DRUG DEVELOPMENT & LUNG DISEASES

SMC LABORATORIES' LUNG DISEASE MODEL CATALOG

Idiopathic Pulmonary Fibrosis (IPF)

Chronic Obstructive Pulmonary Disease (COPD)

Acute Lung Injury (ALI) /
Acute Respiratory Distress Syndrome (ARDS)

Systemic Sclerosis-associated Idiopathic Lung Disease (SSc-ILD)

Silicosis

SMC Laboratories,Inc.

THE MODEL LINEUP

Idiopathic Pulmonary Fibrosis (IPF)

Bleomycin-induced model

Chronic Bleomycin-induced model

Chronic Obstructive Pulmonary Disease (COPD)

PPE-induced model

Acute Lung Injury (ALI) / Acute Respiratory Distress Syndrome (ARDS)

LPS-induced model

Systemic Sclerosis-associated Idiopathic Lung Disease (SSc-ILD)

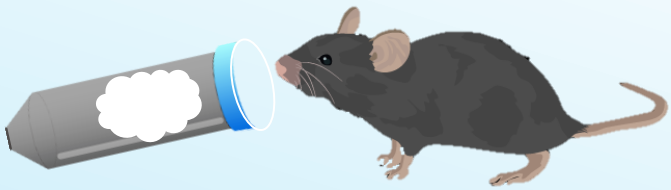
Bleomycin-induced model

Silicosis

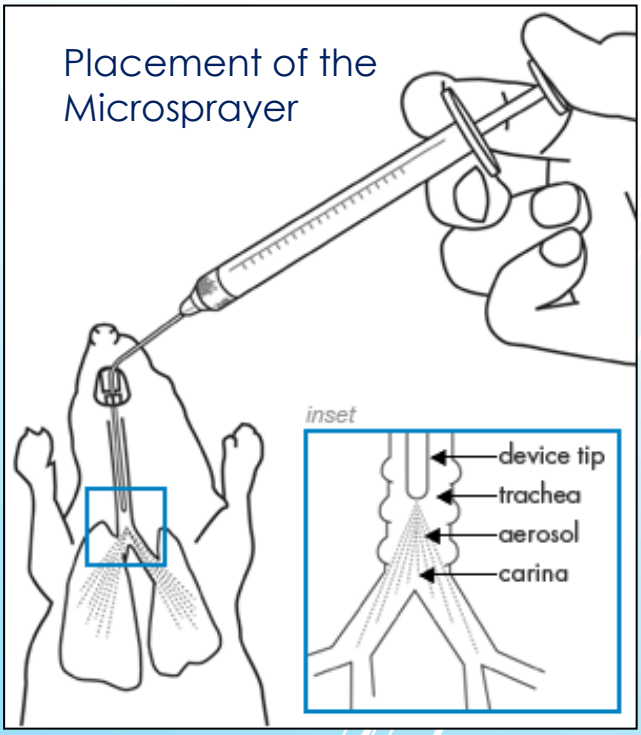
Silica-induced model

Administration via Microsprayer®

Anesthesia wish Isoflurane



Female C57BL/6J



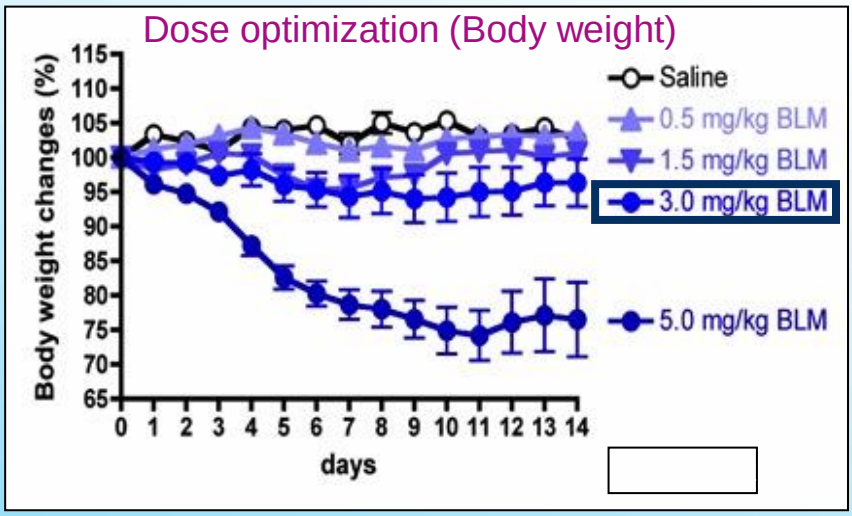
- ✓ Particle size: 16-22 µm (analyzed with water)
- ✓ Administrable volume: 25-250 µl

Using the intra tracheal administration route to induce the disease in the model mouse results in a more uniform phenotype than other administration routes. The inducing agent is administered in the trachea, so that the loss is close to Zero and the Microsprayer® nebulizes the solution with a low particle size so that the agent is distributed evenly through the lung lobes.

"It is recommended to avoid highly viscous or frothy vehicles when administering IT because they are difficult to aerosolize and mice tend to have breathing difficulties after administration."

