

Title: THE FGFR4-388ARG VARIANT PROMOTES LUNG CANCER PROGRESSION BY N-CADHERIN INDUCTION

Authors and affiliations

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SUPPLEMENTARY INFO

Supplementary Table S1. Description of the driver molecular alterations and FGFR1 and FGFR4 mRNA expression of our lung cell line panel **Figure S1**. Related to **Figure 1**. ADC=Adenocarcinoma, SCC=Squamous Cell Carcinoma, TN="Triple Negative" (referring to the absence of alterations in KRAS, EGFR or ALK), I=Immortalized.

Cell line	Histology	Driver mutation	Reference for driver mutation
H2009	ADC	KRAS p.G12A	COSMIC
HCC827	ADC	EGFR E746-E750 del	(Helfrich, Raben et al. 2006)
Calu-1	SCC	KRAS p.G12C	COSMIC
H520	SCC	TN	COSMIC, (Helfrich, Raben et al. 2006)
H226	SCC	TN	COSMIC, (Helfrich, Raben et al. 2006)
NL20	I	TN	COSMIC

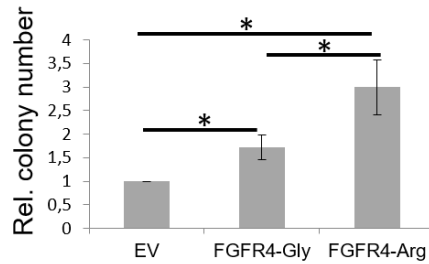
Supplementary Table S2. Characteristics of the resected lung cancer patient cohort.

	Whole cohort	Gly	Arg	p-value
Subjects, n	65	43	22	
Gender				
Male	63 (96.9%)	42 (97.6%)	21 (95.5%)	0.624
Female	2 (3.1%)	1 (2.4%)	1 (4.5%)	
Median age, years	65.0 [54.0-78.0]	66.0 [52.0-79.0]	64 [54.0-77.0]	0.726
Smoking status				
Smoker	34 (52.3%)		12 (54.5%)	0.832
Ex-smokers	29 (44.6%)		9 (40.9%)	
Never-smokers	2 (3.1%)	22 (51.1%)	1 (4.9%)	
Squamous cell carcinoma	34 (52.3%)	20 (46.5%)	11 (50.0%)	0.068
Adenocarcinoma	20 (30.7%)	1 (2.0%)	10 (45.4%)	
Large cell carcinoma	11 (17%)	23 (53.4%)	1 (4.6%)	
Stage I-II	34 (52.3%)	10 (23.3%)	11 (50.0%)	0.790
Stage III	31 (47.7%)	10 (23.3%)	11 (50.0%)	
		23 (53.4%)		
		20 (46.6%)		

p-values were calculated using the Chi-Square test to study the differences between the FGFR4-388gly (Gly) and FGFR4-388Arg (Arg) variants expressing patient subgroups.

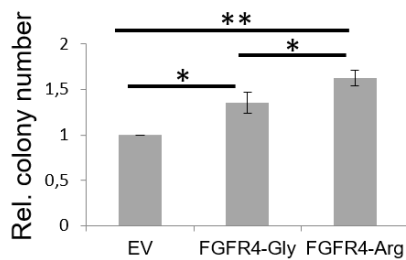
Clonogenicity assay

Immortalized lung epithelial cell line
NL20

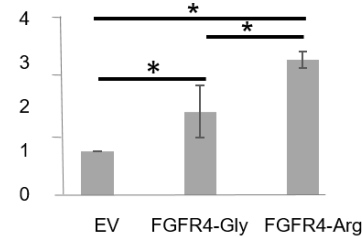


Squamous cell carcinoma cell lines

H226

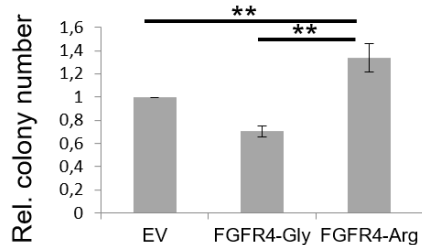


Calu-1

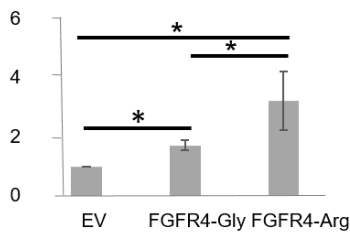


Adenocarcinoma cell lines

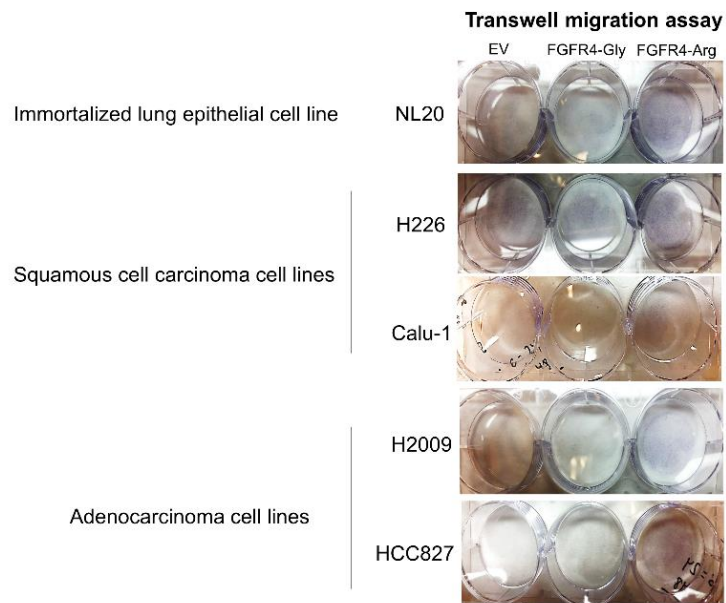
H2009



HCC827



Supplementary Figure S1. Clonogenicity assays for FGFR4-388Gly- and FGFR4-388Arg-overexpressing NL20, H226, Calu-1, H2009 and HCC827 cells. The value for each replicate was normalized to that for the empty vector control in each experiment, and the mean of all normalized replicates is presented. P values (Student's t-test) are indicated by asterisks (* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$). EV=Empty vector control, FGFR4-Gly=FGFR4-388Gly overexpressing, FGFR4-Arg=FGFR4-388Arg overexpressing, FBS=fetal bovine serum.



Supplementary Figure S2. FGFR4-388Arg overexpression increases migration in NSCLC cell lines. Representative images of the wells after the migration assay exhibiting the purple stained migrated cells, fixed in the bottom of the well. EV=Empty vector control, FGFR4-Gly=FGFR4-388Gly overexpressing, FGFR4-Arg=FGFR4-388Arg overexpressing, I=immortalized, SCC=Squamous cell carcinoma, ADC=Adenocarcinoma.