

Supplementary Materials

Potential Onco-Suppressive Role of miR122 and miR144 in Uveal Melanoma Through ADAM10 and C-Met Inhibition

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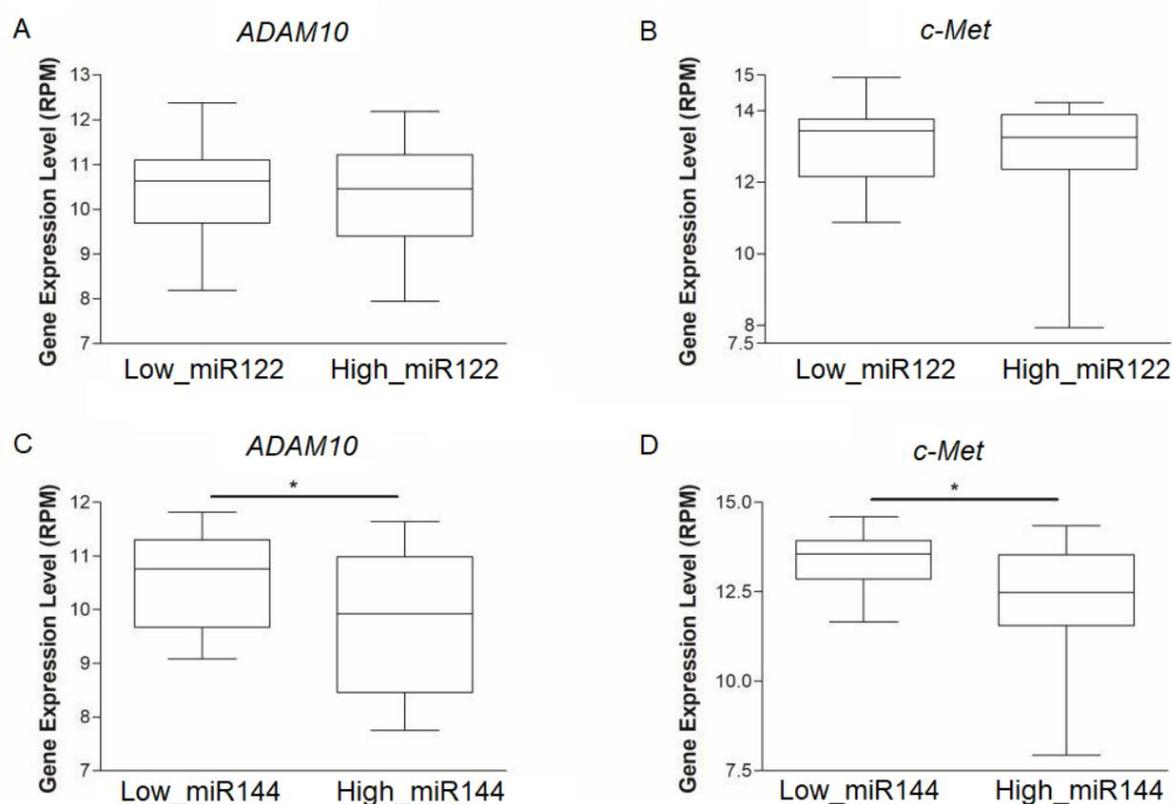


Figure S1. Differential expression of miR122 and 144 and correlation to *ADAM10* and *c-Met*: box and whiskers chart shows that the low expression of miR144 (bottom quartile) significantly associates with high *ADAM10* or *c-Met* expression (* $p < 0.05$). Analysis of *ADAM10* and *c-Met* expression was performed comparing patients with high miR144 (19) with patients with low miR144 (19), and patients with high miR122 (37) with patients with low miR122 (19).

