Supplementary Materials for

SaRCC1, a Regulator of Chromosome Condensation 1 (RCC1) Family Protein Gene from *Spartina alterniflora*, Negatively Regulates Salinity Stress Tolerance in Transgenic *Arabidopsis*

Table S1 List and information of primers used in this study.

Primer names	Forward primer (5'–3')	Reverse primer (5'–3')	usage
AtActin2	CTTCCTCAATCTCATCTTCTTCCGCTC	ATACCTCTCTTGGATTGTGCTTCATC	Semi- and qRT-PCR normalization
AtRCC1-2	GTCTCGATGGTGTGTCCATGAA	TCAGTTGGGATGACTAAAACCGGG	Semi RT-PCR expression
qAtRCC1-2	GAGAAATCATTGGTGAAGTAGCTCC	ACGCGAATTGGCACCAGCGGGGC	qRT-PCR expression
SaTubulin	GAAGGTGATGAGGGTGATGAGT	TTCAAGCAAACAAGCCTTCATA	qRT-PCR normalization
SaRCC1	AAAGGAGCCCAAGAAGGTCAAGGCAT	GCTACAACATGAACAGCTCCACAAGA	Semi RT-PCR expression
qAtUVR8	TACTGGTGGTGAGAAAATGTCAATGGTT	GTCAATGCCATTGTATGTCTCCAACCT	Semi- and qRT-PCR expression
qSaRCC1	ATGACGTGCACTGCAACCTCCGTGT	TGAGGCTGTCTGCATTCCTCAATTTG	qRT-PCR expression
HPT	CTCTGATAGAGTTGGTCAAGAC	GATATGTCCTGCGGGTAAATAG	PCR verification for transgenic plants
SaRCC1-sense	CGAAGGATCCATGGATATAGATGACGTG	CTAGGGTACCTCTGTCAGTTGGTTAACTGGC	*35S*: *SaRCC1* overexpression
SaRCC1-antisense	GCTCGGTACCATGGATATAGATGACGTGC	TAGAGGATCCTCTGTCAGTTGGTTAACTGGC	*35S*: Antisese-*SaRCC1* overexpression
SaRCC1-GFP	CTTAAGTCCGGAGCTAGCTCTAGAATGGATATAGATGACGTGC	CTCGCCCTTGCTCACCATGGATCCACTGGCTTTTCCCTGAACT	Subcellular localization
AtHMGB1-mCherry	CGCTTCTAGAATGGCTGACAAGAAGATTAG	AATCCCCGGGGGCCTTTGACATGTGGACGAT	nucleus-localized marker
AtPEX7-mCherry	CGCTTCTAGAATGCCGGTGTTCAAAGCTCC	AATCCCCGGGACTGGCTCTAGGATCCATCCC	peroxisome-localized marker
qAtHY5	ACAAGCGGCTGAAGAGGTTGTTG	TCTGGTTCTCGTTCTGAAGAGT	qRT-PCR expression
AtRD29A	TCTGAAGAACGAATCTGATAT	GCCGGAAATTTATCCTCTTCTGA	qRT-PCR expression
AtRD29B	TACATCAGCCATAACTAACAAA	TGCCCGTAAGCAGTAACAGATC	qRT-PCR expression
AtCOR47	AGTGAAGGAGAACAAGATTACTC	TCTTCATCGCTCGAAGAGGAAG	qRT-PCR expression
AtCOR15A	AACGAGGCCACAAAGAAAGC	CAGCTTCTTTACCCAATGTATCTGC	qRT-PCR expression
AtRD22	CACCGATGCAGAAGTACAAAAT	CCGTTCTCTCCCTCGAGCGGAA	qRT-PCR expression
AtABI2	GGGAAAGTAATCCGGTGGAACGG	TAAGATGAGACAATCATCTTCTT	qRT-PCR expression
AtP5CS1	TCACCGACATGGGAGTGCACACAC	TGCCCGTGCTTACCCCCACCTCT	qRT-PCR expression

Table S2 Accession numbers of UVR8 and RCC1 proteins used in Fig. 1.

