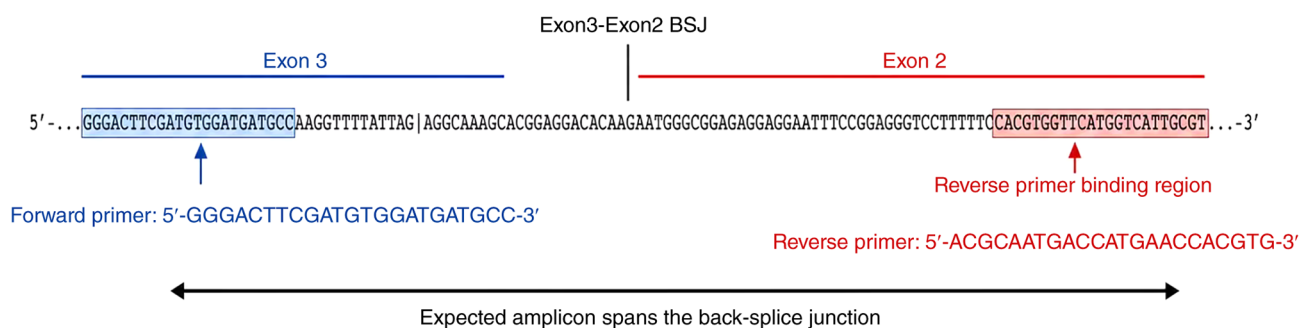


Figure S1. Schematic illustration of circRNA_013145-specific qPCR primers and siRNA/shRNA target sites spanning the Exon3-Exon2 BSJ. (A) Divergent qPCR primer pair used for circRNA_013145 detection. The forward primer is located in Exon 3, and the reverse primer is located in Exon 2. The expected amplicon spans the Exon3-Exon2 BSJ, thereby allowing specific amplification of the circular transcript. (B) siRNA and As-delivered shRNA target site for circRNA_013145. The target sequence crosses the Exon3-Exon2 BSJ, which improves specificity for circRNA_013145 and decreases the likelihood of targeting the linear aspartate β -hydroxylase transcript. Only the targeting portion of the shRNA sequence is shown. circ, circular; si, small interfering; sh, short hairpin; BSJ, back-splice junction; q, quantitative; Ad, adenovirus.

A circRNA_013145 qPCR primer overlap



B circRNA_013145 siRNA/shRNA target overlap

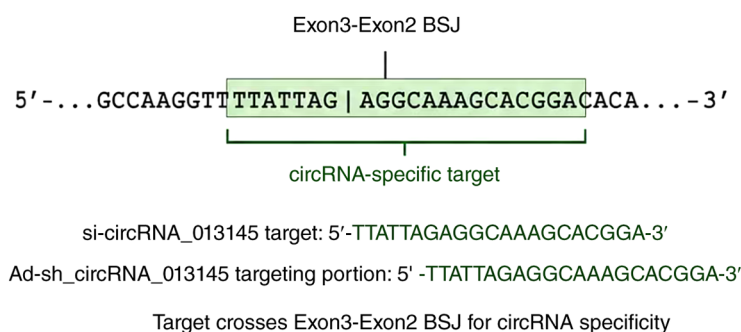


Figure S2. Identification of differentially expressed circRNAs in penile tissue of DMED rats. (A) Distributions of circRNAs in the DMED and control group samples was nearly the same after normalization. (B) Variation of circRNA expression between DMED and control group. Dots outside the green lines indicate circRNAs with logarithmized expression changes >2 -fold between groups. (C) Distributions of the differentially expressed circRNAs in the microarray analysis. (D) circRNA expression profiles in DMED and control tissue. Red, high relative expression; green, low relative expression. circ, circular; DMED, diabetes mellitus-induced erectile dysfunction.

