaagaggcaattcatgttattagctgt	atcttcttagtcgcatagtcctctctggtatggc
CESA7 ^{TM3} _CESA4	atattttcagccggcactgtcctttggtatgct	ttggtttgggtcggattgatcctttgtgctcaag
CESA7 ^{CT} _CESA4	ttgctttgggtaagaattgatccgttcttgccgaaa	-
CESA7 ^{NT} _CESA8	gtttccagtcacgacgttgtaaacgacggccag	gagtctaggctcccatctgtgagatatgtggagat
CESA7 ^{ZN} _CESA8	aatctagatggacaattctgcaacacttgggtgaa	cgttgaggcaatccttacaagcgtctcagaggaagc
CESA7 ^{VR1} _CESA8	cagtgaagactcgttacgatgagaatgtgttgat	acagaatctgggtctacacagccactctcgcggaaa
CESA7 ^{TM1} _CESA8	ctgatcgacgaggcacgggatgtgcttccggttg	ttgatcagttccctaatggttccctattgaacgt
CESA7 ^{CR1} _CESA8	cttgatcagttcccgaatgggtctctattaaccga	agacaagcactttacggatatgaaccaccaaagggt
CESA7 ^{VR2} _CESA8	cgacaagctctgtatGGTtacagtcaccttcaaaa	acgctcatcaaagaagctatccatgtcataagctgc
CESA7 ^{CR2} _CESA8	gtgctcctaaagaggcaattcatgtcattagctgt	atcttccttagccgacatagtcctctctggtatggc
CESA7 ^{TM3} _CESA8	atattttcagccggcactgtcctttggtacggt	cttgtttgggtcgtatcgcctttgtgctcaag
CESA7 ^{CT} _CESA8	ttgctttgggtaagaattaatcctttcgtctcaaaa	-
CESA4 ^{NT} _CESA7	gtttccagtcacgacgttgtaaacgacggccag	aatctagatggacaattctgcaaagtctgtggcgat
CESA4 ^{ZN} _CESA7	tcattctcggctaagatttgtgagatatgtggagat	cagtgaagactcgttacaaacgccacaaaggctct
CESA4 ^{VR1} _CESA7	caatgcaacactctttacaagcgtctcagaggaagc	ctgatcgacgaggcacggcaacctctctggagaaaa
CESA4 ^{TM1} _CESA7	ctagatgctgaagcaagacagccactctcgcggaaa	cttgatcagttcccgaatggttccgattaaccga
CESA4 ^{CR1} _CESA7	cttgatcagttcccgaatggttccctattgaacgt	cgacaagctctgtatGGAtacgaacctcggatcgc
CESA4 ^{VR2} _CESA7	agaccagcattatacGGAtatgaaccaccaaagggt	gtgctcctaaagaggcaattcatgttattagctgt
CESA4 ^{CR2} _CESA7	tcggtgatcaaagaagctatccatgtcataagctgc	atattttcagccggcactgtcctttggtatgct
CESA4 ^{TM3} _CESA7	atcttcttagtcgcatagtcctctctggtatggc	ttgctttgggtaagaattgatccgttcttgccgaaa
CESA4 ^{CT} _CESA7	ttggtttgggtcggattgatcctttgtgctcaag	-
CESA8 ^{NT} _CESA7	gtttccagtcacgacgttgtaaacgacggccag	aatctagatggacaattctgcaacacttgggtgaa
CESA8 ^{ZN} _CESA7	gagtctaggctcccatctgtgagatatgtggagat	cagtgaagactcgttacgatgagaatgtgttgat
CESA8 ^{VR1} _CESA7	cgttgaggcaatccttacaagcgtctcagaggaagc	ctgatcgacgaggcacgggatgtgcttccggttg
CESA8 ^{TM1} _CESA7	acagaatctgggtctacacagccactctcgcggaaa	cttgatcagttcccgaatgggtctctattaaccga
CESA8 ^{CR1} _CESA7	ttgatcagttccctaatggttccctattgaacgt	cgacaagctctgtatGGTtacagtcaccttcaaaa
CESA8 ^{VR2} _CESA7	agacaagcactttacggatatgaaccaccaaagggt	gtgctcctaaagaggcaattcatgtcattagctgt
CESA8 ^{CR2} _CESA7	acgctcatcaaagaagctatccatgtcataagctgc	atattttcagccggcactgtcctttggtacggt
CESA8 ^{TM3} _CESA7	atcttccttagccgacatagtcctctctggtatggc	ttgctttgggtaagaattaatcctttcgtctcaaaa

