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Antagonism between miR-21 and 338-3p in prostate cancer invasion through regulation of matrix metalloproteinase 9

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MicroRNAs

-19-25 nucleotides.

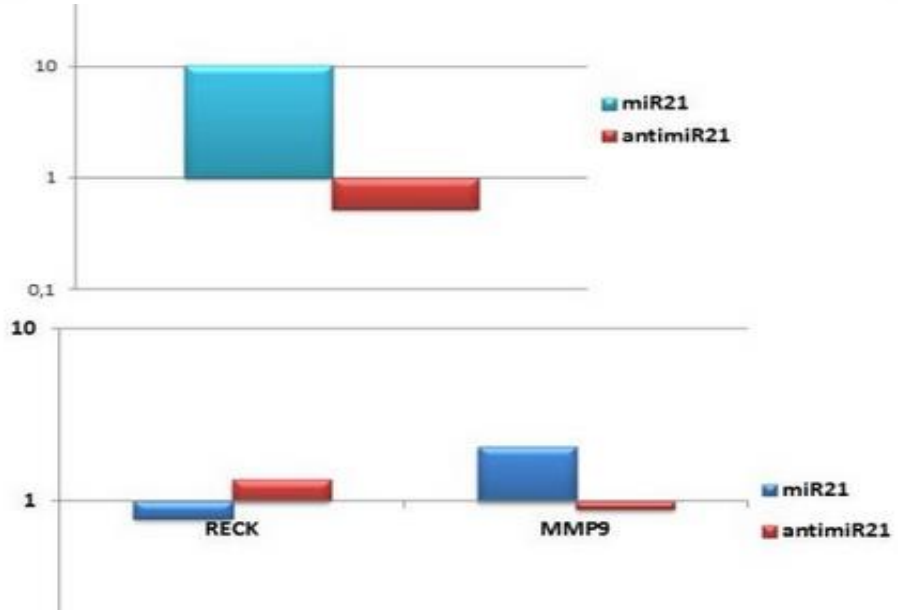
- Non coding RNAs

-Regulates gene expression

- Post transcriptional level

Ha M, Kim VN. *Regulation of microRNA biogenesis*. Nat Rev Mol Cell Biol. 2014 Aug;15(8):509-24

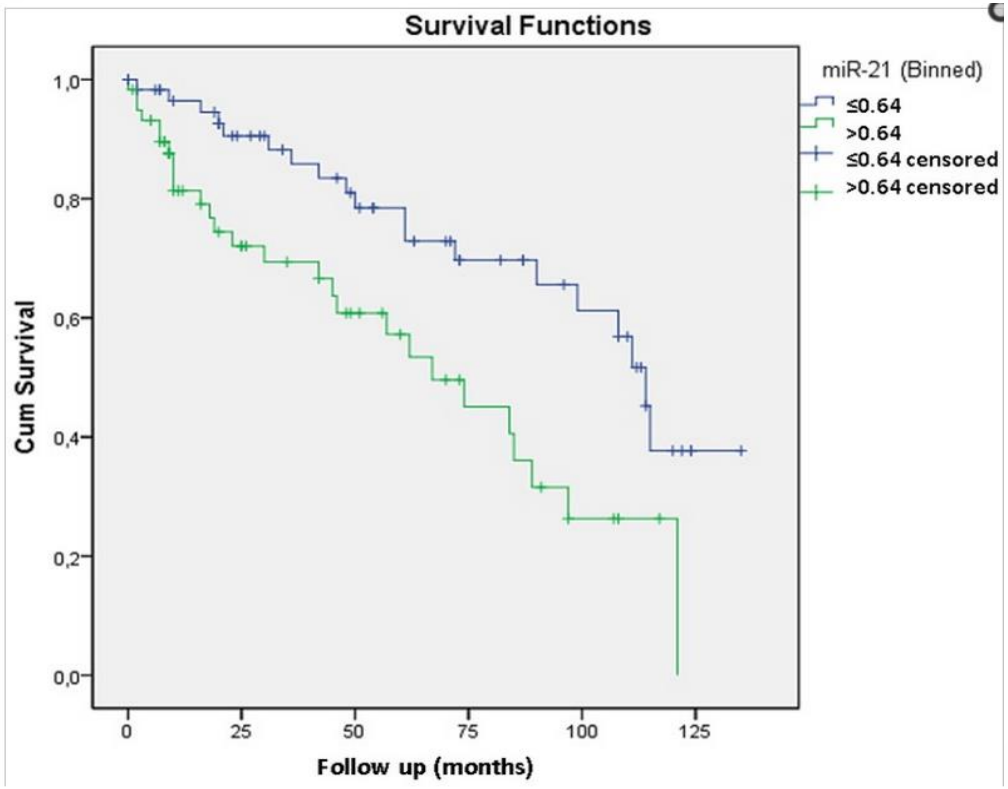
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	↓miR-21 and ↓RECK	↑miR-21 and ↓RECK	↓miR-21 and ↑RECK	P value
Pathological Stage				
pT2	52.0%	8.3%	53.3%	**0.025**
pT3	48.0%	91.7%	46.7%	

