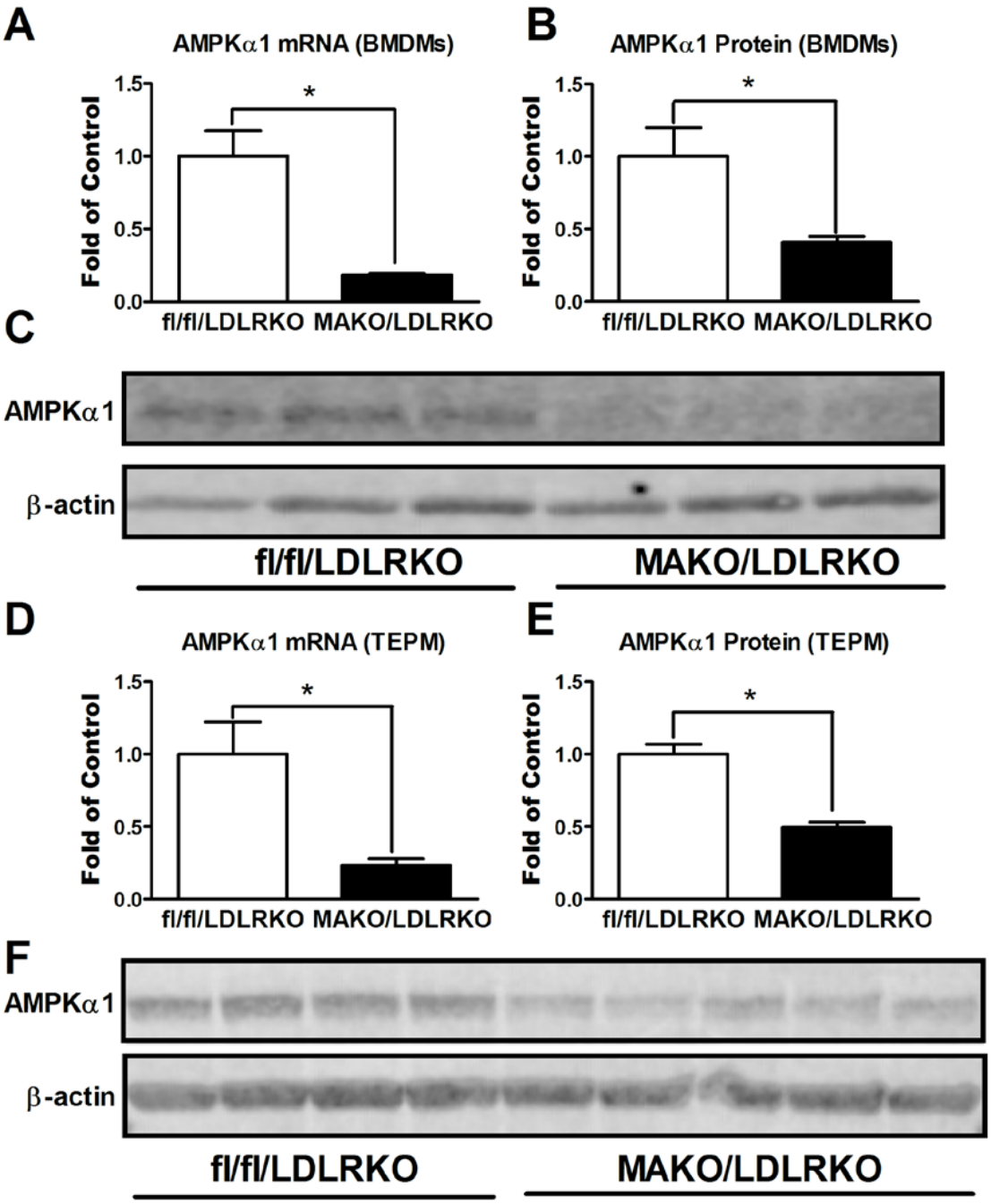
