Microvascular improvement and fibrosis regression by the angiotensin II type 2 receptor agonist buloxibutid (C21) in pulmonary hypertension associated with pulmonary fibrosis

Johan Raud[1], Jimmie Hofman[1], Johanna Gräns[1], Dan Salvail[2], Göran Tornling[3]

[1]Vicore Pharma AB, Stockholm, Sweden

[2]IPS Therapeutique Inc., Sherbrooke, Canada

[3]Division of Immunology and Respiratory Medicine, Department of Medicine Solna, Karolinska Institutet, Stockholm, Sweden

Introduction

The angiotensin II type 2 receptor (AT_2R) is highly expressed on alveolar epithelial cells type 2 (AEC2) and represents a tissue-protective mechanism that counteracts tissue injury through anti-inflammatory, anti-fibrotic and vasodilatory signalling (Figure 1).

In the recent Phase 2a open-label AIR trial, the selective oral AT_2R agonist buloxibutid (also known as C21) stabilized and improved lung function in patients with idiopathic pulmonary fibrosis (IPF)[1].

Given that pulmonary vascular pathology is a key determinant of morbidity and mortality in fibrotic lung disease, we investigated the effects of buloxibutid on both vascular remodeling and pulmonary fibrosis in the rat Sugen-hypoxia model. This model recapitulates key features of pulmonary arterial hypertension (PAH) and shares important pathophysiological characteristics with pulmonary hypertension associated with interstitial lung disease (PH-ILD).

Results

Hemodynamic Assessments

Compared with normoxic controls, Sugen-hypoxia caused significant right ventricular (RV) hypertrophy, increased pulmonary artery (PA) and RV pressures, and impaired cardiac output (Figure 2 and Table 1).

As previously described[2], buloxibutid treatment reduced the RV hypertrophy as reflected by reduced Fulton's index, and was associated with lower mean PA and RV pressures (Table 1).

These hemodynamic improvements were accompanied by improved cardiac output, while mean arterial pressure was not affected by buloxibutid (Table 1).

Vascular Remodeling

Sugen-hypoxia induced significant thickening of the vessel wall across all vessel sizes (Figure 3), driven predominantly by expansion of the muscular layer. In parallel, vessel luminal opening was significantly reduced across all sizes (Figure 4).

These two parameters are directly and mechanistically linked: as the muscular wall expands inward it encroaches on the lumen, raising vascular resistance and pulmonary artery pressure, which in turn imposes an increasing pressure load on the right ventricle of the heart.

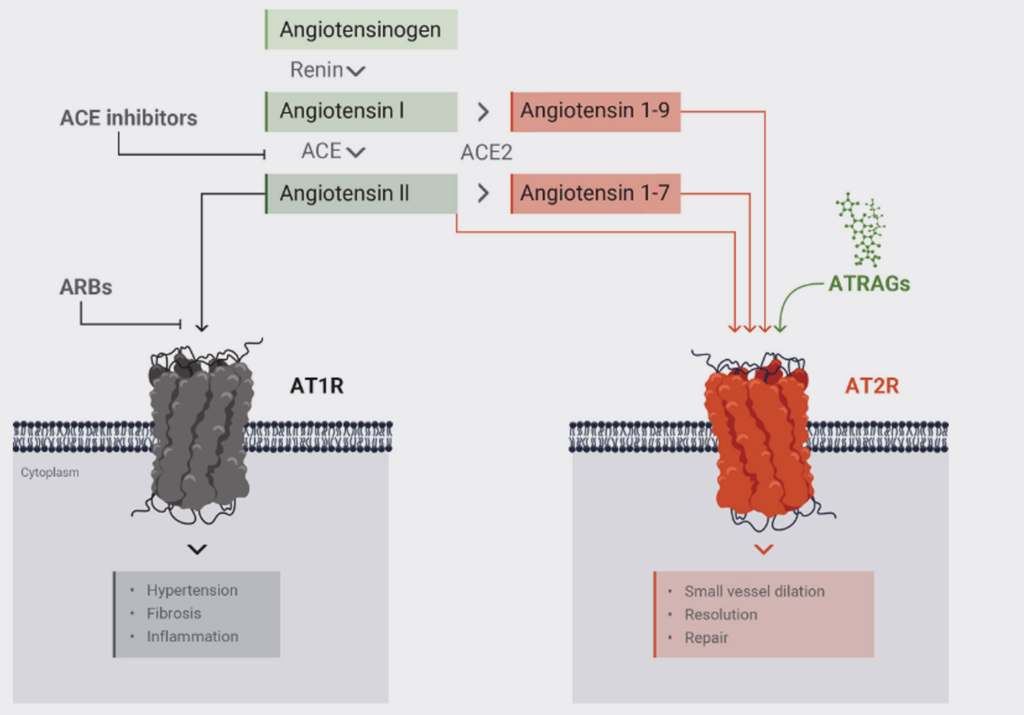
Treatment with buloxibutid (both doses combined) significantly and consistently caused a partial reduction of vessel wall thickening and increase in luminal opening across all vessel sizes (Figure 3 and 4). Given that treatment was initiated only after pulmonary hypertension was already established, these effects suggest a genuine disease-modifying action on the vascular remodeling process rather than a vasodilatory effect.