Target sites of miR122-5p and miR144-3p on *ADAM10* and *c-Met*

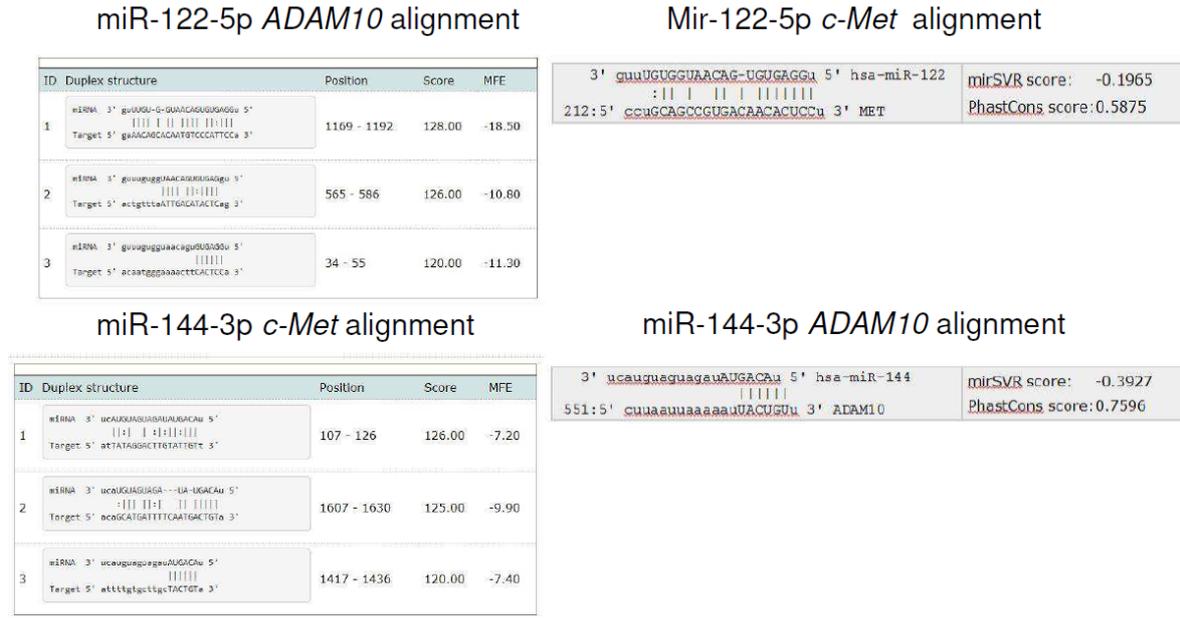
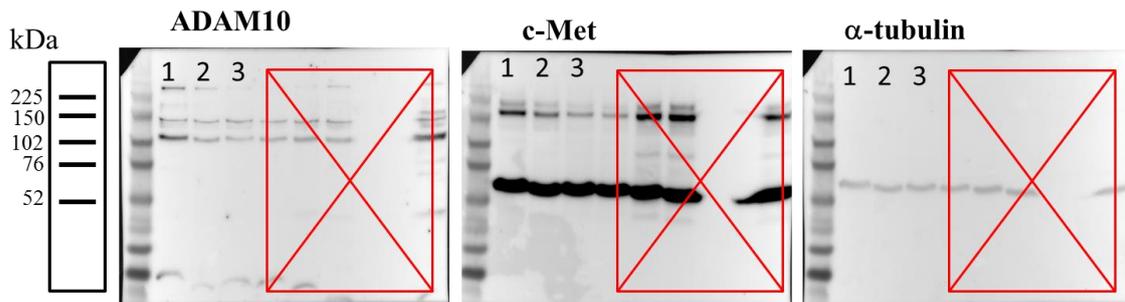
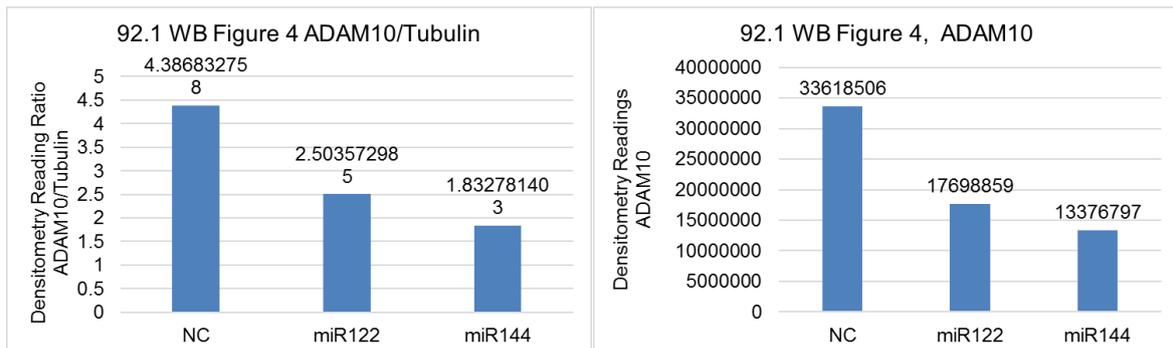


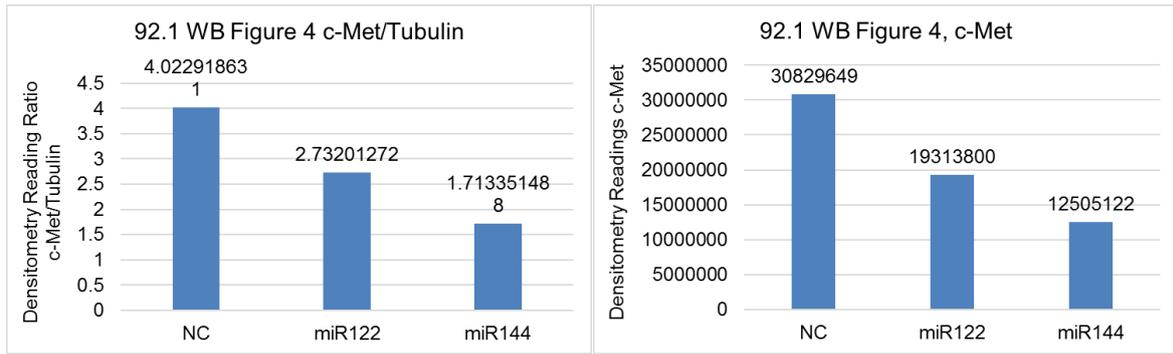
Figure S2. Potential target sites of miR122 and miR144 on *ADAM10* and *c-Met* by MIRANDA and miRBASE tools.

92.1

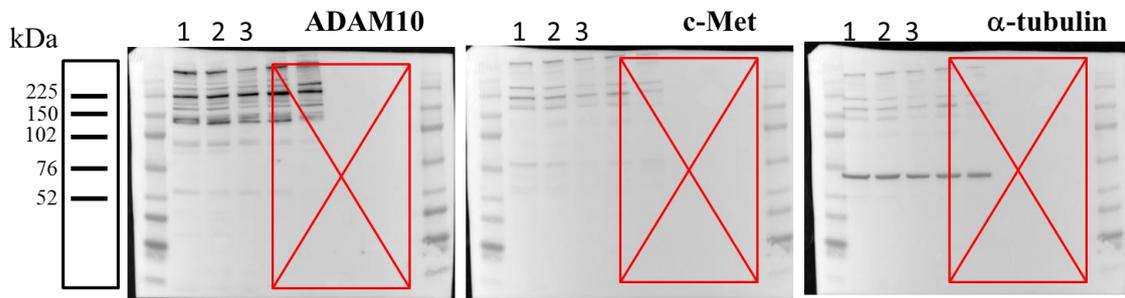


(A)

