

Allele Specific MicroRNA Mediated Regulation of a Glycolysis Gatekeeper PDK1 in Cancer Metabolism

Sugarniya Subramaniam, Varinder Jeet, Jennifer H. Gunter, Judith A. Clements and Jyotsna Batra

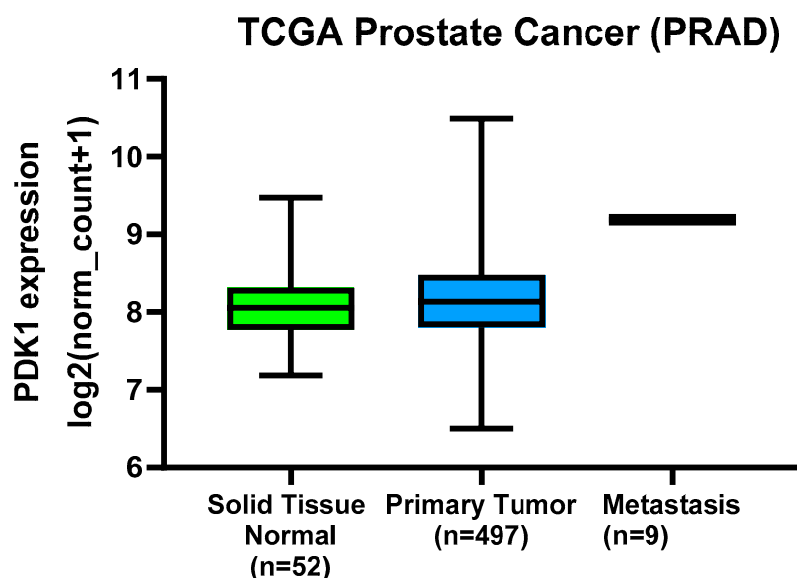


Figure S1. Gene expression of PDK1 in solid tissue normal, primary tumor and metastasis from The Cancer Genome Atlas (TCGA) dataset.

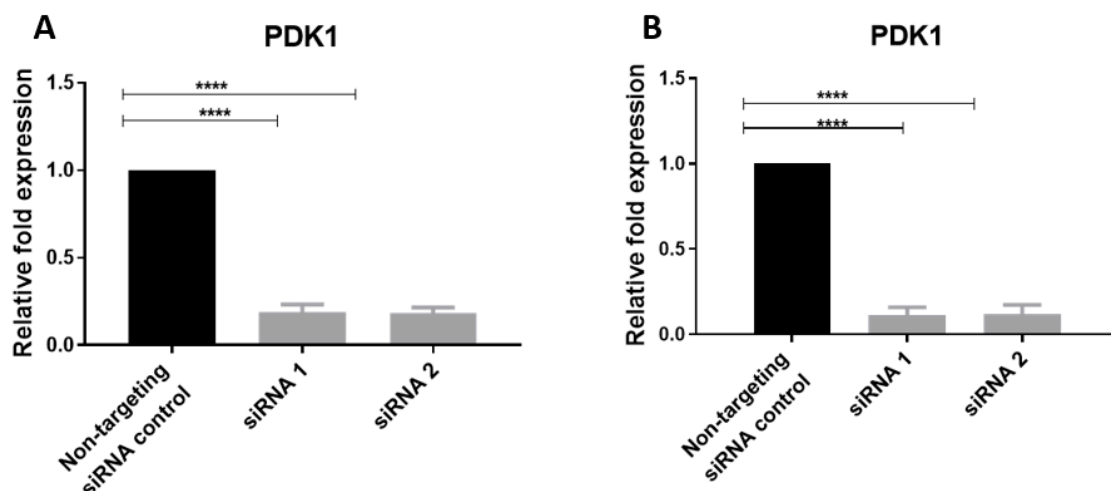
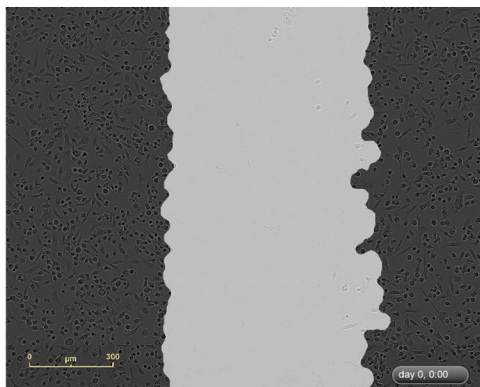
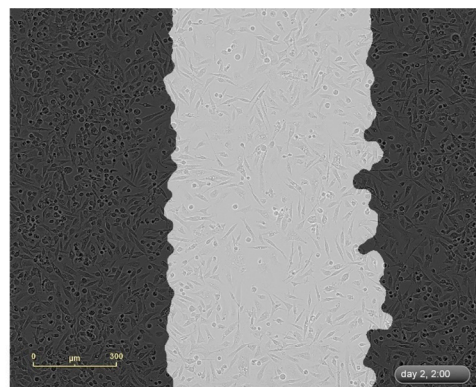