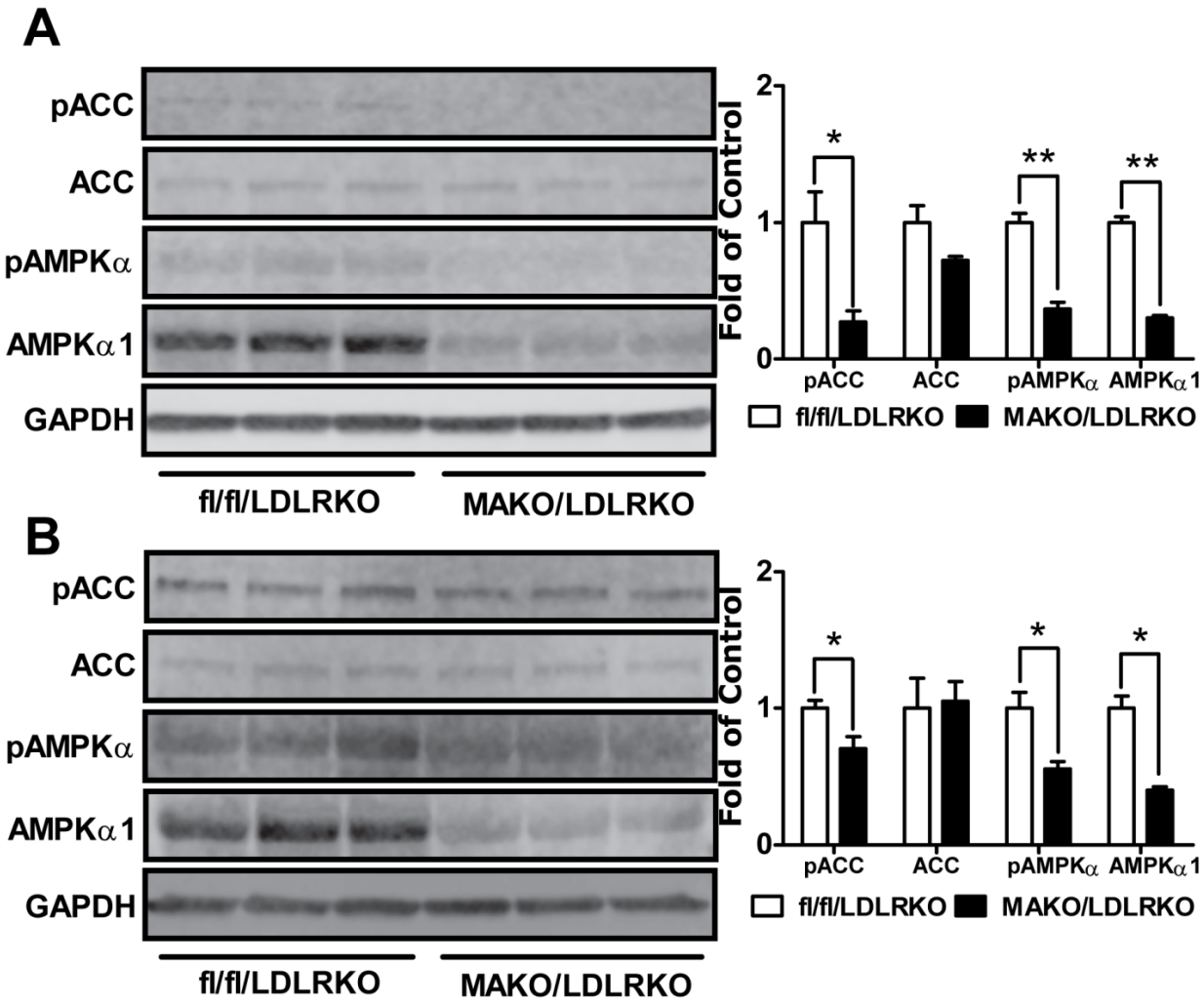
SUPPLEMENTARY DATA