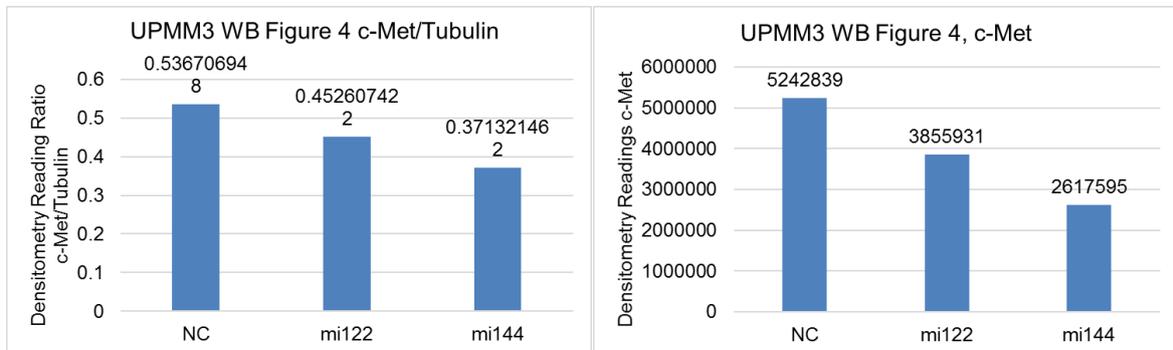
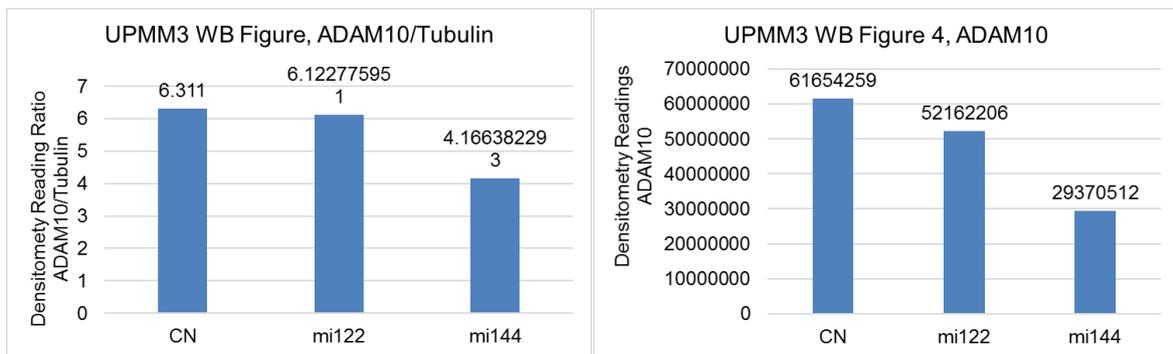




UPMM3



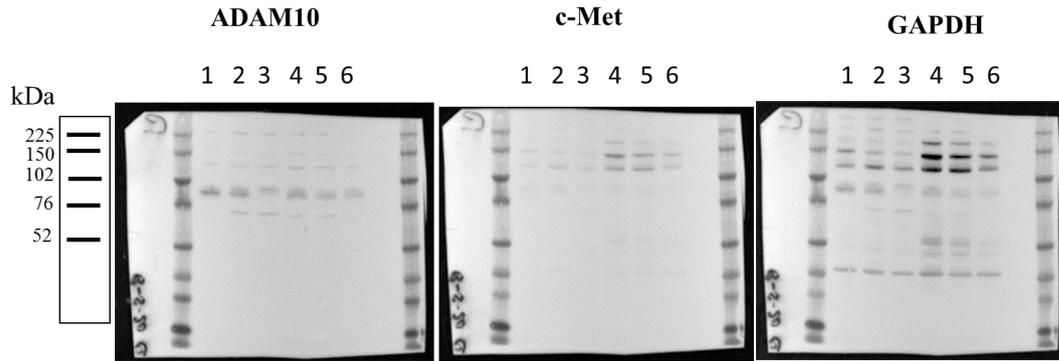
(B)



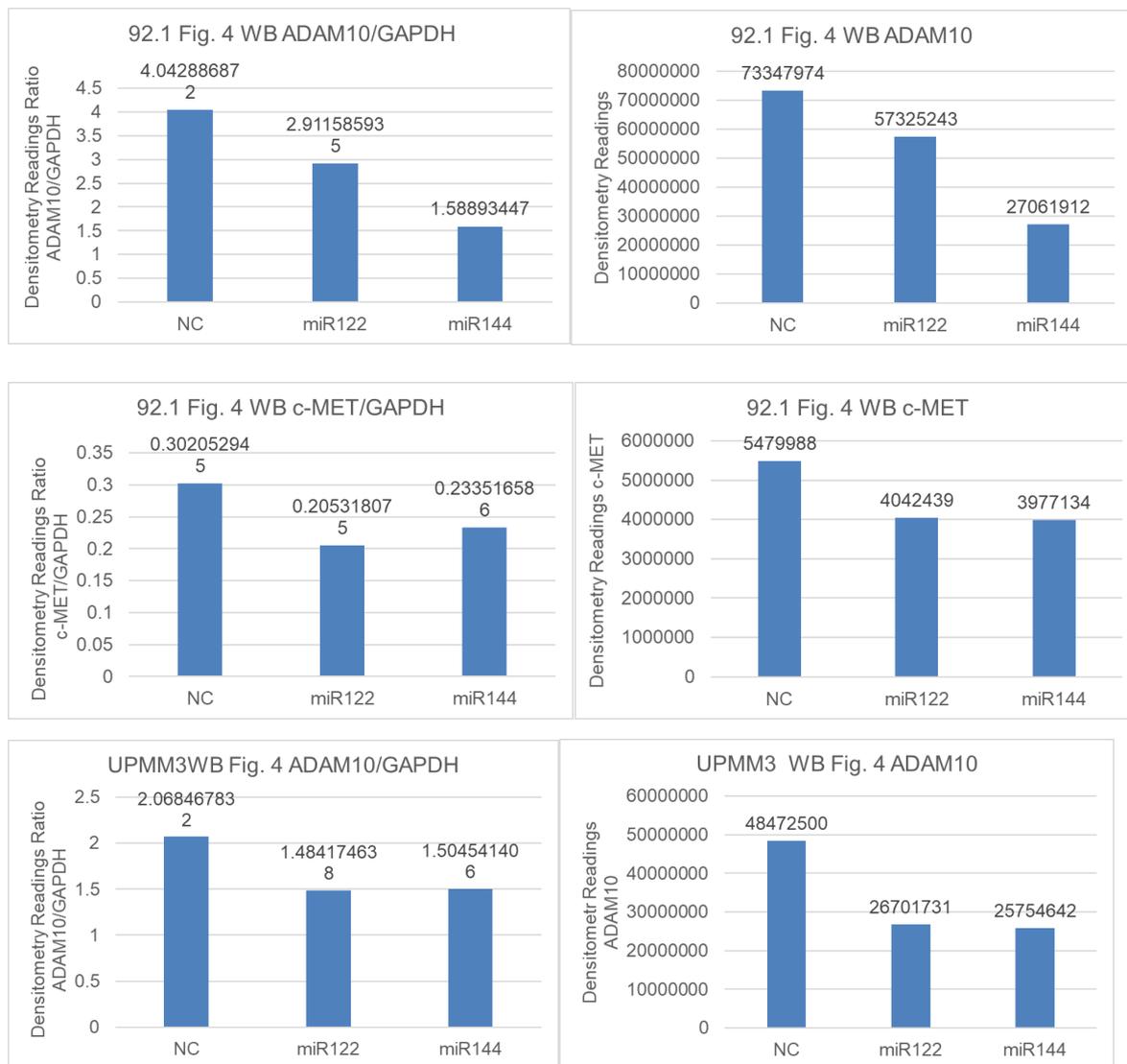
(C)

