An Essential Role for Interleukin 6 in Lung Carcinogenesis

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Abstract

Lung cancer is the leading cause of cancer related deaths worldwide. Epidemiological studies have shown that patients with chronic obstructive pulmonary disease (COPD) have a higher risk of developing lung cancer, when compared to smokers without COPD. Moreover, we showed that a bacterial lysate-induced COPD-like airway inflammation promotes lung carcinogenesis in a K-ras mutant (CC-LR) mouse model. In our CC-LR mouse model, particularly high levels of interleukin 6 (IL-6) were observed. IL-6 is up-regulated in many types of inflammatory diseases and cancers, such as non-small cell lung cancer. Therefore, we genetically ablated IL-6 in the CC-LR mice, and found an essential role for IL-6 in cancer promotion in this K-ras induced model of lung cancer. In this study, we focused on blocking the IL-6 pathway using a monoclonal anti-IL-6 antibody therapy. CC-LR mice were injected intraperitoneally with a 20 mg/kg dose of monoclonal anti-IL-6 rat IgG1, twice a week, for a period of eight weeks. Anti-IL-6 treated mice were also exposed to the aerosolized bacterial lysate, weekly for a period of 8 weeks. Treatment with anti IL-6 antibody did not change the quantity of inflammatory cells in bronchoalveolar lavage fluid (BALF) of CC-LR mice. However, it not only inhibited intrinsic lung cancer development by ~78% (4.6-fold) in the absence of COPD-like inflammation, but also inhibited the tumor promoting effect of extrinsic NTHi-induced COPD-like airway inflammation by ~74% (3.9-fold). Here we propose a new therapeutic strategy for treatment of lung cancer patients with K-ras mutations, and possible preventive strategy in patients with COPD, using anti-IL-6 therapy.

Introduction

Several studies have shown that patients with COPD have a higher risk of developing lung cancer when compared to smokers without COPD (1-3). Furthermore, we have shown that COPD-like airway inflammation, induced by a bacterial lysate of non-typeable *Haemophilus influenzae* (NTHi), promotes lung carcinogenesis in a K-ras mutant (CC-LR) mouse model (4). We observed a shift from a macrophage to a neutrophil dominant inflammatory response in CC-LR mice after exposure to NTHi. Moreover, an evident increase in inflammatory cytokines and chemokines levels, especially IL-6, was observed in brocheoalveolar lavage fluid (BALF) of CC-LR mice, even before exposure to NTHi (4) .

IL-6 is a cytokine, with multiple functions, involved in several biological processes such as hematopoiesis, immune regulation, inflammation and oncogenesis (5). This cytokine is deregulated in many types of cancer including breast, ovarian, prostate and lung cancer, and it is involved in the proliferation and differentiation of malignant tumor cells (5-6). Classic IL-6 signaling pathway involves binding of IL-6 to the gp130 receptor and the membrane-bound IL-6 receptor (IL6-R). However, there is an alternative and particular trans-signaling pathway for IL-6, involving a soluble form of the IL-6 receptor (sIL-6R) (7). Binding of IL-6 to either of its receptors causes the activation of Janus kinase (JAK) tyrosine kinases, and this leads to the activation of transcription factors, such as the signal transducer and activator of transcription (STAT) 3.

Rationale

In this study, we focused on testing a monoclonal anti-IL-6 antibody therapy in our mouse model of lung cancer in presence and absence of COPD-like inflammation, to preclinically evaluate the effect of blocking IL-6 on prevention and treatment of lung cancer. This could provide the basis for rationally directed therapy in patients at high risk for lung tumor development (COPD) as well as patients with established stage lung cancer.

Objectives

•Evaluate the changes in the inflammatory cell profile of lung microenvironment due to anti-IL-6 therapy.
•Evaluate the change in tumor burden in response to anti-IL-6 therapy.

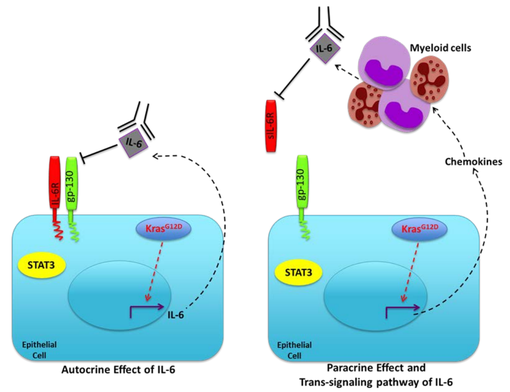

Figure 1. Effect of anti-IL-6 monoclonal antibody therapy.

