



Supplementary Figure 3. *Atg16l1* knockout promotes macrophage inflammatory responses, glycolysis, and mitochondrial metabolism disorders. Proinflammatory gene expression in activated *Atg16l1^{fl/fl}* and *Atg16l1^{ΔMφ}* BMDMs treated with CM from PAOA-stimulated hepatocytes. (B) ELISA results showing the levels of TNF-α, IL-6, and IL-1β in the supernatants of activated *Atg16l1^{fl/fl}* and *Atg16l1^{ΔMφ}* BMDMs treated with CM from PAOA-stimulated hepatocytes. (C) The extracellular acidification rate (ECAR) of stimulated *Atg16l1^{fl/fl}* and *Atg16l1^{ΔMφ}* BMDMs was assessed by a Seahorse assay; n=3/group. (D1 and D2) The oxygen consumption rate (OCR) of stimulated *Atg16l1^{fl/fl}* and *Atg16l1^{ΔMφ}* BMDMs was assessed by a Seahorse assay before and after the sequential addition of oligomycin, FCCP, rotenone, and antimycin; n=3/group. (E) The protein expression levels of ATG16L1 in liver tissues, hepatocytes and BMDMs from *Atg16l1^{fl/fl}* and *Atg16l1^{OE}* mice. (F) Schematic diagram showing that primary hepatocytes with or without PAOA stimulation isolated from *Atg16l1^{fl/fl}* mice were cocultured with LPS-primed primary BMDMs from *Atg16l1^{fl/fl}* and *Atg16l1^{OE}* mice. (G) The expression of proinflammatory genes in activated *Atg16l1^{fl/fl}* and *Atg16l1^{OE}* BMDMs treated with CM from PAOA-stimulated hepatocytes. (H) ELISA results showing the levels of TNF-α, IL-6, and IL-1β in the supernatants of activated *Atg16l1^{fl/fl}* and *Atg16l1^{OE}* BMDMs treated with CM from PAOA-stimulated hepatocytes. (I) The ECAR of stimulated *Atg16l1^{fl/fl}* and *Atg16l1^{OE}* BMDMs was assessed by a Seahorse assay; n=3/group. (J1, J2) The OCRs of stimulated *Atg16l1^{fl/fl}* and *Atg16l1^{OE}* BMDMs were assessed by a Seahorse assay before and after the sequential addition of oligomycin, FCCP, rotenone, and antimycin; n=3/group. (K) The expression of genes that affect the IFN-I response. (L) Western blotting was performed to analyze the expression levels of the inflammatory proteins p-p65 (Ser536), p65, p-IkBa, IkBa, TNF-α, IL-6, cleaved IL-1β, and pro-IL-1β in stimulated *Atg16l1^{fl/fl}* and *Atg16l1^{OE}* BMDMs. ATG16L1, autophagy-related protein 16-like 1; BMDMs, bone marrow-derived macrophages; PAOA, palmitic acid- and oleic acid; CM, conditioned media. The data are expressed as the mean±SD. *P<0.05 (unpaired t test or ANOVA).