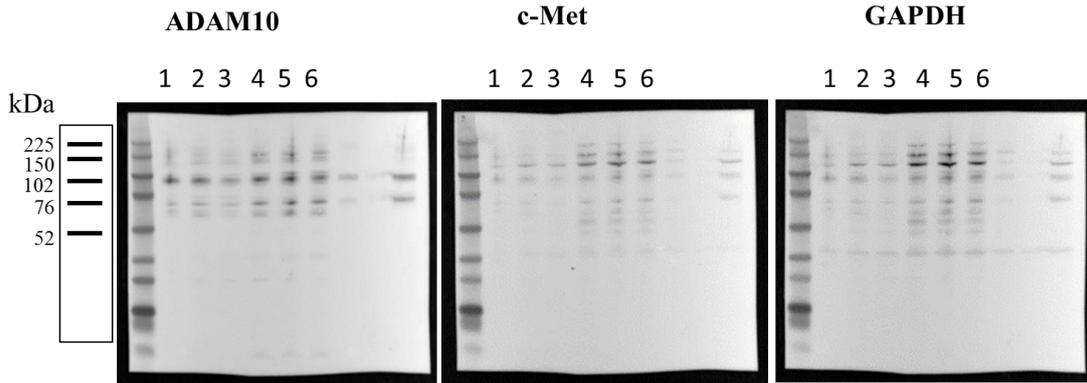
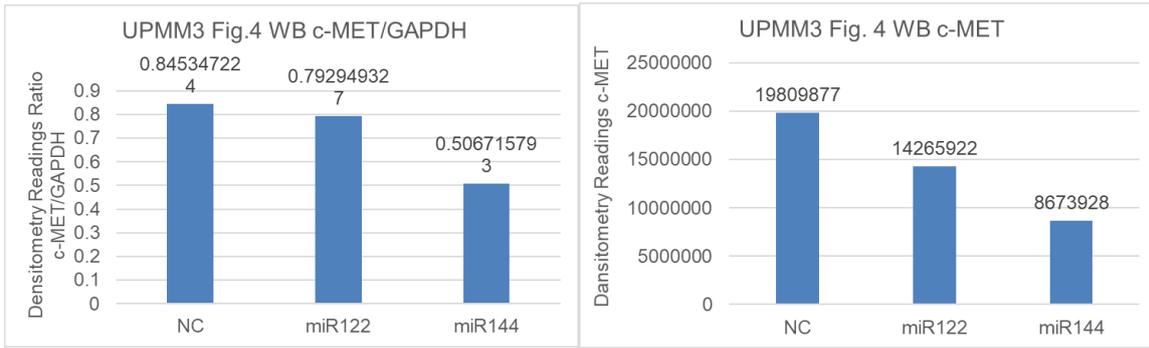
Figure S3. (A) Whole blots showing all the bands with molecular weight markers and densitometry presented in Figure 4. (B) 92.1 cell lysates probed with anti-ADAM10, anti-c-Met and anti- α -tubulin antibodies. 1: irrelevant miRNA mimic as negative control (NC); 2: miR122; 3: miR144. (c) UPMM3

cell lysates probed with anti-ADAM10, anti-c-Met and anti- α -tubulin antibodies. 1: irrelevant miRNA mimic as negative control (NC); 2: miR122; 3: miR144.

