A study of the expression of miR185, miR29a and their targets in IPF and lung cancer: a BALF study.

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Idiopathic pulmonary fibrosis: diagnosis, management and new therapies

BMC Medicine

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Idiopathic pulmonary fibrosis and cancer: do they really look similar?

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- Common risk factors
- Poor response to treatment
- Poor prognosis
- Similar anatomical distribution

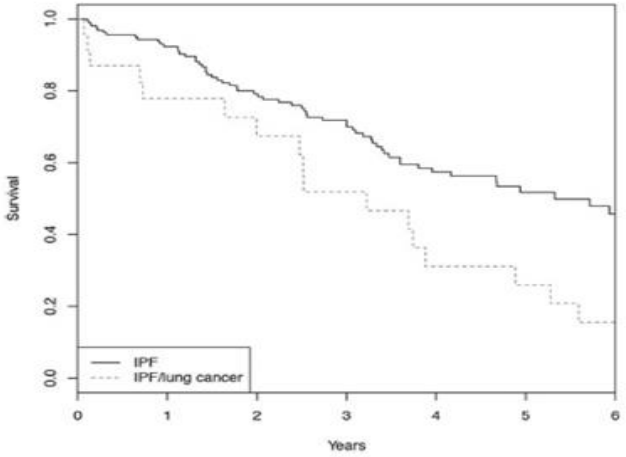
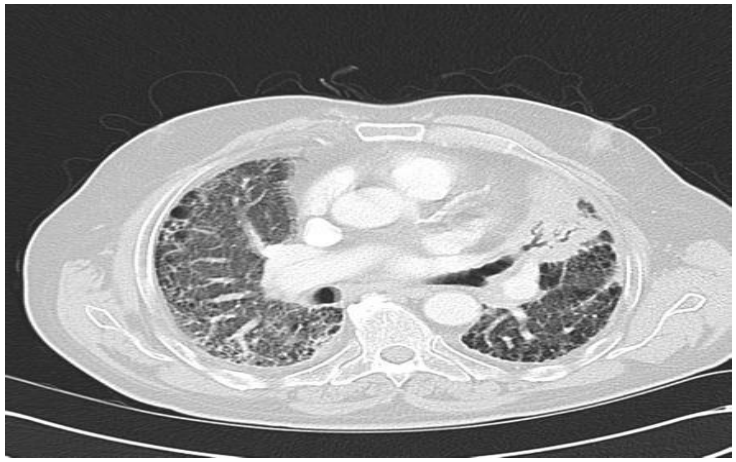


Figure 3 – *Survival curve of patients with IPF with and without LC (time 0 is diagnosis of IPF for both groups). One-y and 3-y survival among the two groups were 78% and 52% in the study group and 92% and 70% in the control group, respectively, by Kaplan-Meier analysis. IPF = idiopathic pulmonary fibrosis. See Figure 2 legend for expansion of other abbreviation.*



The Impact of Lung Cancer on Survival of Idiopathic Pulmonary Fibrosis
Tomassetti et al *Chest, Volume 147, Issue 1, January 2015, Pages 157-164*

Vancheri *BMC Medicine* (2015) 13:220
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- Genetic changes (microsatellite instability, telomere shortening, telomerase impairment).
- Epigenetic changes (promoter hypermethylation/global hypomethylation).
- Abnormal expression of micro RNAs.
- Cellular and molecular aberrances:

✓ Altered cell-cell communication

✓ EMT

✓ Delayed apoptosis

✓ Pathways (Wnt/b-catenin, PI3K/AKT, JAK/STAT, Tyrosine kinases and others….)

Vasilakis et al.Am J Respir Crit Care Med Vol 162. pp 1115–1119, 2000

Vancheri BMC Medicine (2015) 13:220

Idiopathic pulmonary fibrosis and lung cancer: a clinical and pathogenesis update

Katerina M. Antoniou[a,b], Sara Tomassetti[c], Eliza Tsitoura[b], and Carlo Vancheri[d]