Supplementary Figure 1. Generation of MAKO/LDLRKO mice with myeloid-specific deletion of α1AMPK in LDLRKO mice. (A)-(C) The α1AMPK mRNA (A) and protein (B and C) levels are significantly decreased in BMDMs. (D)-(F) The α1AMPK mRNA (d) and protein (E and F) levels are significantly decreased in thioglycolate-elicited peritoneal macrophages (TEPMs). Peritoneal and bone marrow-derived macrophages (BMDMs) were isolated and cultured as described in the Methods. mRNA and protein levels were measured by real time RT-PCR and immunoblotting, respectively. Data are expressed as mean ± SEM, n=6-8 * p< 0.05 vs. fl/fl/LDLKO control.



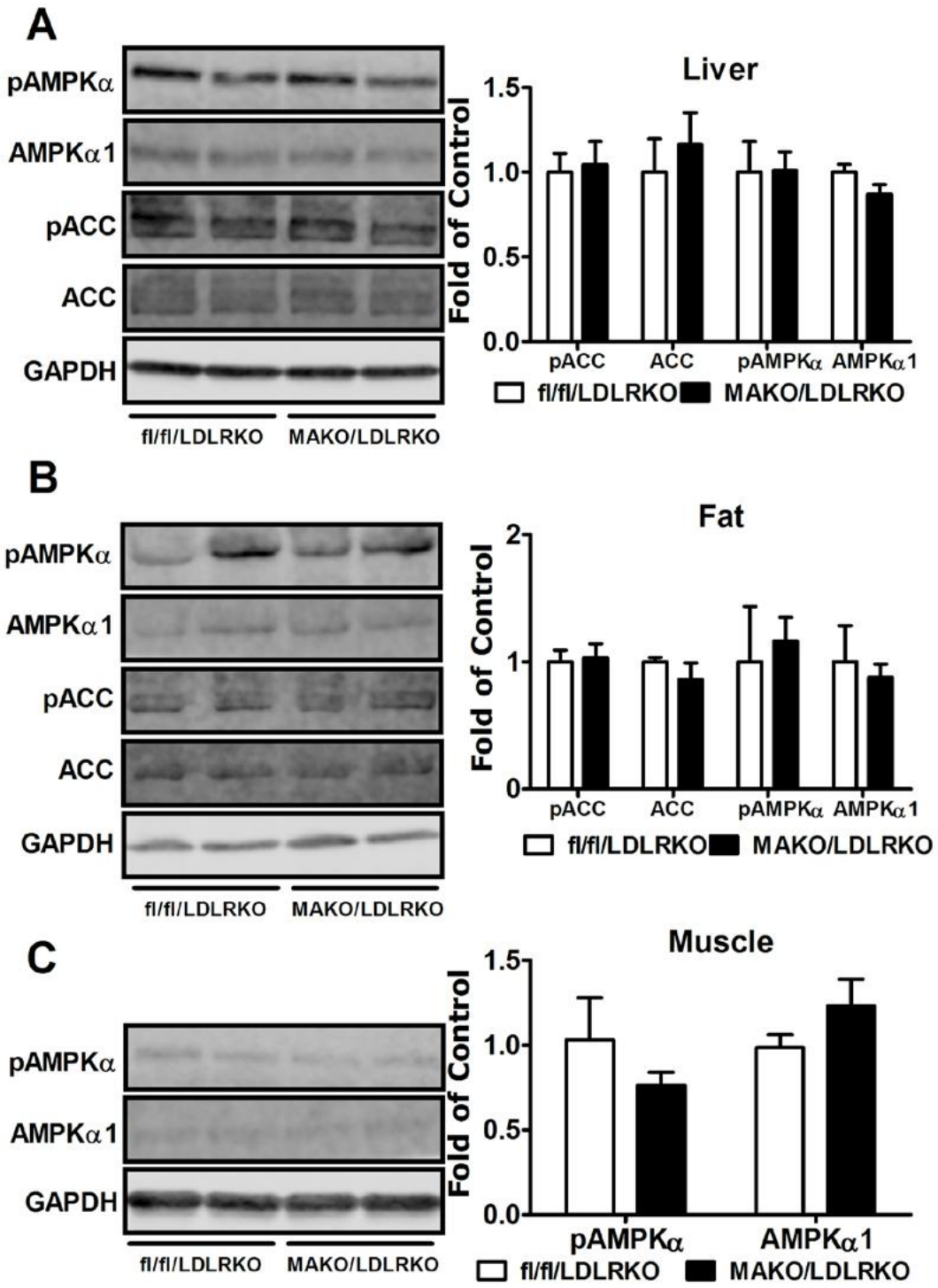
SUPPLEMENTARY DATA

Supplementary Figure 2. Myeloid deletion of α1AMPK suppresses AMPK signaling. Myeloid deletion of α1AMPK suppresses basal phosphorylation of αAMPK and ACC (A) and AICAR stimulated phosphorylation of αAMPK and ACC (B). Blots are shown on left panel and quantitation is shown on right panel in each figure. BMDMs were isolated and cultured as described in the Methods. Protein levels were measured by immunoblotting. Data are expressed as mean ± SEM, n=3 * p< 0.05 vs. fl/fl/LDLKO control.