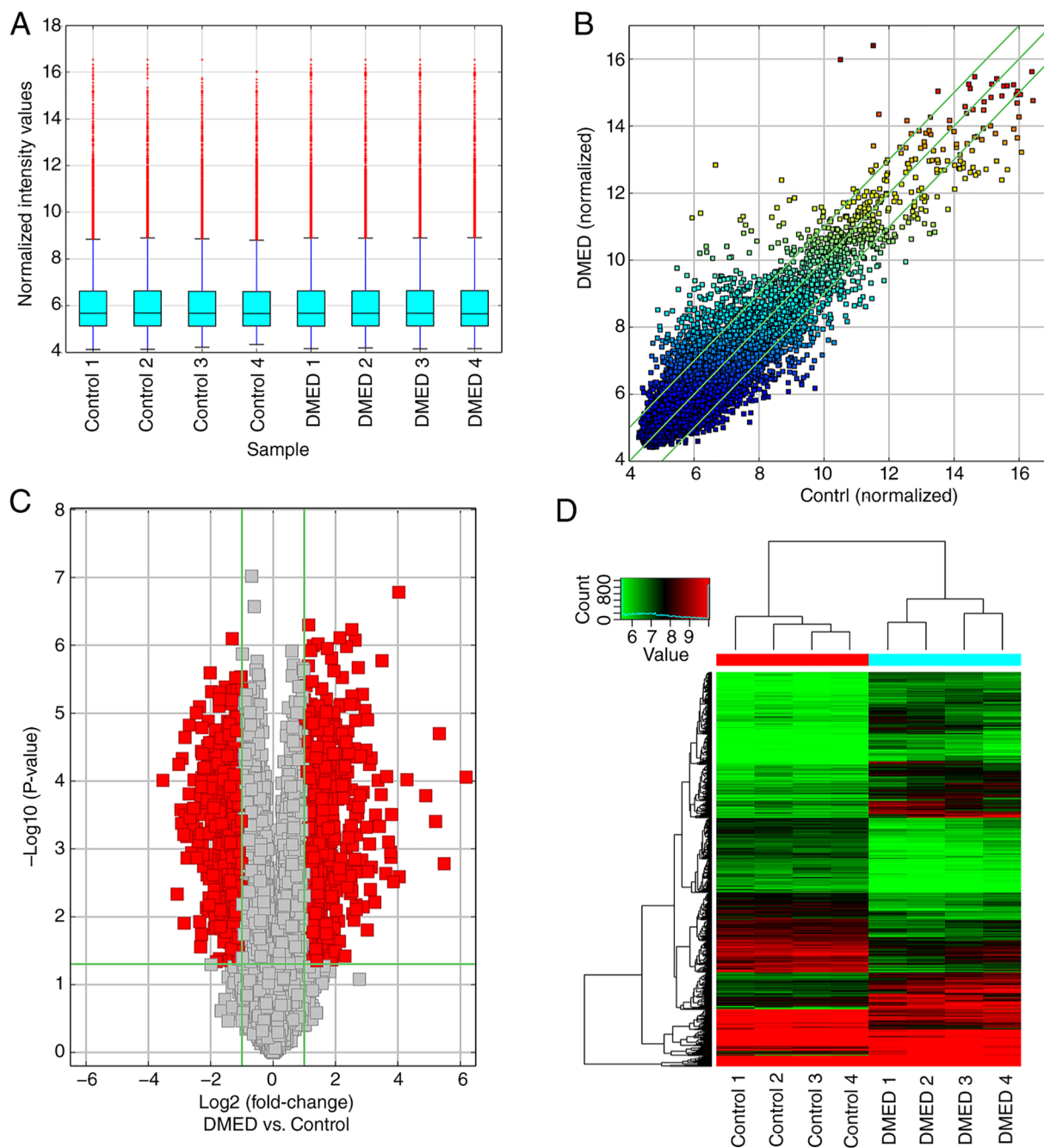


Figure S3. Verification of the expression of differentially expressed circRNAs by reverse transcription-quantitative PCR. Levels of (A) circRNA_013145, (B) circRNA_008695, (C) circRNA_010964, (D) circRNA_31436, (E) circRNA_013139, (F) circRNA_010502, (G) circRNA_26971, (H) circRNA_003748, (I) circRNA_011493 and (J) circRNA_012403 were measured in penile tissues of diabetes mellitus-induced erectile dysfunction rats. *P<0.05, **P<0.01 vs. control. circ, circular.

