

Supplementary Material

Toll-like Receptor 2 Deficiency Abrogates Diabetic and Obese Phenotypes while Restoring Endothelial Function via Inhibition of NOX1

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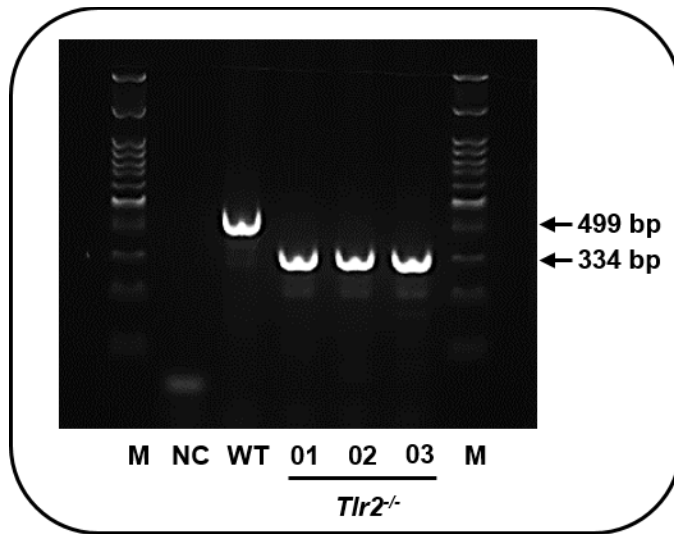


Figure S1: Genotyping of TLR2 knockout mice. Mouse genomic DNA was isolated using buffer containing 100 mM Tris-HCl (pH 8.5), 5 mM EDTA (pH 8.0), 200 mM NaCl and 0.2% SDS. PCR products were separated on 1.5% agarose gel, with a 334 bp band indicating presence of mutant allele. A 499 bp product from PCR indicates wild type (WT) allele.

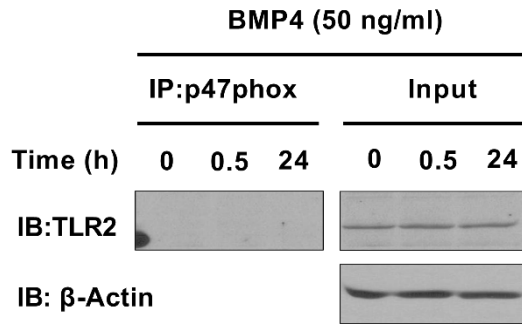


Figure S2: TLR2 does not bind with p47phox in BMP4 treated BAECs. Bovine aortic endothelial cells (BAECs) were treated with BMP4 at 50 ng/ml for 30 min and 24 h, and the cells were harvested for co-immunoprecipitation (co-IP). The co-immunoprecipitated TLR2 by p47phox was determined by western blot analysis, and normalized to β -Actin. Shown are representative co-IP blots of TLR2 by anti-NOX1 immunoprecipitation in BMP4 stimulated endothelial cells, indicating lack of binding between TLR2 and p47phox.

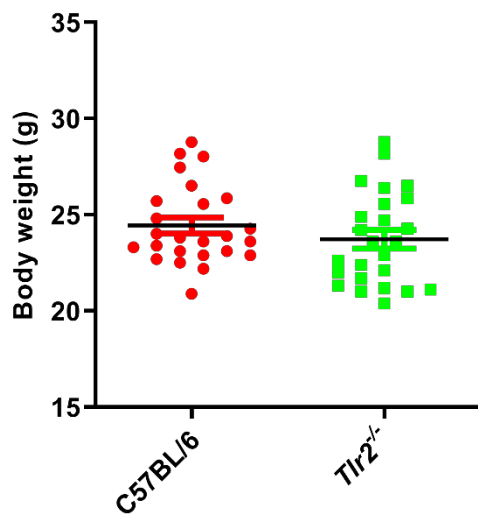


Figure S3: Body weight is not different between WT and TLR2 knockout mice at baseline.

Body weights of WT and TLR2 knockout mice was measured at 8 weeks old. Data are shown as Mean \pm SEM; n = 25.

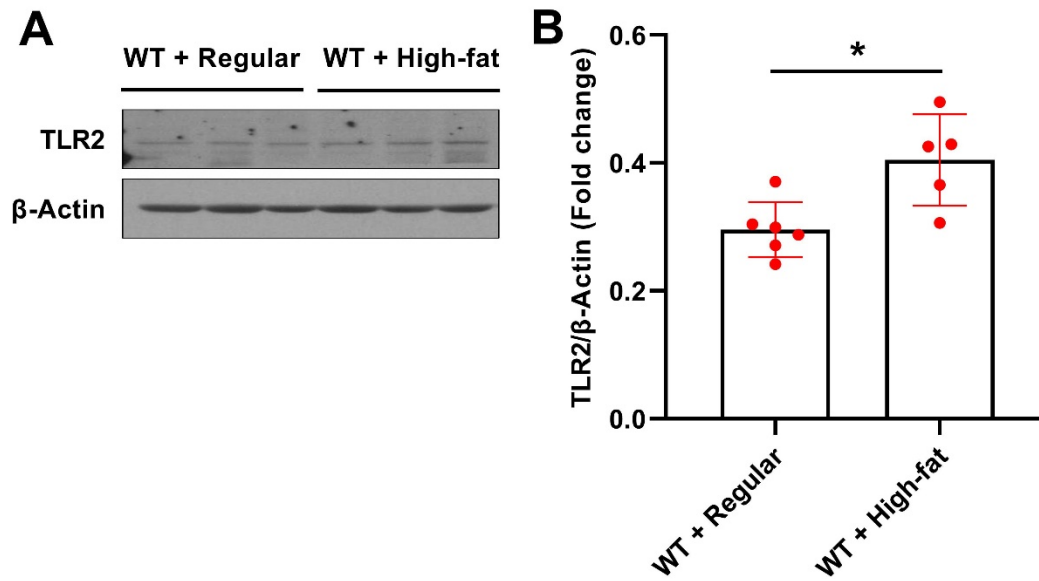


Figure S4: High-fat diet induces upregulation of aortic TLR2 protein expression in mouse aortas. Eight weeks old wild-type (WT) mice were fed high-fat diet for 6 weeks. **(A)** Representative western blots of TLR2/β-Actin protein expression in high-fat fed WT mice. **(B)** Grouped densitometric data of TLR2 protein expression from (A) (n = 5-6). Data are shown as Mean ± SEM. *p<0.05.