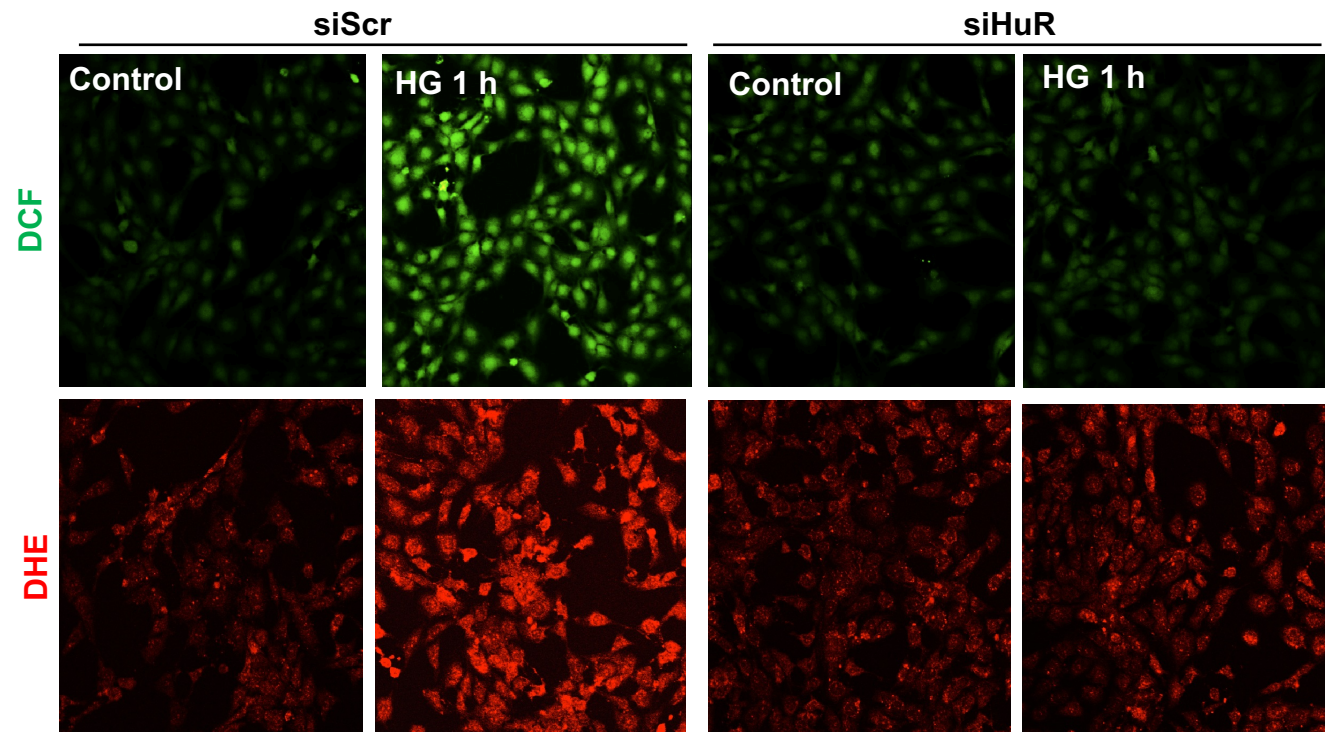
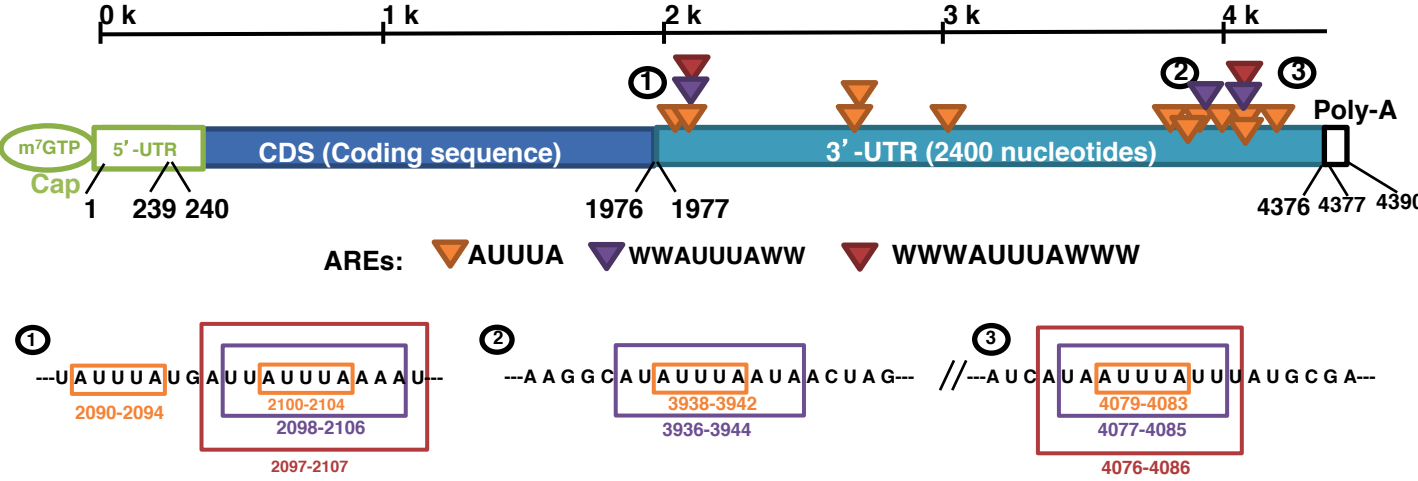