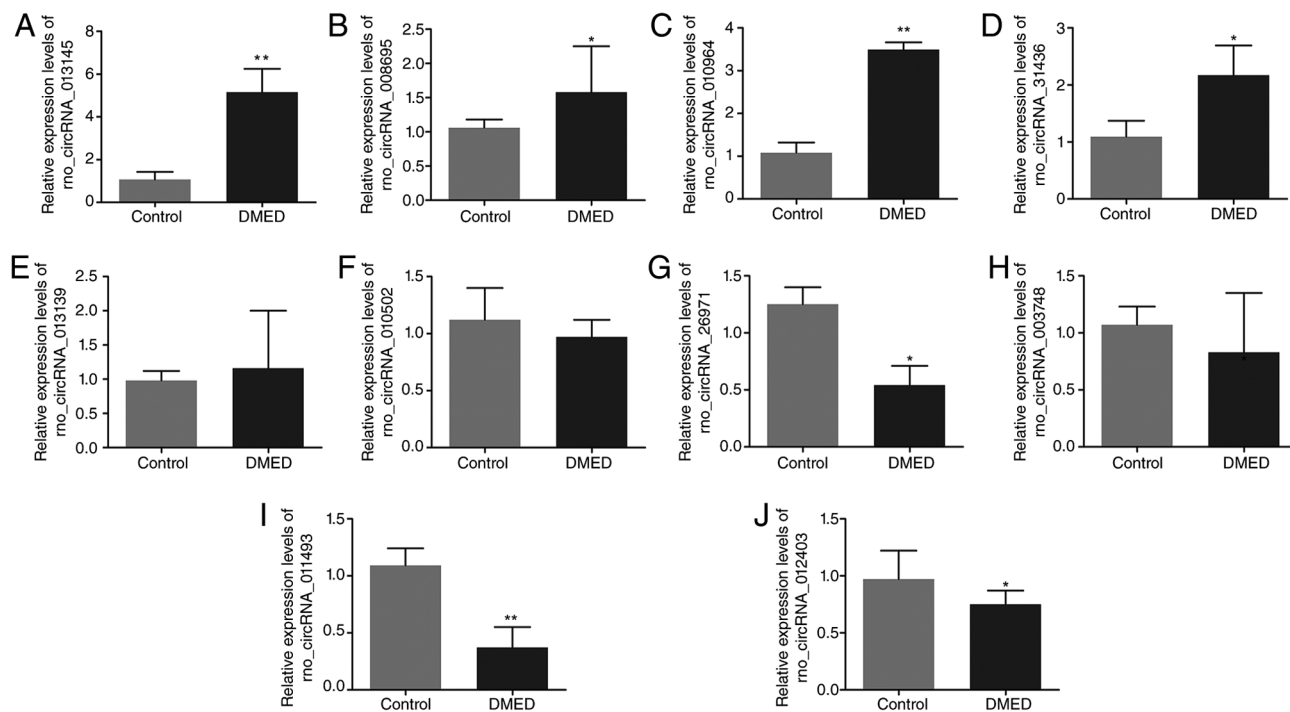


Figure S4. Potential interactions between circRNA_013145 and candidate miRNAs were predicted. The squares indicate the presence of conservation annotation for the predicted MRE among the aligned species provided by the Arraystar database. circRNA_013145 was predicted to bind (A) rno-miR-759, (B) rno-miR-217-3p, (C) rno-miR-1843b-5p, (D) rno-miR-3594-5p and (E) rno-miR-185-5p. The 2D structure demonstrated the MRE sequence, the target miRNA seed type and the 3'pairing sequence. Local AU showed the AU content 30 nt upstream and downstream of the seed sequence. The red bars represent A/U and high accessibility, while the black bars represent G/C and low accessibility of the seed. Accessibility extent is indicated via the height of the bar. Position represents the most likely relative MRE position on the linear presentation of circRNA_013145. circ, circular; miR, microRNA; MRE, microRNA response element; AU, adenine/uracil; nt, nucleotide; M, miRNA sequence; T, target sequence.



Figure S5. Visualization of the circRNA_013145-miR-mRNA regulation network. Nodes represent potential target miRs for circRNA_013145 and mRNAs for rno-miR-185-5p, rno-miR-217-3p, rno-miR-1843b-5p, rno-miR-759, and rno-miR-3594-5p, respectively. circ, circular; miR, microRNA.

