# Competition for XPO5 binding between DICER mRNA, pre-miRNA and viral RNA regulates human DICER levels

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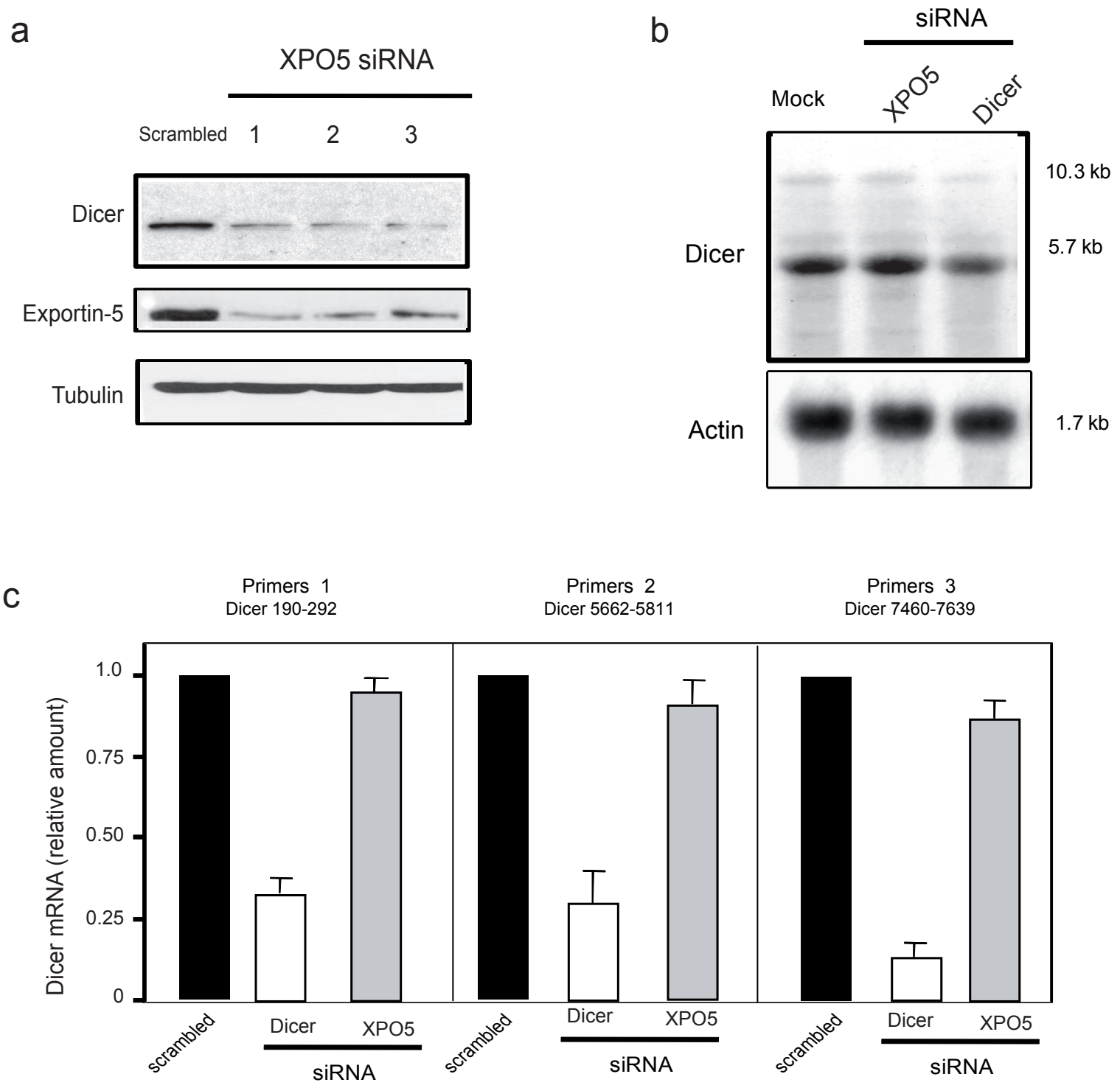
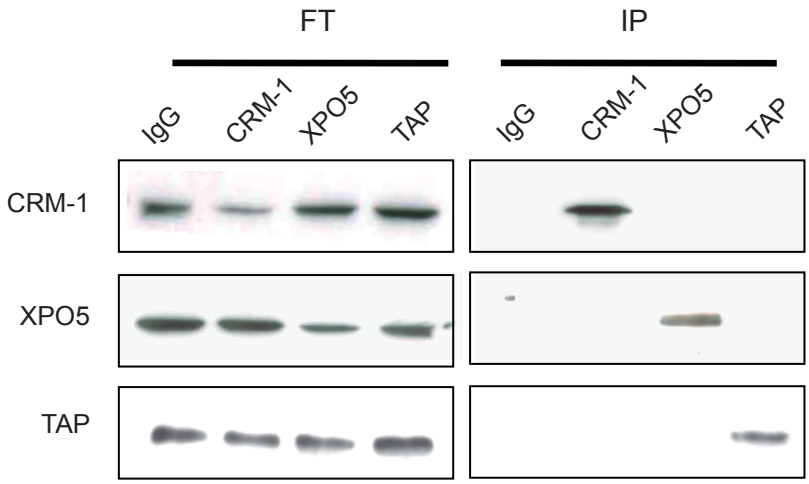
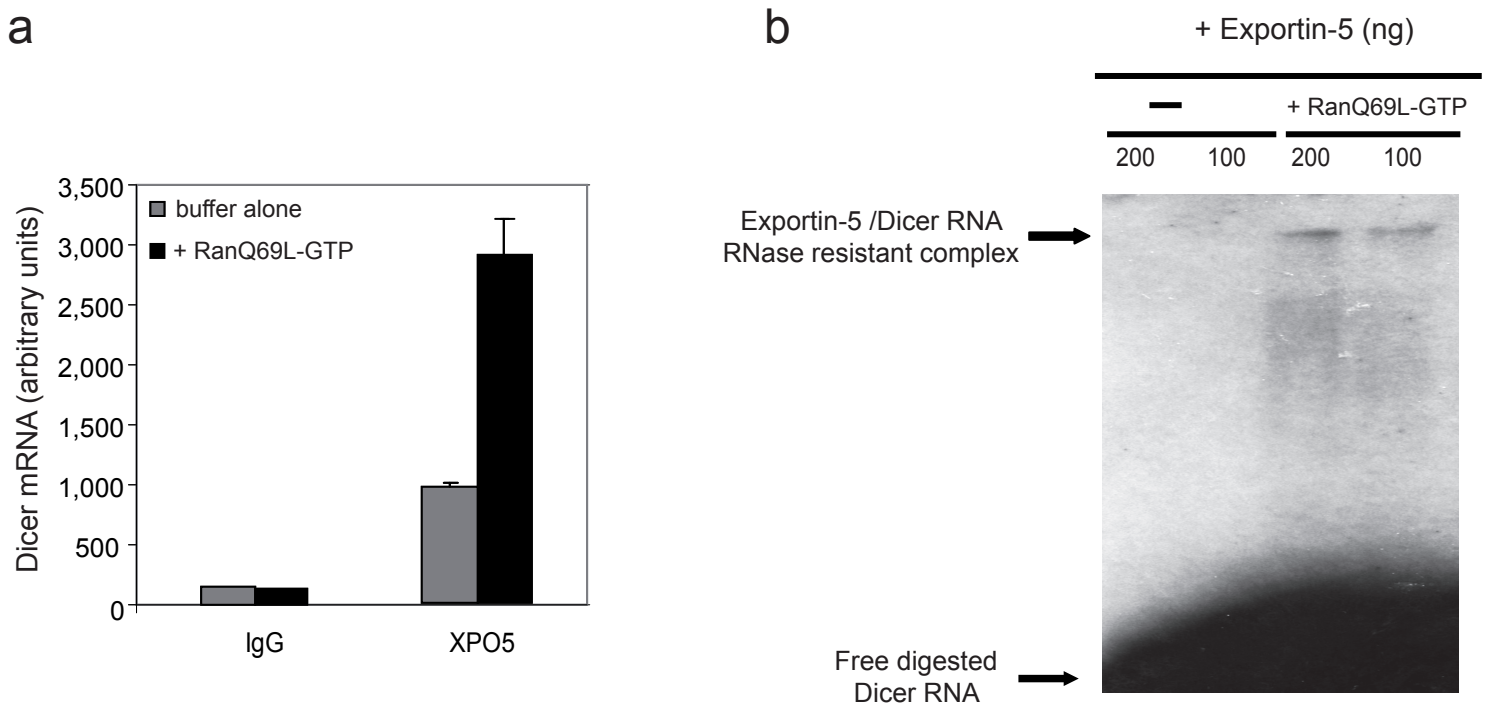