Supplementary Figure 1. HuR is required for HG-induced increase in ROS generation. MCs cells were transiently transfected with siRNA against HuR. 48 hours after transfection, MCs were treated with high glucose or control for 1 hour. Then the cells were immediately incubated with DCF or DHE dye at working concentration for 30 mins, and subjected to confocal imaging.

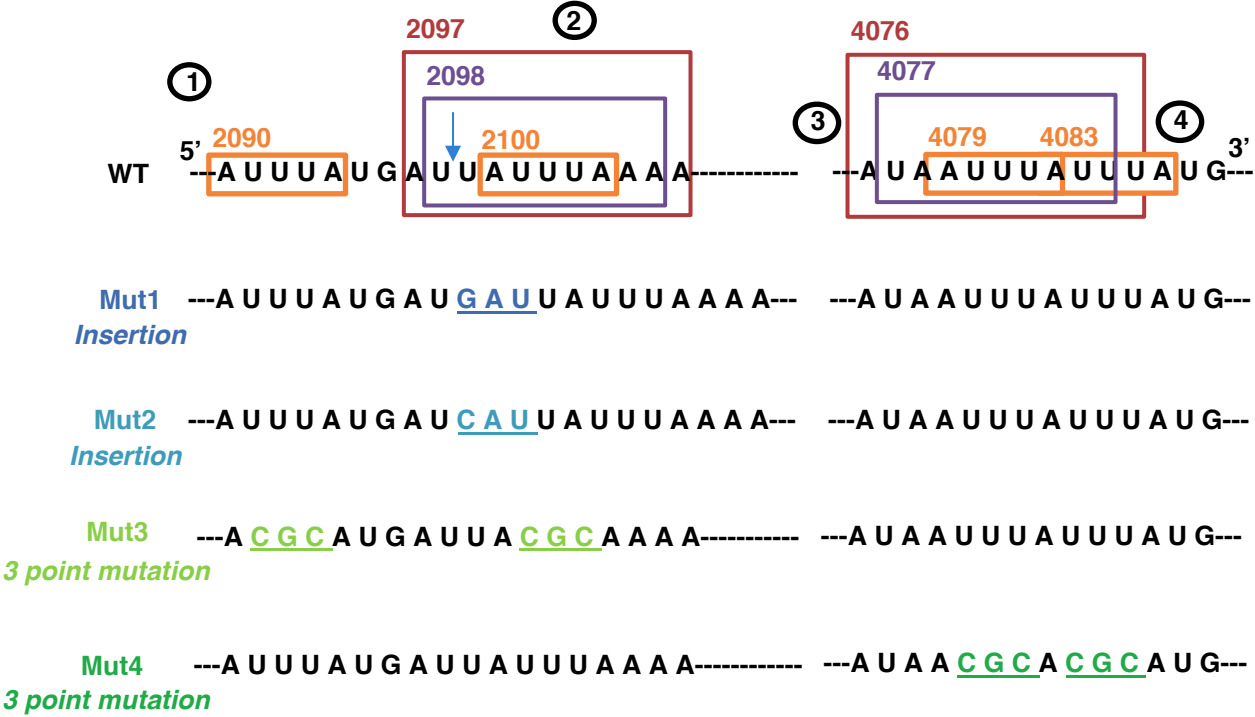


Supplementary Figure 2. Nox4 mRNA 3' untranslated region (3'-UTR) contains HuR binding AU-rich elements (AREs).