Epigenetic mechanism	IPF	Cancer	Effect
Noncoding RNAs			
miR-21	Upregulated [71,72]	Upregulated [43,60]	Oncomir and promising serum biomarker for IPF
			Targets tumor suppressors including *PTEN*. Amplifies the TGF-b signaling pathway by targeting SMAD7
Let-7d	Downregulated [74▪]	Upregulated or downregulated [73]	Can act as an oncogene and a tumor suppressor, in IPF its downregulation results in upregulation of HMGA2 and promotion of EMT and TGF-b pathways
miR-210	Upregulated [78▪]	Upregulated [79]	Hypoxia inducible, promotes fibroblast hyperproliferation
miR-17–92 family	Downregulated [88]	Upregulated [86,87]	Targets tumor suppressors including *PTEN*, *CDKN1A/p21*$^{waf1/cip1}$ causing increased cell proliferation, inhibition of apoptosis, angiogenesis, EMT transformation
			Targets DNMT1
miR-29 family	Downregulated [81]	Downregulated [82,85▪]	Targets ECM proteins-like collagens targets DNMT3A/B leading to accumulation of aberrant epigenetic marks

Global Methylation Patterns in Idiopathic Pulmonary Fibrosis

Einat I. Rabinovich[1], Maria G. Kapetanaki[1], Israel Steinfeld[2], Kevin F. Gibson[1], Kusum V. Pandit[1], Guoying Yu[1], Zohar Yakhini[2,3], Naftali Kaminski[1]*

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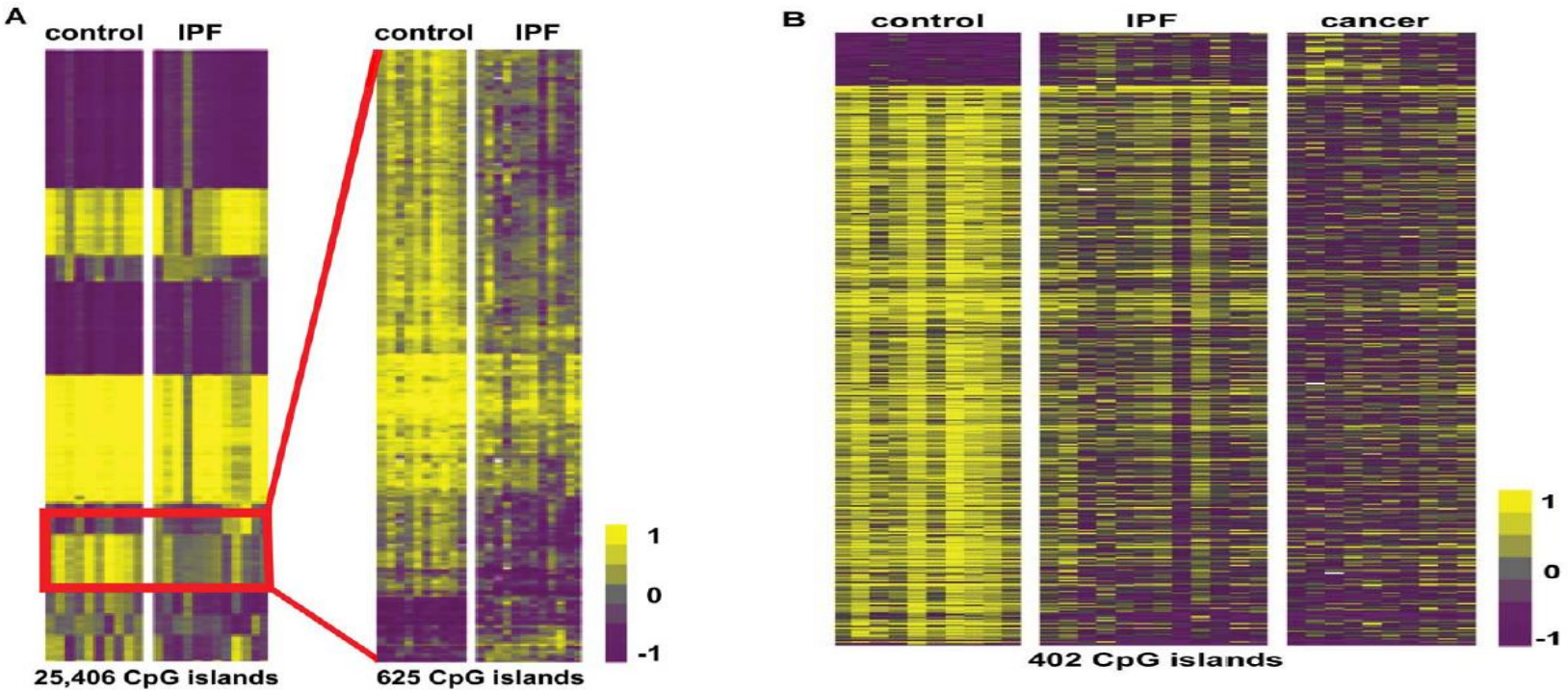


•65% of the CPG islands with altered methylation in IPF are also modified in LC.

