

Supplementary Information

for

Protein phosphatase 4 (PP4) functions as a critical regulator in tumor necrosis factor (TNF)- α -induced hepatic insulin resistance

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Supplementary Fig. S1

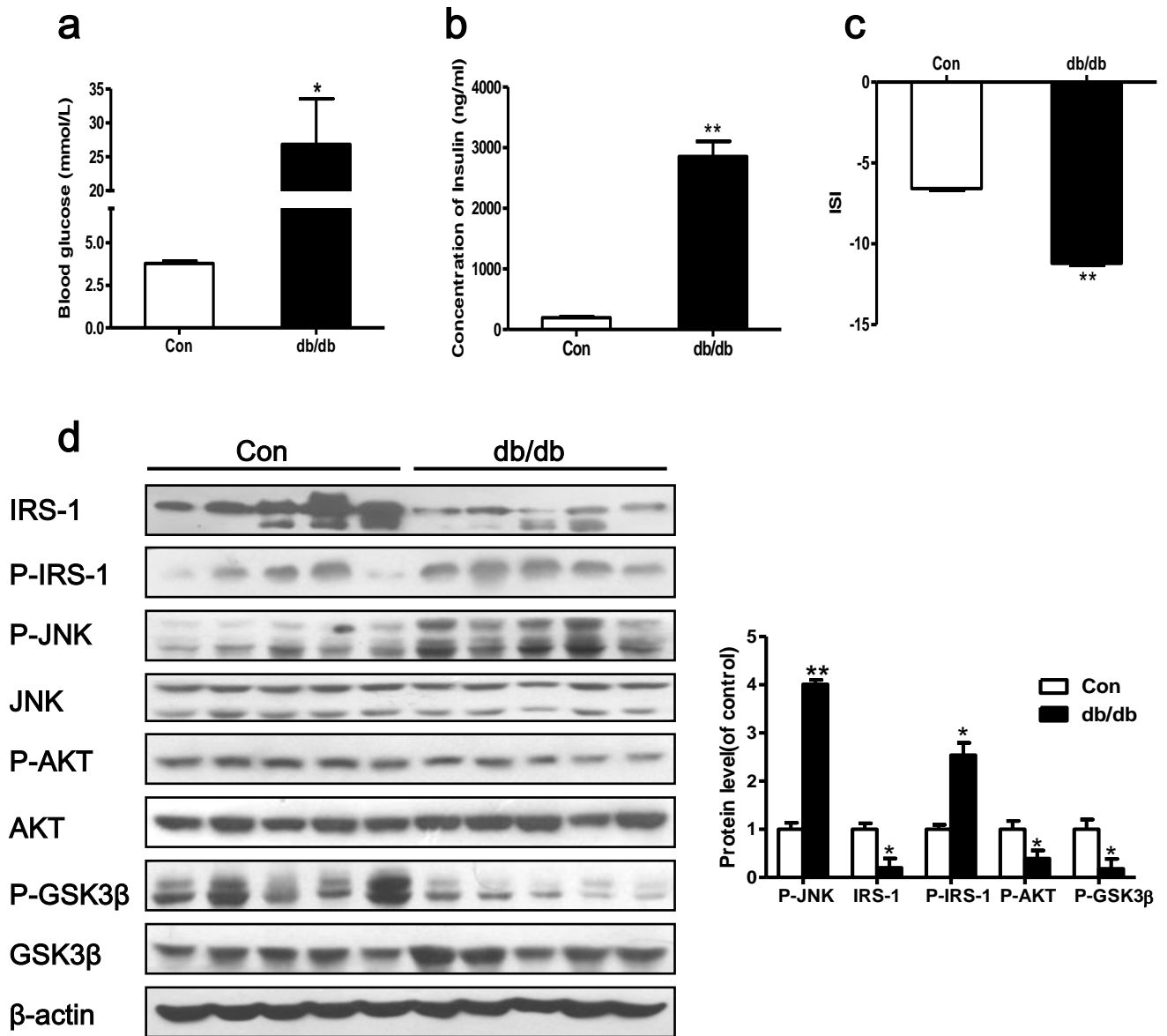


Figure S1. db/db mice exhibited obvious features of insulin resistance. The levels of blood glucose (**a**) and insulin concentration (**b**) were increased, whereas the insulin sensitive index (ISI) was decreased (**c**). The insulin signaling pathway was disordered in the livers of db/db mice, assessed by increased phosphorylation of IRS-1 (ser 307) and JNK, reduced expression of IRS-1 and AKT/GSK3β phosphorylation (**d**). Data represent the means \pm S.D., (n=10 mice *in vivo*). *P<0.05 and **P<0.01 versus control. Con — control.