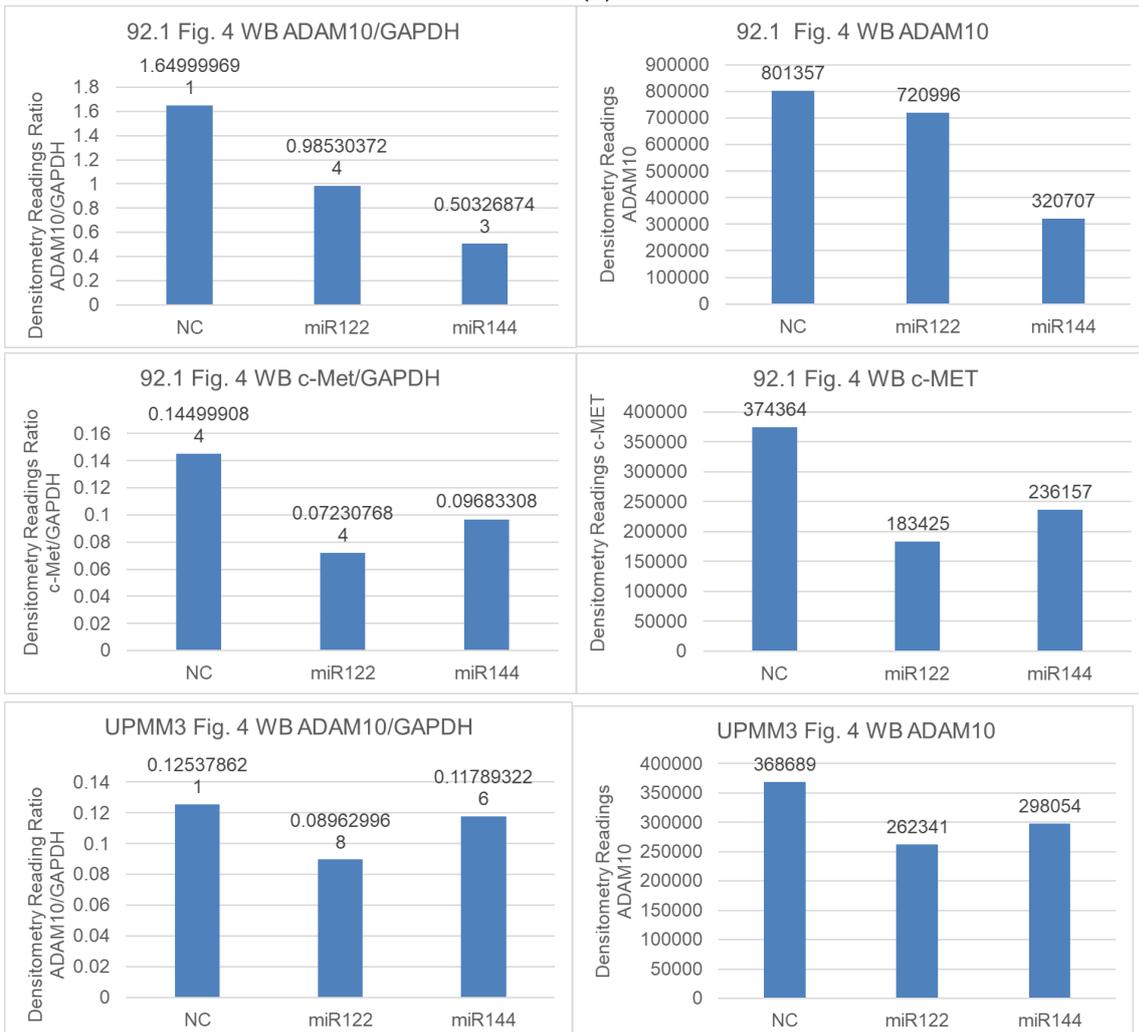


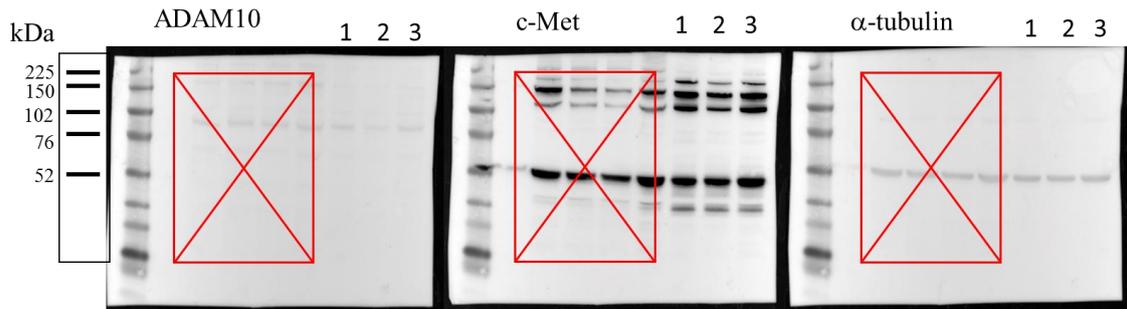
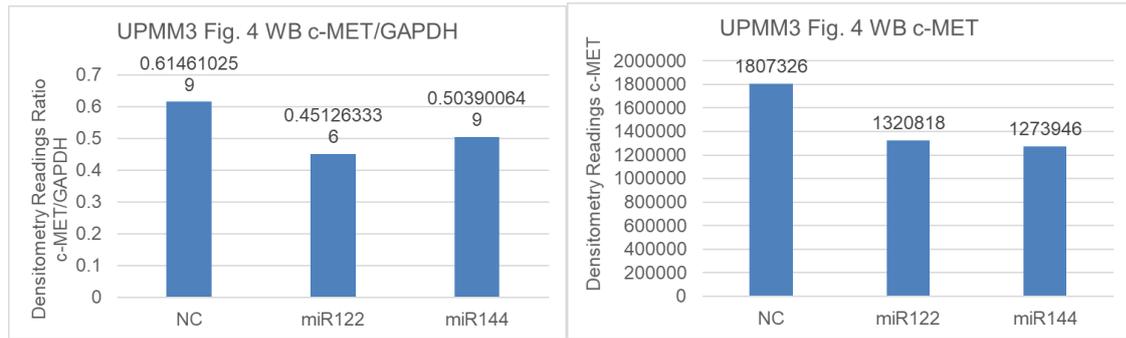
(A)