•The methylation profile of IPF shared similarities with both groups, but were more similar to LC group.

MiR-185/AKT and miR-29a/Collagen 1a pathways are activated in IPF BAL cells

Eliza Tsitoura[1,2], Athol U. Wells[3], Kostantinos Karagiannis[1], Ismini Lasithiotaki[1], Eirini Vasarmidi[1,4], Eleni Bibaki[1,4], Chara Koutoulaki[1], Hiroe Sato[3], Demetrios A. Spandidos[2], Nikolaos M. Siafakas[1], George Sourvinos[2] and Katerina M. Antoniou[1,4]

Oncotarget, Vol. 7, No. 46

We have previously identified significant decrease of **miR185** and **mir29**a in IPF BAL consistent to the profibrotic profile of lung macrophages in IPF. However there are few data published regarding LC.
Is there common markers in the BALF of LC?

miR185 :
• down-regulated both in IPF and LC
• associated with deregulation of cell cycle and cell proliferation
•alleviate TGFb-induced EMT and col(v) overexpression.
•Inhibits the PI3K-AKT pathway by targeting DNMT1 which activates PTEN through methylation of its promoter.

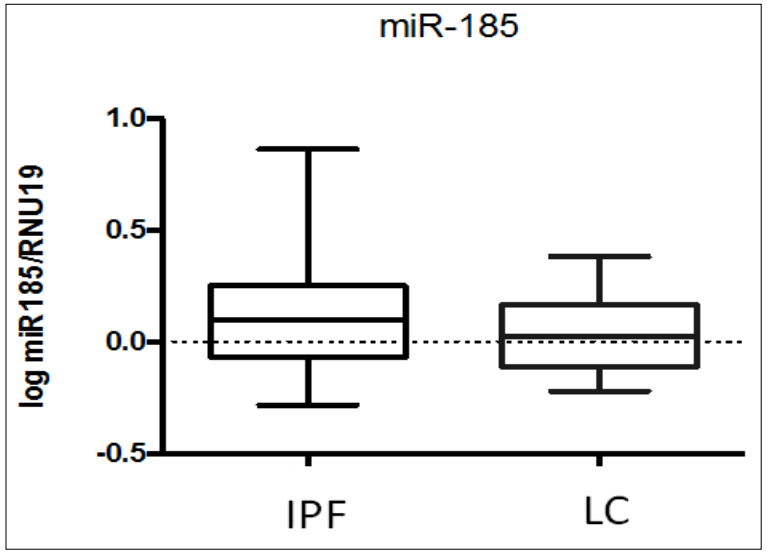
miR29a:
• down-regulated both in IPF and LC
•Targets DNMTs
•Reduces the expression of COL1a1 through inhibiting the phosphorylation of AKT.

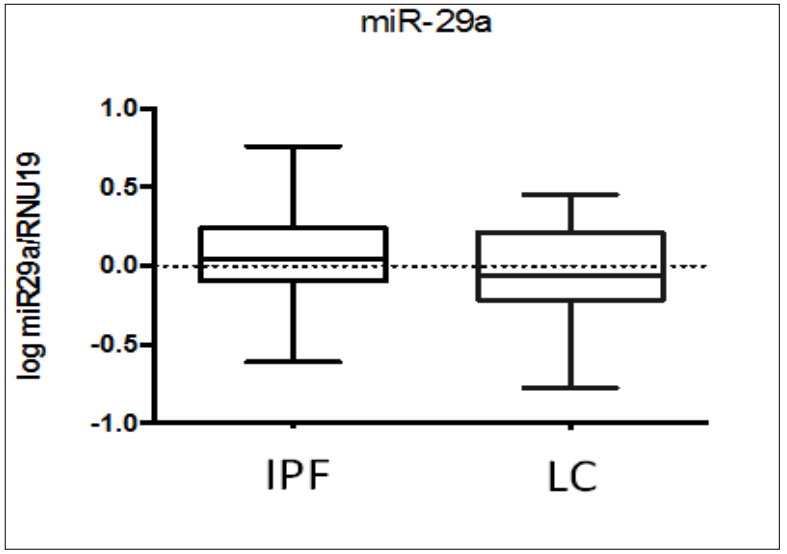
METHODS

- BALF cells from: 57 IPF and 32 LC patients.
- MiR-185, miR29a and corresponding mRNA targets (DNMT1,DNMT3b,AKT1,AKT2) were analysed by RT-PCR.
- Expression levels were evaluated according to:
  - •Disease
  - •Age
  - •smoking status
  - •PFTs
  - •Side of endobrochial lesion
  - •Cytology
  - •pathology results