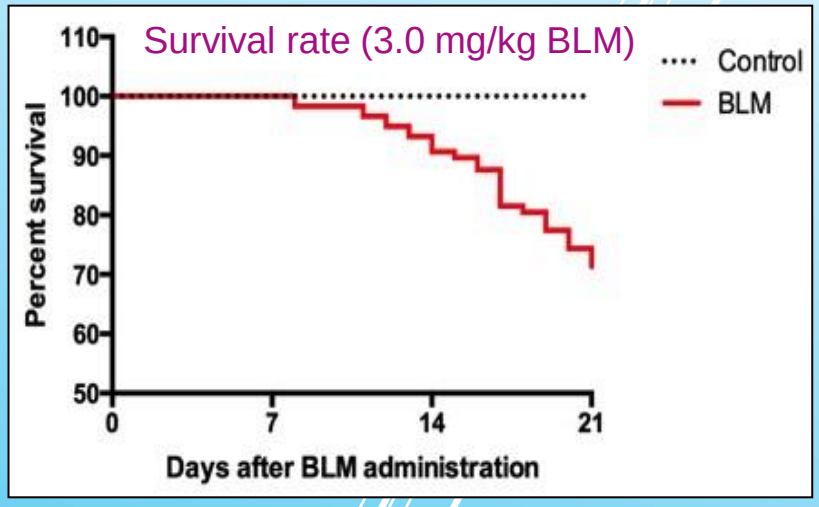
– SMC Laboratories' Research Team

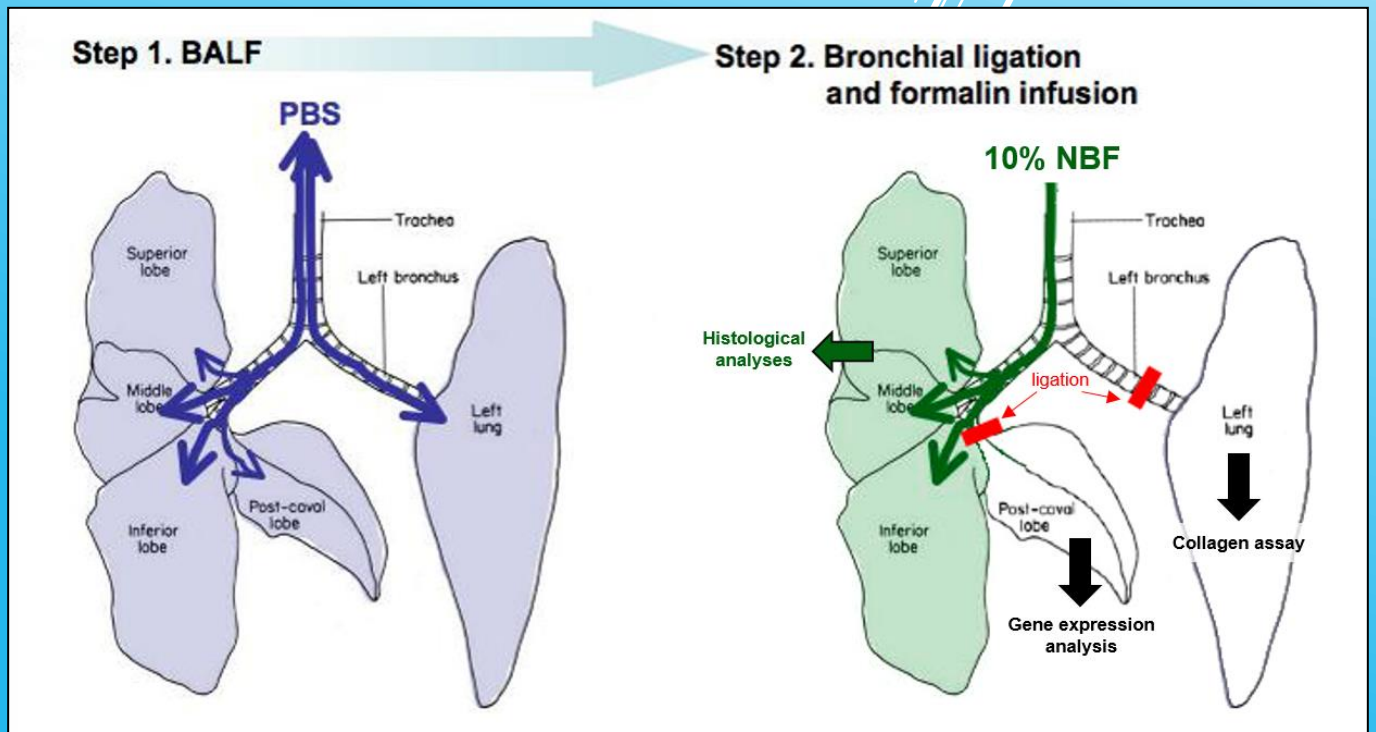
Optimization & Sampling of Bleomycin-induced Lung Models



Our model has been optimized for the right balance between severity of fibrosis development…

… and the survival of the mice at the take-down time at 21 days after induction, when the fibrosis peaks.

