

**Critical role of the cyclic AMP-protein kinase A pathway in hyperglycemia-induced
epigenetic activation of fibrogenic program in the kidney**

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Table S1. Oligonucleotide sequences used in the study

RT-PCR primers	Forward 5'-3'	Reverse 5'-3'
mb2M	ACCGGCCTGTATGCTATCCAGAAA	ATTTCAATGTGAGGCGGGTGGAAAC
mGlut-1	CTACAGTGTATCCTGTTGCCCTTC	CTTCTTTCATCTCCTGCAGGTCTC
mFN	CGAGGTGACAGAGACCACAA	CTG GAG TCA AGC CAG ACA CA
mCol IV	AGGGTTACCTGGAGAAAAAGGG	TGGTCTCCTTTCTGTCCCTTC
mTgfb1	GGAGCAACATGTGGAACCTCT	CCTGTATTCCGTCTCCTTGG
mTgfb3	GATGAGCACATAGCCAAGCA	ATTGGGCTGAAAGGTGTGAC
mCtgf	GTGTGCACTGCCAAAGATGGTG	CAGCTTGACCCTTCTCGGGAA
mHk2	GAAGAGCTTTTCGACCACATTGT	CCTTGAATCCCTTTGTCCACTT
mPfk	AGATGGGTGCTAAGGCTATGAAC	AGTCTGTCTGGTCCTTCAGC
ChIP primers	Forward 5'-3'	Reverse 5'-3'
Tgfb1 promoter	ATCCACTGAGCACATAATGGCAC	TCATGTCCCTCCAACCCACA
Tgfb1 TSS	CCCACGCTAAGATGAAGACAGTGCCCC	ACGGCCTGGGGCGCGAAG
Tgfb3 promoter	CATGCACACACACACTGCTTTCTCTAT	TTACCACGTGGAACCAGAGGACTCA
Tgfb3 TSS	GCGTTACTCCGTCTCCGCAAA	CGATACGTCATGGGAGGGAGGTATAAAATT
Ctgf promoter	GAGTGTCAAGGGGTCAGGATCA	GGAGTGGATCTGGCTGAGTCT
Ctgf TSS	GGGGAGGAATGTGAGGAATGTC	TCTGGAGGAGGTCCGGTCT
EMSA probes	Forward 5'-3'	Reverse 5'-3'
Canonical CRE	GACGCGTGACGTCACAACAAGC	GCTTGTGTGACGTCACGCGTC
Canonical κB	AGTTGAGGGGACTTTCCAGGC	GCCTGGGAAAGTCCCCTCAACT
mCtgf κB	GGTGGGAGGAATGTGAGGAATGTCCCTG TTTGTGTAGGA	TCCTACACAAACAGGGACATTCCTCACATTC CTCCCCACC
mTgfb3CRE	TATACCTCCCTCCCATGACGTATCGGCCTG GGGTG	CACCCCAGGCCGATACGTCATGGGAGGGAG GTATA

Table S2. Gene names in representative Ingenuity pathways that are enriched in genes with HG-induced CBP and HAC peaks in the promoters.