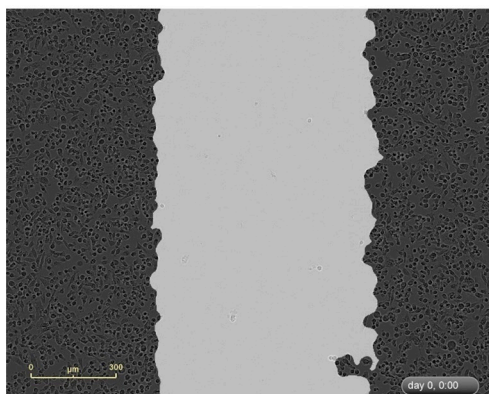
Figure S2. Efficiency screening of PDK1 knockdown in prostate cancer cells. (A) LNCaP cells showed 80% knockdown efficiency with siRNAs 1 and 2; (B) PC3 cells exhibited 90% knockdown efficiency at the mRNA level. Data presented as Mean \pm SEM, $n = 3$ (biological replicates), One-way ANOVA, Tukey's *post-hoc* test **** $p < 0.0001$.



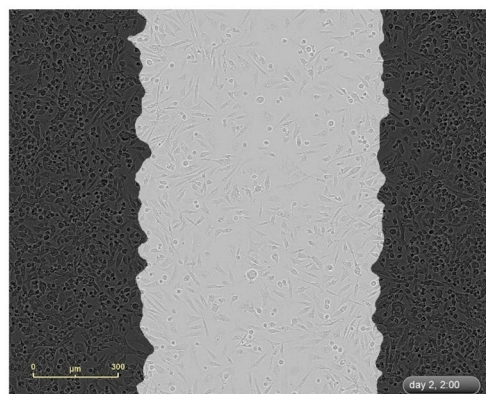
Non-targeting siRNA control 0 hours



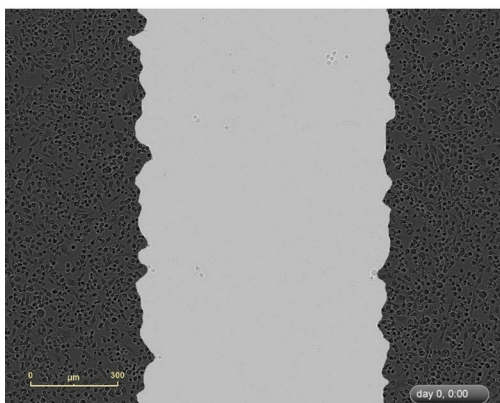
Non-targeting siRNA control 48 hours