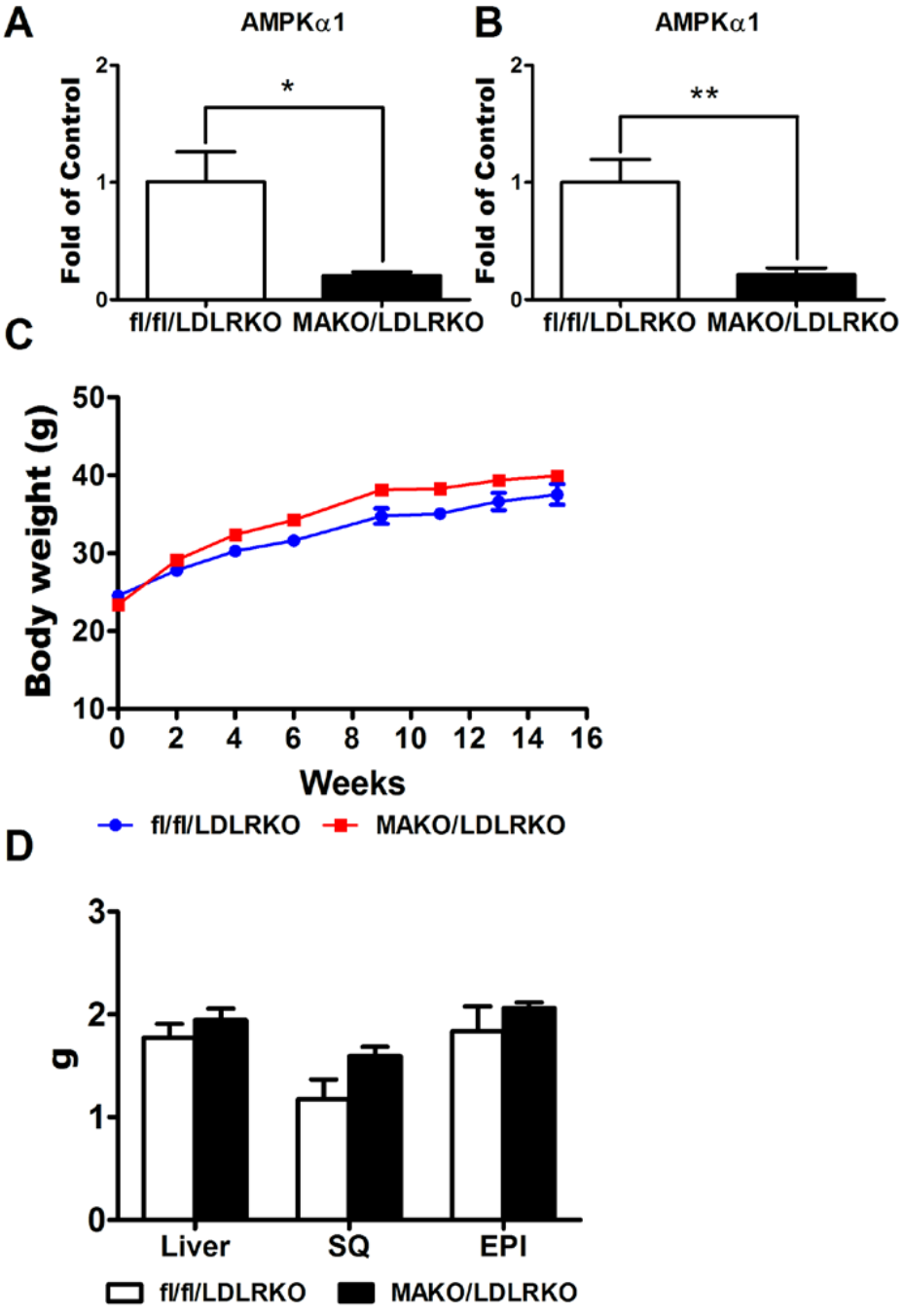
SUPPLEMENTARY DATA

Supplementary Figure 3. Myeloid deletion of α1AMPK does not alter α1AMPK expression and signaling in liver (A), fat (B) and skeletal muscle (C). Tissues were dissected from MAKO/LDLRKO and control mice and processed into lysates. Protein levels were measured by immunoblotting.



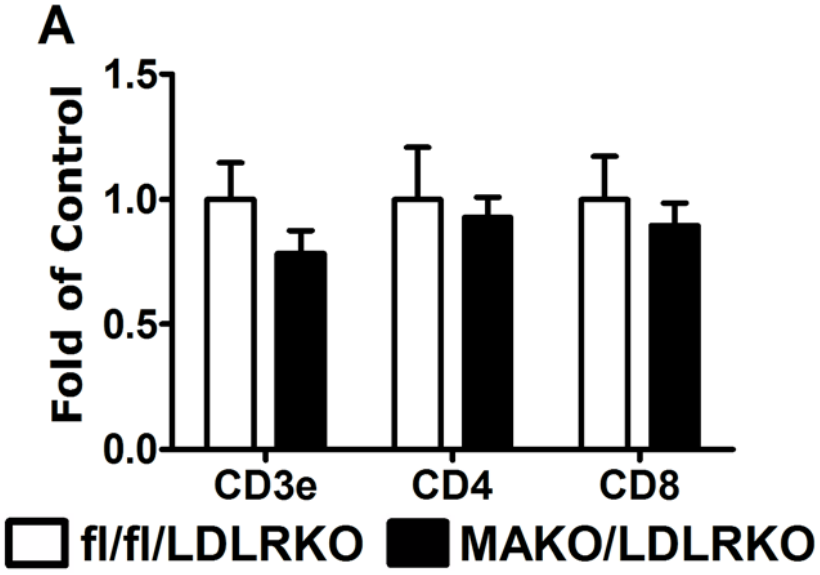
SUPPLEMENTARY DATA

Supplementary Figure 4. Myeloid deletion of α1AMPK decreased α1AMPK mRNA expression in peritoneal macrophages (A) and aorta macrophages (B) from MAKO/LDLRKO mice fed with an atherogenic diet for 16 weeks. (C) Myeloid deletion of α1AMPK does not change body weight (C) and fat pad mass (D). 8-week old male MAKO/LDLRKO and control mice were fed with an atherogenic diet for 16 weeks. Peritoneal and aorta macrophages were isolated as described in Method. mRNA and protein levels were measured by real time RT-PCR and immunoblotting, respectively. Body weights (C) were measured weekly and fat pads (D) were dissected and weighted at the end of study. Data are expressed as mean ± SEM, n=6-8.