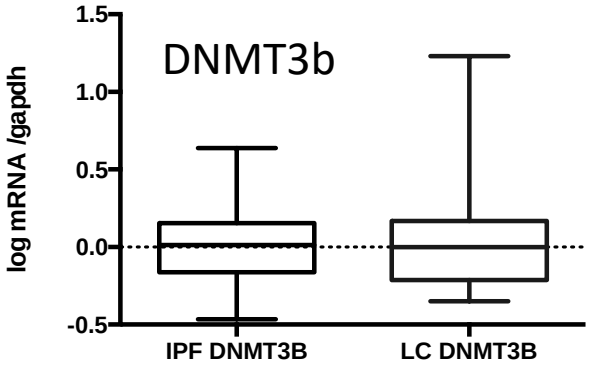
Demographics	**LC (32)**	**IPF (n=57)**
Age(p=0.01)	67.7	72
Gender(m/f)	26/6	46/11
Pyrs (p=0.001)	70,2	37.4
Non smoker/smoker	2/26	13/ 43
PFTs	**LC**	**IPF**
FVC%	ND	78.6
FEV1%	ND	84,8
%	ND	83,8
TLC	ND	73,6
TLCO/SB	ND	52,3
KCO	ND	88,5
BALF %	**LC**	**IPF**
Macrophages	81,3	76,6
Lympocytes	10,2	11,5
Neutrophils	8	9,3
Eosinophils (p=0.01)	1,3	1,8