Supplementary Fig. S2

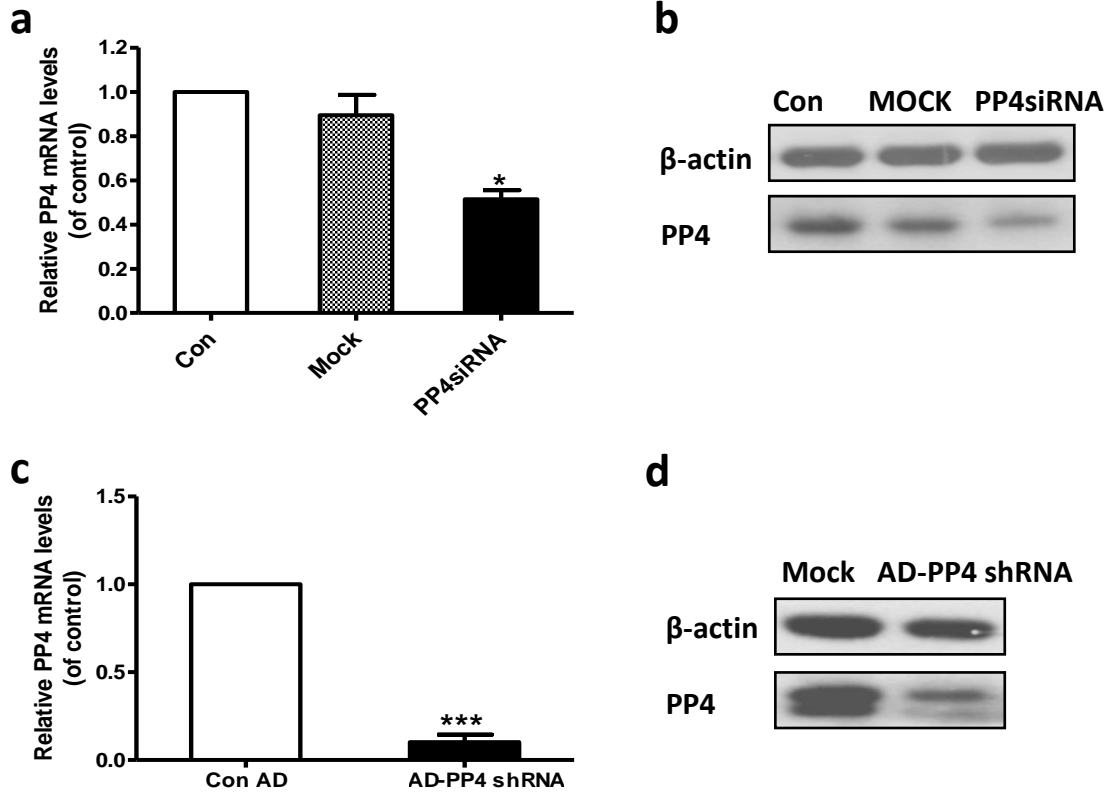


Figure S2. Expression of PP4 was decreased by PP4 siRNA and AD-PP4 shRNA. In order to knock down the expression of PP4, HepG2 cells and mouse primary hepatocytes were transfected with PP4siRNA or AD-PP4 shRNA, respectively. By applying quantitative real-time PCR and western blot analysis, we detected both PP4 siRNA (**a**, **b**) and AD-PP4 shRNA (**c**, **d**) could reduce the expression of PP4. Data represent the means \pm S.D., (n=3 independent experiments in vitro). *P<0.05 and ***P<0.001 versus control. Con – control; Con AD – control AD.

Supplementary Table S1. Oligonucleotides of PP4siRNA

Gene Name	Sequences (5' > 3')
PP4C-728s	GGUACAAGUGGCACUUCATT
PP4C-728as	UGAAGUGCCACUUGUACCTT

Supplementary Table S2. List of oligonucleotide primers for quantitative real-time PCR

Gene Name	Forward primer (F) Reverse primer (R)	Primer' Sequences (5' > 3')
18s rRNA	<i>F</i>	GTAACCCGTTGAACCCCAT
18s rRNA	<i>R</i>	CCATCCAATCGGTAGTAGCG
<i>Human PP4</i>	<i>F</i>	CTGACCCAGAAGACACCACA
<i>Human PP4</i>	<i>R</i>	TGAGCACCGTCTCATTGAAG
<i>Human G6Pase</i>	<i>F</i>	TACGTCCTCTTCCCCATCTG
<i>Human G6Pase</i>	<i>R</i>	CCTGGTCCAGTCTCACAGGT
<i>Human PEPCK</i>	<i>F</i>	GAGAGAACTCCAGGGTGCTG
<i>Human PEPCK</i>	<i>R</i>	CCTTGGAGATGCTGAAAAGC
<i>Human PGC-1α</i>	<i>F</i>	CACCAGCCAACACTCAGCTA
<i>Human PGC-1α</i>	<i>R</i>	ACGTCTTTGTGGCTTTTGCT
<i>Mouse PP4</i>	<i>F</i>	CAATCATGAGAGTCGCCAGA
<i>Mouse PP4</i>	<i>R</i>	GATGGAAGGGGAAAGACCTC
<i>Mouse G6pase</i>	<i>F</i>	TCTGTCCCGGATCTACCTTG
<i>Mouse G6pase</i>	<i>R</i>	GTAGAATCCAAGCGCGAAAC
<i>Mouse Pepck</i>	<i>F</i>	TCTGAGATCTCTGATCCAGACC
<i>Mouse Pepck</i>	<i>R</i>	GAAGTCCAGACCGTTATGCAGC
<i>Mouse Pgc-1α</i>	<i>F</i>	GCCTATGAGCACGAAAGGC
<i>Mouse Pgc-1α</i>	<i>R</i>	TCACACGGCGCTCTTCAATT
<i>Mouse Tnf-α</i>	<i>F</i>	CACAGAAAGCATGATCCGCG
<i>Mouse Tnf-α</i>	<i>R</i>	ACTGATGAGAGGGAGGCCAT