- Reis ST et al *miR-21 may acts as an oncomir by targeting RECK, a matrixmetalloproteinase regulator in prostate cancer*. BMC Urol. 2012 May 29;12:14.

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Leite KR et al. *Controlling RECK miR21 Promotes Tumor Cell Invasion and Is Related to Biochemical Recurrence in Prostate Cancer.* J Cancer. 2015 Jan 21;6(3):292-301.

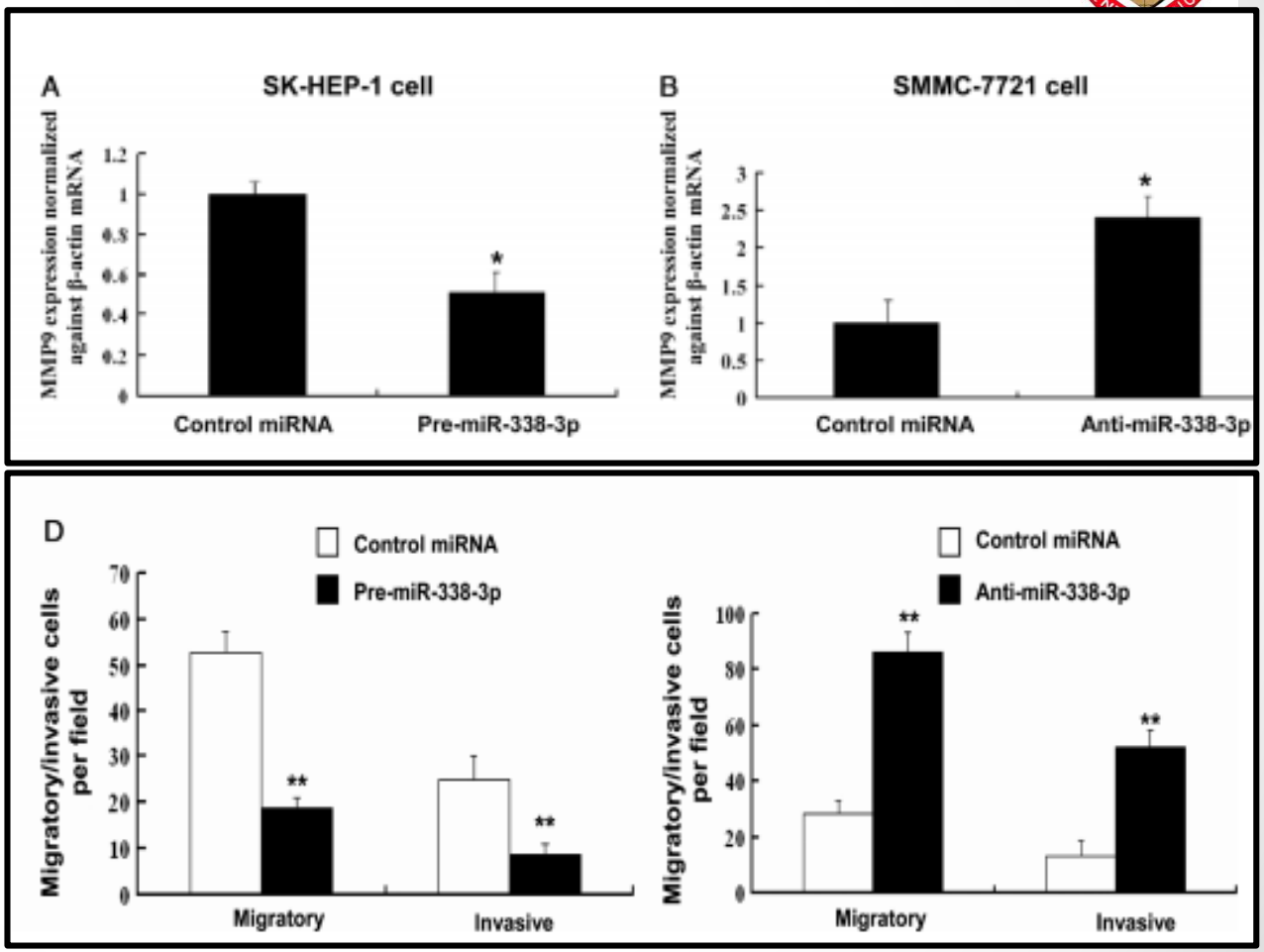
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miR-338-3p