Protein names	Species	Accession number	References
SaRCC1	*Spartina alterniflora*	This study	
SbUVR8	*Sorghum bicolor*	XP_021307437.1	NCBI
DoUVR8	*Dichanthelium oligosanthes*	OEL17715.1	NCBI
SiUVR8	*Setaria italica*	XP_004982469.1	NCBI
AtaRUG3	*Aegilops tauschii subsp. strangulata*	XP_020146593.1	NCBI
BdUVR8	*Brachypodium distachyon*	XP_003562310.1	NCBI
ObUVR8	*Oryza brachyantha*	XP_006650287.1	NCBI
CsUVR8	*Cannabis sativa*	XP_030485099.1	NCBI
OeUVR8	*Olea europaea var. sylvestris*	XP_022884180.1	NCBI
PdUVR8	*Phoenix dactylifera*	XP_008794204.2	NCBI
EgUVR8	*Elaeis guineensis*	XP_010938025.1	NCBI
PaUVR8	*Prunus avium*	XP_021816775.1	NCBI
PpeUVR8	*Prunus persica*	XP_007217882.1	NCBI
PduUVR8	*Prunus dulcis*	XP_034208971.1	NCBI
ZmRCC1	*Zea mays*	NP_001159191.1	NCBI
AtPROTON1	*Arabidopsis thaliana*	AT1G19880.1	Tair
AtSAB1	*Arabidopsis thaliana*	AT1G27060.1	Tair
AtPARF-1	*Arabidopsis thaliana*	AT1G65920.1	Tair
AtRCC1-1	*Arabidopsis thaliana*	AT1G69710.1	Tair
AtPRAF1	*Arabidopsis thaliana*	AT1G76950.1	Tair
AtRCC1-2	*Arabidopsis thaliana*	AT3G02300.1	Tair
AtRCC1-3	*Arabidopsis thaliana*	AT3G02510.1	Tair
AtRCC1-4	*Arabidopsis thaliana*	AT3G03790.3	Tair
AtRCC1-5	*Arabidopsis thaliana*	AT3G15430.1	Tair
AtRCC1-6	*Arabidopsis thaliana*	AT3G23270.1	Tair
AtRCC1-7	*Arabidopsis thaliana*	AT3G26100.2	Tair
AtRCC1-8	*Arabidopsis thaliana*	AT3G47660.1	Tair
AtRCC1-9	*Arabidopsis thaliana*	AT3G53830.1	Tair
AtTCF1	*Arabidopsis thaliana*	AT3G55580.1	Tair
AtRCC1-10	*Arabidopsis thaliana*	AT4G14368.1	Tair
AtRUG1	*Arabidopsis thaliana*	AT5G08710.1	Tair
AtRCC1-11	*Arabidopsis thaliana*	AT5G11580.1	Tair
AtRCC1-12	*Arabidopsis thaliana*	AT5G12350.1	Tair
AtRCC1-13	*Arabidopsis thaliana*	AT5G16040.1	Tair
AtRCC1-14	*Arabidopsis thaliana*	AT5G19420.2	Tair
AtRCC1-15	*Arabidopsis thaliana*	AT5G42140.1	Tair
AtRUG2	*Arabidopsis thaliana*	AT5G48330.1	Tair
AtRUG3	*Arabidopsis thaliana*	AT5G60870.1	Tair
AtUVR8	*Arabidopsis thaliana*	AT5G63860.1	Tair
AtUVR1 (AtUVH3)	*Arabidopsis thaliana*	AT3G28030.1	Tair
AtUVR2	*Arabidopsis thaliana*	AT1G12370.2	Tair
AtUVR3	*Arabidopsis thaliana*	AT3G15620.1	Tair
AtERCC1 (AtUVR7)	*Arabidopsis thaliana*	AT3G05210.1	Tair
BpUVR8	*Betula platyphylla*	AHY02156.1	Li et al., 2018
CmUVR8	*Chrysanthemum morifolium*		Yang et al., 2018
CqUVR8	*Colobanthus quitensis*	QEM23329.1	Contreras et al., 2019
CrUVR8	*Chlamydomonas reinhardtii*	AKS29686.1 (Cre05.g230600)	Tilbrook et al., 2016
MdUVR8	*Malus domestica*	KU095866	Zhao et al., 2016
MpUVR8.1	*Marchantia polymorpha*	Mapoly0023s0125.1	Soriano et al., 2018
MpUVR8.2	*Marchantia polymorpha*	Mapoly0023s0125.2	Soriano et al., 2018
OsUVR8a	*Oryza sativa Japonica Group*	Os02g0554100,	Idris et al., 2020
OsUVR8b	*Oryza sativa Japonica Group*	Os04g0435700	Idris et al., 2020
PeUVR8	*Populus euphratica*	AKJ54489.1	Mao et al., 2015