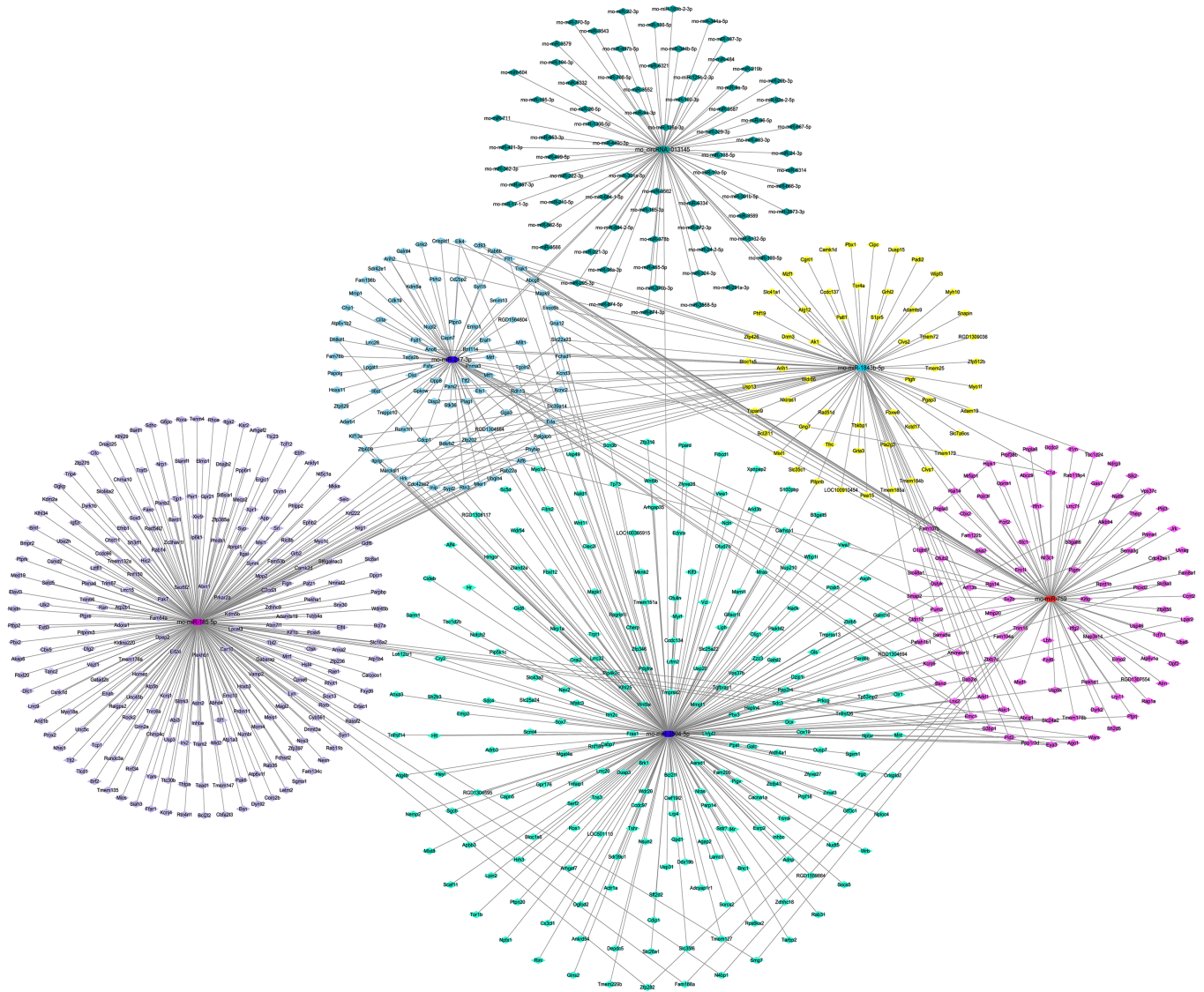


Figure S6. Gene Ontology, KEGG pathway, and PPI network analysis of the target genes of miR-185-5p. (A) Biological process, (B) molecular function, (C) cellular component, (D) KEGG pathway enrichment and (E) PPI network of the target genes of miR-185-5p. Nodes indicate target proteins, and edges represent interactions between two targets. KEGG, Kyoto Encyclopedia of Genes and Genomes; PPI, protein-protein interaction; miR, microRNA.