Canonical Pathways	Molecules
EIF2 Signaling	SOS2,PIK3C2A,RPL23,RPS5,PIK3CB,RPL26,RPS13,RPS19,RPL8,EIF3E,RPS11,RPL14,RPL19,RPL27A,RPS29,RPS6,PIK3R3,RPL11,RPS18,EIF3A,RAF1,RPL35,RPL22,PABPC1,EIF4G2,EIF5,RPS12,RPL7,NRAS,RPLP0,RPS9,EIF2AK4,PIK3R2,PAIP1,PPP1R15A,PPP1CB,EIF3H,EIF3K,RPS27A,RPL15,EIF3F,GSK3B,RPS27L,EIF3M,RPS10,RPS26,EIF2B3,EIF4A2,PIK3R1,EIF3B,EIF3I,RPS20,EIF2AK1,RPL28,EIF2S2,RPL38,RPS15A,Rpl22I1,RPL37,RPL3,GRB2,EIF2B1,RPS4Y1,RPL31,EIF4A1,EIF4A3,RPL18A,RPL17,RRAS2,RPL21,ATM,RPL7L1,AKT2,SOS1,EIF3D,EIF2S3,PDPK1,RPS17,MAP2K2,MAPK1,RPS23,RPL36AL,EIF4G3,RPS28,Rpl36a,EIF2B4,EIF2AK3,RPL10
Regulation of eIF4 and p70S6K Signaling	PPP2R5E,SOS2,PIK3C2A,RPS5,PIK3CB,RPS13,RPS19,EIF3E,PPP2R5A,RPS11,PPP2CB,PAIP2,RPS6KB1,RPS29,RPS6,PIK3R3,RPS18,EIF3A,RAF1,MAPK12,PABPC1,EIF4G2,RPS12,NRAS,RPS9,PIK3R2,PAIP1,EIF3H,PPP2R5D,EIF3K,RPS27A,EIF3F,RPS27L,EIF3M,MAPK14,RPS10,RPS26,ITGA3,EIF2B3,IRS1,EIF4A2,PIK3R1,EIF3B,EIF3I,RPS20,EIF2S2,RPS15A,ITGB1,GRB2,EIF2B1,RPS4Y1,EIF4A1,EIF4A3,EIF4EBP1,MTOR,RRAS2,ATM,PPM1L,AKT2,SOS1,EIF3D,EIF2S3,PDPK1,RPS17,MAP2K2,MAPK1,RPS23,EIF4G3,MKNK1,RPS28,EIF2B4
PI3K/AKT Signaling	PPP2R5E,SOS2,PTEN,YWHAQ,PIK3CB,INPP5B,PPP2R5A,PPP2CB,SYNJ2,RPS6KB1,PIK3R3,YWHAG,JAK2,CTNNB1,RAF1,IKBK,FKB2,NRAS,PIK3R2,CDKN1B,CCND1,PPP2R5D,BCL2L1,LIMS1,YWHAZ,FOXO3,GSK3B,YWHAQ,MCL1,ITGA3,GSK3A,PIK3R1,GAB1,YWHAH,ITGB1,GRB2,HSP90AA1,TSC1,YWHAB,EIF4EBP1,SYNJ1,FOXO1,RPS6KB2,NFKB1,MTOR,CHUK,GDF15,RRAS2,PPM1L,TYK2,AKT2,SOS1,RELA,PDPK1,GAB2,BCL2,MAP2K2,MAPK1,RHEB,MDM2,HSP90AB1