- Low expression in cancer liver cells

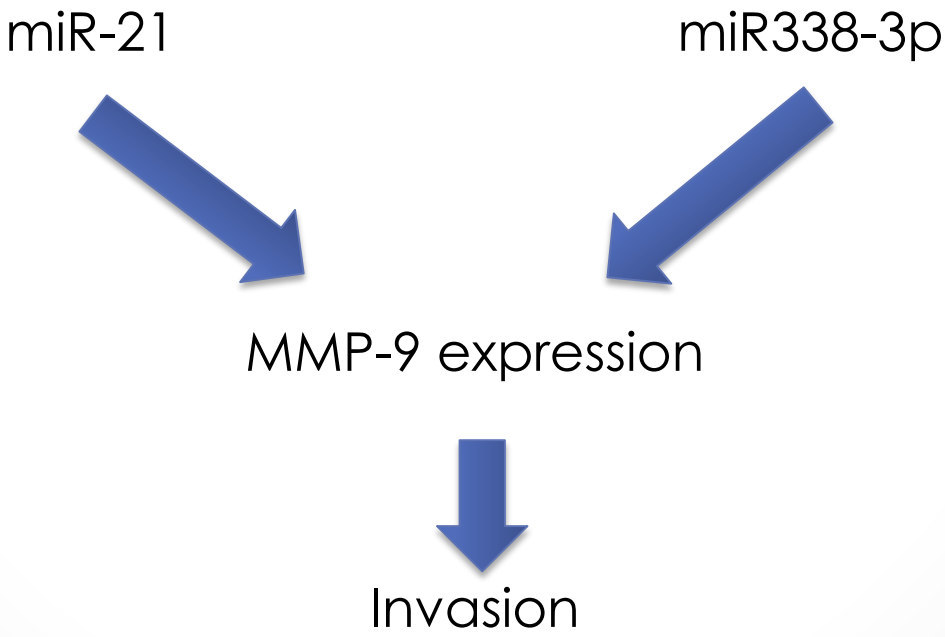
- increase in MMP-9 expression and invasion

- Smoothened (Hedgehog cascade component)



Xiao-Hui Huang et al. miR-338-3p suppresses invasion of liver cancer cell by targeting smoothened. J Pathol 2011; 225: 463–472

Objectives



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Material and methods

DU145, PC3 and LnCaP prostate cancer cell lines – $6x10^4$ cells

mir21, mir338-3p or their antagonists (lipofectamin)

MMP-9 and RECK mRNA expression (2 $^{-\Delta\Delta CT}$ – Livak et al, 2001)

MMP-9 detection was conducted with ELISA

Matrigel Assay:

$3x10^4$ cell (after transfection – 50microL of Matrigel)