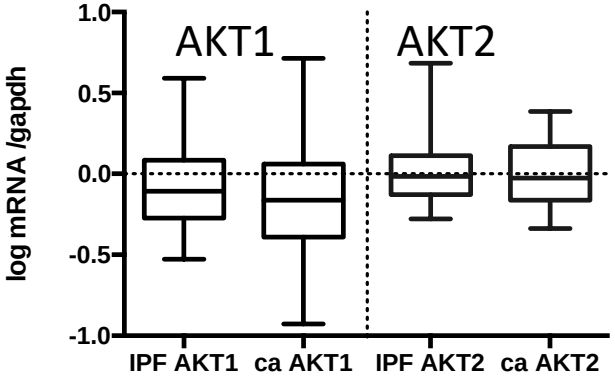
RESULTS



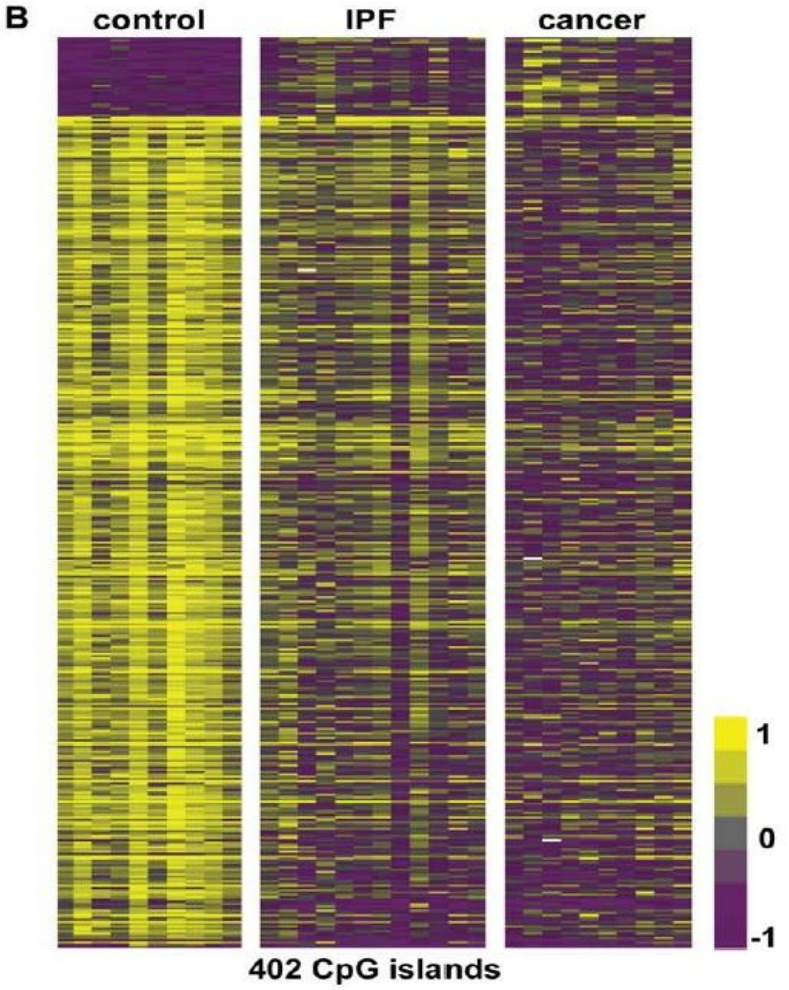


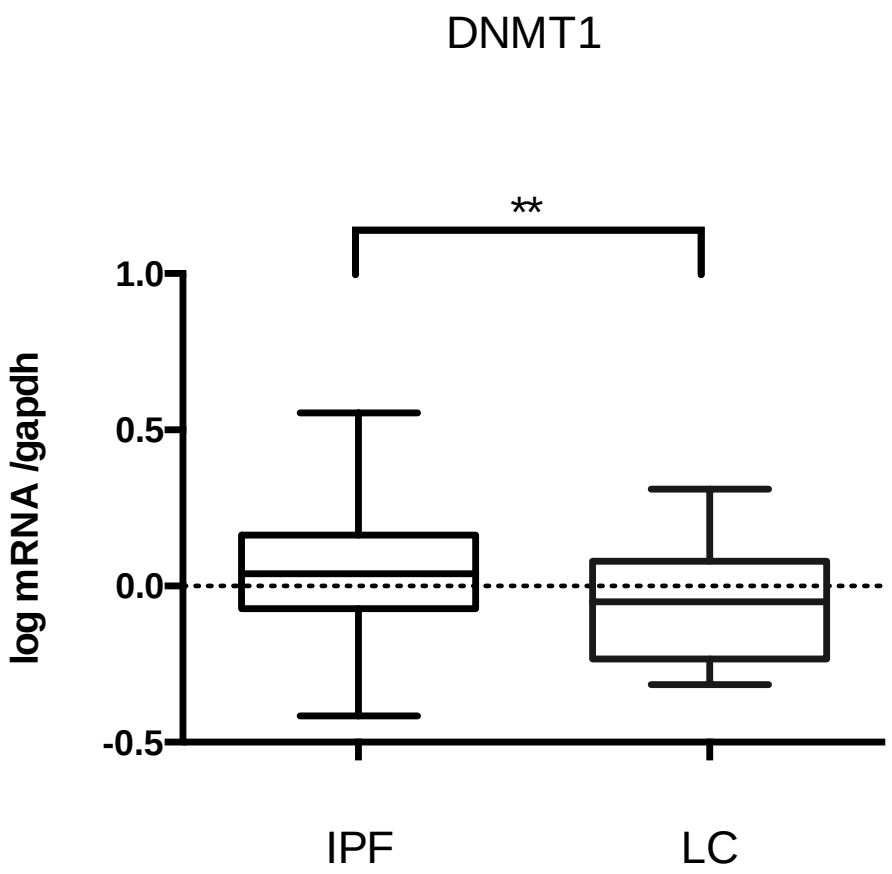
Mir185: inverse correlation with age (R -0.5 p=0.009) and PYrs (R -0.45 p=0.02)



