

Supplementary Figure Legends

Figure S1. miRNAs microarray profiling analysis demonstrates differential expression of miRNAs in mucosal melanoma (MM) tissues and normal mucosal nevi. (A) Hierarchical clustering heat map based on statistically significant dysregulated miRNAs in 10 MM as compared to three normal mucosal nevi. The miRNAs with a fold change ≥ 2 and P -value ≤ 0.05 were regarded as significantly different. (B) Volcano plot of significantly dysregulated miRNAs in MM tissues compared to mucosal nevi tissues.

Figure S2. The effects of overexpression of miR-23a-3p on mucosal melanoma cells. (A) HMOVII and VMRC-MELG cells were transfected with miR-23a-3p mimic or control (miR-Ctrl), and miR-23a-3p expression was quantified by qRT-PCR at 48 h after transfection. (B) HMOVII and VMRC-MELG cells were infected with lentiviral Lenti-pMIR-23a or Lenti-pMIR-Ctrl, and the miR-23a-3p expression was quantified at 48 h after infection. (C) VMRC-MELG cells were stained with PI at 24 and 48 h after transfection with miR-23a-3p mimic or miR-Ctrl, and cell cycle distribution was analyzed by flow cytometry. (D) Cell apoptotic death events were measured by Annexin V-FITC/PI staining and flow cytometry at 24 h after transfection with miR-23a-3p mimic or miR-Ctrl. $*P < 0.05$, $**P < 0.01$, $***P < 0.001$.

Figure S3. Expression of ADCY1 in mucosal melanoma fresh-frozen tissues and cell lines. (A) ADCY1 mRNA and miR-23a-3p expression was measured by qRT-PCR in 20 fresh-frozen MM tissues and was normalized to the mean level in normal mucosal nevi, and ADCY1 mRNA levels were significantly higher in MM tissues with low miR-23a-3p expression than in those with high miR-23a-3p expression. (B) ADCY1 mRNA expression was measured by qRT-PCR in three MM cell lines, and was normalized to the mean level in HEMs. GAPDH was used as an endogenous control. $*P < 0.05$, $**P < 0.01$, $***P < 0.001$.

Figure S4. ADCY1 knockdown regulates EMT-related markers in mucosal melanoma. (A) HMOVII and VMRC-MELG cells were transfected with two ADCY1 siRNAs, and ADCY1 mRNA expression was quantified by qRT-PCR at 48 h after transfection. (B) HMOVII and VMRC-MELG cells were stably transfected with ADCY1-specific shRNA, and ADCY1 mRNA expression was quantified by qRT-PCR at 96 h after transfection. (C) Western blot analysis of EMT-related markers in cells transfected with two ADCY1 siRNAs.