24hs



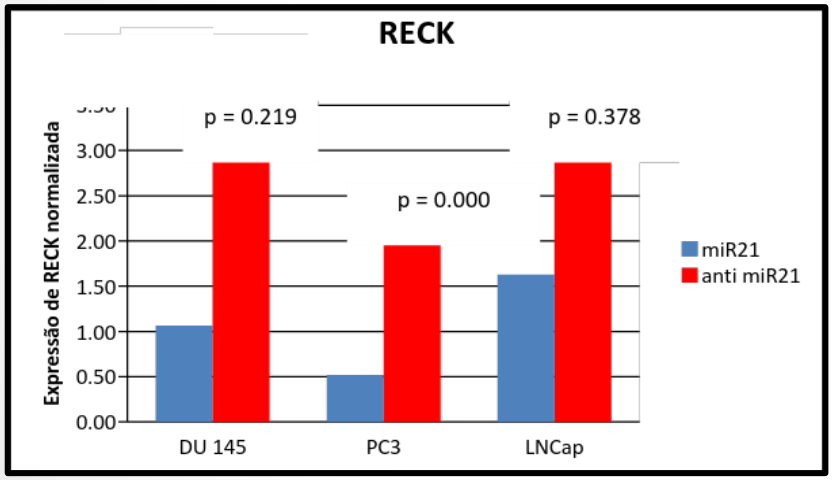
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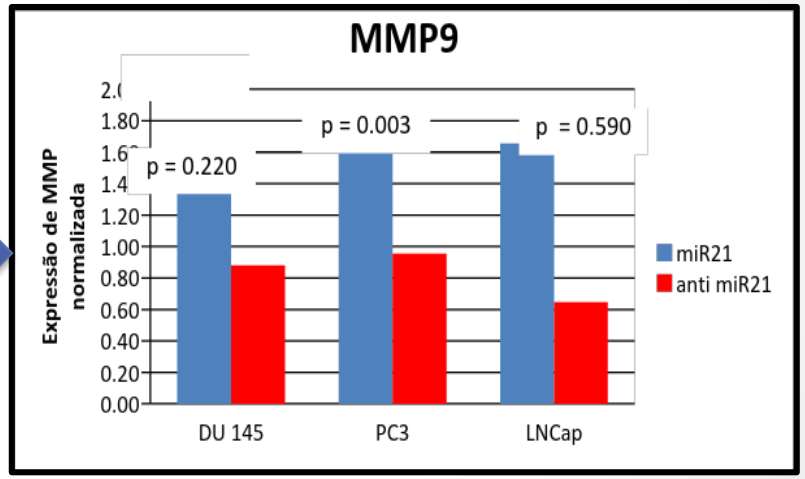