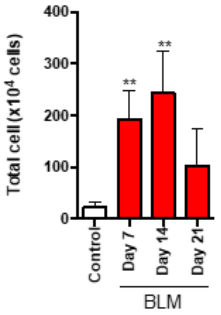




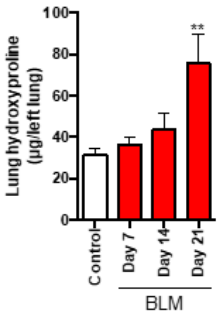
IPF - BLM-induced model -

BALF Analysis: Hydroxyproline | Ashcroft Score | Gene Expression

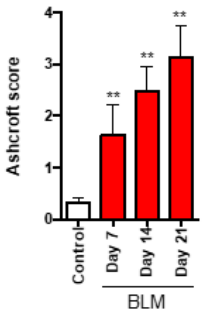
BALF total cells



Lung hydroxyproline content

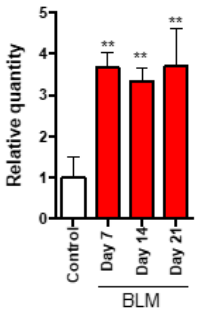


Ashcroft score

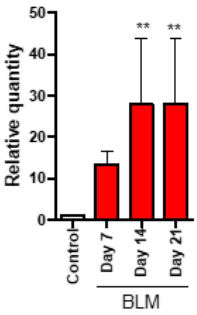


Gene expression

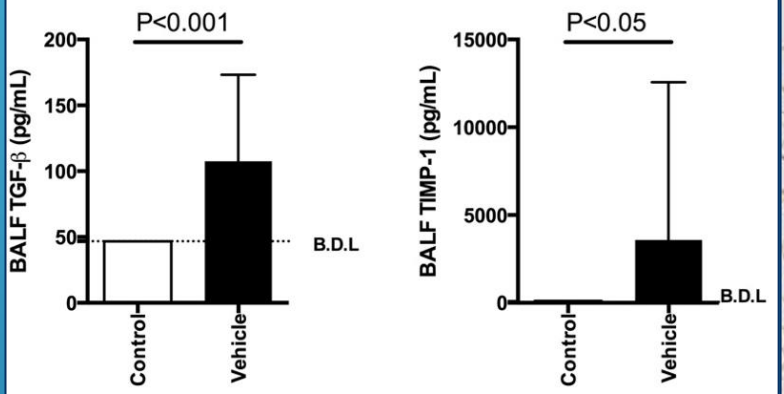
Collagen Type 1 gene



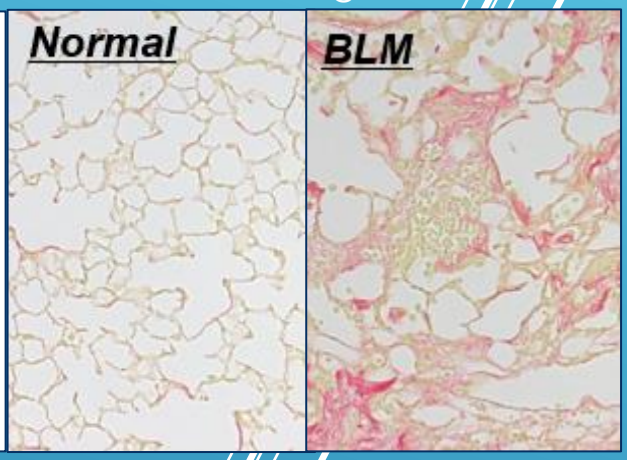
TIMP-1 gene



BALF Cytokine Analysis



Sirius Red Staining



BLM i.t.

Disease Progression

Day 0	Day 7	Day 14	Day 21	Day 28
Direct damage initially to alveolar epithelial cells	Increase of ・**neutrophils** ・**lymphocytes**	**Fibroblast** proliferation **Extracellular matrix** synthesize	**Development of fibrosis** (biochemically and histologically)	**Maximal responses**

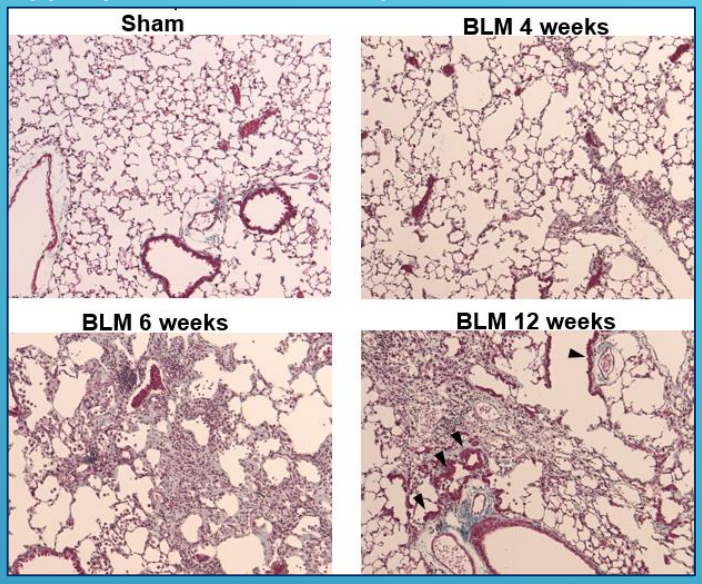
Inflammation phase → Fibrosis phase

TIMELINE

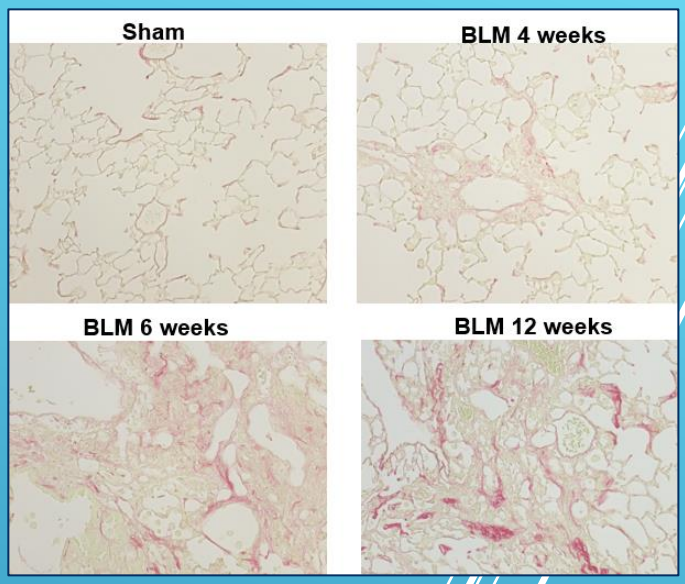
1-2 months	0.5 month	3 months
Discussion ・ NDA (If needed)	**Contract** ・Study protocol ・Quote ・Contract	**Preparation** ・ Test compound ・ Pregnant mice ・ Disease induction	**Study & Report** ・Treatment ・Analyses ・Report