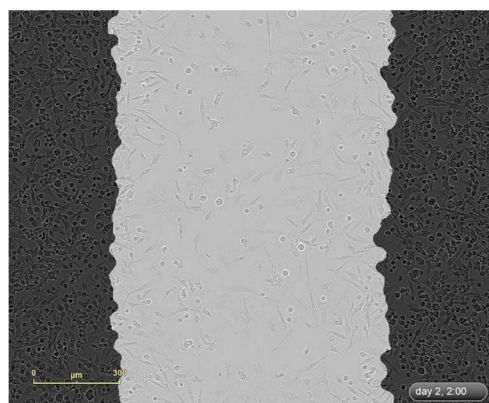
siRNA 1 0 hours



siRNA 1 48 hours

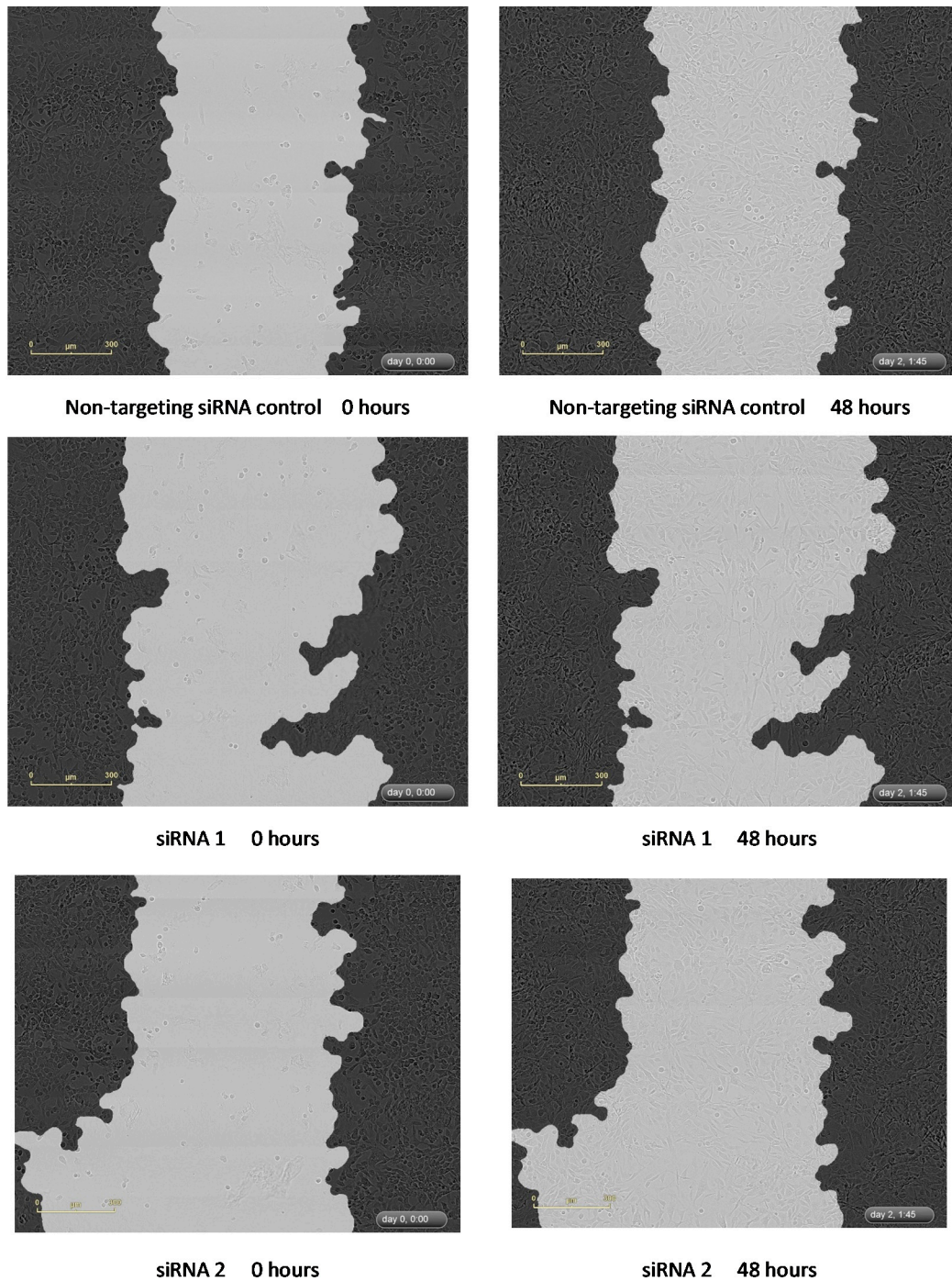


siRNA 2 0 hours



siRNA 2 48 hours

(A)



(B)

Figure S3. PDK1-mediated migration and invasion of prostate cancer cells. Representative images are shown for non-targeting siRNA control cells and siRNA treated cells at time points 0 h and 48 h. (A) PDK1 suppression reduced the number of percentage of migrating cells in the PC3 wound closure. (B) The percentage of cells that invaded through the wounded area reduced with PDK1 suppression.