CESA8 ^{CT_CESA7}	cttgttgggttcgatcgcacctttgtgctcaag	-
CESA4 ^{CRS_CESA7}	cgacaagctctgtatGGAtacgaacctccggtatcg	tcaactttgatggaagaaggaggattgccggaggcg
CESA4 ^{CRS_CESA8}	agacaagcactttacggatacgaacctccggtatcg	tctactcttatggagaatggaggattgccggaggcg
CESA4 ^{VRS_CESA7}	agaccagcattatacGGAtatgaaccaccaaagggt	tcaactttgatggaagaaggaggattgccggaggcg
CESA4 ^{VRS_CESA8}	agaccagcattatacGGAtacagtcaccttcaaaa	tctactcttatggagaatggaggattgccggaggcg
CESA4 ^{LOOP_CESA7}	cttgatcagttcccgaatggttcctattgaacgt	atattttcagccggcactgtcctttgtggtatgct
CESA4 ^{LOOP_CESA8}	cttgatcagttcccgaatggttcctattaaccga	atcttccttagccgacattgtcctttgtggtatgct
CESA7 ^{CRS_CESA4}	agaccagcattatacGGAtatgaaccaccaaagggt	tgcagcttgatggagaacgggtggttctcctcgta
CESA7 ^{CRS_CESA8}	agacaagcactttacggatatgaaccaccaaagggt	tctactcttatggagaatggtggttctcctcgta
CESA7 ^{VRS_CESA4}	cgacaagctctgtatGGAtacgaacctccggtatcg	tgcagcttgatggagaacgggtggttctcctcgta
CESA7 ^{VRS_CESA8}	cgacaagctctgtatGGTtacagtcaccttcaaaa	tctactcttatggagaatggtggttctcctcgta
CESA7 ^{LOOP_CESA4}	cttgatcagttcccgaatggttcctattaaccga	atcttcttagtctcatagtcctctctggtatggc
CESA7 ^{LOOP_CESA8}	cttgatcagttcccgaatggttcctattaaccga	atcttccttagccgacatagtcctctctggtatggc
CESA8 ^{CRS_CESA4}	agaccagcattatacGGAtacagtcaccttcaaaa	tgcagcttgatggagaacggcggttcccgactct
CESA8 ^{CRS_CESA7}	cgacaagctctgtatGGTtacagtcaccttcaaaa	tcaactttgatggaagaaggcggttcccgactct
CESA8 ^{VRS_CESA4}	agacaagcactttacggatacgaacctccggtatcg	tgcagcttgatggagaacggcggttcccgactct
CESA8 ^{VRS_CESA7}	agacaagcactttacggatatgaaccaccaaagggt	tcaactttgatggaagaaggcggttcccgactct
CESA8 ^{LOOP_CESA4}	ttgatcagttccctaaatggttcctattaaccga	atcttcttagtctcattgtcctttgtggtacggt
CESA8 ^{LOOP_CESA7}	ttgatcagttccctaaatggttcctattgaacgt	atattttcagccggcactgtcctttgtggtacggt
CESA8 ^{VR1_CESA4}	cgttgaggcaatccttacaacgccacaaaggctct	ctagatgctgaagcaagagatgtgctttcggttg
CESA4 ^{VR1_CESA8}	caatgcaacactctttacgatgagaatgtttgat	acagaatctggtgctacacaacctctctggagaaaa

Table S2. List of primers used in this study.

For each swap, upto 6 primers were used. Two general primers, MF (GTTTTCCAGTCACGACGTTGTAACGACGGCCAG) and MR (CAGAAACAGCTATGACCATGTAATACGACTACTA) were used in all swaps. Either 2 or 4 swap specific primers were used for each swap. Primer BF and CR have complementary sequences BR and CR respectively. Here we have listed sequence for BF and CF primers only.