Pulmonary fibrosis

In parallel with the vascular and hemodynamic changes, Sugen-hypoxia increased pulmonary fibrosis (measured as lung collagen content) by 20 % (P<0.05), and this was completely reversed by the high dose of buloxibutid (p<0.01) (Figure 5).

Correlation analyses demonstrated that the increased vascular patency/openness was inversely correlated with pulmonary collagen burden (Figure 6).

Figure 1. Renin-Angiotensin System and AT2R agonist buloxibutid (C21)



- Angiotensin II (formed mainly in the lung) activates AT1 and AT2 receptors with similar potency
- AT1R is widely expressed, while AT2R is mainly expressed on alveolar epithelial cells, but is upregulated at sites of disease/tissue injury
- AT1R effects include increase in blood pressure, the basis for ACEi and ARB development
- AT2R is a tissue protective mechanism
- Buloxibutid is a potent (IC50 5.7 nM) and selective (2900x vs AT1R) AT2R agonist

Figure 2. Echocardiography and pulmonary and systemic artery pressures in rat Sugen-Hypoxia model

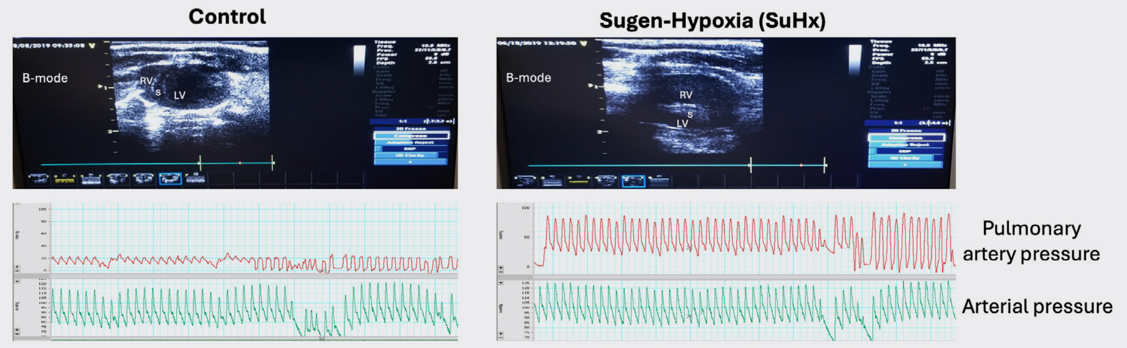
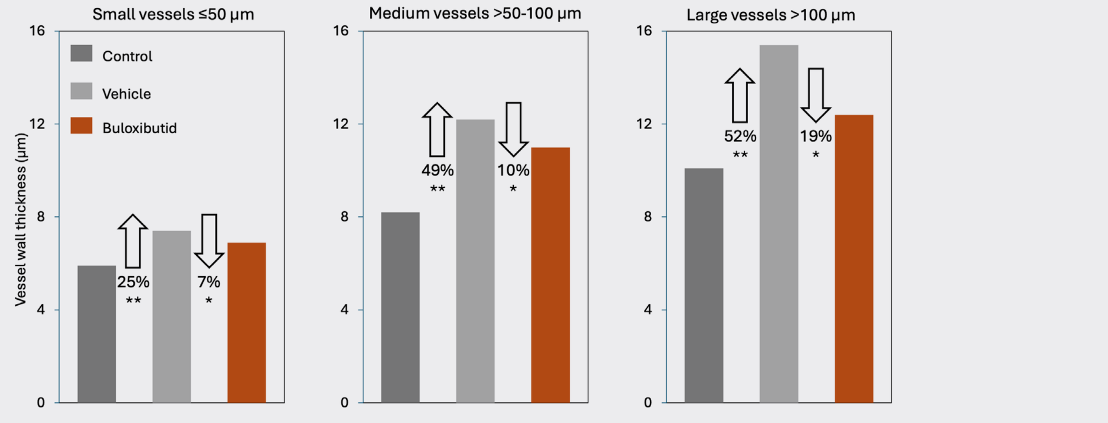