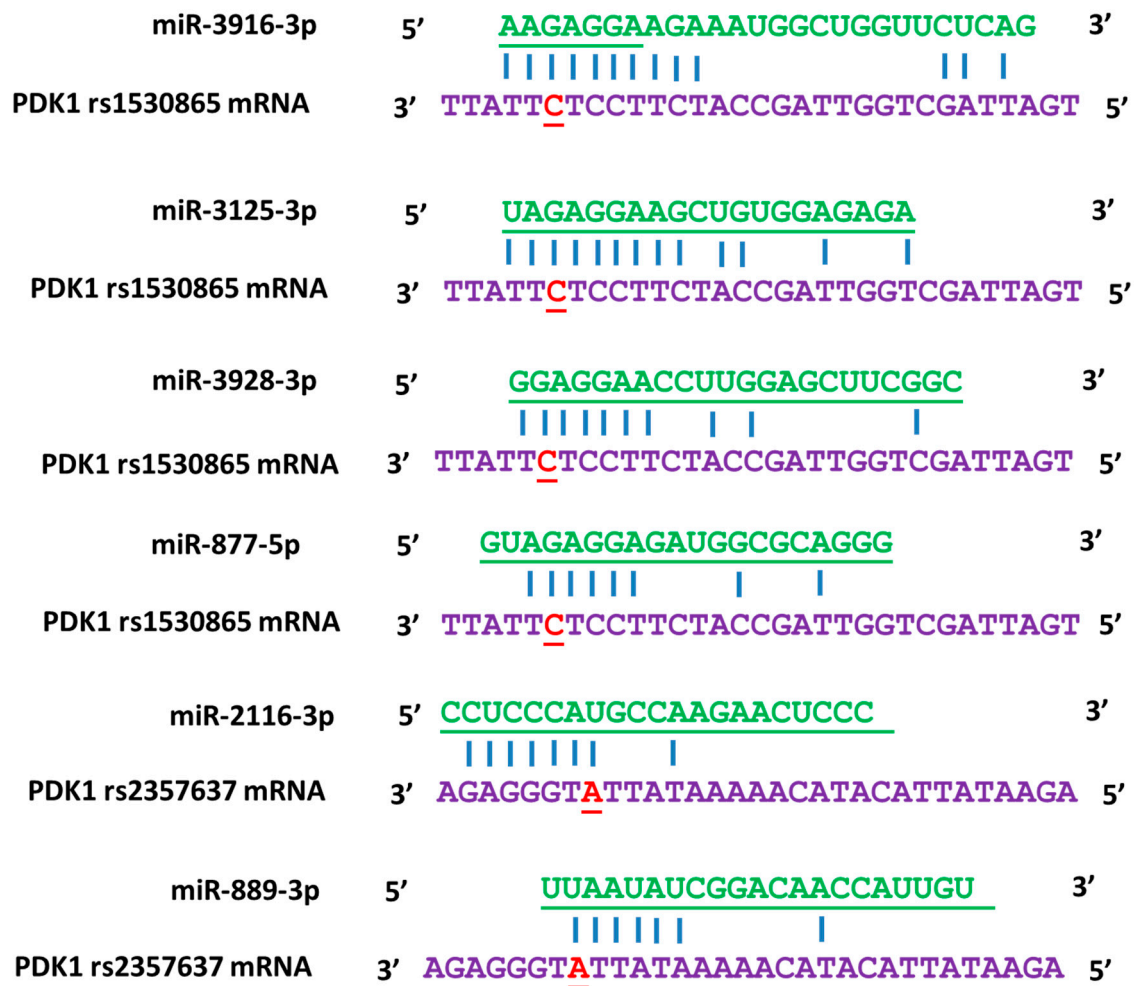


Figure S4. Schematic diagram showing the miRNA-mRNA nucleotides affinities for PDK1 rs1530865 and rs2357637 SNPs with the SNPs underlined and miRNA seed regions.