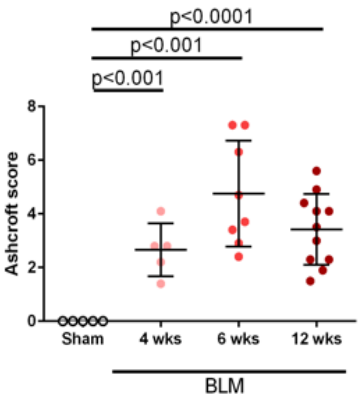
IPF - Chronic BLM-induced model -

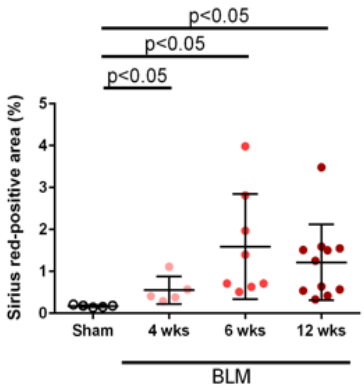
Hyperplastic alveolar epithelial cells

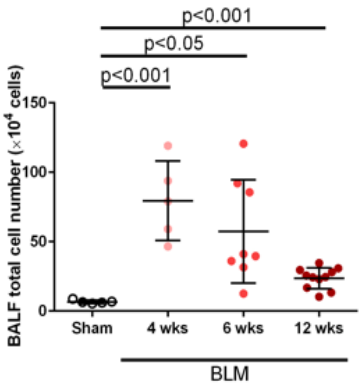


Sirius Red positive area



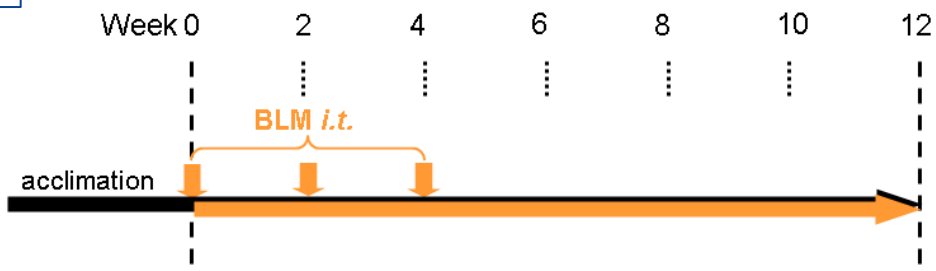






Disease Induction

C57BL/6J, female, 6 weeks of age at induction



TIMELINE

1-2 months	0.5 month	3 months
Discussion ・NDA (If needed)	**Contract** ・Study protocol ・Quote ・Contract	**Preparation** ・Test compound ・Disease induction	**Study & Report** ・Treatment ・Analyses ・Report

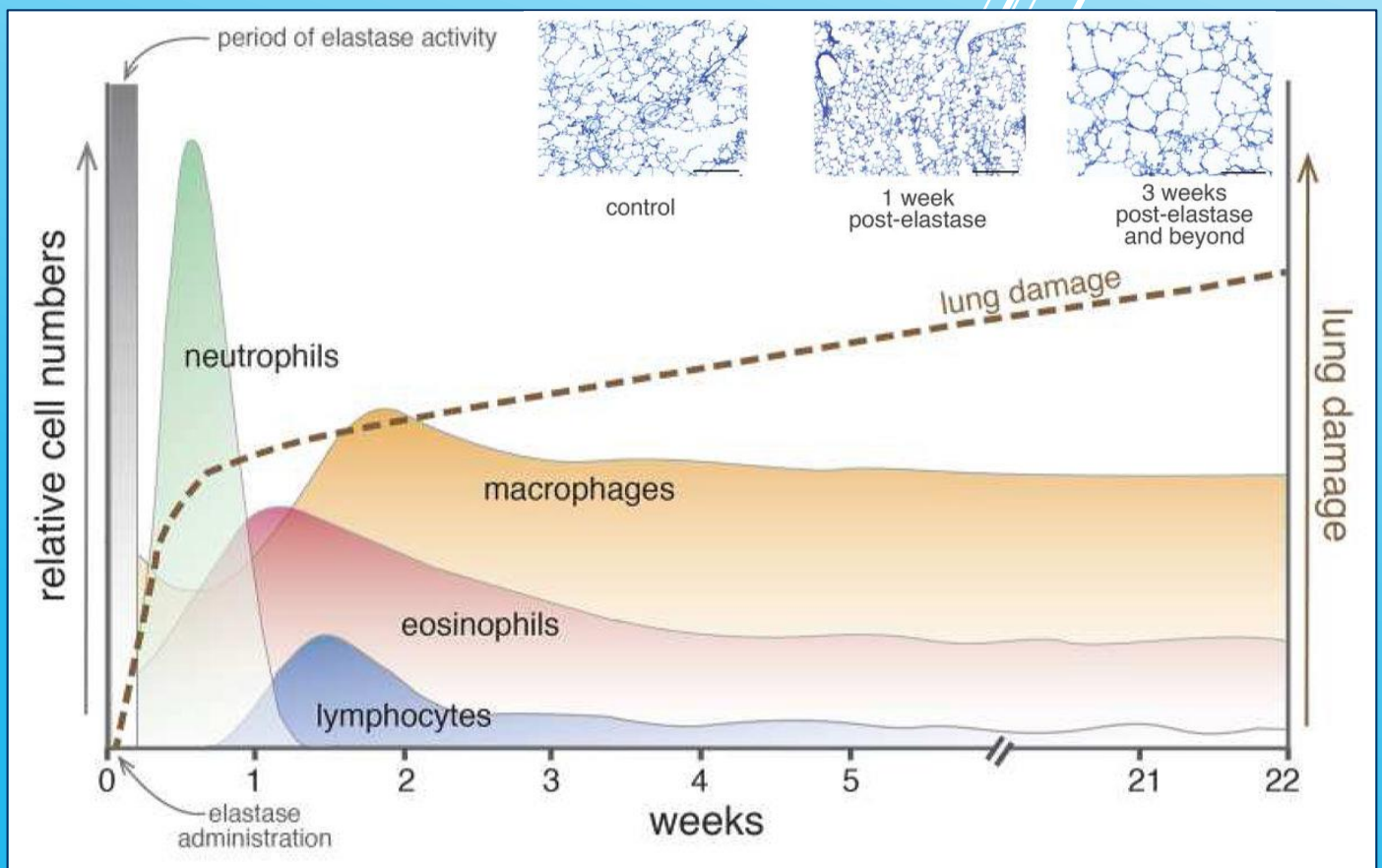
Chronic Obstructive Pulmonary Disease (COPD)

- COPD is recognized as a major cause of death in developed countries and is characterized by chronic, slowly progressive airway obstruction and destruction.

- Histological and morphological characteristic features are compatible with those of panacinar emphysema.

- Induced morphological and functional changes are detectable in the long term and the lesion severity can be modulated enzyme dose.

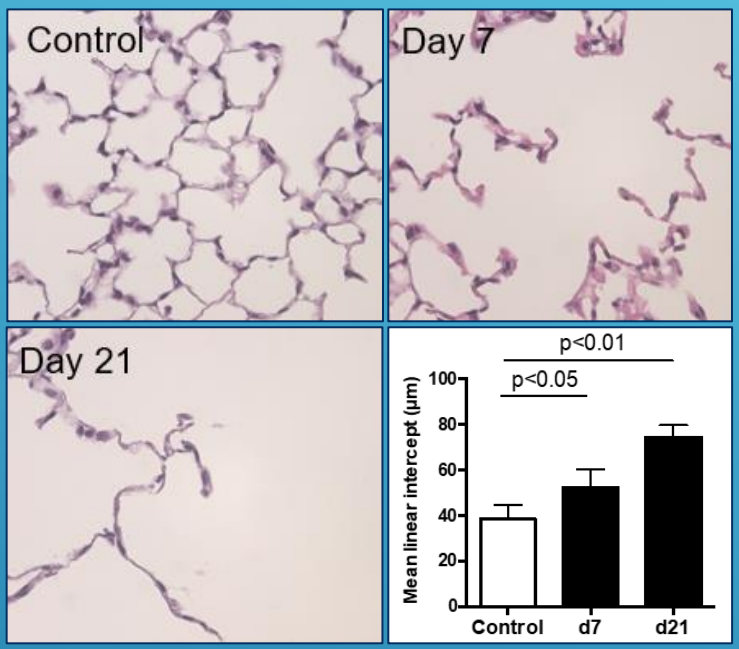
- The cellular response following elastase challenge is characterized by a swift influx of neutrophils, succeeded by the recruitment of other immune cells.