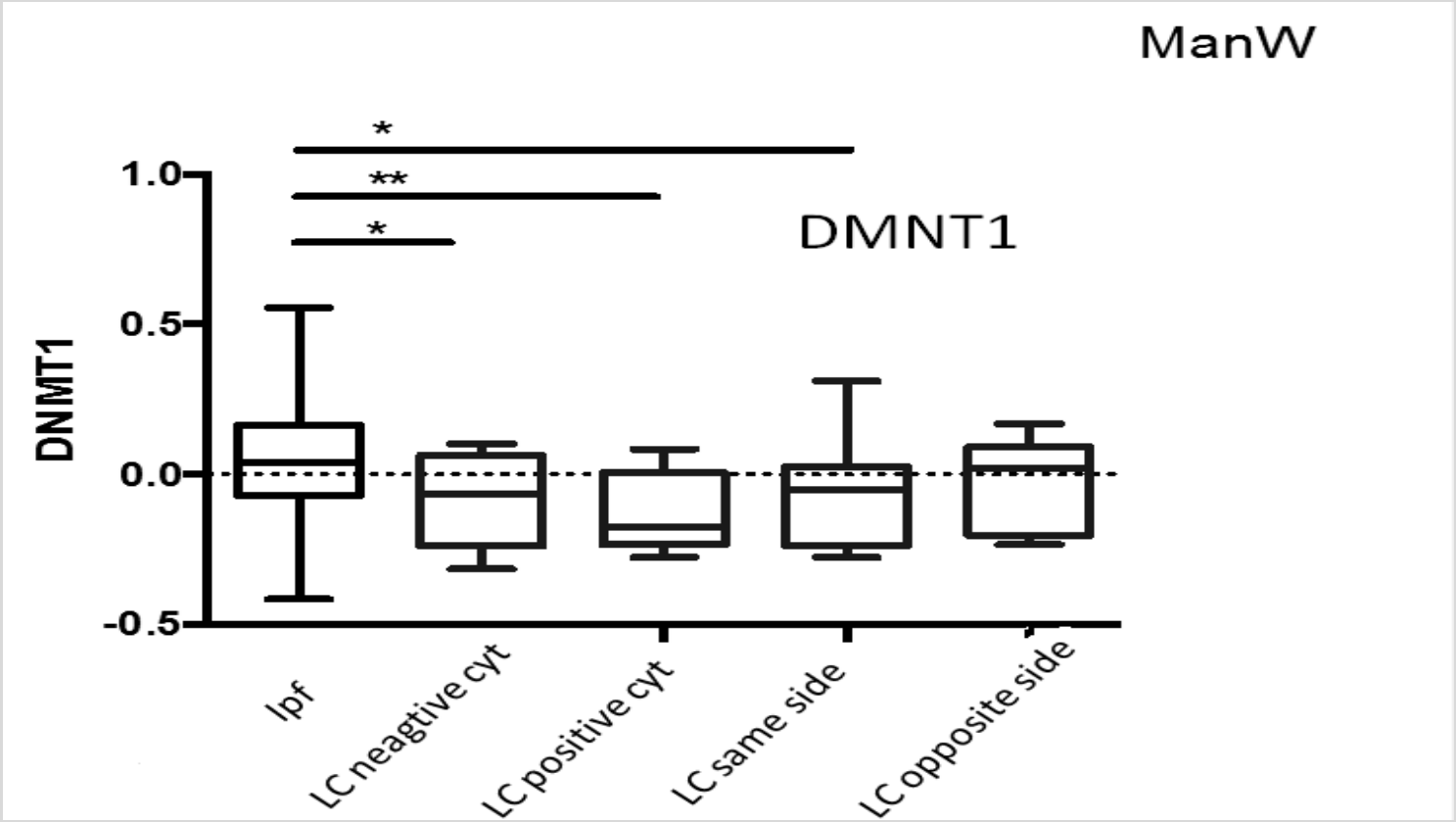


RESULTS


B
control
IPF
cancer
1
0
-1
402 CpG islands


DNMT1
**
1.0
0.5
0.0
-0.5
log mRNA /gapdh
IPF
LC

RESULTS


ManW
DMNT1
DNMT1
1.0
0.5
0.0
-0.5
*
**
*
lpf
LC neagtive cyt
LC positive cyt
LC same side
LC opposite side

Conclusion

- Similarities between IPF and LC were noticed at the level of miRNAs (miR-185 and miR-29a) with known implication in both diseases and their targets.
- However, DNMT1 seems to be downregulated in LC and further reduced in the presence of malignant burden in BALF.
- More targets of the two miRNAs at the level of mRNA and protein are planned to be estimated, such as col1a1 and Cox2a.
- Moreover, further evaluation of BALF cells profile are going to be estimated between the two diseases using flow cytometry.

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