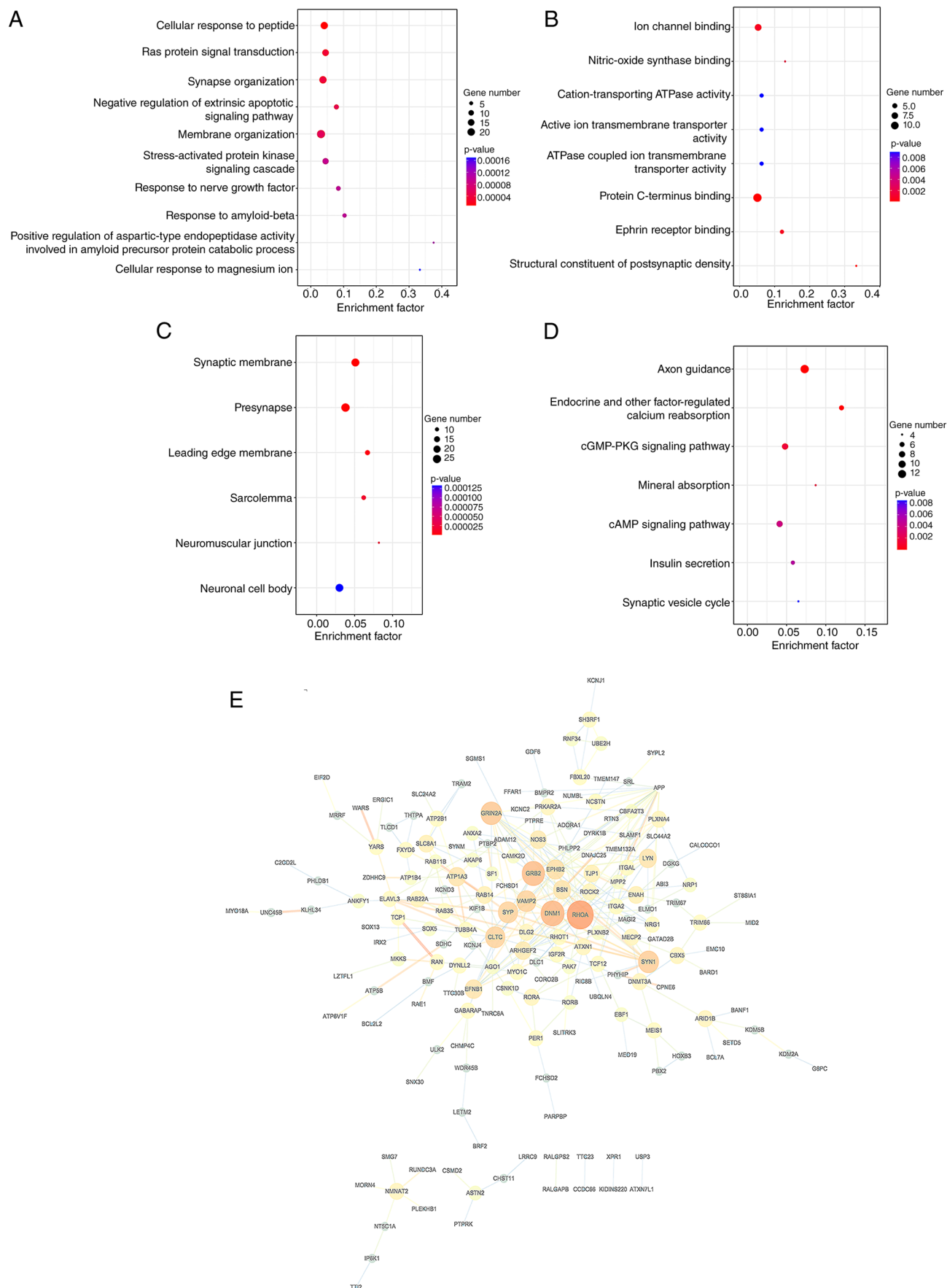


Figure S7. Validation of circRNA_013145 knockdown efficiency following siRNA transfection or Ad shRNA delivery alone. RT-qPCR of circRNA_013145 expression in (A) CCSMCs and (B) HUVECs after transfection with si-NC or siRNAs targeting circRNA_013145. (C) RT-qPCR analysis of circRNA_013145 expression in rat penile tissue following delivery of Ad-sh_NC or Ad-sh_circRNA_013145. * $P < 0.01$ vs. si-NC or Ad-sh_NC. circ, circular; si, small interfering; Ad, adenovirus; sh, short hairpin; RT-q, reverse transcription-quantitative; CCSMC, corpus cavernosum smooth muscle cells; HUVECs, human umbilical vein endothelial cell; NC, negative control.