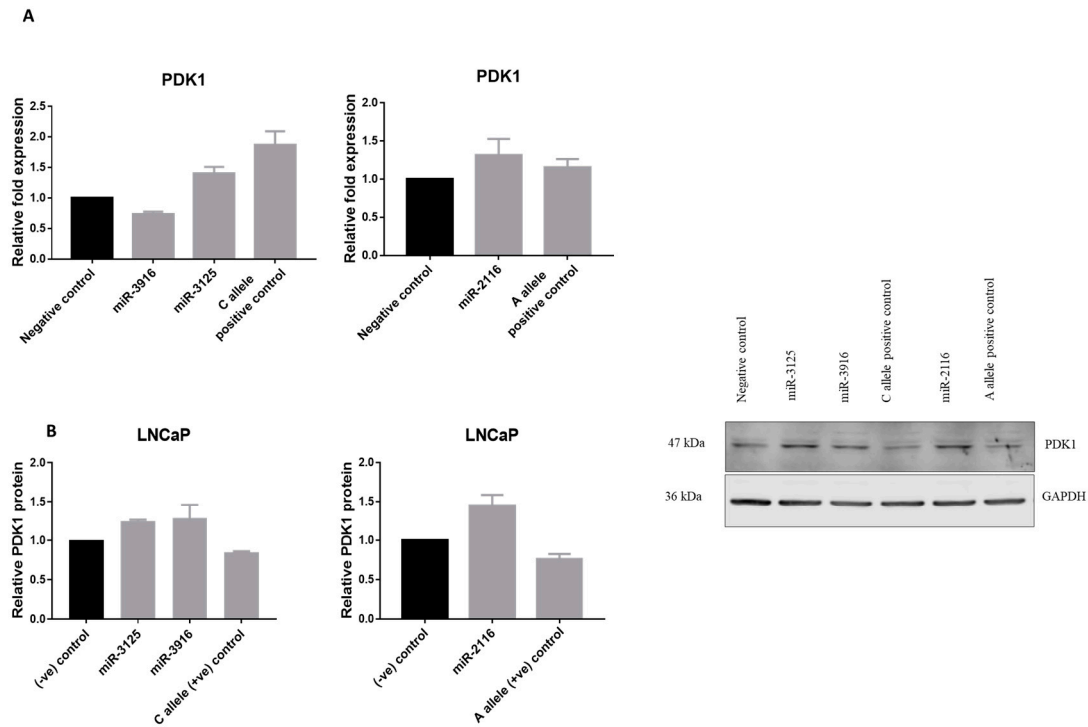


Figure S5. MiR-3916, miR-3125 and miR-2116 did not affect PDK1 mRNA expression and PDK1 protein expression in LNCaP cells. (A) Overexpression of miR-3916, miR-3125 and miR-2116 had no effect on PDK1 mRNA level in LNCaP cells. (B) miR-3916, 3125 and miR-2116 did not alter PDK1 protein in LNCaP cells. Bar graphs in B indicates densitometry analysis of the western blots. Mean \pm SEM, $n = 3$; a representative western blot is shown.

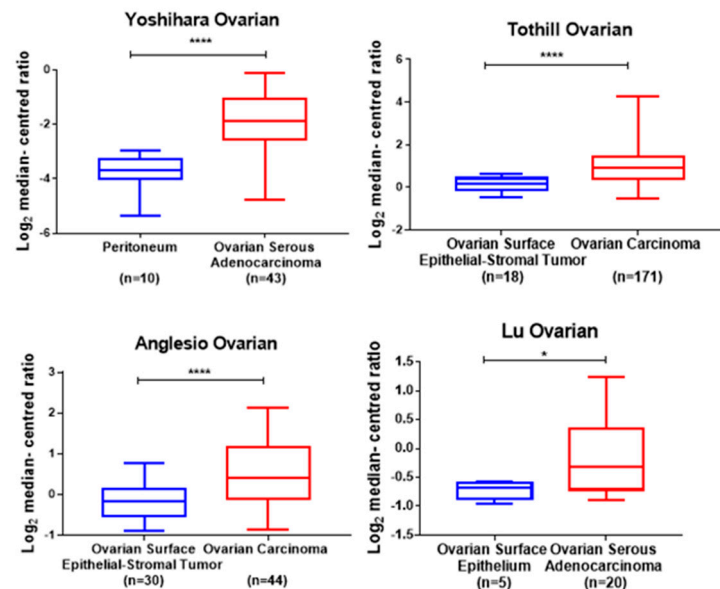


Figure S6. PDK1 expression is up-regulated in ovarian cancer tissues. PDK1 mRNA expression is up-regulated in ovarian carcinoma in four ovarian datasets. (Source-Oncomine, unpaired t -test, **** $p < 0.0001$, ** $p < 0.01$, * $p < 0.05$).