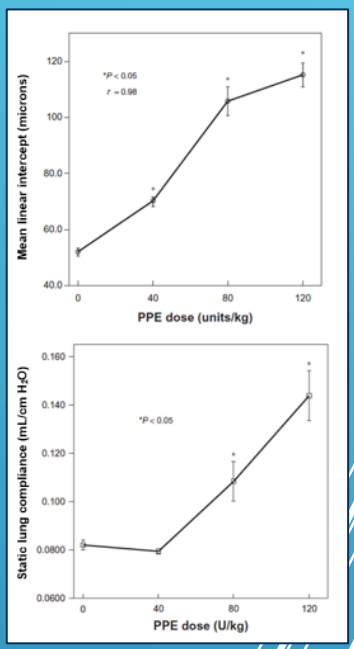
COPD - PPE-induced model -

- ✓ Effective destruction of bronchial alveolar epithelium within 21 days after PPE administration
- ✓ Adjustment of disease severity in the model by dosing concentration of PPE

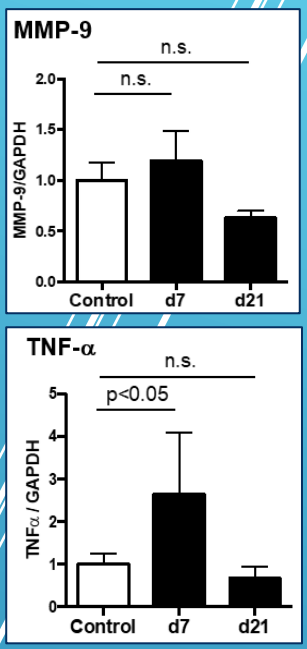
Mean Linear Intercept



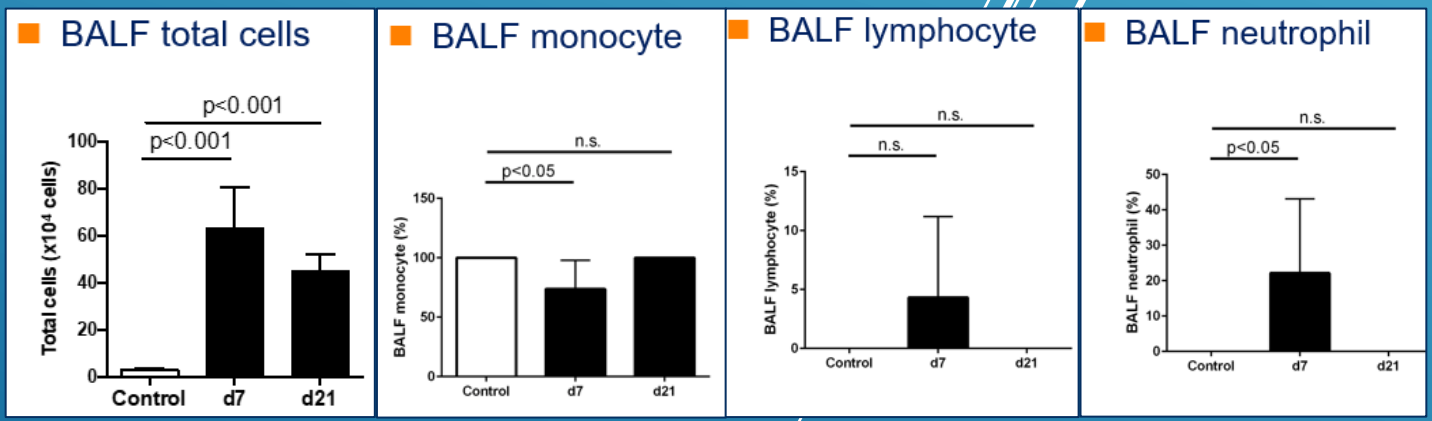
Dose dependency



Gene Expression



Analysis of immune cells in Bronchial Alveolar Lavage



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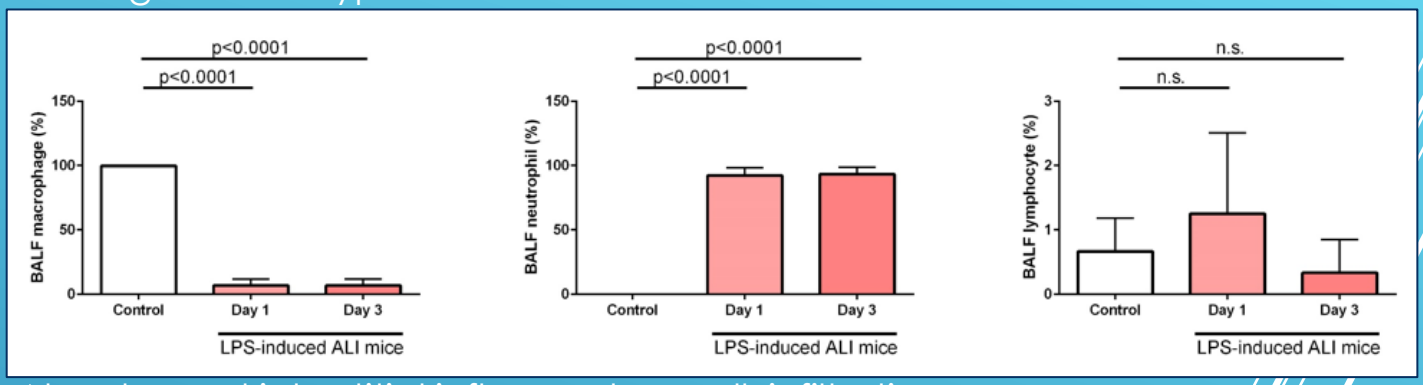
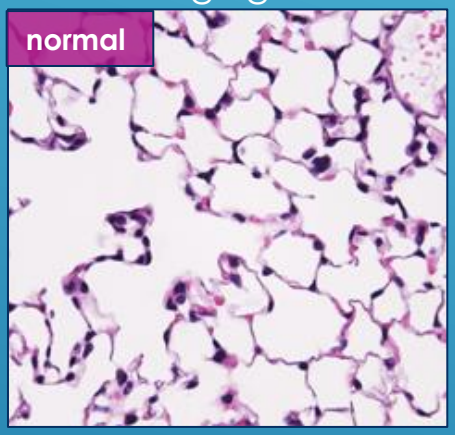
Acute Lung Injury / Acute Respiratory Distress Syndrome