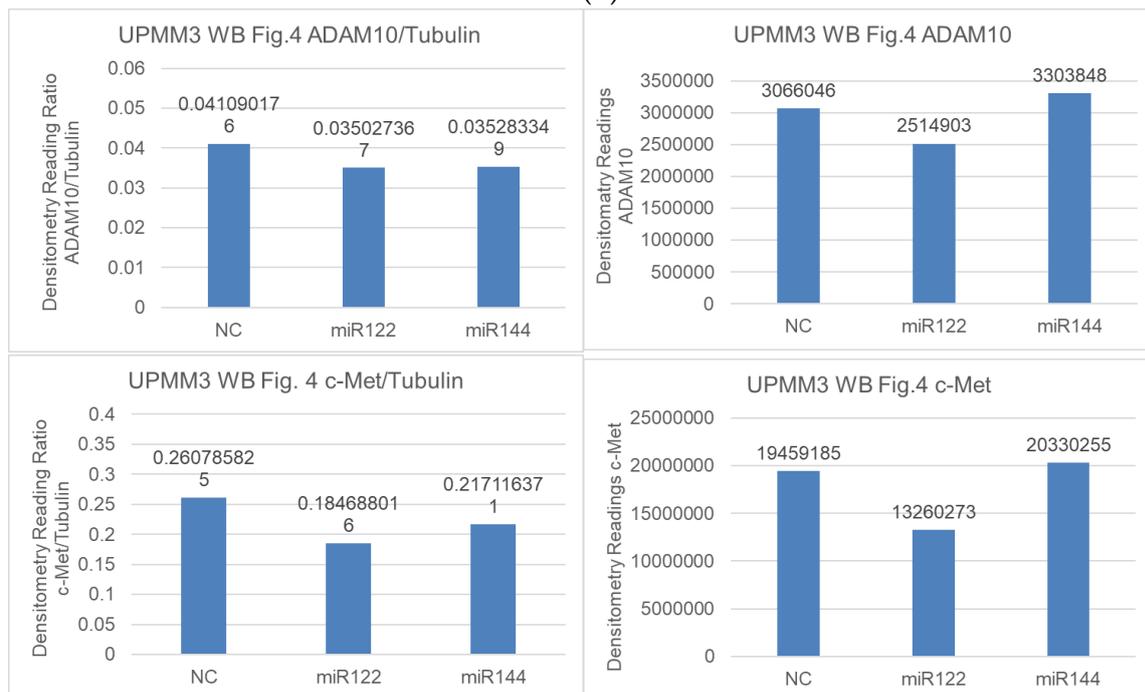


(B)





(C)



(D)

Figure S4. (A) Whole blots showing all the bands with molecular weight markers and relative densitometry used to obtain histogram in Figure 4 panel B. (B) 92.1 and UPMM3 cell lysates probed with anti-ADAM10, anti-c-Met and anti GAPDH antibodies. 92.1: 1: Negative Control (NC); 2: miR122; 3: miR144. UPMM3: 4: Negative Control (NC); 5: miR122; 6: miR144. (C) 92.1 and UPMM3 cell lysates probed with anti-ADAM10, anti-c-Met and anti GAPDH antibodies. 92.1: 1: Negative Control (NC); 2: miR122; 3: miR144. UPMM3: 4: Negative Control (NC); 5: miR122; 6: miR144. (D) UPMM3 cell lysates probed with anti-ADAM10, anti-c-Met and anti α -tubulin antibodies. 1: Negative Control (NC); 2: miR122; 3: miR144.