Figure 1

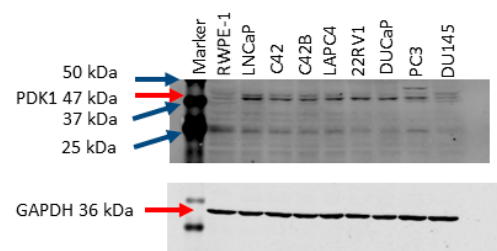


Figure 4

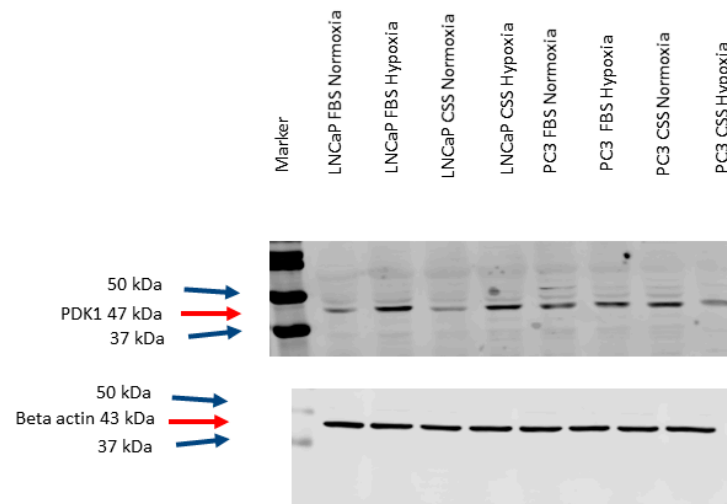


Figure 6B

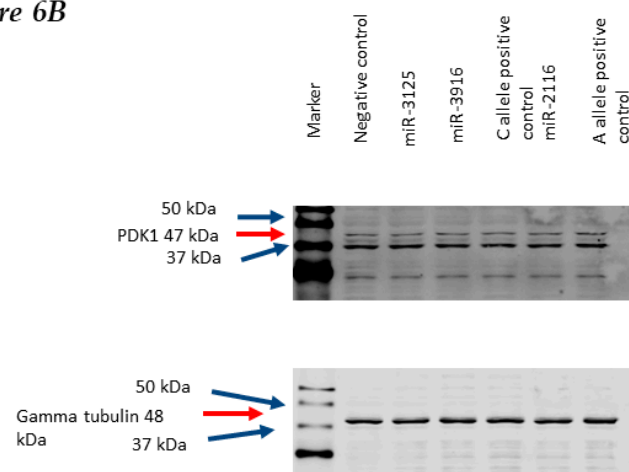
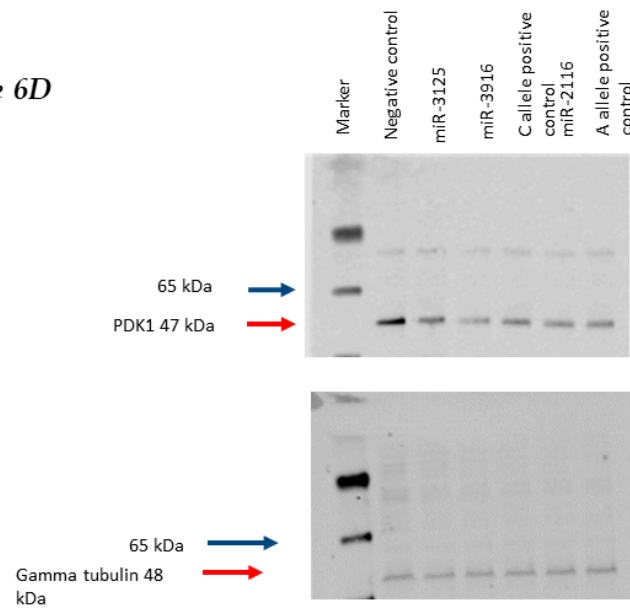
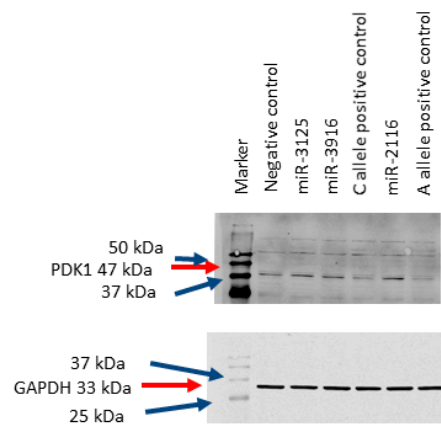


Figure 6D



Supplementary Figure 2B



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