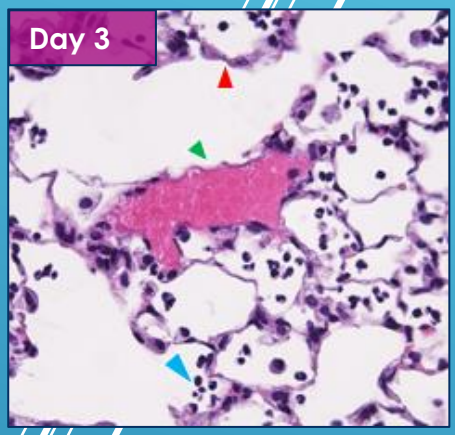
- LPS-induced model -

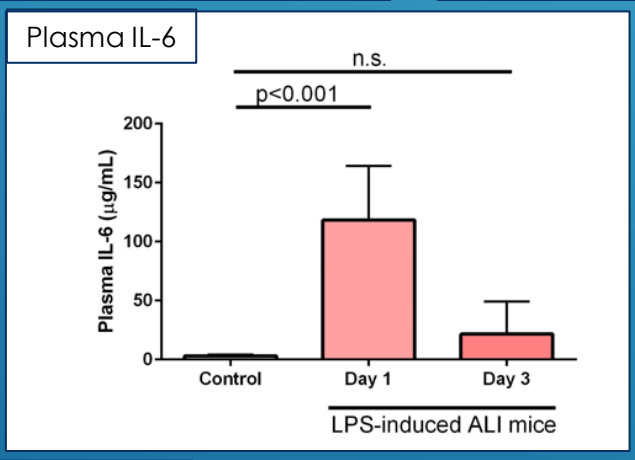
Changes of cell types in BALF

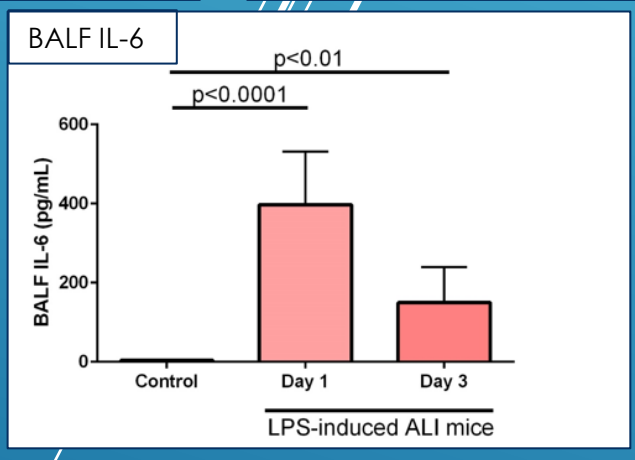


Alveolar and interstitial inflammatory cell infiltration, hemorrhaging and thickening of the alveolar walls









TIMELINE

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Systemic Sclerosis-associated Interstitial Lung disease

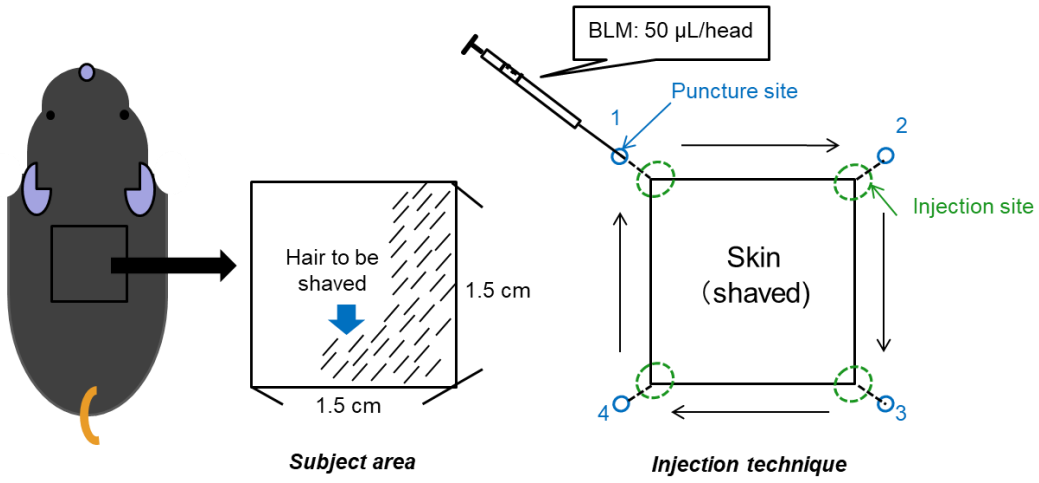
Key Characteristics in the SSc-ILD model

■ Dermal thickness and collagen content in the skin are increased in bleomycin injected mice.

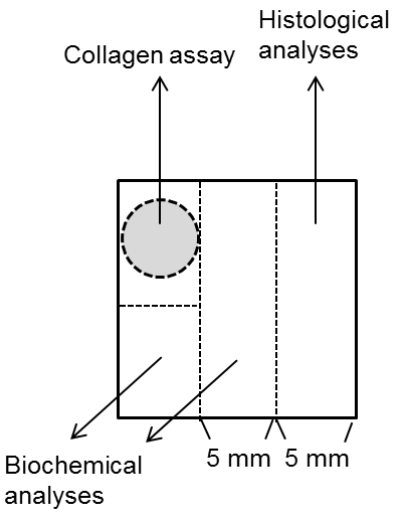
■ Not only skin is affected, but also the fibrotic region and collagen content in the lung are increased by indirect bleomycin exposure.