Figure S1

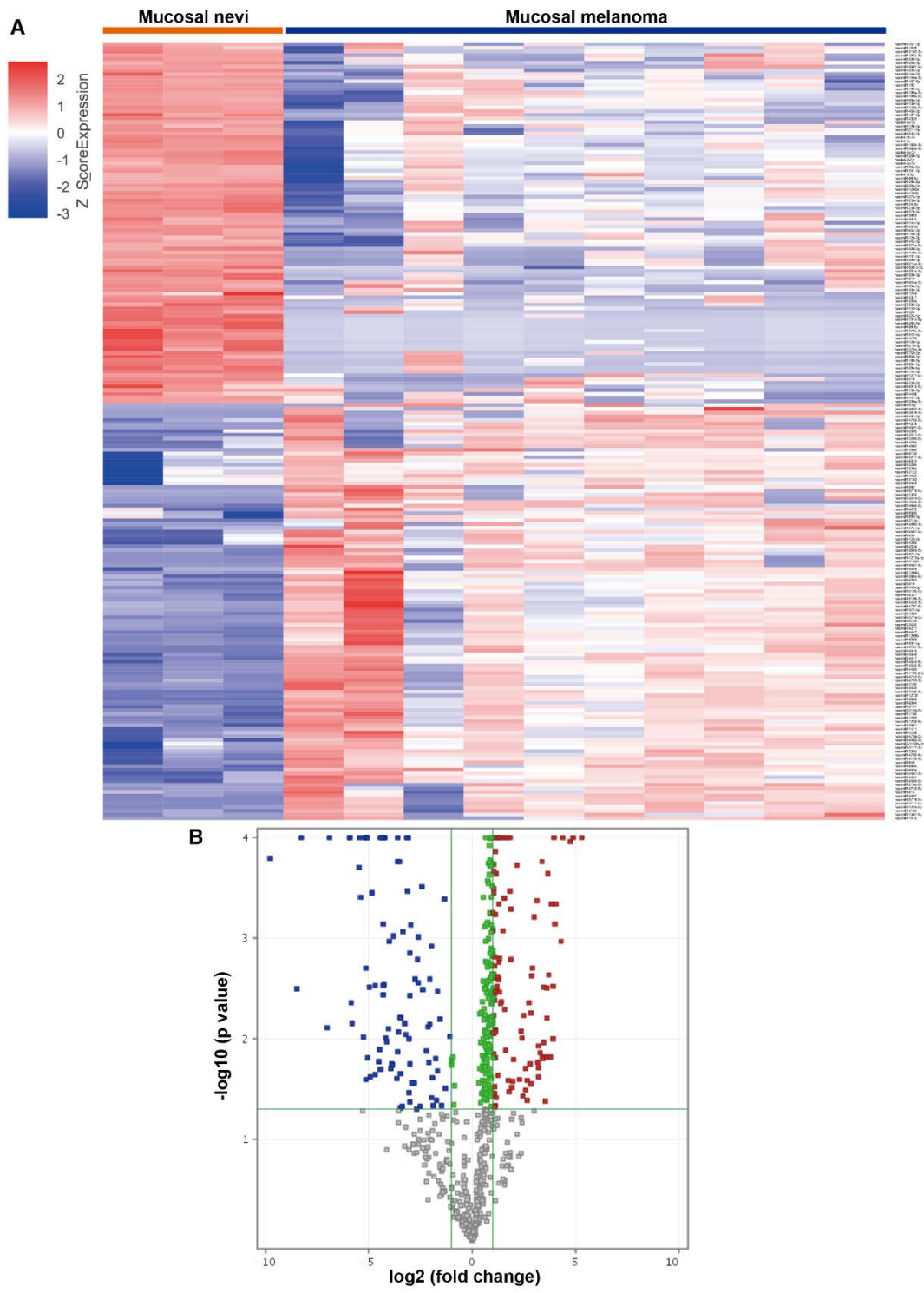


Figure S2

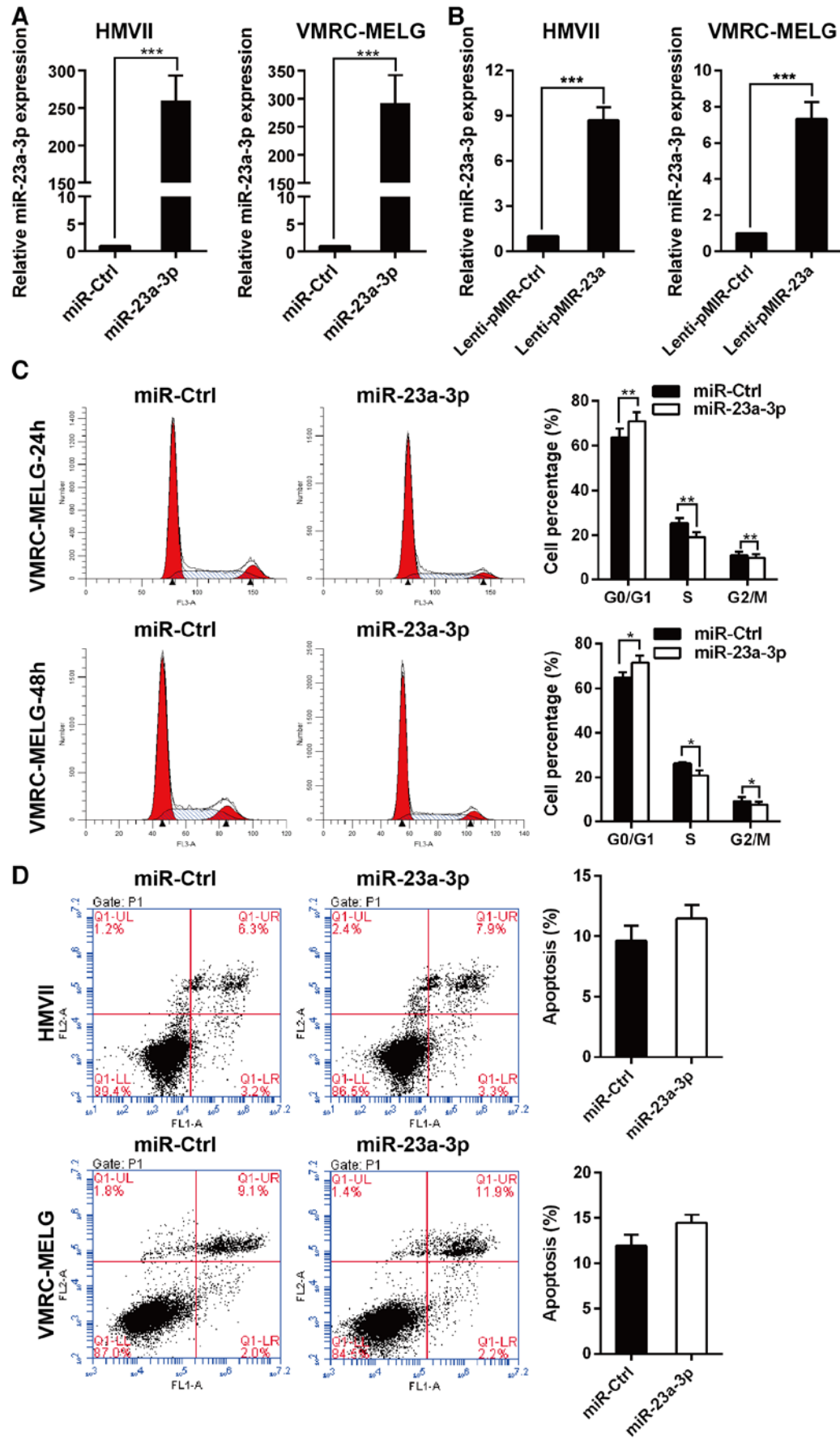


Figure S3