miR-21

Supresses RECK mRNA expression

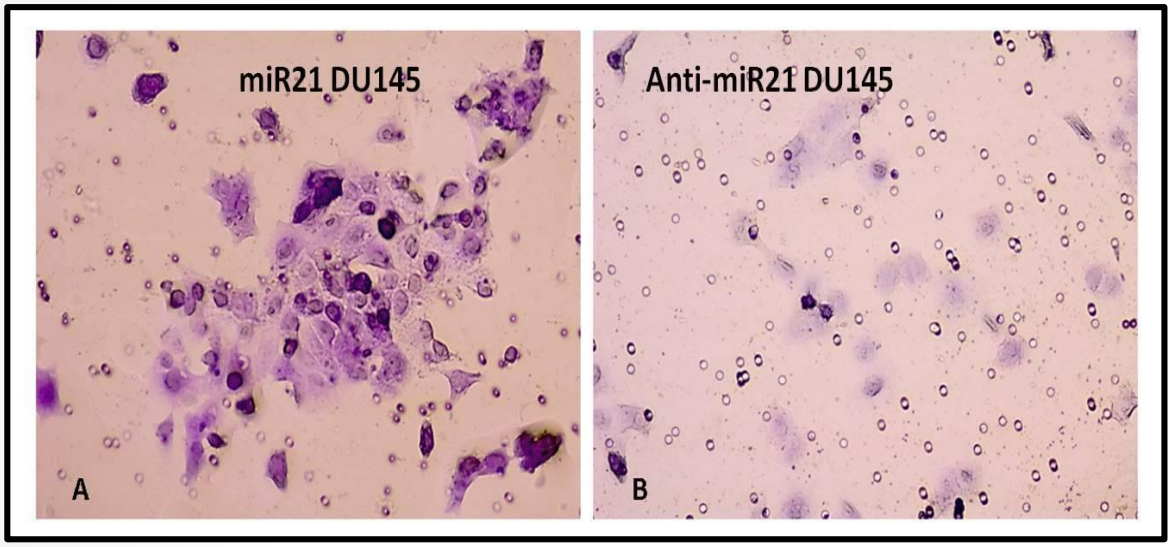
- PC3 (*)
- Du145
- LncaP





(*) p=0.00

miR-21 – Matrigel Assay



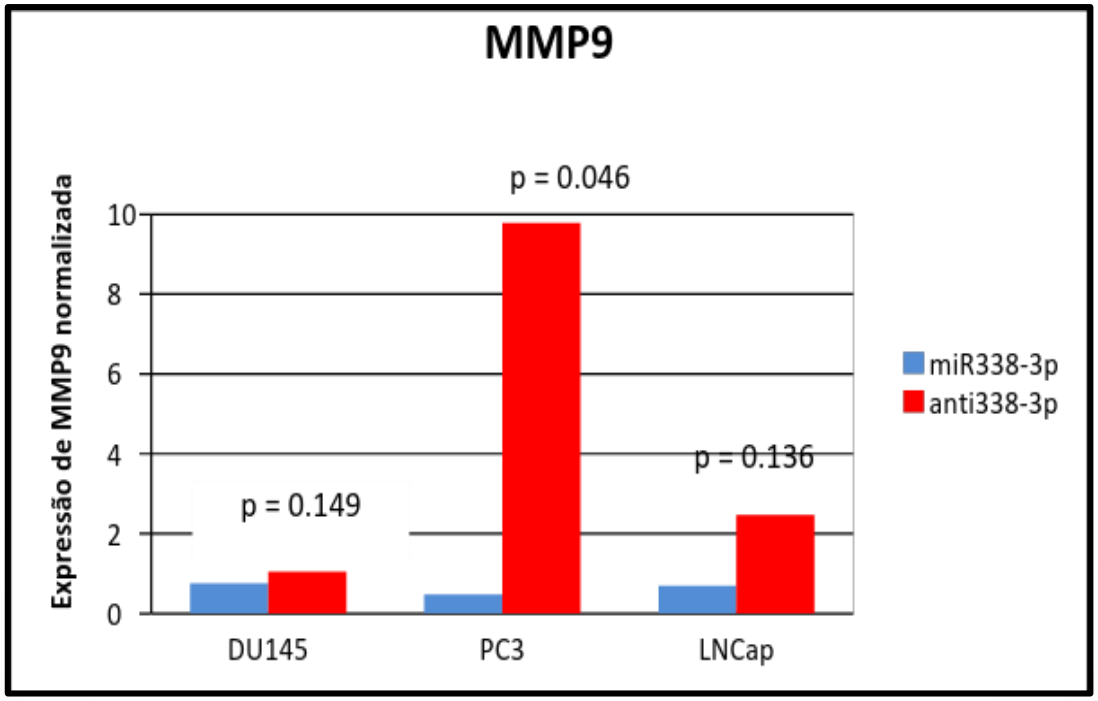
J Cancer. 2015 Jan 21;6(3):292-301

Increased Invasion

DU145 *p= 0.015*
PC3 *p= 0.012*

	MMP-9 – ELISA ASSAY			
	DU145	P-value	PC3	P- value
miR-21	0.140	0.101	0.128	0.06
AntimiR-21	0.053	0.359	0.052	0.217