PpUVR8.1	*Physcomitrella patens*	Pp3c10_2230	Soriano et al., 2018
PpUVR8.2	*Physcomitrella patens*	Pp3c3_15300	Soriano et al., 2018
SlUVR8	*Solanum lycopersicum*	Solyc05g018620	Li et al., 2018
VvUVR1	*Vitis vinifera*	VIT_07s0031g02560	Loyola et al., 2016
ZmDEK47 (ZmRUG3)	*Zea mays*	NP_001144762.1	Cao et al., 2021
ZmUVR8	*Zea mays*	GRMZM2G003565	Fernández et al., 2020

Figure S1 Nucleotide and predicted peptide sequence of the *SaRCC1* gene. ORF is shown in upper-case letters; 3'- and 5'-UTRs are in lower-case letters. The predicted amino acids are displayed with single-letter symbols below their capitalized codon using DNAMAN.

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-121                                    gacgaggacaaaggaacaaggaggcaactccattccgcaggtcaaa
       agaggagaggagcactcaaagctccgacctttccaggaatcctcccggaagattgcaccattctctccaggggtc
1      ATGGATATAGATGACGTGCACTGCAACCTCCGTGTTGTTGGTGTGCCAACGAAAAGTGCAATTTACATATGGGGC
1       M  D  I  D  D  V  H  C  N  L  R  V  V  G  V  P  T  K  S  A  I  Y  I  W  G
76     TATAACCAGAGTGGGCAGACGGCACGAAAGGGCAAGGAATGCTACTTGAGGATTCCCAAGAGCCTCCCTCCCAAG
26      Y  N  Q  S  G  Q  T  A  R  K  G  K  E  C  Y  L  R  I  P  K  S  L  P  P  K
151    CTGTTCAAATTGAGGAATGCAGACAGCCTCAGGTGGACTGACATTGCATGTGGCCGTGAGCACACTGCTGCAGTT
51      L  F  K  L  R  N  A  D  S  L  R  W  T  D  I  A  C  G  R  E  H  T  A  A  V
226    GCTTCTGACGGATCACTCTTCACCTGGGGTGCTAATGAGTTTGGCCAGTTGGGAGATGGTACAGAAAAGAGTTCA
76      A  S  D  G  S  L  F  T  W  G  A  N  E  F  G  Q  L  G  D  G  T  E  K  S  S
301    AAGGAGCCCAAGAAGGTCAAGGCATTGGAGACTGAATTTGTGAAATCAGTATCCTGCGGTGCACATTGTACAGCT
101     K  E  P  K  K  V  K  A  L  E  T  E  F  V  K  S  V  S  C  G  A  H  C  T  A
376    GCCATTGCTGAGCCTCGAGAAAGTGATGGAACAGTATCAAAAAACAGGCTTTGGGTTTGGGGACAAAATCAGGGT
126     A  I  A  E  P  R  E  S  D  G  T  V  S  K  N  R  L  W  V  W  G  Q  N  Q  G
451    TCAGACAACCCTCGCCTATTCTGGGGAGATTTTACACCAAACACAGTGATCCAGCAGGTTTCTTGTGGAGCTGTT
151     S  D  N  P  R  L  F  W  G  D  F  T  P  N  T  V  I  Q  Q  V  S  C  G  A  V
526    CATGTTGTAGCTTTATCCGAAGATGGTCTCCTGCAAGCTTGGGGGTACAATGAGTACGGTCAGCTTGGCAGAGGT
176     H  V  V  A  L  S  E  D  G  L  L  Q  A  W  G  Y  N  E  Y  G  Q  L  G  R  G
601    TGTACTTCTCAAGGACTTCAGGGAGCTCGTGTACTAAATGCTTATGCAAGATTCCTTGATGATGCTCCAGAGCTA
201     C  T  S  Q  G  L  Q  G  A  R  V  L  N  A  Y  A  R  F  L  D  D  A  P  E  L
