

Supplementary Materials: RNase L Suppresses Androgen Receptor Signaling, Cell Migration and Matrix Metalloproteinase Activity in Prostate Cancer Cells

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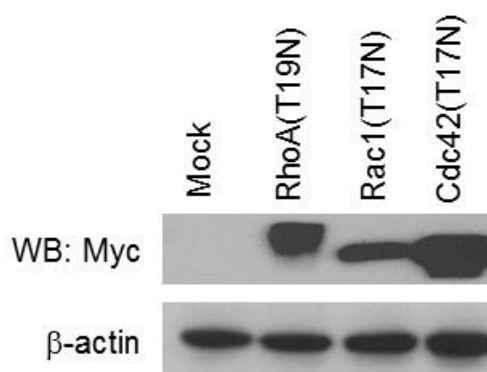


Figure S1. Expression of RhoA (T19N), Ras-related C3 botulinum toxin substrate 1-guanosine—Rac1 (T17N) and cell division control protein 42 homolog—Cdc42 (T17N) in PC3 cells compared to cells receiving vector alone (mock). Cell lysates were analyzed by immunoblot analysis using anti-Myc antibody and normalized to β -actin levels.

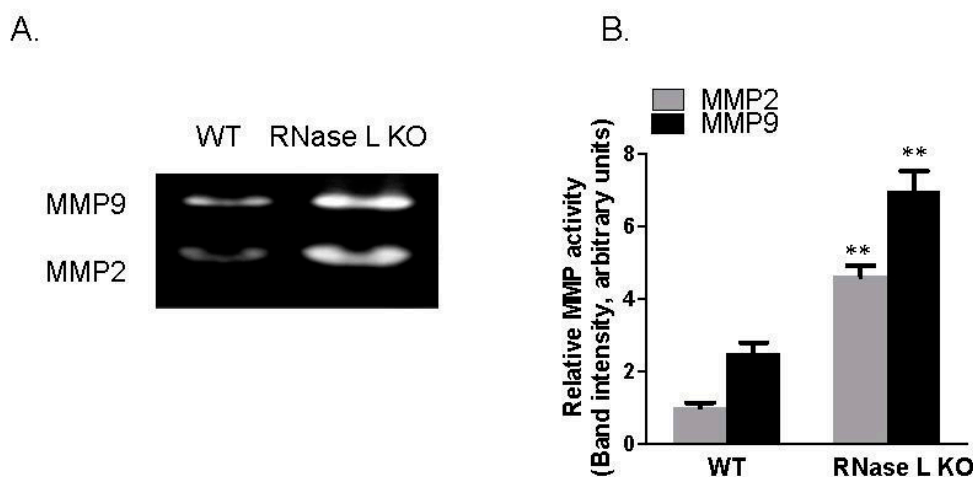


Figure S2. Increased matrix metalloproteinase (MMP)-2 and MMP-9 activities in RNase L KO mouse embryonic fibroblasts (MEFs). (A) Gelatin zymography analysis of MMP-2 and MMP-9 activities in conditioned media harvested from Wild type (WT) and RNase L KO MEFs. Data shown is representative of three independent experiments. (B) Quantitative analysis of MMP-2 and MMP-9 activities in WT and RNase L KO MEFs. Data shown are mean values \pm standard error of mean (SEM) from three independent experiments. Student's *t*-test was used to determine *p*-values. * *p* < 0.01, ** *p* < 0.001, and compared to control WT MEFs.

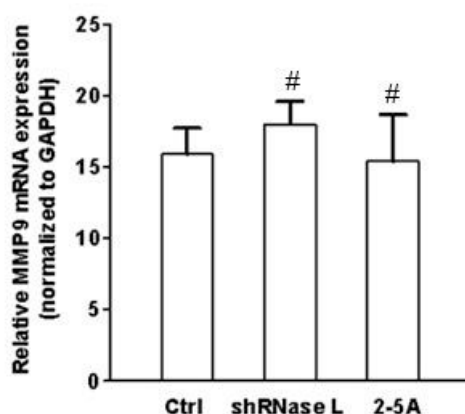


Figure S3. Effect of RNase L on *MMP-9* mRNA levels. Reverse transcriptase-polymerase chain reaction (RT-PCR) analysis of messenger RNA (mRNA) levels in DU145 cells expressing control or RNase L short hairpin RNA (shRNA) or transfected with 2-5A complexed with lipofectamine 2000 and added to cells to activate RNase L normalized to glyceraldehyde-3-phosphate dehydrogenase (*GAPDH*) mRNA levels. Student's *t* test was used to determine *p* values. # not significant, and compared to control cells.

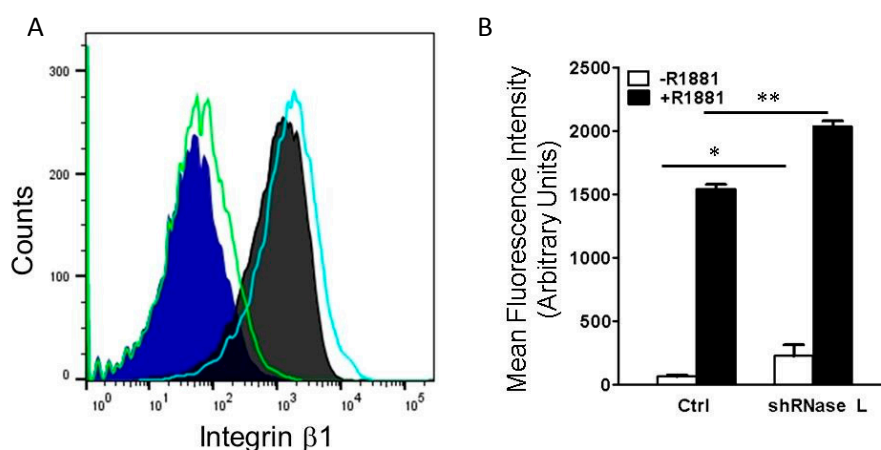


Figure S4. Effect of androgens on cell surface expression of integrin $\beta 1$. LNCaP cells expressing control shRNA or RNase L shRNA were treated with R1881 (1 nM) and analyzed by flow cytometry for surface staining with antibodies against integrin $\beta 1$ and Alexa-488 conjugated secondary antibodies. (A) Representative histograms, and (B) Bar graphs for the mean fluorescence intensity of at least three independent experiments for integrin $\beta 1$ are shown. Student's *t*-test was used to determine *p*-values. * $p < 0.01$, ** $p < 0.001$ and compared to cells expressing control shRNA; Ctrl: control.