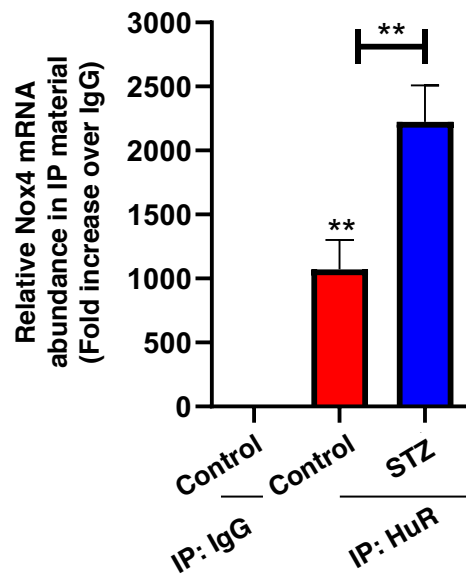
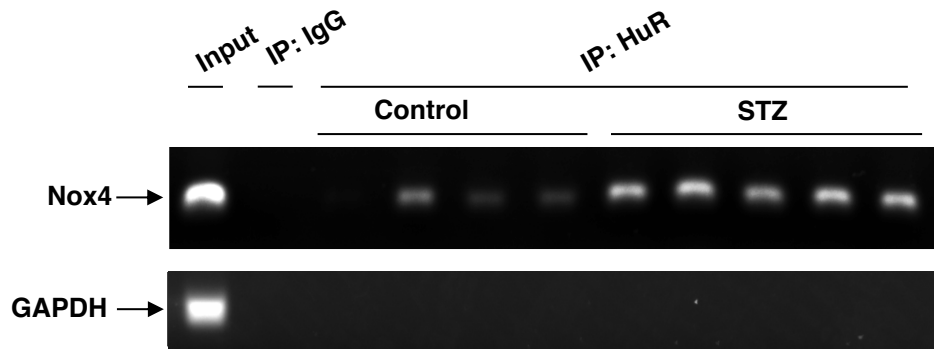


Online database for the investigation of AREs: <http://rna.tbi.univie.ac.at/cgi-bin/AREsite.cgi>

