Relationship between mRNA-microRNA interactions and forced vital capacity in patients with idiopathic pulmonary fibrosis

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INTRODUCTION

- Idiopathic pulmonary fibrosis (IPF) is a progressive fibrosing interstitial lung disease characterized by decline in lung function.
- MicroRNAs are small non-coding RNA molecules with functions in gene silencing or post-transcriptional gene regulation. Altered microRNA expression has been implicated in the pathogenesis of IPF.[1]
- Further investigation is needed to understand the relationships between messenger RNAs (mRNAs) and microRNAs and progression of IPF.

AIM

- To investigate the relationship between mRNA-microRNA interactions and forced vital capacity (FVC) in patients with IPF.

METHODS

Subjects

- The cohort was drawn from the Idiopathic Pulmonary Fibrosis Prospective Outcomes (IPF-PRO) Registry, a multicenter US registry that enrolled patients with IPF that was diagnosed or confirmed at the enrolling center in the past 6 months.[2]
- These analyses were based on samples taken at enrollment from 272 subjects who had whole blood mRNA and plasma microRNA sequencing data that met quality control filters.

Analyses

- T-tests were used to determine differential mRNA and microRNA expression between subjects with FVC % predicted in the lowest tertile (<63.7% predicted; n=90) and the highest tertile (>76.8% predicted; n=92).
- We then used Pearson correlation to identify negatively correlated mRNA-microRNA pairs among:
  - mRNA transcripts with an absolute fold change >1 and p≤0.05 for the difference between lowest versus highest tertiles of FVC % predicted.
  - microRNAs with p≤0.05 for the difference between lowest versus highest tertiles of FVC % predicted.
- Functional and network analyses were used to visualize top mRNA-microRNA connections.
- mRNA-microRNA interaction analyses were performed in R using miRComb;[3] p-values were adjusted for multiple testing.
- Pathways analysis was performed using Ingenuity Pathway Analysis (QIAGEN Inc.). Databases searched were miRTarbase, microCOSM, mirDB, targetScan, and mirWalk2.

CONCLUSIONS

- We identified a number of mRNA-microRNA pairs that were differentially expressed in patients with IPF in the lowest versus the highest tertile of FVC % predicted.
- This supports the idea that microRNA regulation may be related to the progression of IPF.
- Ongoing studies will assess whether circulating microRNAs and their related mRNAs are associated with a greater risk of disease progression in patients with IPF.

RESULTS

Subjects

Baseline characteristics of subjects in the highest and lowest tertiles of FVC % predicted

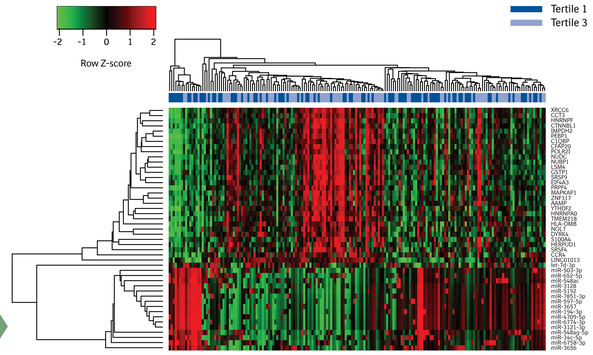
	Tertile 1: FVC <63.7% predicted (n=90)	Tertile 3: FVC >76.8% predicted (n=92)
Age, years	69.5 (64.0, 73.0)	71.0 (65.0, 75.5)
Male	70 (78%)	64 (70%)
White	83 (92%)	87 (95%)
Smoking		
Past	58 (64%)	64 (70%)
Never	32 (36%)	27 (29%)
Current	0	1 (1%)

Values are median (Q1, Q3) or n (%).

Differential expression of mRNAs and microRNAs

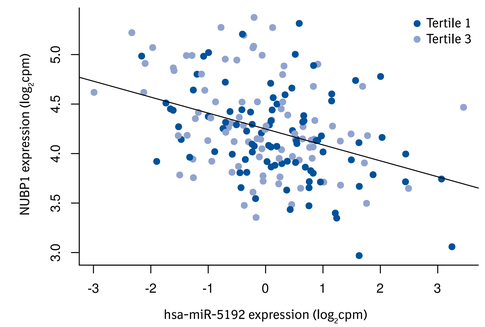
- Of 35628 mRNAs and 2576 microRNAs sequenced, 2441 and 214, respectively, met the criteria for differential expression between subjects in the lowest versus the highest tertile of FVC % predicted.
- A cluster heatmap showed sub-clusters of expression among the top mRNA-microRNA pairs from the differentially expressed mRNAs and microRNAs.

Top mRNA-microRNA pairs from the mRNAs and microRNAs differentially expressed between subjects in the highest and lowest tertiles of FVC % predicted



- The mRNA-microRNA pair with the strongest negative correlation was the nucleotide binding protein 1 (NUBP1) transcript and the microRNA hsa-mir-5192 (r=-0.37; p=1.03e-07):

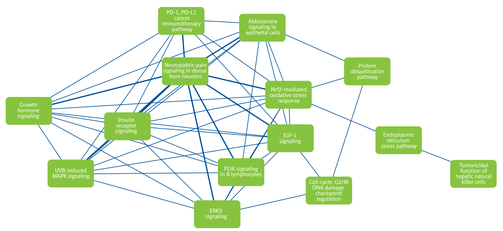
mRNA-microRNA pair with the strongest negative correlation



cpm, counts per million.

Pathways analysis

mRNA-microRNA pairs with a confirmed connection in ≥1 database searched

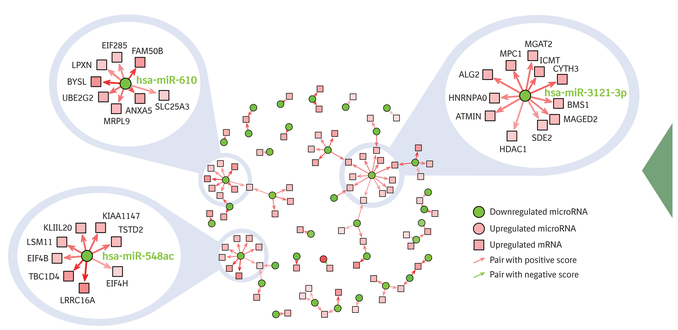


Pathways analysis suggested alterations in pathways previously associated with the pathogenesis of IPF or lung injury: aldosterone signaling,[4] Nrf2-mediated antioxidant response,[5] and IGF-1 signaling[6]

IGF-1, insulin-like growth factor 1;
Nrf2, nuclear factor E2-related factor 2.

Network analyses

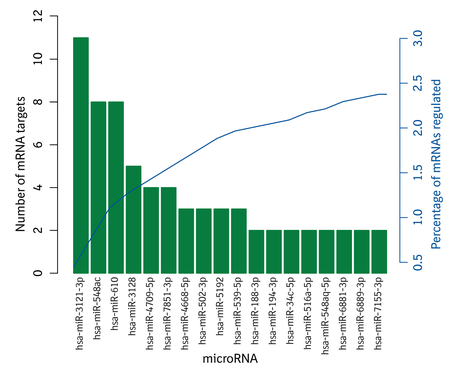
Network of mRNA-microRNA interactions with adjusted p≤0.05



Darker shades in squares or circles indicate stronger up- or downregulation. Darker shades of arrows indicate connections were found in a greater number of the databases searched.

Functional analyses

MicroRNAs with ≥2 mRNA targets with adjusted p≤0.05, and cumulative percentage of 2441 differentially expressed mRNAs regulated by each microRNA



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