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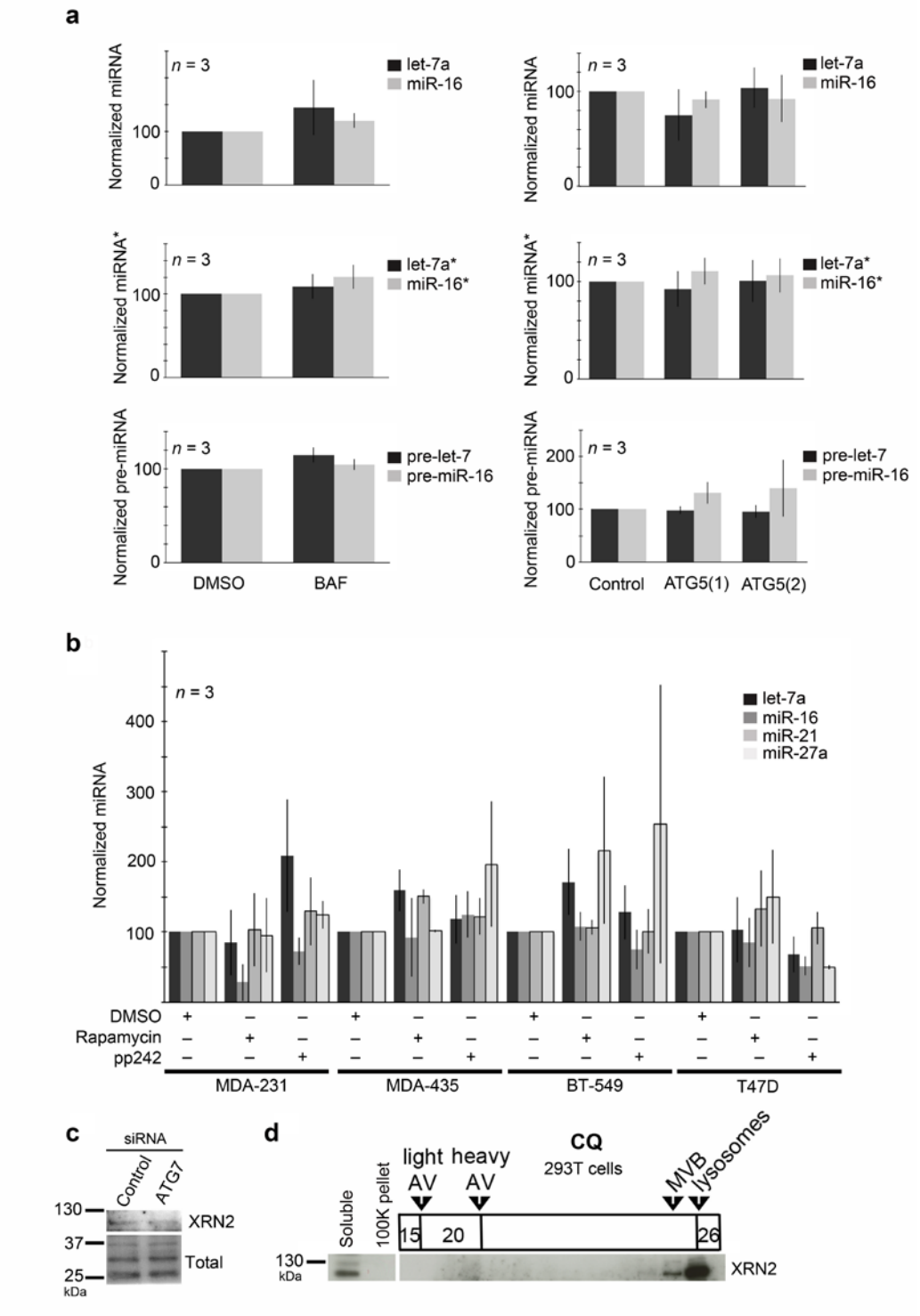

Figure S1 Short-term modulation of autophagy does not affect miRNA levels and XRN2 is not regulated by autophagy. (**a**) RT-qPCR analysis of levels of miRNA (miR-16, let-7a), and the corresponding miRNA* or pre-miRNA in HeLa cells treated with DMSO (control) or BAF for 12 h. ATG5(1) and ATG5(2) represent results with two independent siRNA. (**b**) RT-qPCR analysis of MDA-231, T47D and MDA-435 cells treated with DMSO, or inhibitors of mTORC1 (rapamycin) or mTORC1/2 (pp242) for 48 h. Error bars represent SEM. (**c**) Western blot of XRN2 in cells treated with siRNA targeting ATG7 or a control siRNA. Total protein stained by Coomassie blue serve as a loading control. (**d**) Western blot analysis of XRN2 in fractions from a discontinuous gradient of Histodenz™ (15%, 20%, 24%, 26%) described to enrich autophagosomes (light AV) and autophagolysosomes (heavy AV). 293T cells were treated with CQ (20 μM, 16 h). Fractions enriched in MVB and lysosomal markers are indicated. Material that was pelleted by centrifugation at 100,000 g (100K pellet) or that remained in solution after the 100,000 g spin (soluble) was not added to the gradient.