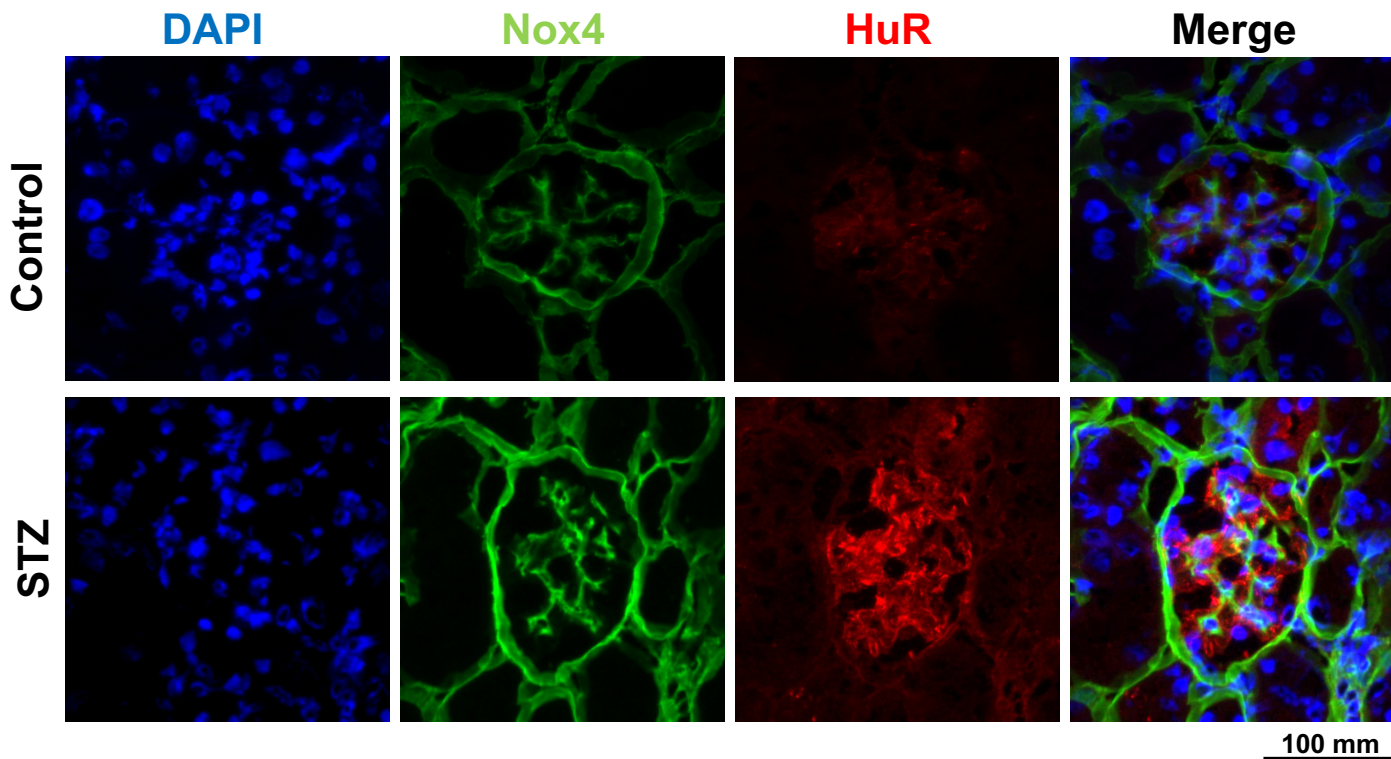
Supplementary Figure 3. The mutations on wild type Nox4 3'UTR-pEZX-MT01 that disrupts ARE regions, by site-direct mutagenesis.



Supplementary Figure 4. Nox4 mRNA is also present in the IP complex with HuR antibody in rat, and the affinity is increased after STZ induction. Kidney cortex tissues were isolated from control and STZ-induced diabetic rats. The total protein lysates were incubated with HuR antibody or IgG as control. The presence of Nox4 were detected by RT-PCR from the RNAs in immunoprecipitated complex.



Supplementary Figure 5. HuR is co-localized with Nox4 in glomerulus, and the intensities for both protein are increased in diabetic animal. Kidney frozen sections were prepared from both control and STZ-induced high blood glucose mice. The sections were stained with Nox4 and HuR simultaneously and images were taken under a confocal microscope. DAPI was used as counter staining for nuclei. Scale bar = 100 mm.



Supplementary Table-1. RNA sequence used in RNA pull down experiments.

RNA mol.	Sequence	AREs Region
Biot RNA1 wt	U <u>AUUUA</u> UGAUU <u>AUUUA</u> AAAU	2089-2108
Biot RNA1 Mut	U <u>ACGCA</u> UGAUU <u>ACGCA</u> AAAU	
Biot RNA2 wt	AAGGCAU <u>AUUUA</u> AUAACUAG	3931-3950
Biot RNA2 Mut	AAGGCAU <u>ACGCA</u> AUAACUAG	
Biot RNA3 wt	AUCAUA <u>AUUUAUUUA</u> AUGCGA	4073-4092
Biot RNA3 Mut	AUCAUA <u>ACGCA</u> <u>ACGCA</u> AUGCGA	

Supplimentary Table-2. Primer sequences for generating hNox4-3UTR mutants

Mutants	Primers	Sequence
Mut-1 (2098GAT)	Forward	GATTATTTAAAATGGAAATGTGAGAATG
	Reverse	ATCATAAATAAGAAAACCTTACTATTC
Mut-2 (2098CAT)	Forward	CATTATTTAAAATGGAAATGTGAGAATG
	Reverse	ATCATAAATAAGAAAACCTTACTATTC
Mut-3 (2091/2101_mut_CGC)	Forward	TTACGCAAAAATGGAAATGTGAGAATGTG
	Reverse	TCATGCGTAAGAAAACCTTACTATTCTTTGG
Mut-4 (4079/4085_mut_CGC)	Forward	CGCATGCGATCATATTAATAGACC
	Reverse	TGCGTTATGATCAGTATAGAGTCAGTATTG