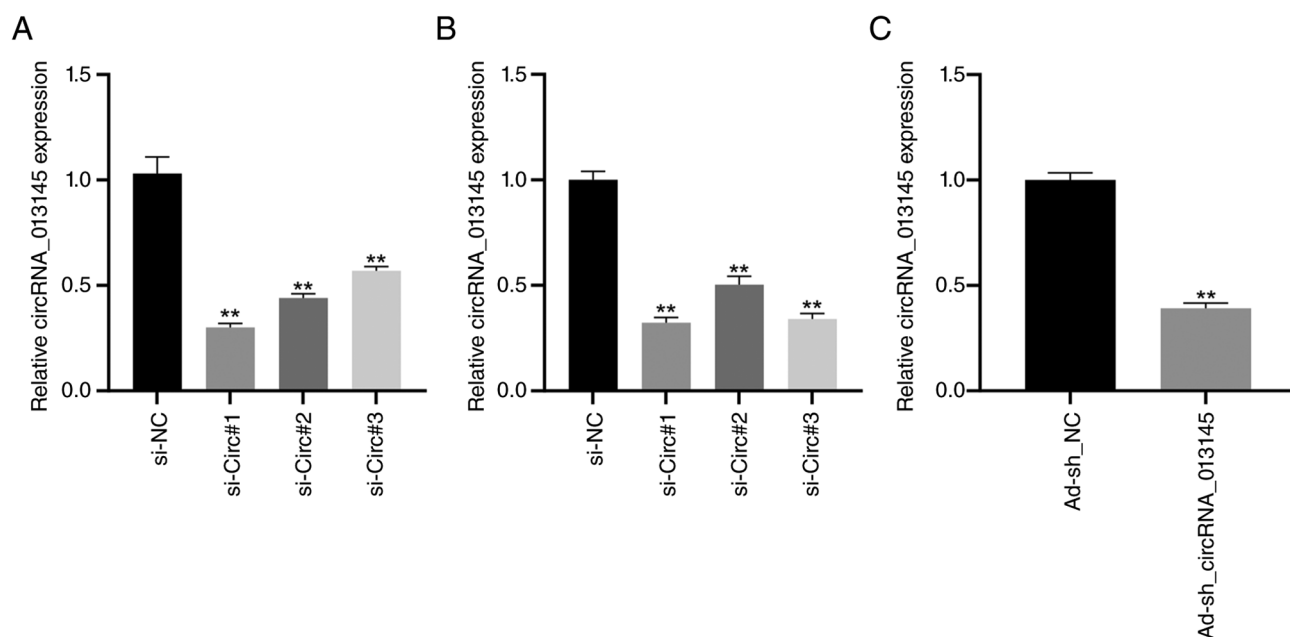


Figure S8. Schematic of upregulation of circRNA_013145 under high-glucose conditions, its sponging effect on miR-185-5p, subsequent regulation of RhoA and the downstream effects on oxidative stress, inflammation, phenotypic transformation, apoptosis and erectile function. circ, circular; miR, microRNA; MDA, malondialdehyde; SOD, superoxide dismutase; eNOS, endothelial nitric oxide synthase; VCAM-1, vascular cell adhesion molecule-1; ICAM-1, intercellular adhesion molecule-1.