676    GTGAAGATTGTTAGGGTGTCATGTGGAGAGTACCATACAGCAGCTATATCAAAAAATGGGGAGGTATATACCTGG
226     V  K  I  V  R  V  S  C  G  E  Y  H  T  A  A  I  S  K  N  G  E  V  Y  T  W
751    GGACTGGGAAGCATGGGGCAGCTTGGGCATTGCTCCCTGCAGTCTGGAGACAAGGAGCTAATCCCTAGGCGAGTT
251     G  L  G  S  M  G  Q  L  G  H  C  S  L  Q  S  G  D  K  E  L  I  P  R  R  V
826    GTTGCCCTTGATGGGATAATAGTCCGAGACGTATCTTGTGGAGGTGTCCACTCTTGTGCTGTGACTCAAAATGGA
276     V  A  L  D  G  I  I  V  R  D  V  S  C  G  G  V  H  S  C  A  V  T  Q  N  G
901    GCTTTGTATGCTTGGGGCGGAGGACACGTGGGTCAGCTGGGAGTTGGACCTCAGAGTTGCTTCTTTTCTTGCTCT
301     A  L  Y  A  W  G  G  G  H  V  G  Q  L  G  V  G  P  Q  S  C  F  F  S  C  S
976    CTTAATGGATCTGAAATGCTGCTACGCAACATTCCAGTCCTGGTCATACCGTCAGGTGTCCGTCTTGCTACCTGT
326     L  N  G  S  E  M  L  L  R  N  I  P  V  L  V  I  P  S  G  V  R  L  A  T  C
1051   GGACACTCTCACACACTTGTATCTATGAAAGATGGCCGTATATACGGGTGGGGCTATAATAGTTATGGCCAGGCA
351     G  H  S  H  T  L  V  S  M  K  D  G  R  I  Y  G  W  G  Y  N  S  Y  G  Q  A
1126   GCAAACGAGAAGTCAACTTACGCTTGGTTCCCCTCTCCAGTTGATTGGTGTGTTGGTGAGGTGAGAAGACTTGCT
376     A  N  E  K  S  T  Y  A  W  F  P  S  P  V  D  W  C  V  G  E  V  R  R  L  A
1201   GCTGGAGGTGGCCATTCAGCTGTACTGACTGATGCATGTTCATTGAAGGAGCTGTGTGAGTTCAAGCTGGCAGAG
401     A  G  G  G  H  S  A  V  L  T  D  A  C  S  L  K  E  L  C  E  F  K  L  A  E
1276   ACTGTAAACATTTCTAACGCTCAGCTAATAGAAGATGTTGCGTCACGGACTGGTGCTGATGCCTTGGCCCGTTTA
426     T  V  N  I  S  N  A  Q  L  I  E  D  V  A  S  R  T  G  A  D  A  L  A  R  L
1351   TGTGAGAAATTAAGGGAACATCCGGATGTGCAAGGAGAATGTGAATTCCTCGAAAAGCAAGTAGATGGAGTTCAG
451     C  E  K  L  R  E  H  P  D  V  Q  G  E  C  E  F  L  E  K  Q  V  D  G  V  Q
1426   GGAAAAGCCAGTTAAccaactgacagatacttgacagtttgaggtatcggccattaatagttattgtggcacttg
476     G  K  A  S  *
ctgtctttaacggtgccatgttattgattcagacattaattgttgtagtcatggcagattgaacagtgtcaatggacttggg
acctggtttttattggtgtataaaccgattggtgtgtctcccatgtgtatcgaaggcgtggaggccctgtgtagatcatgaa
cttagtgtacttttgttgaactaaatggaaacttgtgcctgggtgaaaaaaaaaaaaaaaaaaaaaaaaaaaaa   +298
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Figure S2 Nucleotide sequence alignment between *SaRCC1* and *AtRCC1-2*. The alignment was performed using DNAMAN software. Primers used for semi RT-PCR qRT-PCR in *SaRCC1* and *AtRCC1-2* were shown by blue arrow and red arrow, respectively.