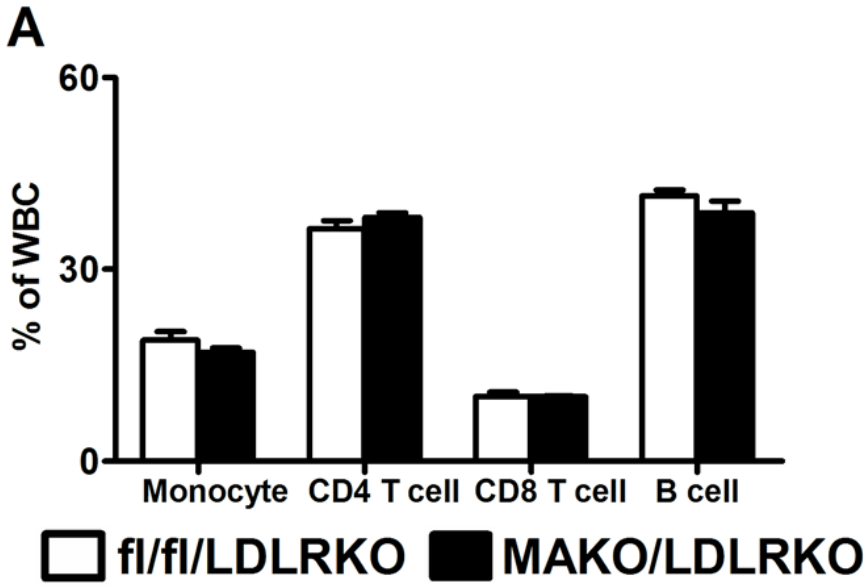
SUPPLEMENTARY DATA

Supplementary Figure 5. Myeloid deletion of α1AMPK does not change the expression of T cell markers in aortas of MAKO/LDLRKO mice. The whole aortas were dissected and used for total RNA extraction. Gene expression was measured by real-time RT-PCR and normalized to cyclophilin. Data are expressed as mean ± SEM, n=6-8.



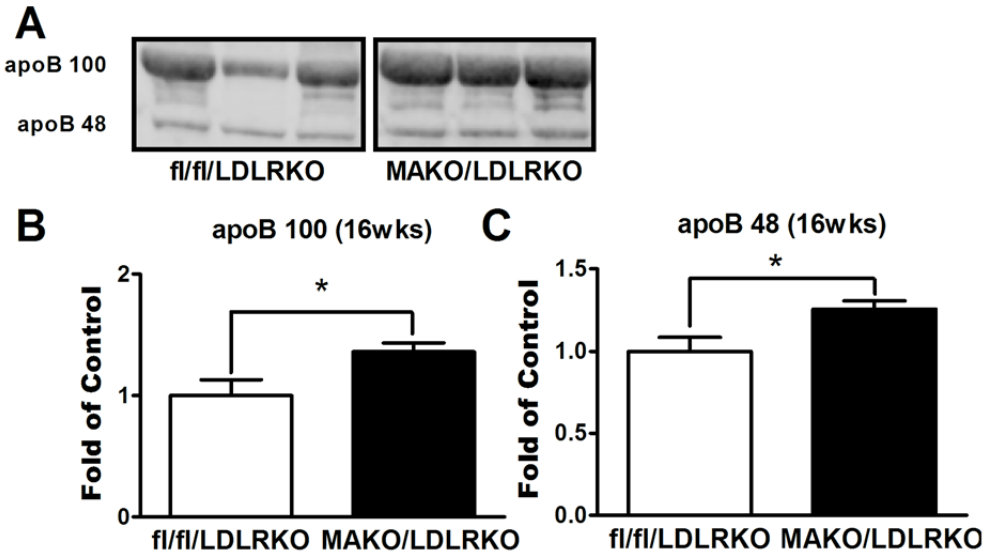
SUPPLEMENTARY DATA

Supplementary Figure 6. Myeloid deletion of α1AMPK does not change the composition of major immune cells in circulation in MAKO/LDLRKO mice. Eight-week old male MAKO/LDLRKO and control mice were fed with an atherogenic diet for 16 weeks. Immune cell composition was analyzed using flow cytometry as we previously described in the Methods. Data are expressed as mean ± SEM, n=6-8.



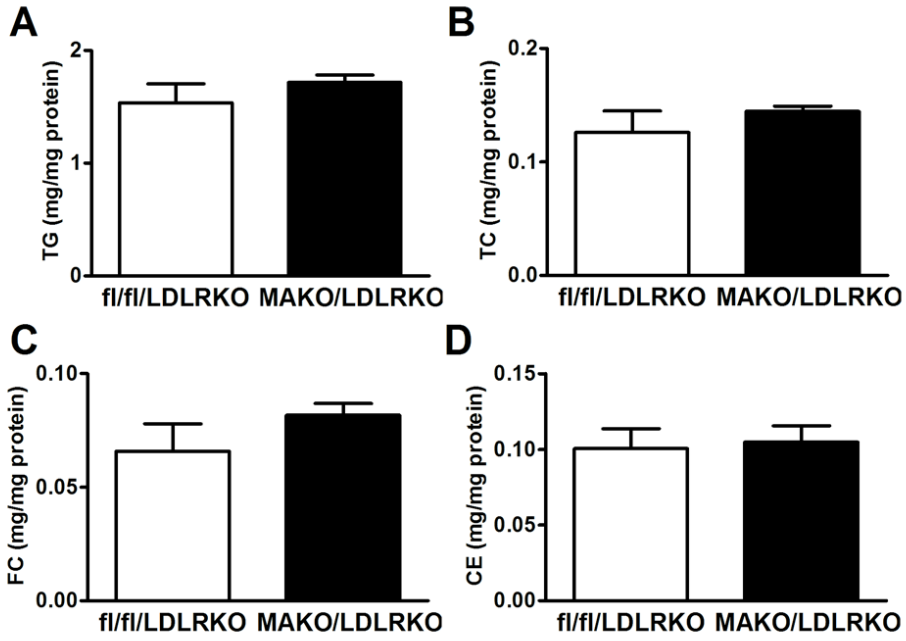
SUPPLEMENTARY DATA

Supplementary Figure 7. Myeloid deletion of α1AMPK up-regulates plasma apoB levels in MAKO/LDLRKO mice fed with an atherogenic diet for 16 weeks. Plasma apoB protein levels were measured by immunoblotting. Data are expressed as mean ± SEM, n=6-8 * p< 0.05 vs. fl/fl/LDLKO control.