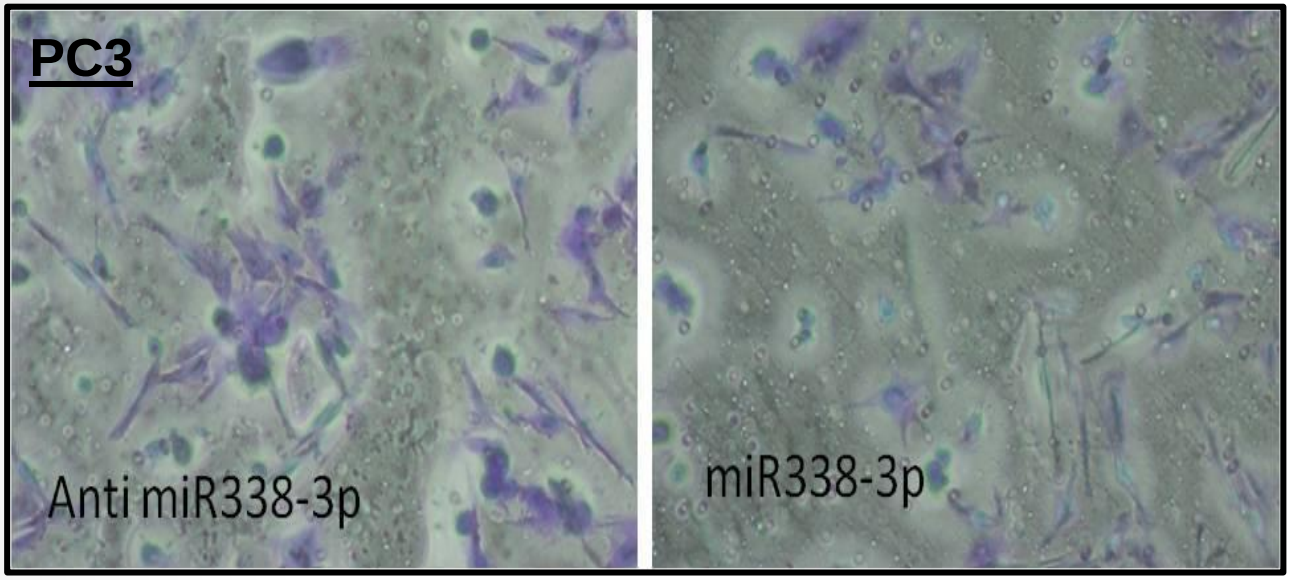
miR-338-3p


MMP9
Expressão de MMP9 normalizada
10
8
6
4
2
0
p = 0.046
p = 0.149
p = 0.136
DU145
PC3
LNCap
miR338-3p
anti338-3p



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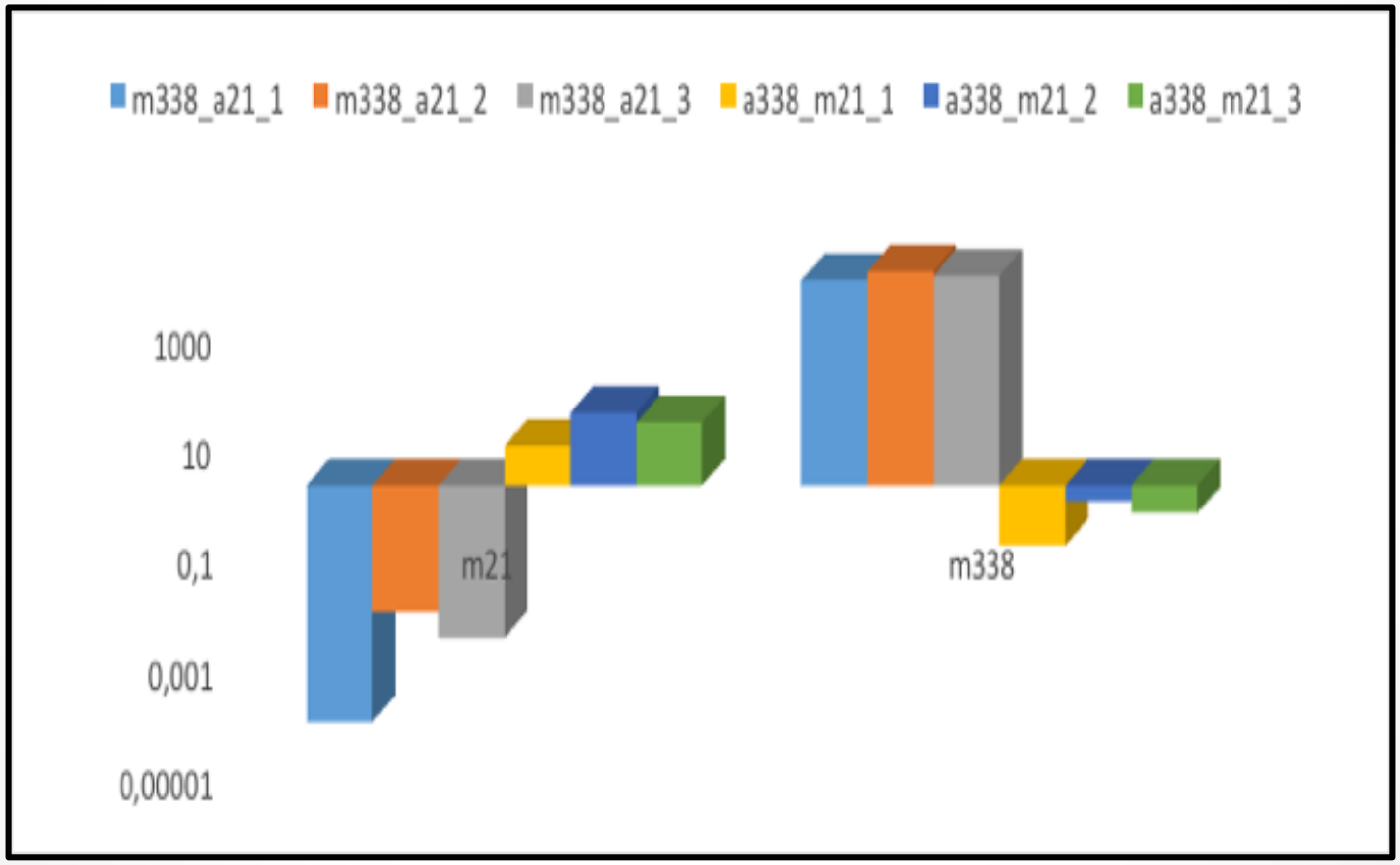