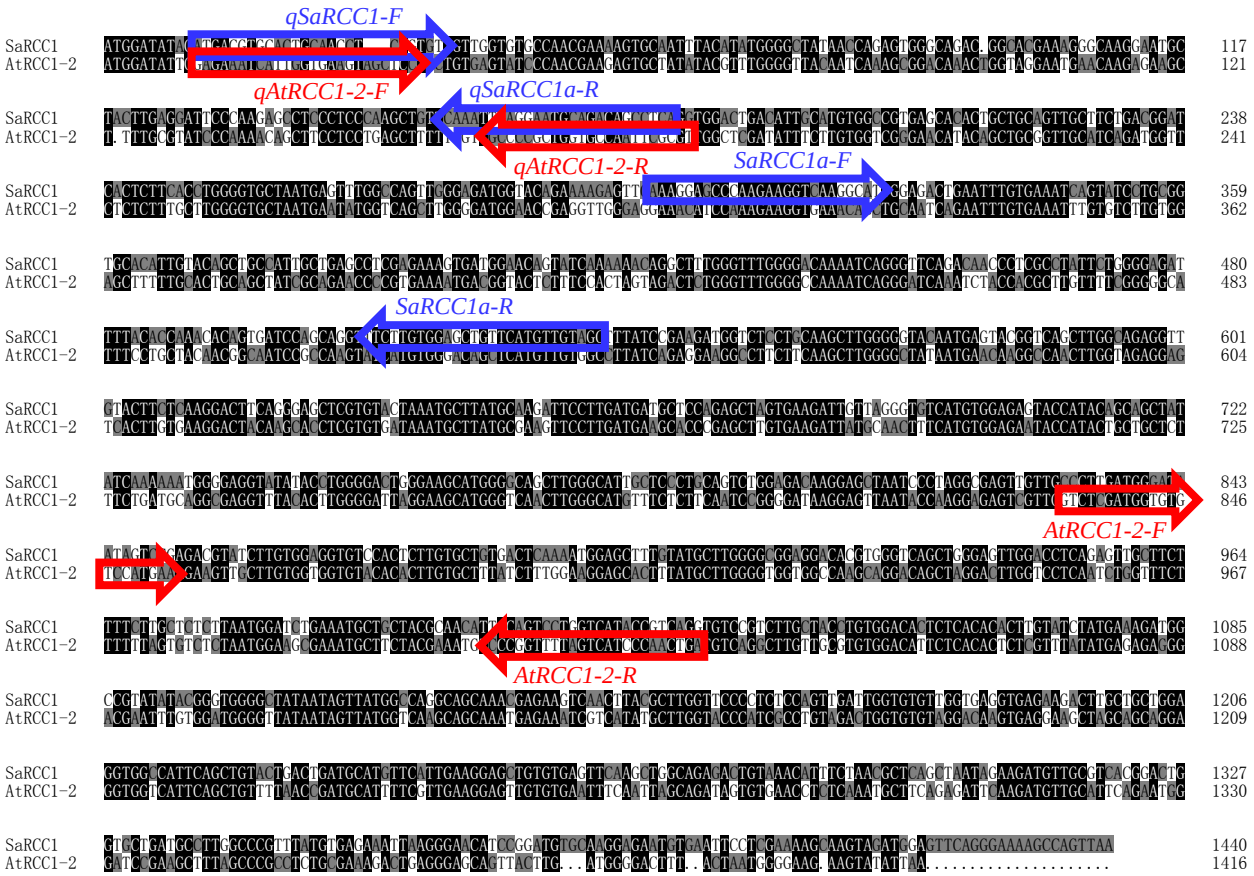


Figure S3 Amino acid sequence alignment between *SaRCC1* and *AtUVR8*. The alignment was performed using DNAMAN software.