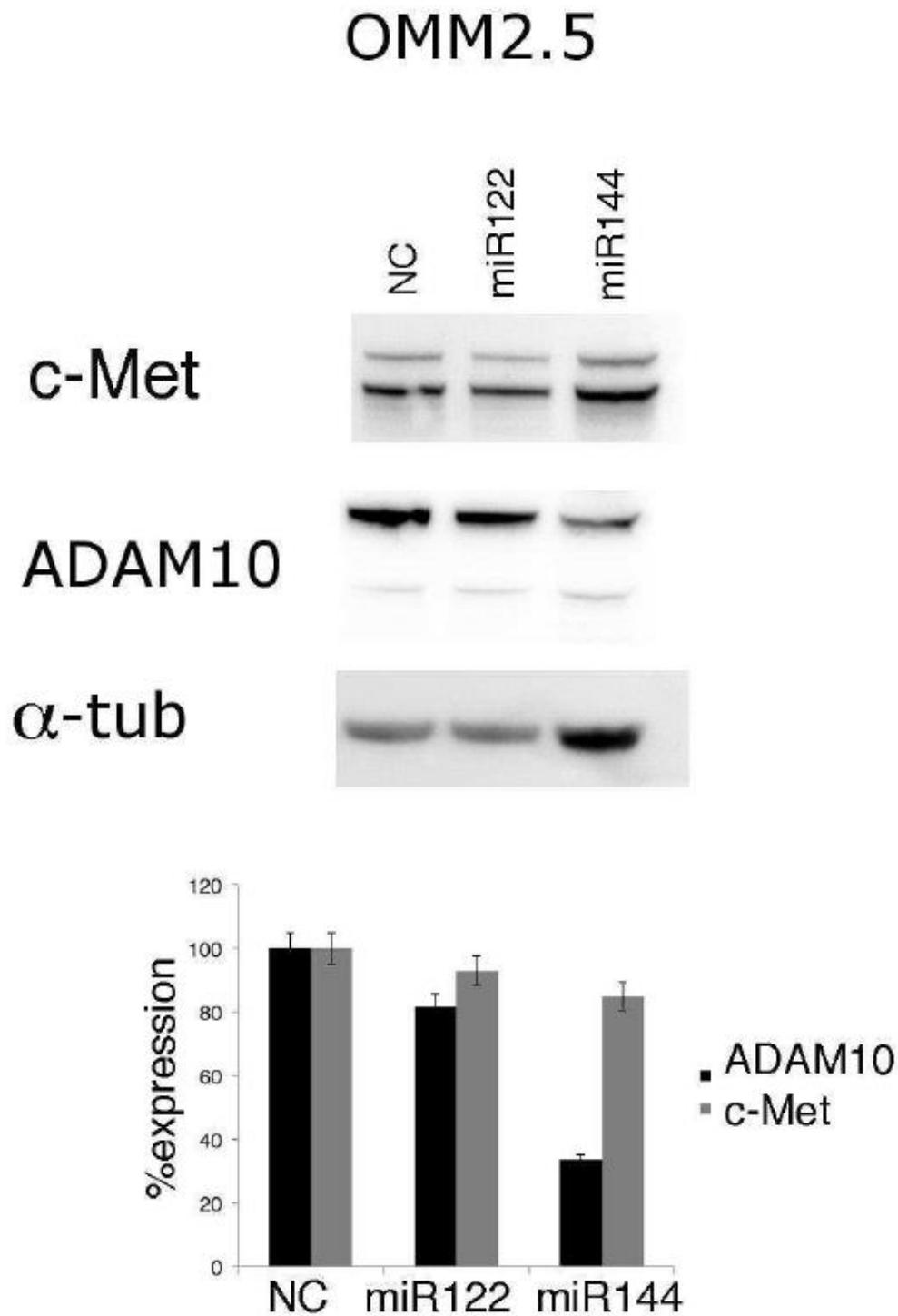
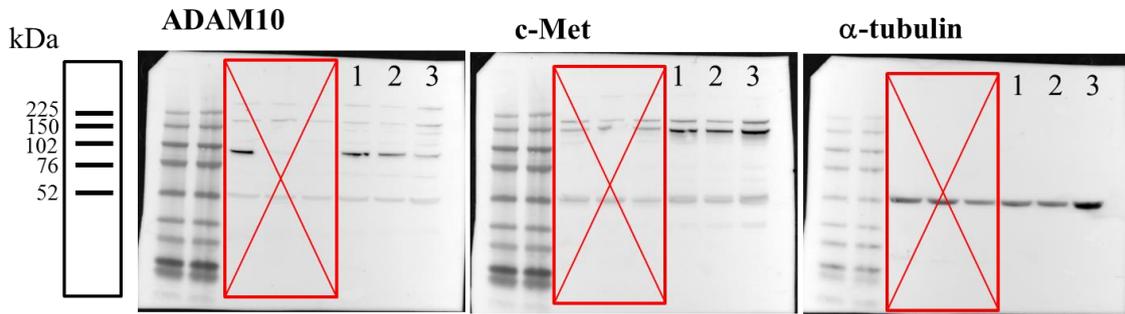
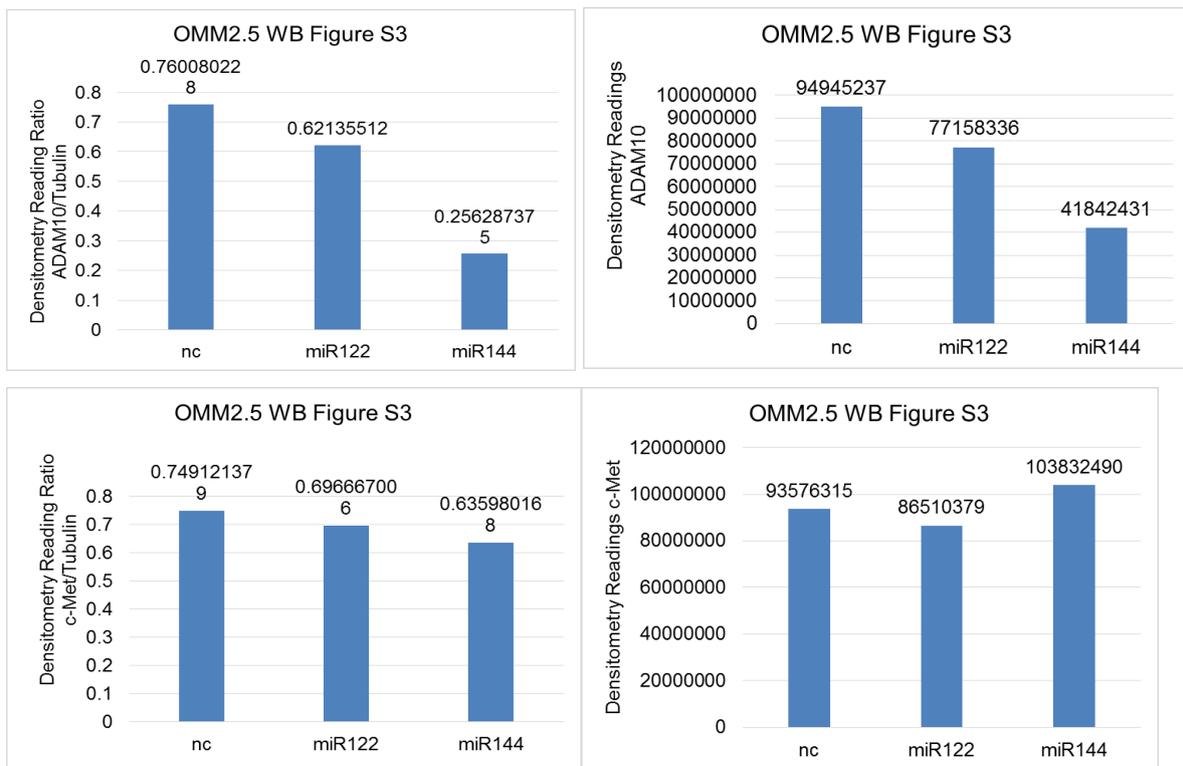


Figure S5. Western blot analysis of ADAM10 and c-Met expression in miR mimic transfected OMM2.5 cells: ADAM10 and c-Met proteins are down-modulated by mimic miR122, or miR144 transfection in OMM2.5 cell lines. Histograms show the quantification of band intensity as percentage expression of the irrelevant miRNA mimic, control (NC). A representative experiment of three is shown.

OMM2.5



(A)



(B)

Figure S6. (A) Whole blots showing all the bands with molecular weight markers and relative densitometry presented in Figure S5. (B) OMM2.5 cell lysates probed with anti-ADAM10, anti-c-Met and anti- α -tubulin antibodies. 1: irrelevant miRNA mimic as negative control (NC); 2: miR122; 3: miR144.