**Supplementary Figure 1: XPO5 siRNA inhibits DICER protein expression but has no effect on Dicer mRNA levels.** a) HeLa cells were transfected with three supplemental siRNA directed against three additional regions of XPO5 mRNA. 48 hours post transfection, cells were analyzed for DICER, XPO5 and tubulin expression by western blot. b) DICER mRNA levels are not affected by XPO5 inhibition as assessed by northern blot. DICER mRNA level was analyzed by northern blot following DICER or XPO5 siRNA transfection. c) Quantitative RT-PCR of DICER mRNA levels using 3 sets of primers recognizing different regions of DICER mRNA following XPO5 inhibition: primers 1 amplifies region 190-292, primers 2 region 5662-5811 (also used in figure 1c) and primers 3 region 7460-7639. Results are represented as DICER mRNA levels compared to cells transfected with a control scramble siRNA.

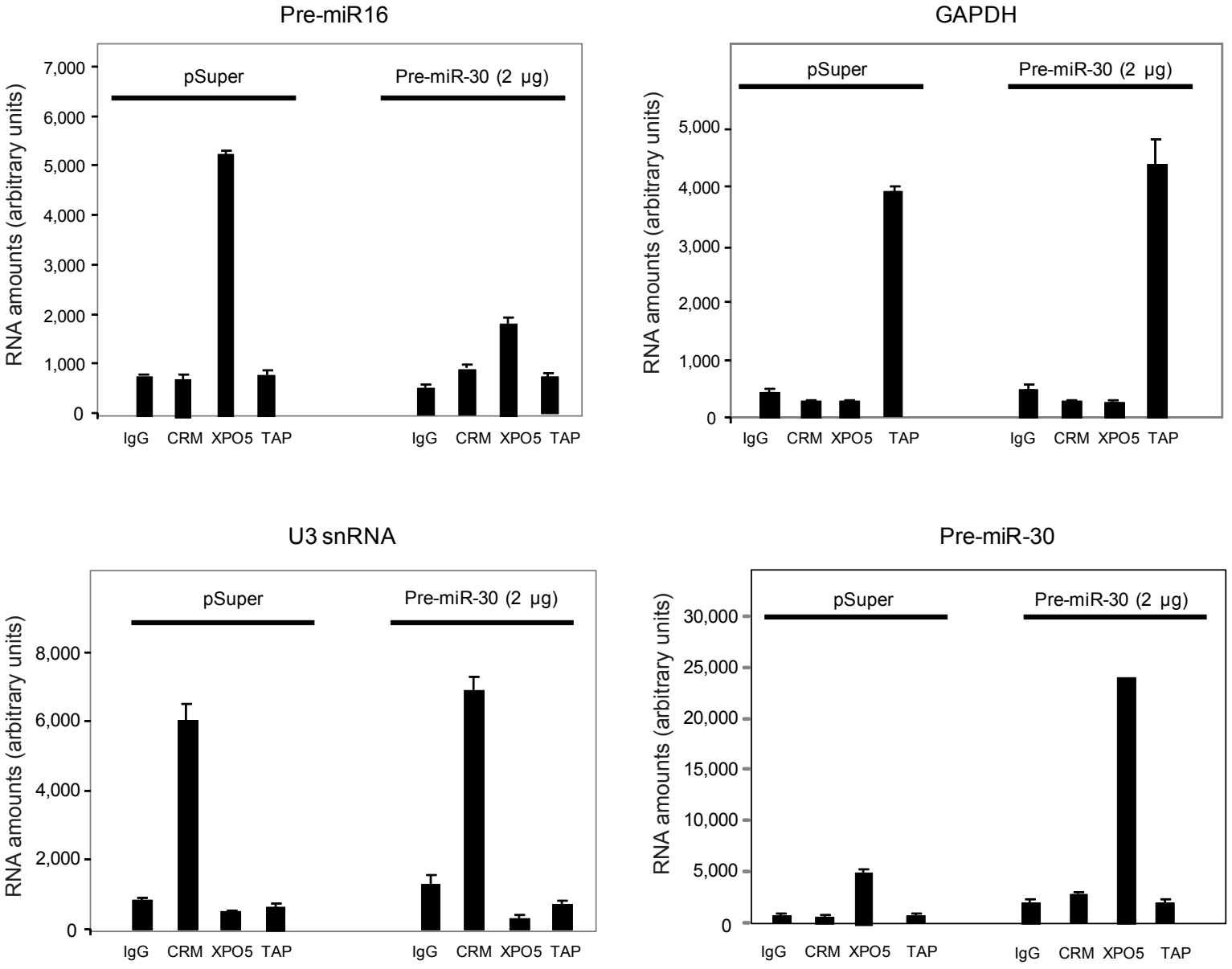



**Supplementary Figure 2: Karyopherin immunoprecitation.** HeLa cell extracts were prepared and subjected to immunoprecipitation using IgG (control), anti-CRM1, anti-XPO5, or anti-TAP/p15 antibodies. After washing, a fraction of the unbound (FT) material and immunoprecipitates (IP) were analyzed by western blot using anti-CRM1, anti-XPO5, or anti-TAP/p15 antibodies.