mTOR Signaling	PPP2R5E,PIK3C2A,PRKAA1,PLD3,EIF4B,PRKAG2,RPS5,PIK3CB,RPS13,RPS19,EIF3E,PPP2R5A,RPS11,PPP2CB,MLST8,RHOT1,RND3,RPS6KB1,PRKCE,RPS29,RPS6,PIK3R3,PRKD3,RPS18,EIF3A,EIF4G2,RPS12,GNB1L,NRAS,RPS9,RPTOR,PIK3R2,RHOB,EIF3H,PPP2R5D,EIF3K,RPS27A,PRKAG1,EIF3F,PRKAB2,RPS27L,EIF3M,RPS10,RPS26,RHOD,RICTOR,IRS1,EIF4A2,RHOQ,PIK3R1,EIF3B,EIF3I,ATG13,RPS20,RPS15A,RPS4Y1,PRKCI,EIF4A1,EIF4A3,TSC1,EIF4EBP1,RPS6KB2,MTOR,RRAS2,ATM,PPM1L,AKT2,RHOG,EIF3D,PDPK1,NAPEPLD,RPS17,HIF1A,PLD2,MAPK1,RPS23,EIF4G3,RPS28,RH,EB,RAC1
Cell Cycle: G1/S Checkpoint Regulation	E2F6,GSK3B,TGFB1,HDAC10,HDAC8,CDKN2A,CUL1,SMAD4,MAX,CCND3,GNL3,SKP2,SKP1,SIN3A,PA2G4,RBL1,CDK6,FOXO1,E2F5,CCNE2,HDAC5,TGFB3,CDK4,RPL11,CDKN2C,ATM,CDC25A,CDK2,HDAC3,HDAC7,ABL1,MYC,CDKN1B,CCND1,CDKN2B,MDM2,PAK1IP1,TFDP1
Insulin Receptor Signaling	GRB10,SOS2,PTEN,PIK3C2A,FOXO4,PRKAG2,PRKACB,PIK3CB,INPP5B,SYNJ2,SH2B2,RPS6KB1,PRKAR2A,PIK3R3,JAK2,CRKL,RAF1,PTPN1,PRKAR1A,NRAS,RPTOR,SGK1,PIK3R2,PPP1CB,PRKAG1,FOXO3,GSK3B,EIF2B3,IRS1,GSK3A,RHOQ,PIK3R1,GAB1,CBL,PPP1R14B,PTPN11,GRB2,EIF2B1,CRK,MAPK8,PRKCI,RAPGEF1,TSC1,EIF4EBP1,SYNJ1,FOXO1,RPS6KB2,MTOR,RRAS2,PPP1R12A,ATM,AKT2,SOS1,PPP1R14C,PDPK1,STXBPA,MAP2K2,MAPK1,PRKACA,EIF2B4,STX4
ERK/MAPK Signaling	PPP2R5E,SOS2,PIK3C2A,MYCN,PRKAG2,PRKACB,RAP1A,PIK3CB,HSPB2,PPP2R5A,PPP2CB,DUSP4,PRKAR2A,PRKCE,MAPKAPK5,PIK3R3,SRC,TLN1,YWHAQ,CRKL,RAF1,ATF1,H3F3A/H3F3B,PTK2,DUSP2,PRKAR1A,HSPB1,MYC,NRAS,PIK3R2,LAMTOR3,RAP1B,PPP1CB,BCAR1,PPP2R5D,DUSP6,ETS1,PRKAG1,YWHAZ,YWHAQ,ATF2,CREB1,ITGA3,PIK3R1,YWHAH,RAPGEF3,ETS2,DUSP1,PPP1R14B,ITGB1,GRB2,CRK,PRKCI,RAPGEF1,YWHAB,EIF4EBP1,PAK2,RRAS2,PPP1R12A,ATM,PPM1L,EP300,PXN,NFATC1,SOS1,PPP1R14C,ATF4,MAP2K2,MAPK1,MKNK1,PRKACA,PAK4,RAC1
Mitochondrial Dysfunction	NDUFA10,VDAC3,COX7B,DHODH,ATP5F1,COX5A,NDUFA1,NDUFAF1,NDUFA5,CYCS,FIS1,NDUFAF2,NDUFA8,GSR,TXN2,SDHD,ATP5A1,ATP5C1,ATPAF1,COX4I1,NDUF57,COX7A2L,HTRA2,MAPK12,NDUFA2,NDUF54,ATP5G1,UQCRC2,NDUFA4,PRDX5,PSENEN,COX6A1,AIFM1,NDUFB2,UQCRCB,ATP5G3,VDAC1,OGDH,CPT1A,GLRX2,NDUFV3,ATP5I,PRDX3,NDUFA13,COX7A2,COX10,COX6B1,UQCRC11,MAPK8,UQCRCF1,ATP5G2,NDUFV2,PSEN1,SDHA,ATP5D,PDHA1,NDUFS1,NCSTN,NDUFB6,UQCRCQ,NDUFS3,PSEN2,ATP5L,BCL2,ACO1,NDUFB11,CASP8,GPD2