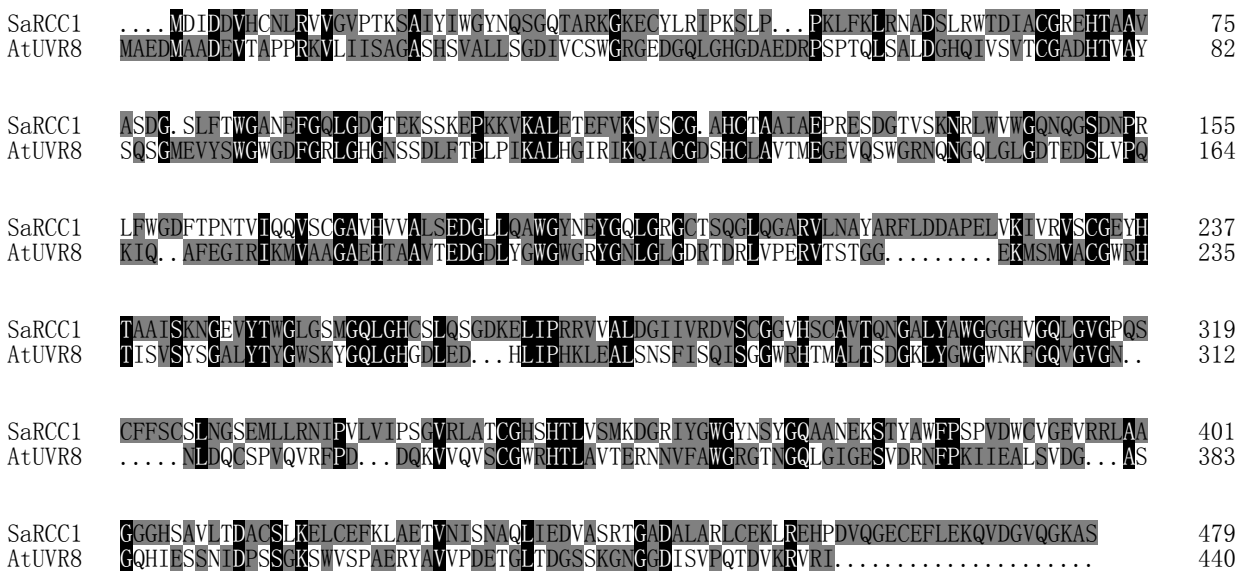


Figure S4 Construction of transgenic *Arabidopsis* for overexpression and antisense-overexpression of *SaRCC1*. (A) Construct of *SaRCC1* used in this study. (B) PCR analyses were performed on transgenic T_2 *Arabidopsis* plants, using *HPTII* gene primer. (C) Semi-quantitative reverse transcription PCR (RT-PCR) analysis of *SaRCC1*, *AtUVR8* and *AtRCC1-2* expression in transgenic *Arabidopsis* plants. RT-PCR was performed using RNA extracted from wild-type (WT), five independent *SaRCC1*-overexpressing, and ten independent *SaRCC1*-antisense-overexpressing lines. Specific primer pairs were used to detect their transcripts. Primers specific for *Arabidopsis* actin (*AtActin2*) were used for the loading control. *hyg*: hygromycin-phosphotransferase gene; *35S*: Cauliflower mosaic virus (CaMV) 35S promoter; Tnos: nopaline-synthase terminator; LB: T-DNA left border; RB: T-DNA right border. M, 250-bp DNA ladder marker; WT, wild-type *Arabidopsis* plants; N, negative control (dH_2O were used as PCR template).