■ This model may provide advantages to test therapeutic interventions for SSc-ILD.

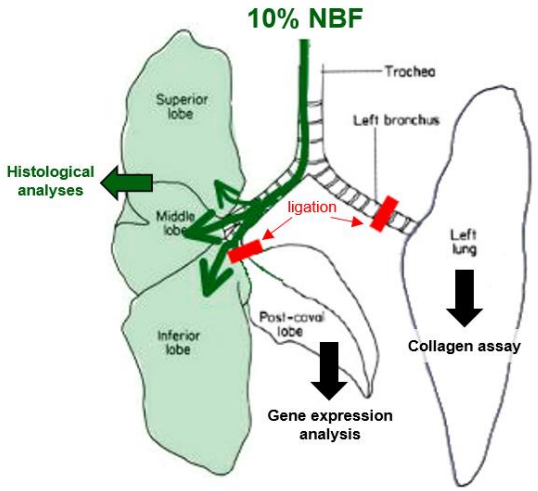
Injection technique



Skin sampling



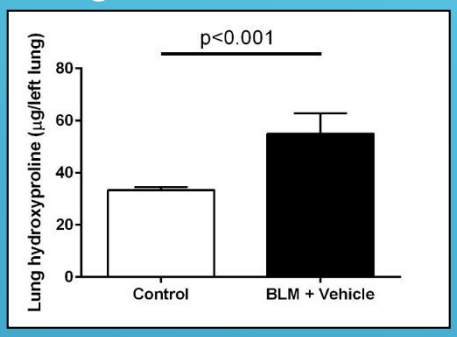
Lung sampling



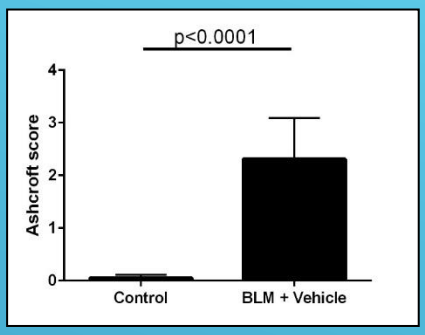
Systemic Sclerosis-associated Interstitial Lung disease