Decreases Invasion

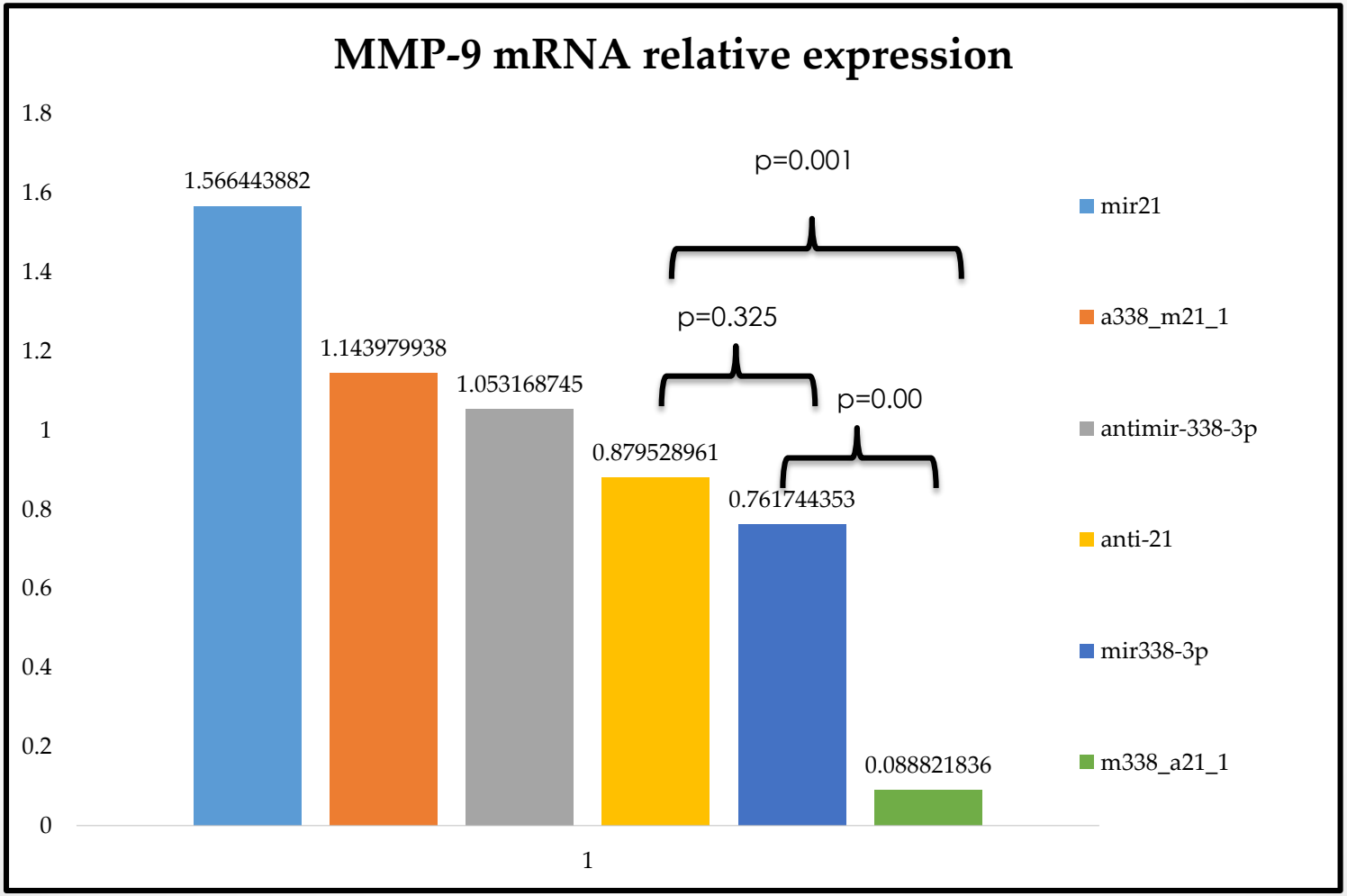
PC3 *p=0.058*
DU145 *p=0.002*
LnCaP *p=0.085*