Protein Kinase A Signaling	AKAP13,MPPE1,GNB1,SMAD4,RAP1A,PTPN13,AKAP1,CDC27,PRKAR2A,PRKCE,PDE12,YWHAQ,CTNNB1,TCF4,ATF1,H3F3A/H3F3B,DUSP2,PTPN23,PTK2,EYA3,GNB2,PLCD3,PRKAR1A,NFKB2,Cal m1,PTPN12,CAMK2G,MAP3K1,RAP1B,PRKAG1,YWHAZ,FLNB,ATF2,AKAP10,CREB1,PTCH1,DUSP11,Hist1h1e,GSK3A,PDE4B,PTPN4,HIST1H1C,HHAT,CREM,HIST1H1D,PDE7A,DUSP1,PTPN11,ROCK1,TCF7L2,PRKCI,ANAPC7,AKAP11,PTPN9,EYA1,CHUK,GNB2L1,MYL12B,VASP,EP300,MYL12A,PTPN3,NFATC1,DUSP22,PTPRJ,MYL6,PDE8A,PTPRK,ITPR3,DUSP5,KDEL2R,PTP4A1,MAP2K2,PRKACA,APEX1,DUSP8,ITPR2,PTEN,TGFB1,SMO,PRKAG2,PRKACB,ROCK2,YWHAQ,GNAI3,ANAPC1,PTPN21,PTPDC1,DUSP4,PTPRS,DUSP16,PRKD3,RAF1,CDC25A,PTPN1,GNB1L,TGFB2,PPP3R1,GN G12,PPP1CB,DUSP6,GSK3B,CDC14B,YWHAQ,CAMK2D,YWHAH,GNAI2,KDEL2R,PPP1R14B,PDE6D,NFAT5,YWHAB,TGFB3,NFKB1,NTN1,PPP1R12A,PTPN2,AKAP8,GLI3,PXN,MYH10,RELA,NFATC3,PPP1R14C,ATF4,GNAS,CDC16,MAPK1
NRF2-mediated Oxidative Stress Response	MAP2K5,ABCC4,DNAJC13,PIK3C2A,DNAJC5,UBE2E3,NQO2,PIK3CB,ENC1,DNAJC21,GSR,DNAJB14,DNAJA4,PRKCE,PIK3R3,GSTT2/GSTT2B,USP14,DNAJB1,PRKD3,RAF1,PMF1/PMF1,BGLAP,SOD1,DNAJC14,MAFF,DNAJC8,NRAS,DNAJC18,PIK3R2,MAP3K1,EPHX1,VCP,JUNB,GSK3B,JUN,MAP2K7,MAPK14,DNAJC17,MAFK,DNAJB12,PIK3R1,DNAJC19,FKBP5,CDC34,MAPK8,PRKCI,GSTP1,CUL3,GCLC,BACH1,DNAJC10,RRAS2,ATM,EP300,TXNRD1,ABCC1,PP1B,ATF4,DNAJA2,MAP2K2,MAPK1,AKR1A1,DNAJB4,UBE2K,KEAP1,GCLM,FOSL1,EIF2AK3,NFE2L2
Cyclins and Cell Cycle Regulation	E2F6,GSK3B,PPP2R5E,TGFB1,HDAC10,HDAC8,CDKN2A,CUL1,CCND3,PPP2R5A,SKP2,SKP1,SIN3A,PPP2CB,PA2G4,CDK6,E2F5,CCNE2,HDAC5,TGFB3,CDK4,CDKN2C,RAF1,ATM,PPM1L,CDC25A,CDK2,HDAC3,HDAC7,ABL1,CDKN1B,CCND1,CDKN2B,PPP2R5D,TFDP1,CCNB1
