

Supplementary information, Figure S3

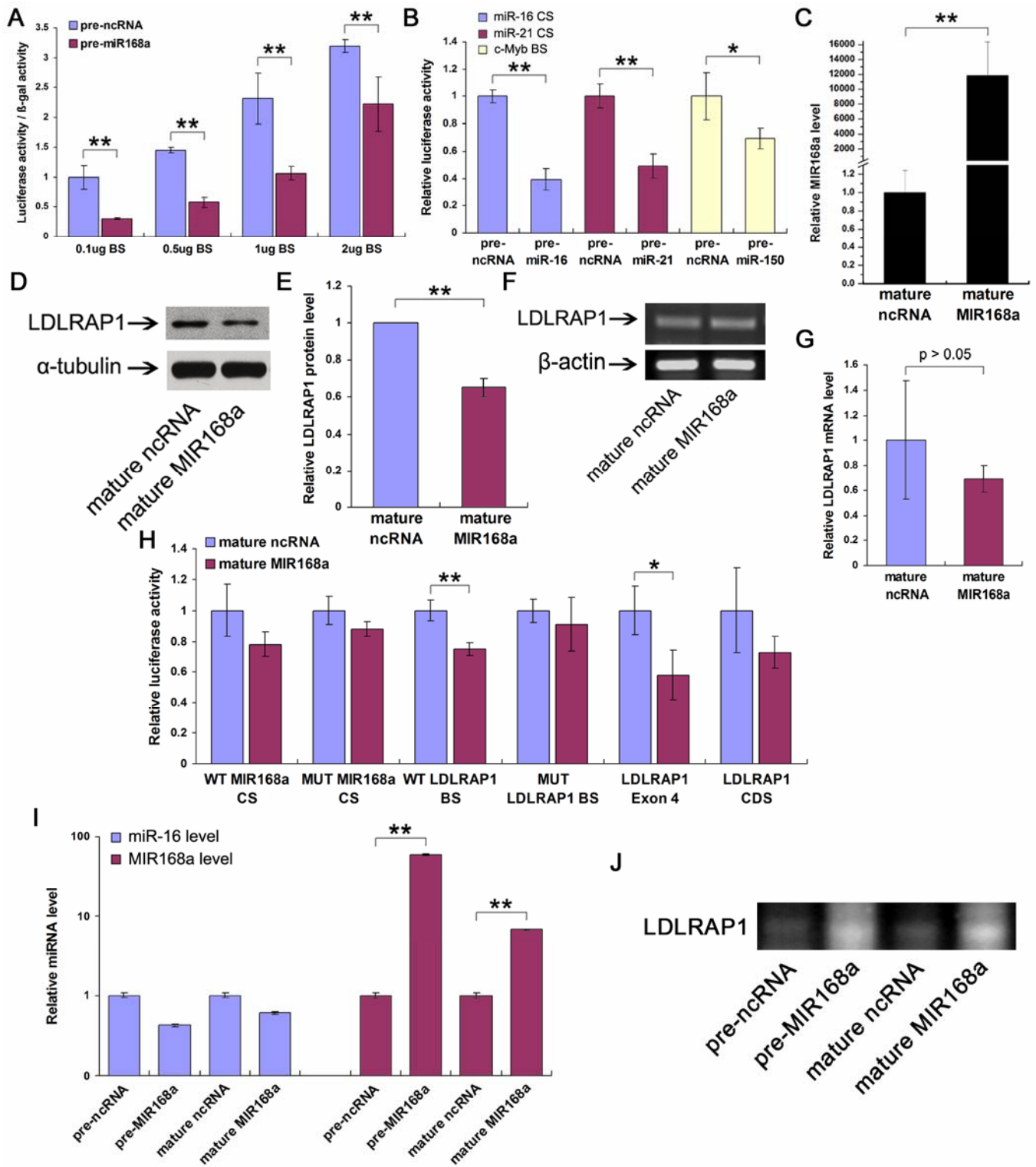


Figure S3 Targeting of LDLRAP1 by MIR168a. **(A)** The correlation between the inhibition rate of MIR168a as a ratio of the levels of MIR168a and binding site (BS)-containing reporter ($n = 3$). **(B)** A luciferase activity reporting assay using endogenous miR-16, miR-21, miR-150 and their binding sites on target genes as controls ($n = 3$). **(C)** qRT-PCR analysis of the levels of MIR168a in mature MIR168a-transfected HepG2 cells ($n = 9$). The HepG2 cells were transfected with $20 \text{ pmol}/10^5$ cells of scrambled mature control oligonucleotides (mature ncRNA) or mature MIR168a. **(D)** Western blot analysis of the levels of LDLRAP1 protein in mature MIR168a-transfected HepG2 cells. **(E)** The quantification of LDLRAP1 protein expression in **D** ($n = 9$). **(F-G)** Semi-quantitative RT-PCR **(F)** and qRT-PCR **(G)** analysis of the levels of LDLRAP1 mRNA in mature MIR168a-transfected HepG2 cells ($n = 5$). **(H)** Luciferase activities in HepG2 cells co-transfected with luciferase reporters described previously and mature MIR168a or ncRNA ($n = 9$). **(I)** The association of MIR168a with AGO2 in HepG2 cells transfected with pre-MIR168a or mature MIR168a ($n = 3$). HepG2 cells were transfected with $20 \text{ pmol}/10^5$ cells of pre-ncRNA, pre-MIR168a, mature ncRNA, or mature MIR168a. The MIR168a in anti-AGO2 immunoprecipitated products was detected by qRT-PCR, with the level of miR-16 serving as the control. **(J)** The levels of LDLRAP1 mRNA associated with AGO2 in HepG2 cells transfected with pre-MIR168a or mature MIR168a. The LDLRAP1 mRNA in anti-AGO2 immunoprecipitated products was detected by semi-quantitative RT-PCR. $*P < 0.05$; $**P < 0.01$.