ECB 2025 Conference
The 3rd International Electronic Conference on Biomedicines
12–15 May 2025 | Online
MDPI

Pulmonary fibrosis and lung cancer: unity and difference of cellular processes

Pershina O.1,2, Pakhomova A.1,2, Rybalkina O.1,2, Uzyanbaev I.1,2, Kogai L.1, Sandrikina L.1,2, Fedorova E.1,2, Sedinova V.1, Zhdanov V.1
Goldberg ED Research Institute of Pharmacology and Regenerative Medicine, Tomsk NRMC of the RAS, Tomsk, Russia 1
Siberian State Medical University, Tomsk, Russia 2

INTRODUCTION AIM

Pulmonary fibrosis and lung cancer are severe lung diseases that are difficult to treat. Previously, pulmonary fibrosis (PF) was classified as an orphan disease. The global COVID-19 pandemic has increased the severity of PF, as lung infections aggravate PF and create the risk of its development. Lung cancer is the most common form of cancer, it leads in the number of deaths compared to other oncopathologies [1,2]. Despite advances in diagnosis, treatment and care for patients with lung cancer and PF, most of them have a poor prognosis. Unfortunately, the average life expectancy after diagnosis for both diseases is up to 5 years. The both diseases tend to be a difficult diagnostic task. In 1965, the article appeared that first proposed a link between PF and lung cancer. Since then, despite the many common genetic, molecular and cellular processes that were subsequently discovered that link both diseases, there is still no clear answer about the contribution of each process to the mechanisms of development [1]. The PF and lung cancer have common risk factors. In addition, PF is a risk factor for the development of lung carcinogenesis. It is known that in patients with mild fibrosis, the risk of developing lung cancer has increased more than sevenfold, patients with fibrosis and lung cancer have a poor prognosis compared to patients with fibrosis alone. One of the features that distinguishes the cancer process from fibrosis may be cancer stem cells (CSC). In modern scientific literature, there is increasing evidence that CSCs are crucial in the occurrence and progression of tumor diseases and may be a promising therapeutic target for various types of cancer. The search for markers of tumor process occurrence is especially important in diseases that themselves may be risk factors for the development of carcinogenesis. At present, identification of CSCs still presents certain difficulties: there are no validated methods for their isolation, there are no specific markers for this class of cells, and none of the markers are specific for tumor cells. Further clinical and experimental studies are needed to determine the role of these changes and to find biomarkers that predict the progression of pulmonary fibrosis and the risk of developing lung cancer. In our opinion, it is promising to study models of combined pathology of pulmonary fibrosis and lung cancer.

METHOD

Male C57BL/6 mice (age, 8-10 weeks) (from the Department of Experimental Biomodels of Tomsk NRMC, Tomsk, Russia) were used in the experiments. All experimental protocols were approved by the IACUC of the Goldberg ED Research Institute of Pharmacology and Regenerative Medicine, Tomsk NRMC.

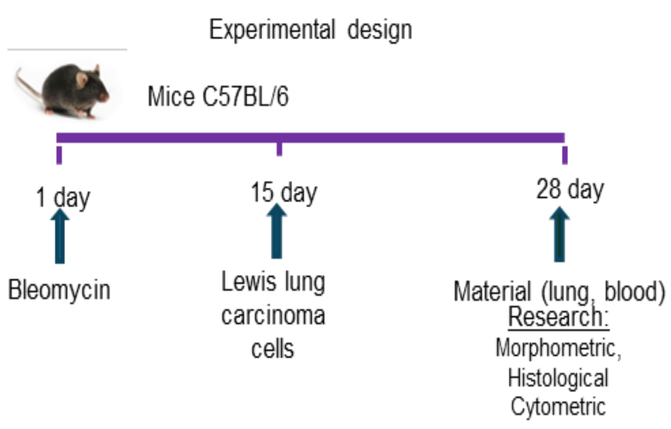

Figure 1. The experimental design of investigation

The study design is shown in Figure 1. It was evaluated the changes in the lung and blood at the modeling of pulmonary fibrosis (PF), lung cancer (LC), PF+LC. Experimental PF was induced by a single intratracheal bleomycin (BLM, Nippon Kayaku Co., Ltd., Tokyo, Japan). The Lewis lung carcinoma (LLC, 400263 CLS Cell Lines. Service, GmbH, Germany) cell line of C57BL strain was used in experiments for induce LC. Mice were injected subcutaneously into the right axillary region with 5×10^6 LLC cells.