G2/M DNA Damage Checkpoint Regulation	KAT2B,YWHAQ,PPM1D,TRIP12,CDKN2A,CUL1,YWHAQ,YWHAH,TOP2B,PKMYT1,SKP2,SKP1,CKS2,YWHAB,MDM4,BRCA1,YWHAQ,PTPMT1,ATM,EP300,ABL1,MDM2,CKS1B,YWHAZ,CCNB1,CHEK1
Apoptosis Signaling	BIRC2,MAP2K7,MCL1,TNFRSF1A,CYCS,BIRC3,LMNA,ROCK1,MAPK8,CASP2,CAPNS1,CAPN7,PRKCE,NFKB1,MAP4K4,CHUK,RRAS2,RAF1,HTRA2,CAPN1,MAP3K14,IKBK,MAPK10,ACIN1,RELA,NFKB2,NRAS,PARP1,CAPN5,BCL2,BIRC6,MAP2K2,MAPK1,AIFM1,BCL2L1,BID,CASP8,DIABLO
ErbB2-ErbB3 Signaling	JUN,GSK3B,SOS2,PTEN,PIK3C2A,GSK3A,PIK3R1,PIK3CB,GRB2,FOXO1,PIK3R3,RRAS2,RAF1,ATM,SP1,TYK2,SOS1,MYC,NRAS,PDPK1,PIK3R2,CDKN1B,CCND1,MAP2K2,MAPK1,STAT5B
Oxidative Phosphorylation	NDUFA10,COX7B,ATP5F1,COX5A,NDUFA1,NDUFV3,ATP5I,NDUFA5,CYCS,NDUFA13,COX7A2,NDUFA8,COX10,COX6B1,UQCRC11,SDHD,ATP5A1,ATPAF1,UQCRCF1,ATP5G2,ATP5C1,NDUFV2,COX4I1,NDUF57,COX7A2L,SDHA,NDUFA2,ATP5D,NDUFS1,NDUFA4,ATP5G1,NDUFA4,UQCRC2,NDUFB6,UQCRCQ,NDUFS3,ATP5L,COX6A1,NDUFB11,UQCRCB,NDUFB2,ATP5G3
TGF-β Signaling	JUN,SOS2,MAPK14,HOXC9,TGFB1,SMURF2,ACVR2A,SMAD4,RNF111,PIAS4,SMAD7,GRB2,MAPK8,PMPEA1,TRAF6,TGFB3,RRAS2,RAF1,SMAD2,MAPK12,EP300,SOS1,SMAD9,SKI,BMPR2,BMPR1A,TGFB2,NRAS,SMAD5,BCL2,SMAD1,TGIF1,MAP2K2,MAPK1,TFE3
TCA Cycle II	DLD,FH,IDH3B,OGDH,SDHA,DLST,IDH3G,SUCLG1,IDH3A,CS,SDHD,MDH1B,ACO1
STAT3 Pathway	MAPK14,SOCS5,NDUFA13,MAPK8,EGFR,SRC,JAK2,RRAS2,RAF1,MAPK12,PTPN2,CDC25A,MAP3K12,SOCS2,TYK2,MAP3K9,IGF2R,IGF1R,BMPR2,BMPR1A,MYC,TGFB2,NRAS,BCL2,MAP2K2,MAPK1,SOCS1,RAC1
NF-κB Activation by Viruses	TRAF2,ITGAV,PIK3C2A,ITGA3,PIK3R1,PIK3CB,ITGB1,PRKCI,PRKCE,NFKB1,CHUK,PIK3R3,RRAS2,PRKD3,RAF1,ATM,MAP3K14,ITGA6,IKBK,IKBKAP,AKT2,RELA,NFKB2,NRAS,PIK3R2,MAP3K1,MAPK1

