# Repair and Regeneration of Vascular Endothelium as a Target for the Treatment of Lung Fibrosis

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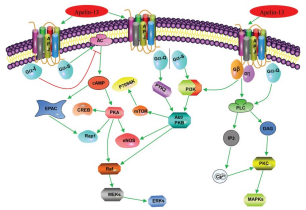
APIE THERAPEUTICS

## ABSTRACT

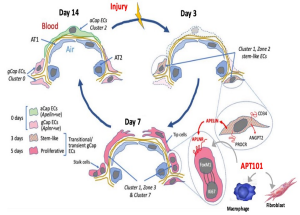
Vascular endothelial niche dysfunction has been identified in idiopathic pulmonary fibrosis (IPF) pathophysiology. Evidence suggests that the apelinergic system, ***APJ (apelin receptor)*** and its ***ligand (apelin)***, highly expressed in the pulmonary vascular endothelial cell subtypes (aerocytes and general capillary, respectively[1]), contributes to alveolar regeneration and that age- or disease-related attenuation of this system contributes to the interstitial lung disease (ILD) and IPF[2]. Furthermore, the apelinergic system is shown to be upregulated in IPF patients[3]. In models of ILD loss of APJ function is associated with more severe disease while promoting APJ activation improves outcomes.

***Novel APJ Agonist, APT101,*** a synthetic, orally bioavailable small molecule APJ agonist designed to to be highly selective of the APJ pathway, away form β-arrestin, to minimize signal internalization and maximize efficacy for chronic treatments. Human Vascular Endothelial cells models and ***acute*** (prophylactic treatment) and ***chronic*** (delayed treatment) Mouse Bleomycin models of lung injury and fibrosis were used to characterize the efficacy of APT101.

## APJ G-PROTEIN SIGNALING PATHWAYS



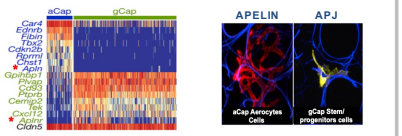
## PROPOSED MECHANISM OF ENDOTHELIAL REGENERATION AND REPAIR IN THE LUNG[1]




Schematic ***of APLN (apelin) and APLNR (APJ)*** localization in transitional endothelial cells and their role in alveolar endothelial regeneration *(Godoy et al., 2021)*. Abbreviations: aCap, alveolar capillary endothelial cells; gCap, general capillary pulmonary endothelial cells.

## PROOF OF CONCEPT

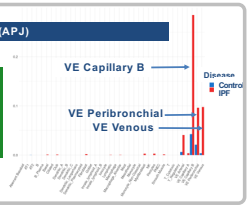
### ALVEOLUS CAPILLARY GENE EXPRESSION HEAT MAP[2]




### APLNR (APJ)

IPF ATLAS[3]
IPF HUMAN scRNA

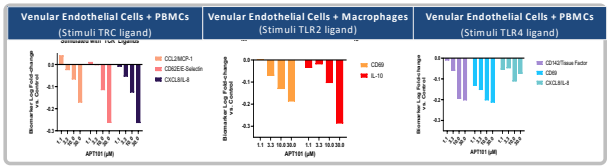
APLNR (APJ) PREGULATED




**Target of APT101, APJ receptor, is upregulated in IPF Patients.**

***APJ located in the lung alveoli capillary cells (g-cap Stems/progenitors cells)***

## HUMAN VASCULAR ENDOTHELIAL CELLS DISEASE MODELS DEMONSTRATE THERAPEUTICS EFFECT OF APT101 APJ AGONIST



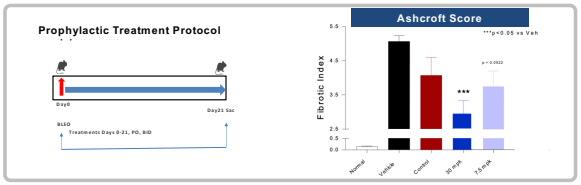

- Anti-inflammatory activity in T-cell mediated vascular inflammation
- Anti-inflammatory and immune modulatory activity
- Modulatory activity in vasculature hemostasis
- Anti-inflammatory activity in monocyte mediated