It was shown that a single intratracheal administration of BLM to mice initially leads to the development of the inflammatory phase of the disease, which lasts about 7-10 days, then passes into the fibrotic phase, which most closely imitates the manifestations of idiopathic pulmonary fibrosis in humans. This model of PF is a standard and well-characterized model to study the mechanisms of pulmonary fibrosis and to test drugs with antifibrotic activity. On the 15th day after the administration of BLM, when fibrosis has already formed, we modeled non-small cell lung cancer by administration of LLC cells using a model of spontaneous metastasis. The material was collected on the 28th day after the administration of BLM.

The material for study was analyzed by histology and cytometry. Histological examination of the lungs was carried out standard method. Lung preparations were fixed in 10% neutral buffered formalin, passed through in-creasing concentrations of alcohol to xylene and embedded in paraffin wax according to the standard method, then sectioned into 5 µm thick slices, and stained with hematoxylin and eosin or Van Gieson's stain. Expression of membrane and intracellular receptors of CSCs, endothelial cells, and fibrocytes derived from the blood and the lungs was studied using mouse monoclonal antibodies following standard flow cytometry protocols. All statistical analyses were carried out by using SPSS statistical software (version 15.0, SPSS Inc., Chicago, IL, USA).

RESULTS & DISCUSSION

The experiments showed that the use of LLC cells reflects pathomorphological changes in the lung tissue corresponding to LC. The histological examination showed that tumor nodules were formed in the animals. In the lungs of mice, the group with PF+LC, a significant acceleration of the spread of metastasis was noted compared to the group with LC only. The formation of combined pathology led to a reliable increase in the mass of tumor nodules and the area of metastases relative to the group with LC without fibrosis. We observed a reliable increase the connective tissue in the lungs of mice from all groups which was more pronounced in the group with combined pathology. We were studied changes in various cell populations in the lung tissue.

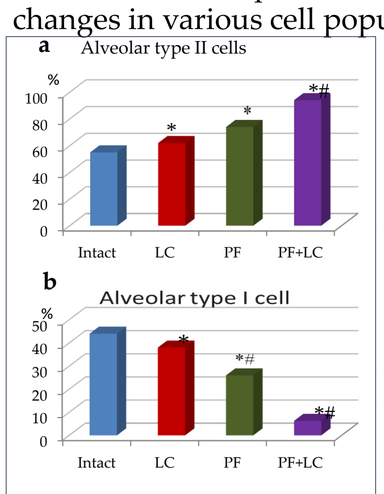

Figure 2. The count (% of total epithelial cells number) of alveolar type II and I epithelial cell in lung of mice

The population of epithelial cells in the lungs increased in all pathologies studied, and more pronouncedly in combined pathology. At the same time, the alveolar type II epithelial cell population is increased, and again more pronouncedly in PF+LC (fig 2a). This circumstance, among other things, confirms the participation of type II cells in the development of PF and LC. The superposition of the two processes enhanced the effect. At the same time, the population of alveolar type I, which is less resistant to damage and which, as a rule, is the first to suffer from any impact, significantly decreased in combined pathology (fig. 2b). The cell population expressing epidermal growth factor increased to a greater extent in PF (fig. 3a). It is known that increased expression of various EGF ligands is associated with the development of fibrosis, and this same marker is identified as an oncogenic factor in non-small cell lung cancer.