Methods

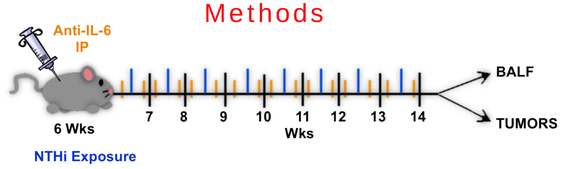

Figure 2. Anti-IL-6 monoclonal antibody therapy and NTHi exposure.

Table 1. Treatment conditions for Anti-IL-6 therapy and NTHi exposure.

Treatment	Conditions	Duration
Anti-IL-6 (Monoclonal Rat IgG_1)	20 mg/Kg	Twice a week/ 8 weeks
NTHi (Aerosolized lysate)	2.5 mg/mL 20 min @ 10 L/min	Once a week/ 8 weeks

Results

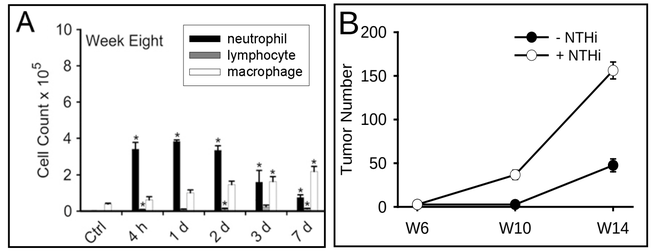

Figure 3. Chronic NTHi exposure induces COPD-like lung inflammation. (A) BALF cell counts at various times after the eighth weekly NTHi exposure with robust neutrophil influx. B) Lung surface tumor numbers in CCSP^{Cre}/LSL–K-ras^{G12D} (CC-LR) mice before aerosol exposure (Week 6), after four and eight weekly exposures to the aerosolized NTHi lysate (Weeks 10 and 14, open circles), or after 4 and 8 weeks without aerosol exposure (Weeks 10 and 14, closed circles) are shown.

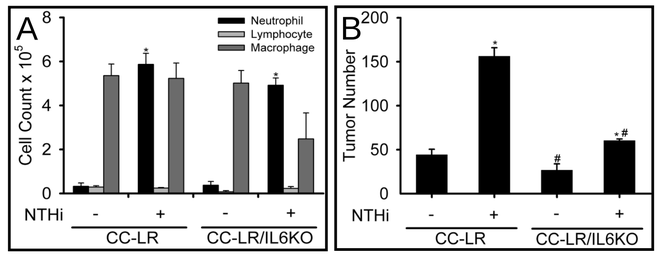

Figure 4. Promoting role of IL-6 in lung carcinogenesis. Lack of IL-6 did not change the BALF inflammatory cell component of CC-LR mice (A), but significantly reduced its surface tumor number (B).

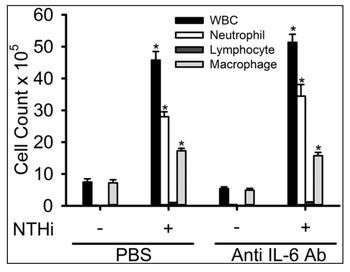

Figure 5. Role of IL-6 blockade in lung inflammation. Total and lineage-specific leukocyte numbers in BALF, 1 day after last NTHi exposure, are shown. A) CC-LR mice with or without 8 weekly NTHi exposure (*p<0.05). B) CC-LR mice with or without 8 weekly NTHi exposure, treated with anti-IL-6 twice a week for 8 weeks (*p<0.05).

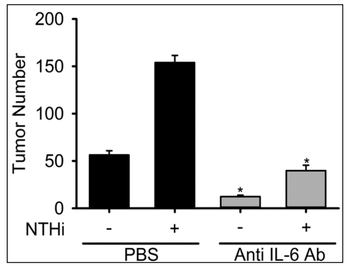

Figure 6. Role of IL-6 blockade in lung carcinogenesis. Lung surface tumor numbers in CC-LR mice were measured. A) Lung tumor number in CC-LR mice with or without 8 weekly NTHi exposure, from six weeks are shown. B) Lung tumor number in CC-LR mice with or without 8 weekly NTHi exposure, treated with anti-IL-6 twice a week for 8 weeks (*p<0.05).

Conclusions

Treatment with anti IL-6 antibody did not change the quantity of inflammatory cells in bronchoalveolar lavage fluid (BALF) of CC-LR mice. However, it not only inhibited intrinsic lung cancer development by ~78% (4.6-fold) in the absence of COPD-like inflammation, but also inhibited the tumor promoting effect of extrinsic NTHi-induced COPD-like airway inflammation by ~74% (3.9-fold). Here we propose a new therapeutic strategy for treatment of lung cancer patients with K-ras mutations, and possible preventive strategy in patients with COPD, using anti-IL-6 therapy.

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