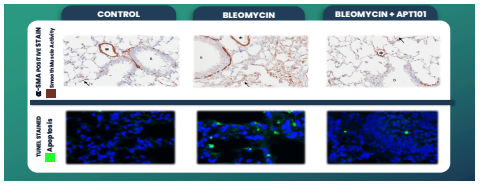
*Ex-Vivo BioMAP Diversity PLUS Primary Human Cell Models*

**ADDITIONAL PROPERTIES OF APT101**

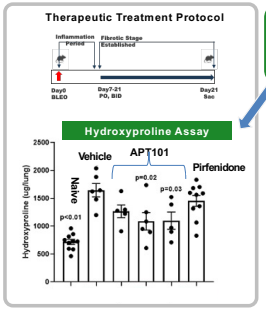
| Human Cells Co-Culture | Stimuli | Biologic Category | Decreased Biomarker(s) |
|---|---|---|---|
| B cells, PBMCs | TCR ligand, α-IgM | Autoimmunity Modulation | sIL-17a, sIL-2 |
| Bronchial Epithelial cells, Dermal Fibroblasts | TNFα, IL-4 | Hemostasis Modulation | tPA |
| Dermal Fibroblasts | IFNγ, TNFα, IL-1β, EGF, FGF, PDGF | Fibrosis and Tissue Remodeling | TIMP2, PAI-1 |

## PROPHYLACTIC TREATMENT WITH APT101 REDUCED FIBROSIS AND APOPTOSIS



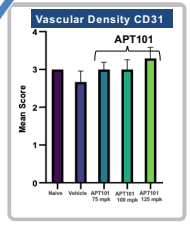

## THERAPEUTIC TREATMENT WITH APT101 REDUCED COLLAGEN BUILD-UP, FIBROSIS AND INFLAMMATION, AND PROTECTS VASCULAR DENSITY



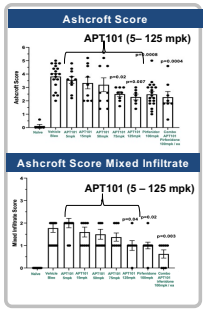

*Inhibition of Collagen Build-Up*

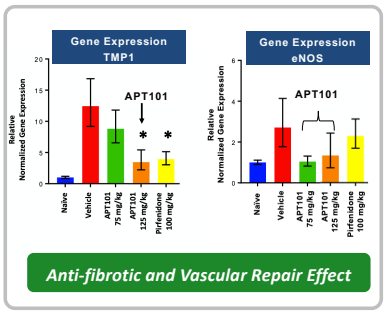
77% APT101

26% Pirfenidone




## APT101 REDUCED GENE EXPRESION OF eNOS AND TMP1




***Anti-fibrotic and Vascular Repair Effect***

## CONCLUSION

- ✓ Novel APJ Agonist APT101, Clinical Lead Candidate, is an orally bioavailable small molecule and acts as a biased ligand to selectively activate APJ G-protein signaling.
- ✓ In human primary vascular endothelial co-cultures, APT101 inhibited the vascular endothelial dysfunction leading to a variety of inflammatory and fibrotic pathogenic mediators.
- ✓ APT101, demonstrate its anti-inflammatory and anti-fibrotic and anti-lung injury effects in the murine bleomycin pulmonary fibrosis model, both prophylactic and therapeutic treatments in a dose related manner
- ✓ Underlying mechanisms include reduced formation of myofibroblasts and apoptosis
- ✓ These results support APJ agonism as a strategy for the treatment of IPF

## ACKNOWLEDGEMENT

NC BIO TECH CENTER
RESEARCH TRIANGLE INSTITUTE

## REFERENCES

1. *"Newly Emergent Apelin Expressing Endothelial Stem-like Cells Orchestrate Lung Microvascular Repair", Godoy RS., et al., bioRxiv:2021.2007.2012.45201 (2021)*
2. *"Capillary cell-type specialization in the alveolus", Gillich et al., Nature, vol. 586, 785–789 (2020)*
3. *IPF ATLAS; http://www.ipfcellatlas.com*
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5. *"Identifying structural determinants of potency for analogs of apelin-13: integration of C-terminal truncation with structure-activity", Zhang, Y et al, Bioorg Med Chem (2014).*