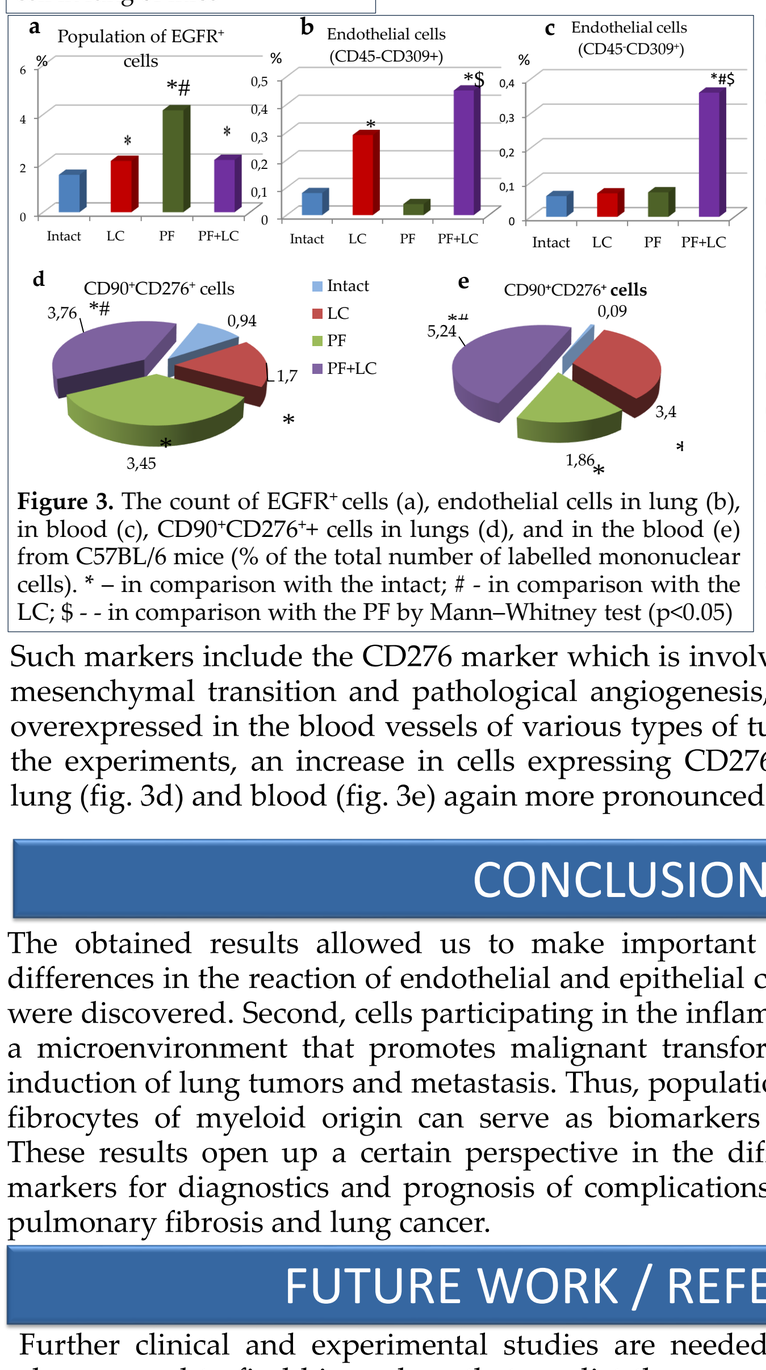

Figure 3. The count of EGFR+ cells (a), endothelial cells in lung (b), in blood (c), CD90+CD276++ cells in lungs (d), and in the blood (e) from C57BL/6 mice (% of the total number of labelled mononuclear cells). * – in comparison with the intact; # - in comparison with the LC; $ - - in comparison with the PF by Mann–Whitney test ($p<0.05$)

Cell populations expressing CD276 and CD274 demonstrated a more pronounced increase in groups of animals with PF and PF+LC. The study of cell populations carrying tumor stem cell markers on their surface showed that the specificity of changes in the lung tissue is generally uniform, the differences mainly concern the degree of expression of changes. We assessed various popular-tions of endothelial cells in the lung and blood. In combined pathology, cell populations expressing CD309 increased in the lungs and blood (fig. 3b, c).

Such markers include the CD276 marker which is involved in the regulation of epithelial-mesenchymal transition and pathological angiogenesis, and the CD90 marker, which is overexpressed in the blood vessels of various types of tumors. According to the results of the experiments, an increase in cells expressing CD276 and CD90 was observed in the lung (fig. 3d) and blood (fig. 3e) again more pronounced in the group with PF+LC.

CONCLUSION

The obtained results allowed us to make important conclusions of this study. First, differences in the reaction of endothelial and epithelial cells to the modeling of PF and LC were discovered. Second, cells participating in the inflammatory and fibrotic reaction form a microenvironment that promotes malignant transformation of cells into tumor cells, induction of lung tumors and metastasis. Thus, populations of CSCs, endothelial cells, and fibrocytes of myeloid origin can serve as biomarkers confirming tumor development. These results open up a certain perspective in the differential use of cells as potential markers for diagnostics and prognosis of complications in patients with severe forms of pulmonary fibrosis and lung cancer.

FUTURE WORK / REFERENCES

Further clinical and experimental studies are needed to determine the role of these changes and to find biomarkers that predict the progression of pulmonary fibrosis and the risk of developing lung cancer.

1. Ballester B. et al. Idiopathic Pulmonary Fibrosis and Lung Cancer: Mechanisms and Molecular Targets. // Int J Mol Sci. 2019. 20(3). P.593.
2. Kato E. et al. Incidence and predictive factors of lung cancer in patients with idiopathic pulmonary fibrosis. //ERJ Open Res. 2018. 4. P. 00111-2016.