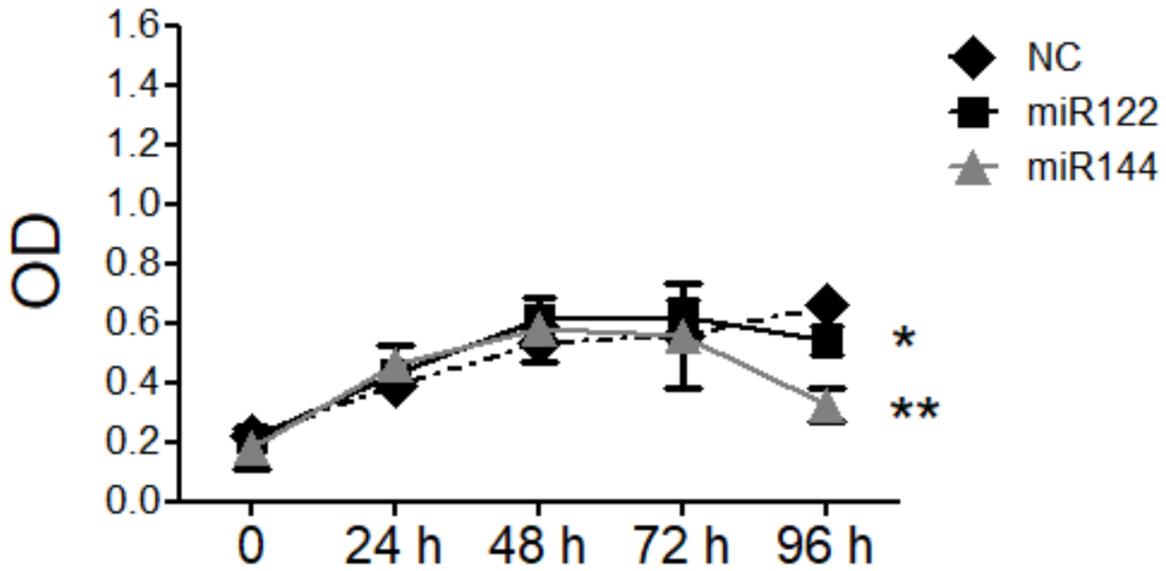
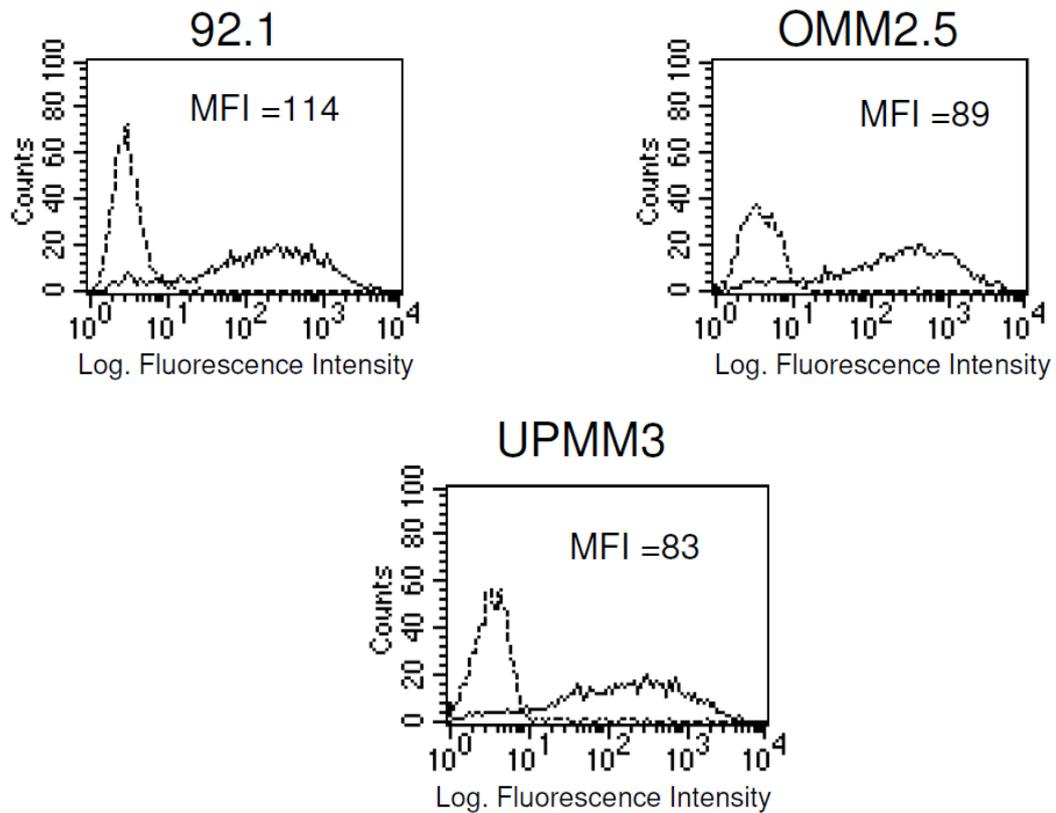


Figure S7. Effects of miR122 and miR144 mimic transfection on UPMM3 proliferation: miR144 and miR122 mimics transfection significantly inhibit UPMM3 cell line proliferation only 96 h after transfection, in respect to irrelevant miRNA mimic control (NC) (* $p < 0.05$; ** $p < 0.01$).



MFI= (sample Mean- CTR Mean)/CTR Mean

Figure S8. Histogram plots of 92.1, OMM2.5 and UPMM3 cell lines transfected with fluorescent oligonucleotides and analysed by flow cytometry: Mean Fluorescence intensity was calculated by the formula: $MFI = (sample\ Mean - CTR\ Mean) / CTR\ Mean$.



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