Integrin $\alpha 6$ promotes esophageal cancer metastasis and is targeted by miR-92b

SUPPLEMENTARY FIGURES AND TABLES



Supplementary Figure 1: H&E stain of the lungs from the control and the integrin α 6 knockdown group. Fewer metastatic nodules could be observed in the lungs of the integrin α 6 silenced mice than these counterparts from the control mice. The bars in the images indicate 2 mm.



30-D

Supplementary Figure 2: Reduced integrin $\alpha 6$ does not affect the luciferase activity. The specific siRNAs (100 nM in total) against integrin $\alpha 6$ and the scramble oligos were delivered into 30-D cells, respectively. Around 20 hr later, the luciferase activity were detected using Luciferase Assay Kit (Promega, WI, USA).



Supplementary Figure 3: Integrin $\alpha 6$ expression is higher in ESCC cell lines than that in normal esophageal cell line. The mRNA level of integrin $\alpha 6$ was detected by RT-qPCR. The data was analyzed using $2^{-\Delta \Delta Ct}$ method.

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Primary antibody	catalog	Vendor	Application
ITGA6	3883-1	Epitomics	WB
ITGA6	NBP1-85747	NOVUS	IHC
ITGA6	561893	BD	FC
GAPDH	G8795	Sigma	WB
FAK Antibody Sample Kit	9330	Cell Signaling	WB
Phospho-Akt (Thr308)	2965	Cell Signaling	WB
Akt (pan)	4685	Cell Signaling	WB

Supplementary Table 1: Primary antibodies used in this investigation

Application	Sequence	
Clone		
ITGA6 3'UTR	F: TTGAGCTCAGAGTGACTACACACAGTACG	
	R: TTGCTAGCTTATTAAAACAACTCTCTCTGGTAC	
microRNA reverse transcription		
miR-92b	CTCAACTGGTGTCGTGGAGTCGGCAATTCAGTTGAGGGAGG	
qPCR detection		
miR-92b	F: ACACTCCAGCTGGGTATTGCACTCGTCCCG	
	R: TGGTGTCGTGGAGTCG	
ITGA6	F: GCTGGTTATAATCCTTCAATATCAATTGT	
	R: TTGGGCTCAGAACCTTGGTTT	
GAPDH	F: TGCACCAACTGCTTAGC	
	R: GGCATGGACTGTGGTCATGAG	
U6	F: CTCGCTTCGGCAGCACA	
	R: AACGCTTCACGAATTTGCGT	
Mutation		
ITGA6 UTR mut	F: AATCACGTTATGAAGGTGATGTTGCCATCCTAC	
	R: TAGTTTAAGACAGCACTTTCCGTTGAAACACAACT	
shRNA oligos		
ITGA6 sh-4 target	GTTTCTGGATTTCATACTGTAA	
ITGA6 sh-5 target	ATCCCGGCCTGTGATTAATATT	
GFP shRNA target	CGAGAAGCGCGATCACATG	

Supplementary Table 2: List of primers and oligos used in this investigation