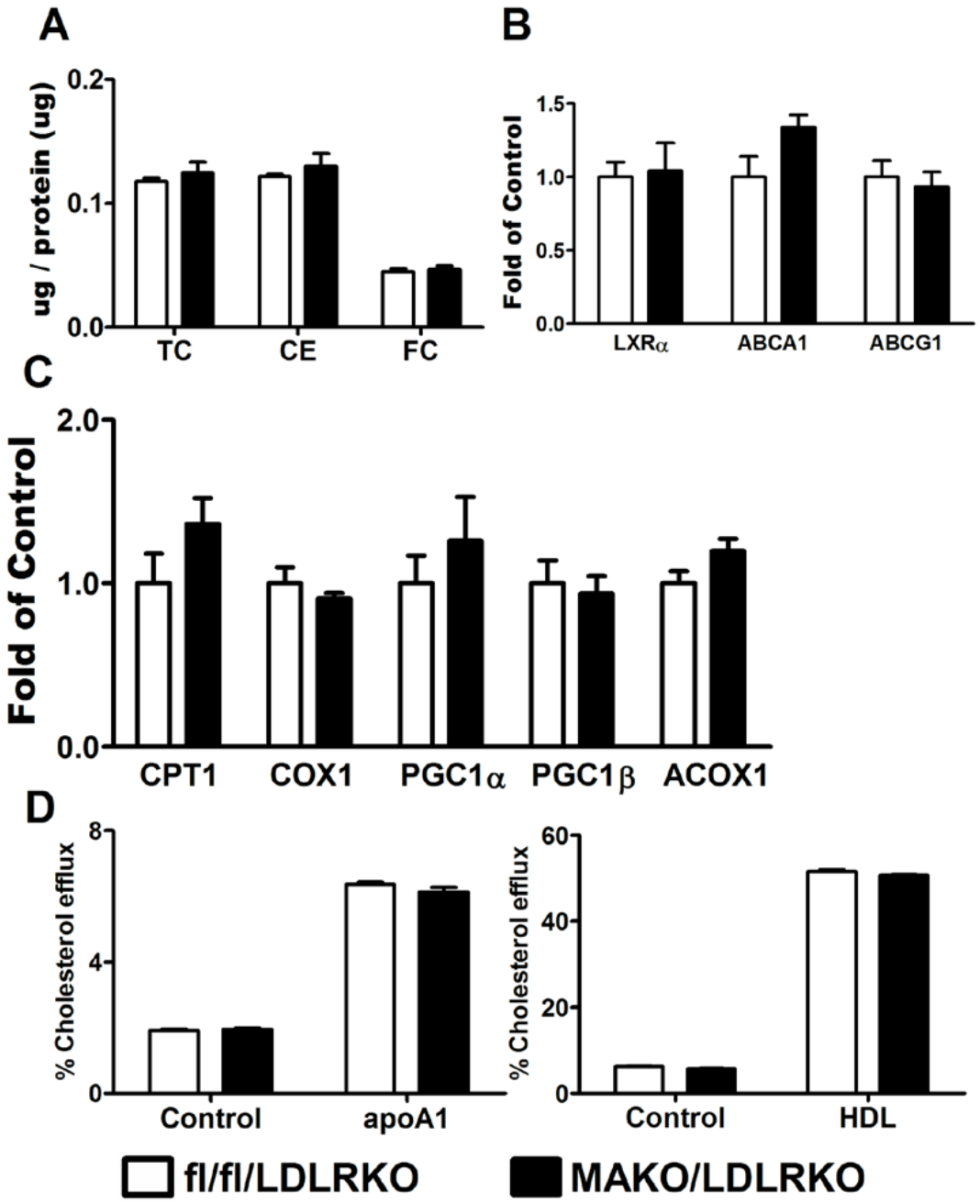
SUPPLEMENTARY DATA

Supplementary Figure 8. Myeloid deletion of α1AMPK does not change hepatic lipid content of MAKO/LDLRKO mice, including TG (A), TC (B), FC (C), and CE (D). Eight-week old male MAKO/LDLRKO and control mice were fed with an atherogenic diet for 16 weeks. Hepatic lipids were measured as described in the Methods. Data are expressed as mean ± SEM, n=6-8.