- Bleomycin-induced model -

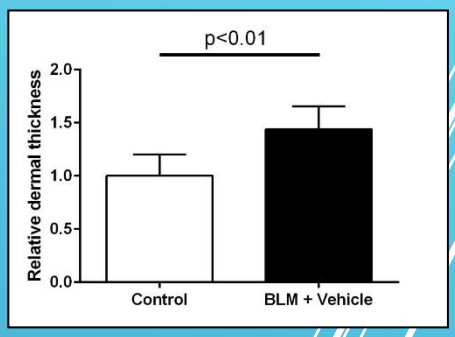
Lung fibrosis

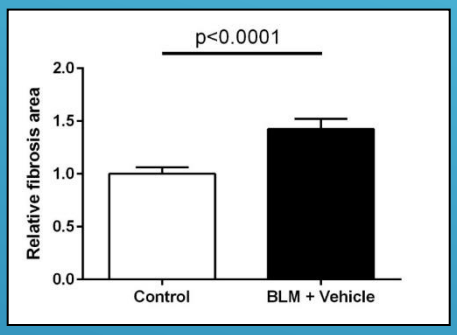


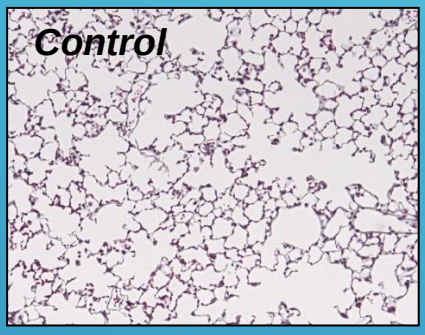
Ashcroft score

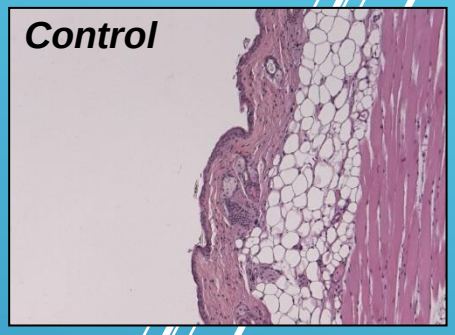


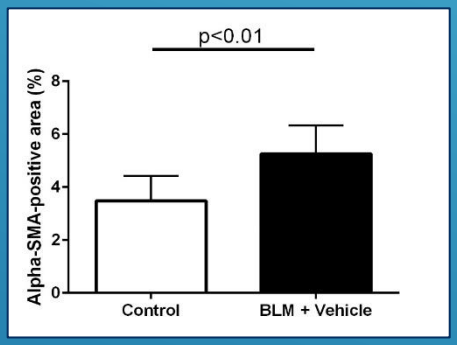
Dermal thickness

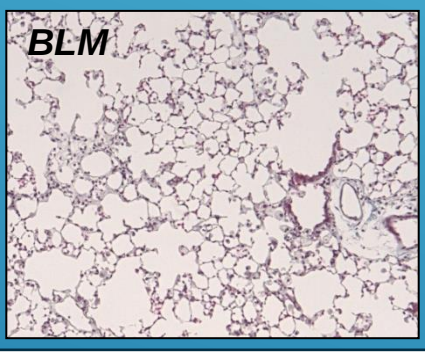


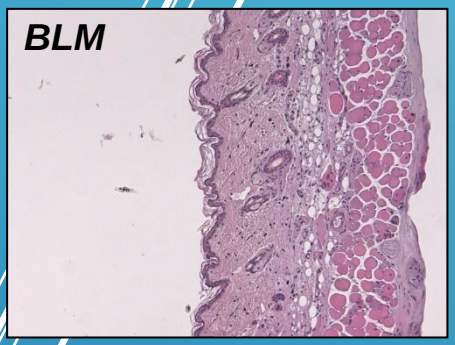












Disease Induction

Female C57BL6/J 7 weeks old

Day 0 — **Day 28**

Bleomycin (4.0 mg/mL, 50 μL), subcutaneous, alternate days, for 4 weeks

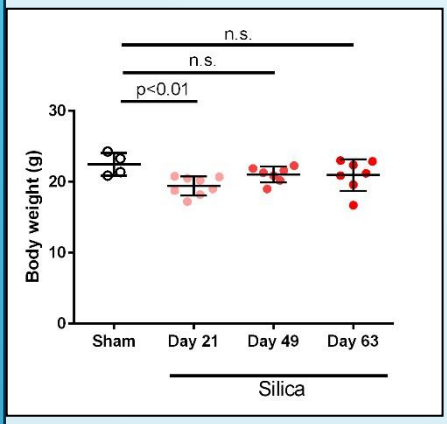
TIMELINE

1-2 months	0.5 month	3 months
Discussion • NDA (If needed)	**Contract** • Study protocol • Quote • Contract	**Preparation** • Test compound • Disease induction	**Study & Report** • Treatment • Analyses • Report

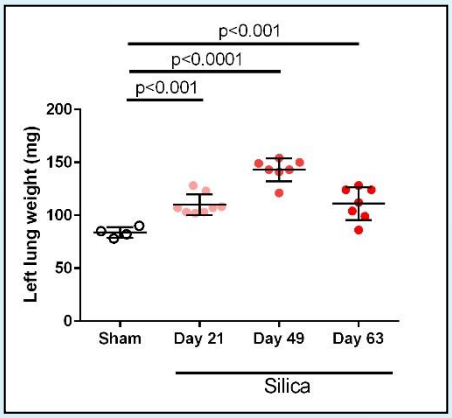
Silicosis

- Silica-induced Model-

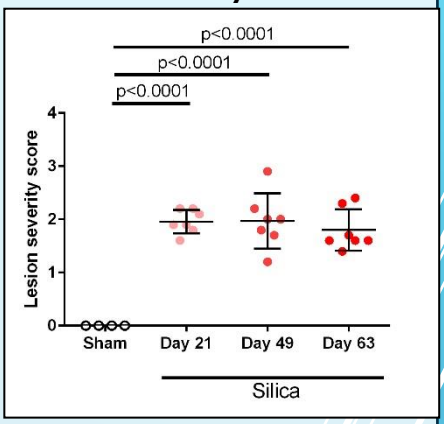
Body weigth



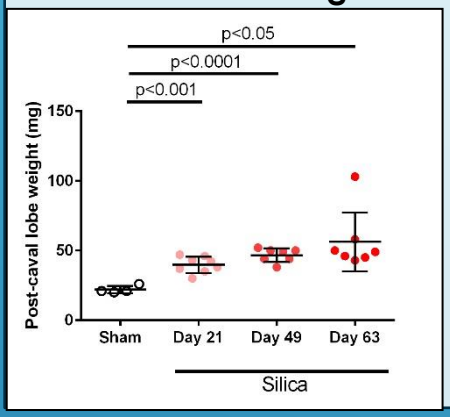
Left Lung weight



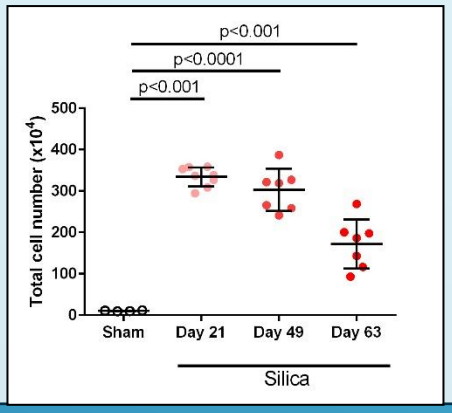
Lesion severity score



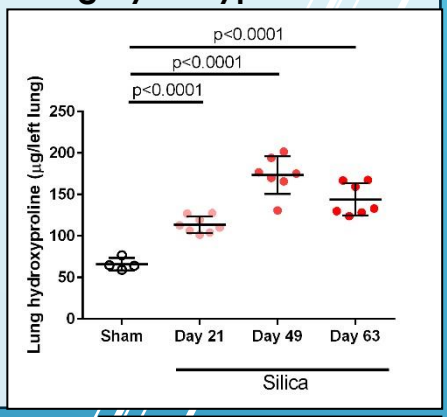
Post-caval lobe weight

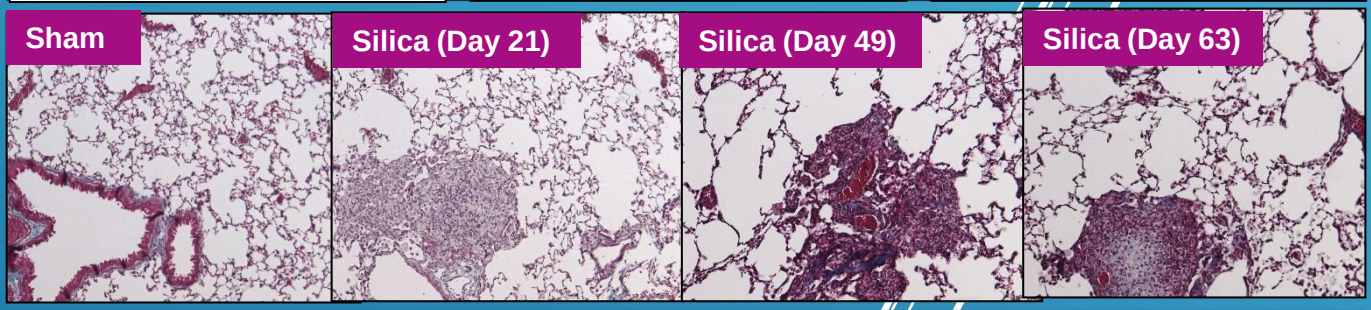


Total cell number



Lung Hydroxyproline





- ✓ Disease induction: Oropharyngeal administration of silica causes inflammation and fibrosis of the lungs
- ✓ Fibrosis feature: **Scattered throughout the lung, nodule formation**

TIMELINE

1-2 months

Discussion
- NDA

(If needed)

Contract
- Study protocol
- Quote
- Contract

0.5 months

Preparation
- Test compound
- Disease induction

4 months

Study & Report
- Treatment
- Analyses
- Report