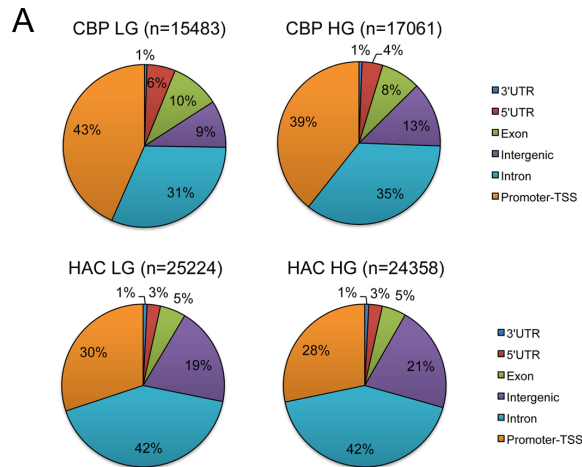


Figure S1. Genomic distribution of CBP and H3K9/14Ac (HAC) peaks. (A) CBP and HAC peaks in genomic features under HG and LG conditions; (B) Genome-wide enrichment of HG-induced CBP and HAC signal peaks.

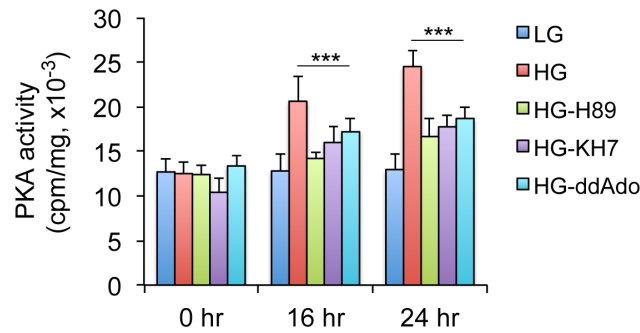
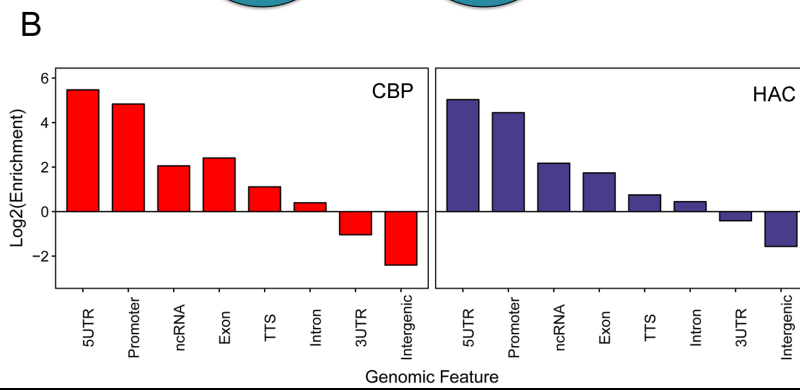


Figure S2. Inhibition of PKA activity by inhibitors specific for adenylyl cyclases or PKA. HG induces PKA activity in MCs at 16 and 24 hours, and the induction is suppressed by PKA inhibitor H89 and adenylyl cyclase inhibitors KH7 and ddAdo. *** P<0.001; n=3.

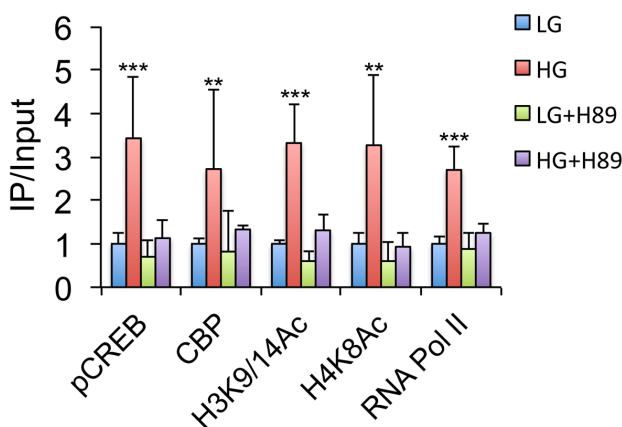


Figure S3. PKA inhibition attenuates high glucose-induced chromatin remodeling in *Tgfb3* promoter. ChIP assays showing that HG-induced pCREB binding, CBP recruitment, H3K9/14 and H4K8 acetylation and RNA Pol II occupation in *Tgfb3* promoter are markedly blocked by H89 treatment. **P<0.01; ***P<0.001 vs. the rest; n=3.