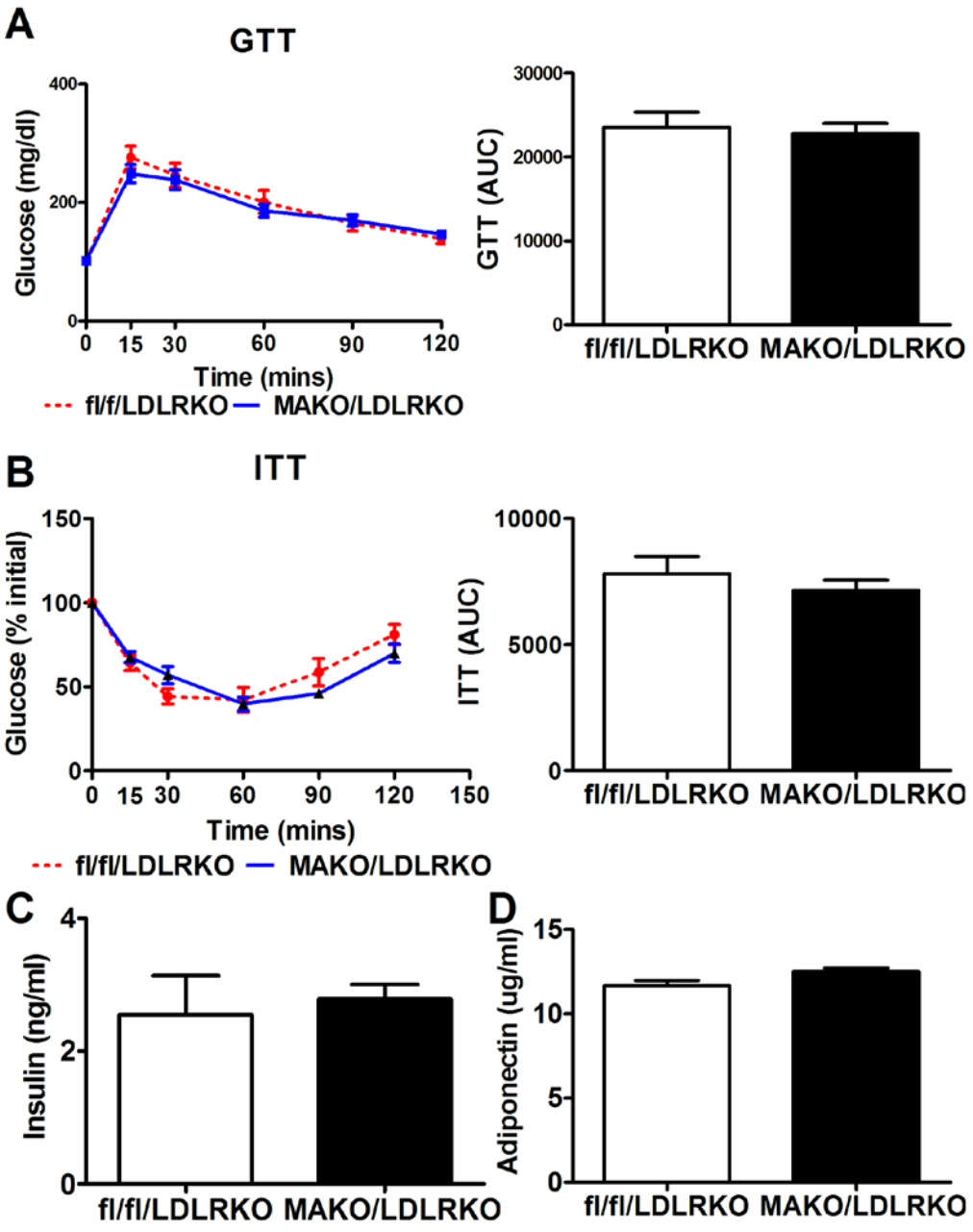
SUPPLEMENTARY DATA

Supplementary Figure 9. Myeloid deletion of α1AMPK does not change lipid metabolism in peritoneal macrophages of MAKO/LDLRKO mice. Myeloid deletion of α1AMPK does not change peritoneal macrophage: (A) cholesterol content, (B) expression of genes involved in cholesterol transport, (C) expression of genes involved in fatty acid oxidation, and (D) cholesterol efflux to apoA1 and HDL. Data are expressed as mean ± SEM, n=6-8.