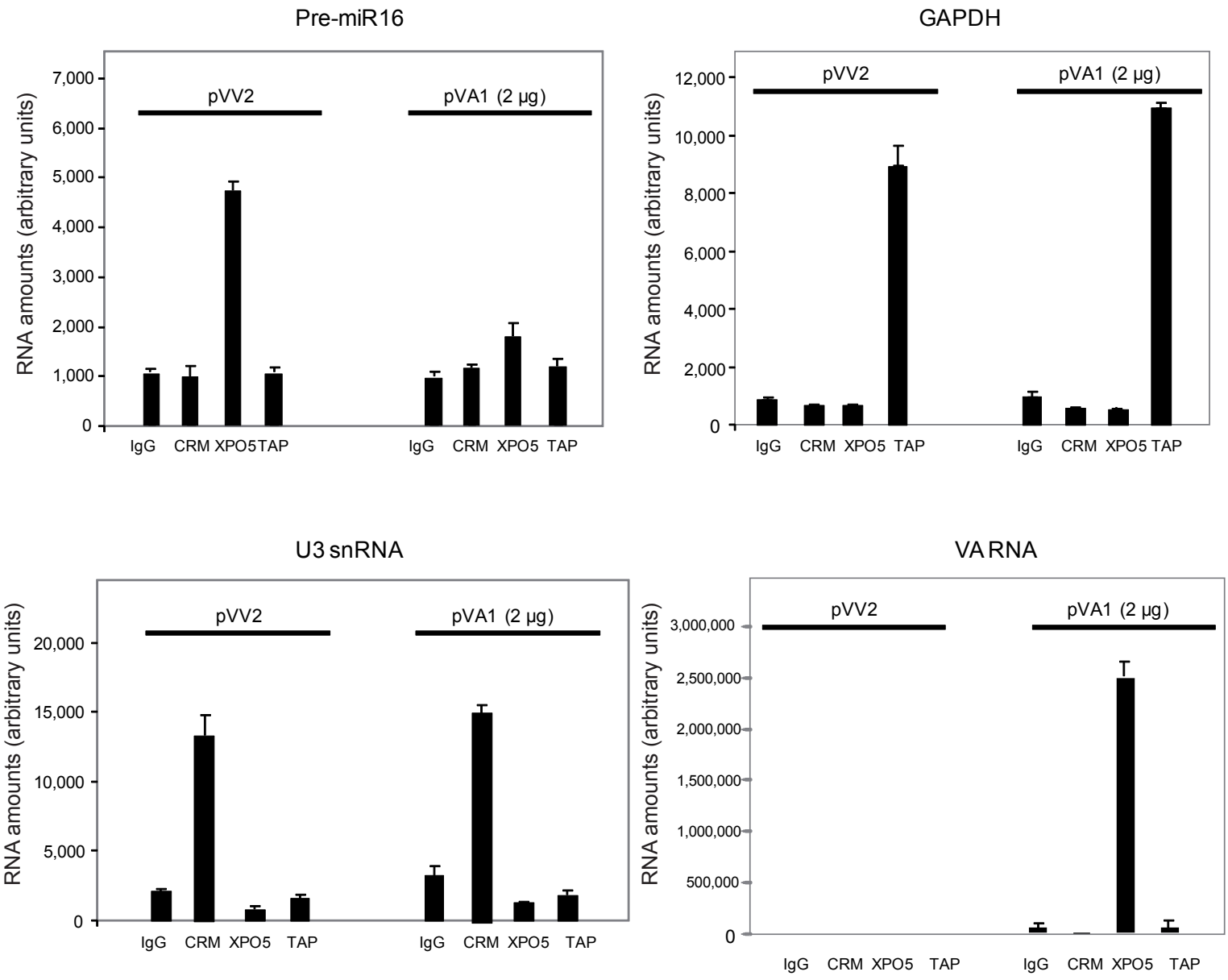

**Supplementary Figure 3: DICER mRNA/XPO5 specific interaction requires RanGTP.** a) Karyopherin immuno-precipitation analysis were realized as described in figure 3, in the absence (grey bars) or presence of RanQ69L-GTP (black bars). b) EMSA was realized as described in Figure 2 in the absence (lanes 1 and 2) or presence of RanQ69L-GTP (lanes 3 and 4).

a




Supplementary Figure 4

b




**Supplementary Figure 4: Pre-miR-30 or VA1 overexpression does not affect amounts of U3 snRNA or GAPDH mRNA recovered from CRM1 and TAP immunoprecipitates.** HeLa cells were transfected with either 2 µg of empty vector (pVV2 or pSuper) or plasmid expressing pre-miR-30 (A) or VA1 (B). 48 hours post transfection, cell extracts were prepared and subjected to immunoprecipitation using IgG (control), anti-CRM1, anti-XPO5, or anti-TAP/p15 antibodies. Purified RNA were reverse transcribed and subjected to qRT-PCR using specific primers for U3 snRNA, pre-miR-16, GAPDH mRNA, pre-miR-30, VA and DICER mRNA as in figure 2. Error bars are expressed as s.d.