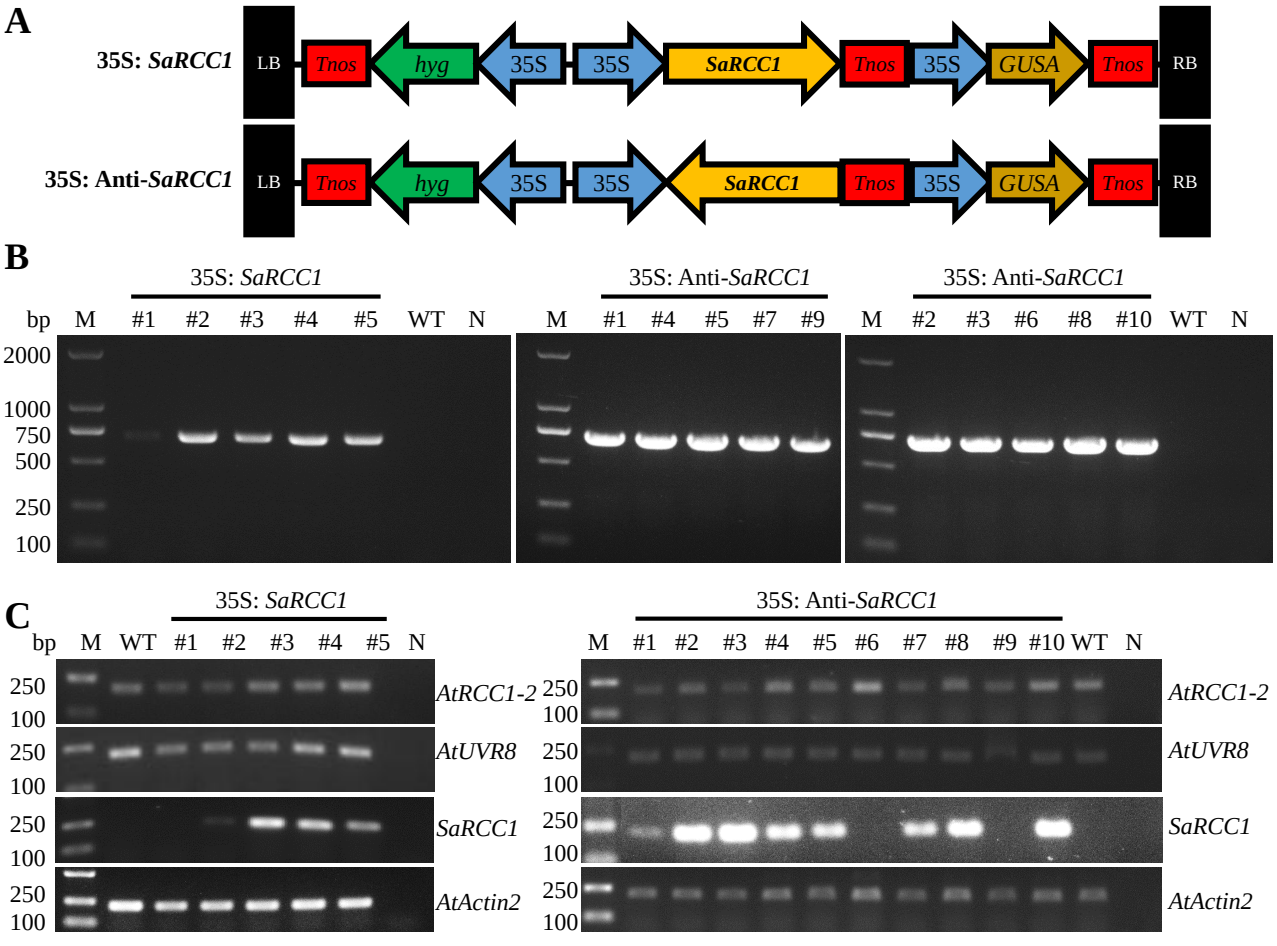


Figure S5 Analysis of osmotic tolerance in *Arabidopsis* wild-type (WT) and *SaRCC1* overexpressing and antisense-overexpressing plants. The phenotype (A), germination rates (B), and root length (**C**) of wild-type (WT) and *SaRCC1* overexpressing and antisense-overexpressing *Arabidopsis* in 1/2 MS medium with or without 200 mM mannitol for 7 d. Different letters indicate significant differences ($p < 0.05$) according Duncan's multiple test.