	MMP-9 – ELISA ASSAY			
	DU145	P-value	PC3	P- value
miR-338-3p	0.04	0.228	0.061	0.066
antimiR-338-3p	0.182	0.259	0.095	0.098

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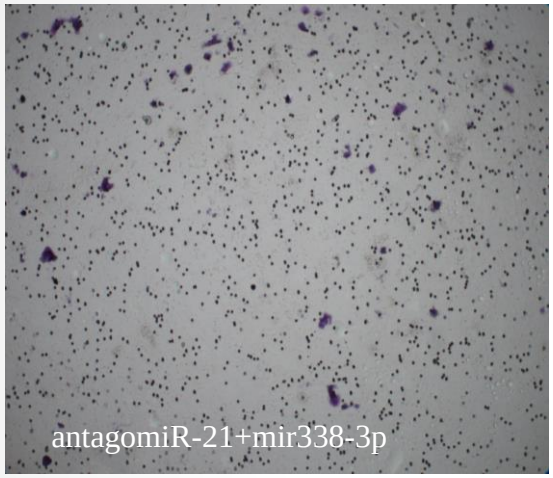
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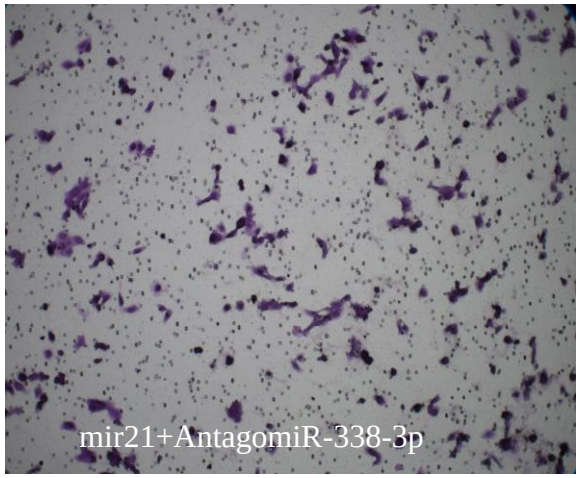




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Number of Cells:
114,67 x 304 (p=0.133)

ELISA FOR MMP-9		
	Mean	
AntimiR-21+ miR-338-3p	2.573	P=0.275
mir-21+anti-mir-338-3p	21.96	

Conclusions:

MMP-9 has its role in Invasion

miR-21 acts as an oncomir, through RECK regulation

miR338-3p acts as a suppressor

Lowest level of MMP-9 expression was achieved with co-transfection

Co-transfection can be performed in an easy way

In vivo study is under development

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