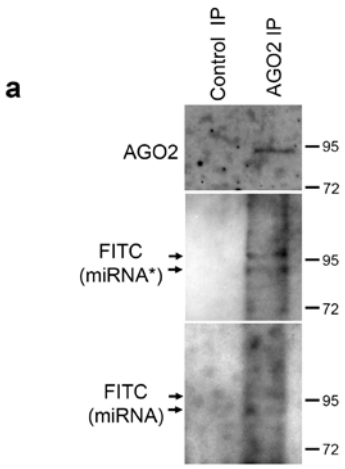

Figure S2 MiRNA and miRNA* added to *in vitro* AGO2-loading assay associate with AGO2. In HeLa cell extracts, following UV-cross-linking and AGO2 immunoprecipitation a fraction of AGO2 eluate was analyzed by western blot with anti-FITC mAb (to detect miRNA or miRNA* labeled 3' with FITC) and anti-AGO2 mAb. Anti-FITC mAb detected bands at masses coinciding with AGO2. Higher band may represent the mass-shift caused by covalent binding of AGO2 to RNA after UV-cross-linking. Similar mass shifts of AGO bound to single-stranded versus double-stranded RNA (miRNA + miRNA* or miRNA + mRNA target) have been described previously[50].

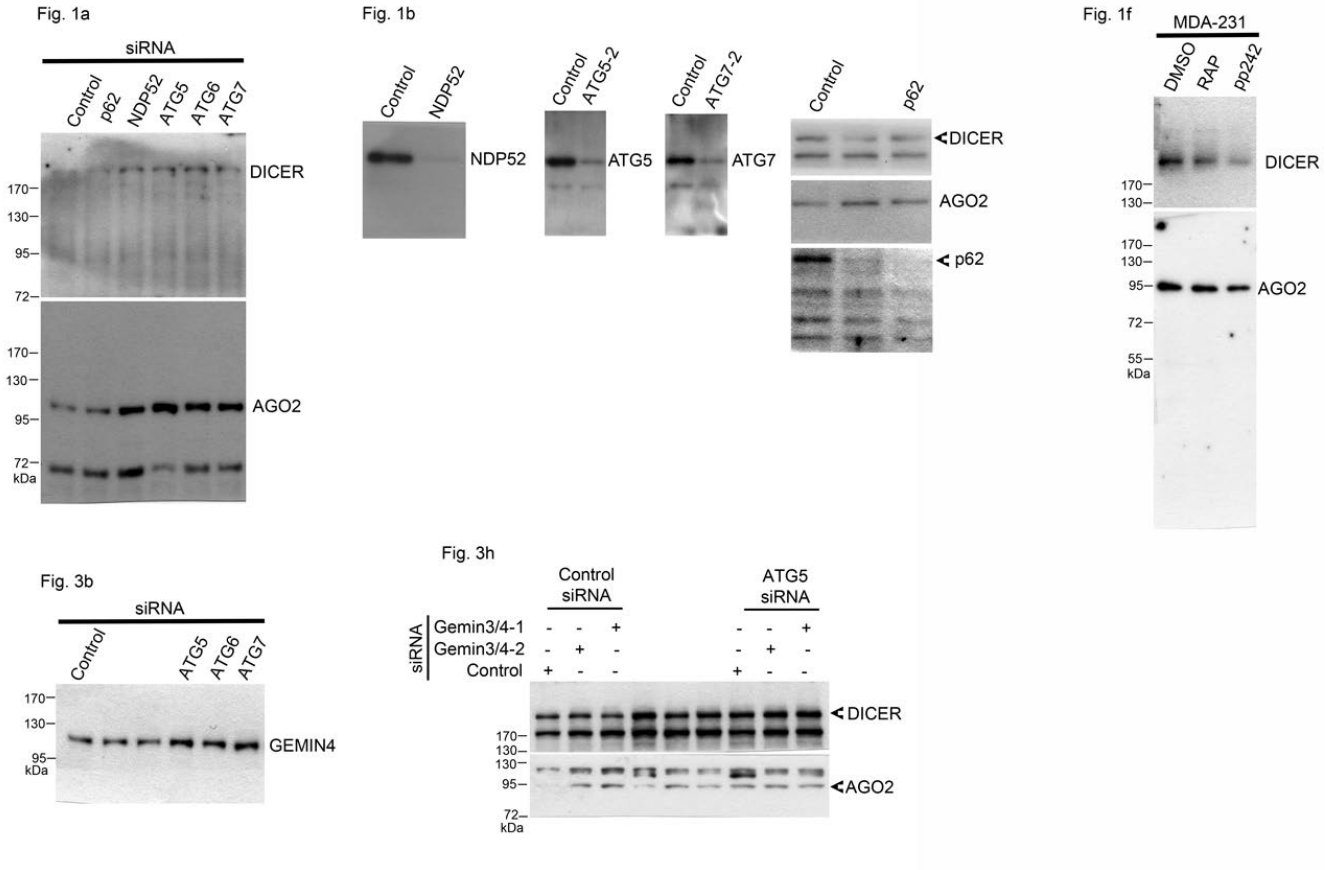

Figure S3 Uncropped scans of blots.