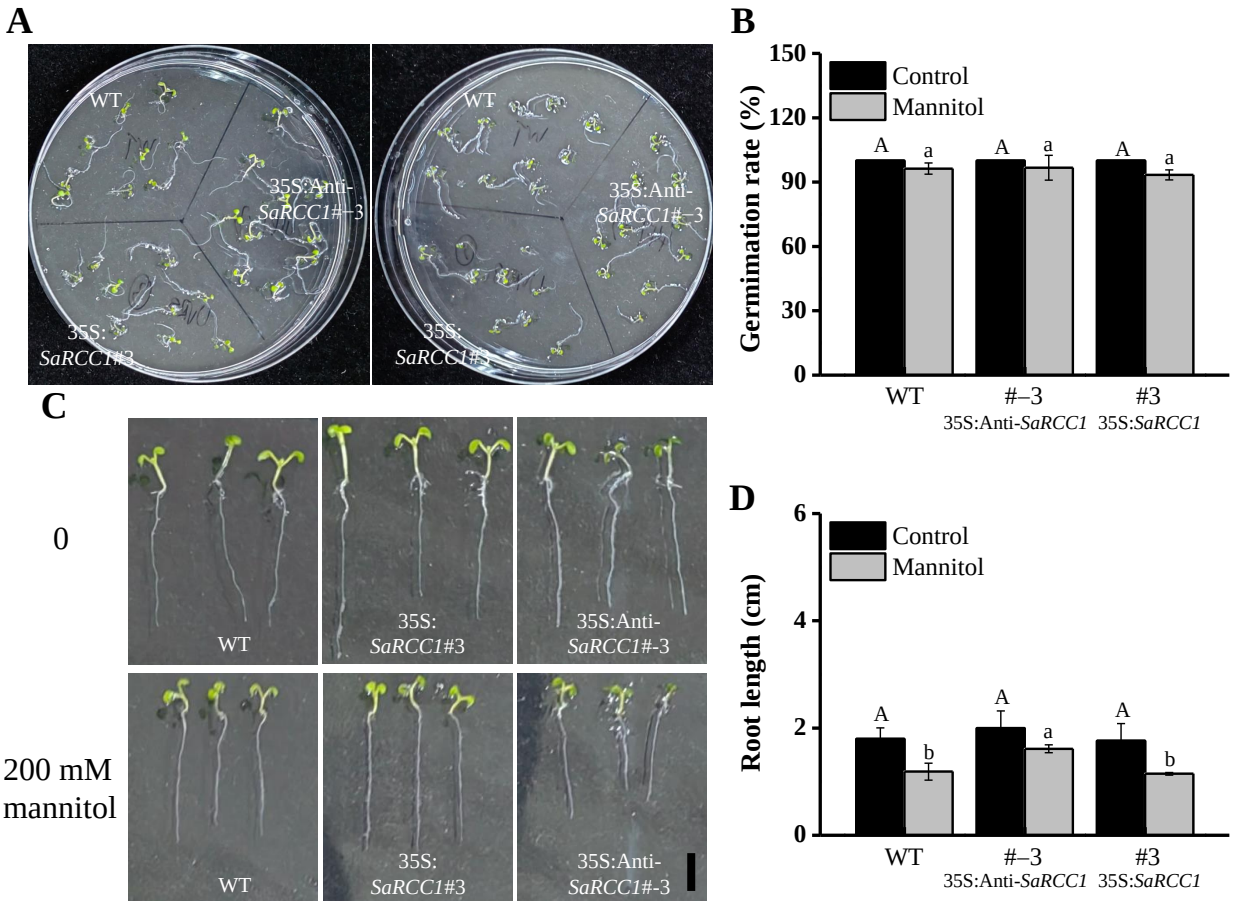


Figure S6. Expression levels of *SaRCC1*, *AtUVR8*, and *AtRCC1-2* genes in wild-type (WT) and transgenic plants under salinity stress, ABA treatment, and osmotic stress conditions. Seven-day-old seedlings were subjected to 100 mM NaCl, 5 μM ABA, or 300 mM mannitol, respectively, 24 h, before determining their expression levels by qRT-PCR. The *actin2* gene was used as an internal reference. The asterisks over the bars indicate the significant differences between the wild-type and *SaRCC1* transgenic *Arabidopsis* lines.