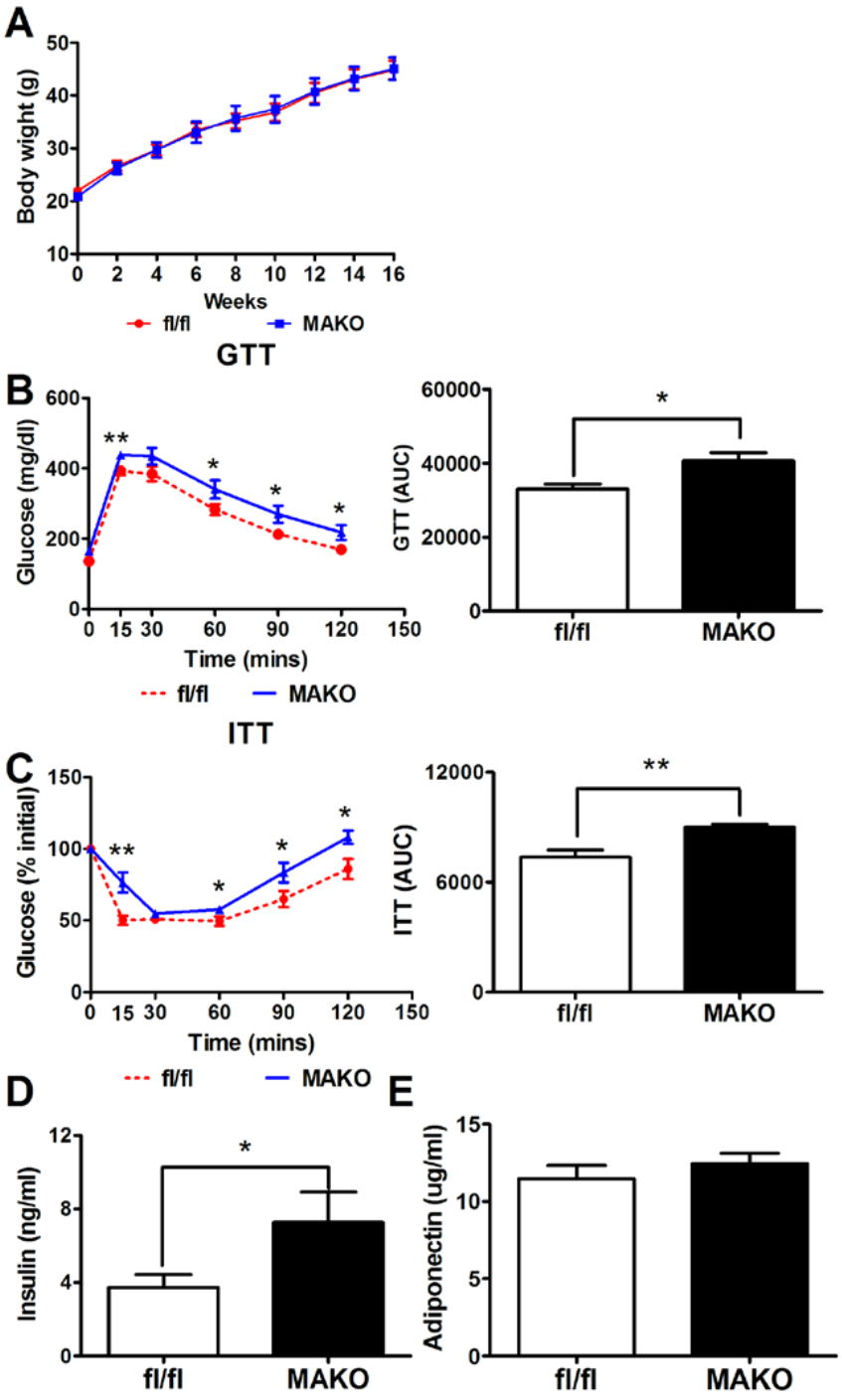
SUPPLEMENTARY DATA

Supplementary Figure 10. Myeloid deletion of α1AMPK does not affect insulin sensitivity of MAKO/LDLRKO mice fed with an atherogenic diet. Myeloid deletion of α1AMPK does not affect glucose tolerance assessed by GTTs (A, right panel: Area Under Curve), insulin sensitivity assessed by ITTs (B, right panel: Area Under Curve), fed insulin levels (C), and adiponectin levels (D). 8-week old male MAKO/LDLRKO and control mice were fed with an atherogenic diet for 16 weeks. Metabolic measures including GTTs, ITTs, insulin and adiponectin levels were conducted as described in Methods. Data are expressed as mean ± SEM, n=6-8.



SUPPLEMENTARY DATA

Supplementary Figure 11. Myeloid deletion of α1AMPK causes insulin resistance in MAKO mice fed with a high-fat diet. (A) MAKO mice showed no change in body weight. However, MAKO mice displayed increased glucose intolerance assessed by GTTs (A, right panel: Area Under Curve), decreased insulin sensitivity assessed by ITTs (B, right panel: Area Under Curve), increased fed insulin levels (C), with no change in adiponectin levels (D). MAKO mice were generated as described in Method. 6-week old male MAKO and their littermate control fl/fl mice were fed with a high-fat diet for 16 weeks. Metabolic measures including GTTs, ITTs, insulin and adiponectin levels were conducted as described in Methods. Data are expressed as mean ± SEM, n=8, * p< 0.05 or p<0.01 vs. fl/fl control.



SUPPLEMENTARY DATA

Supplementary Figure 12. Myeloid deletion of α1AMPK increases expression of IL6 and TNFα in adipose tissue of MAKO mice fed with a high-fat diet. 6-week old male MAKO and their littermate control fl/fl mice were fed with a high-fat diet for 16 weeks. Gene expression was measured by real-time RT-PCR and normalized to cyclophilin. Data are expressed as mean ± SEM, n=8, * p< 0.05 vs. fl/fl control.