Table 1. Cardiovascular effects of Sugen-hypoxia challenge and buloxibutid treatment in the rat Sugen-Hypoxia model

	Vehicle vs. Control	Buloxibutid vs. Vehicle
RV hypertrophy (Fulton's index)	↑ 162% (p<0.01)	↓ 10% (p<0.05)
Mean RV pressure	↑ 133% (p<0.01)	↓ 25% (p<0.05)
Mean PA pressure	↑ 137% (p<0.01)	↓ 25% (p<0.05)
Cardiac output	↓ 43% (p<0.01)	↑ 22% (p<0.05)
Mean arterial pressure	↓ 4% (NS)	↑ 11% (NS)

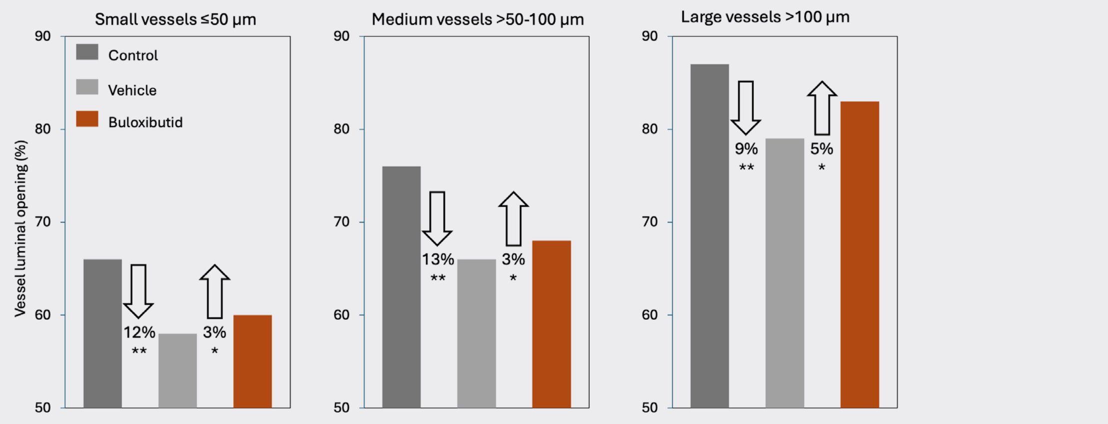
Both doses of buloxibutid combined (n=20).

Figure 3. Sugen-hypoxia – Vessel wall thickness



(*p<0.05, **p<0.01).

Figure 4. Sugen-hypoxia – Vessel luminal opening



Vessel lumen to vessel diameter ratio (*p<0.05, **p<0.01).

Figure 5. Lung collagen (picrosirius red staining).

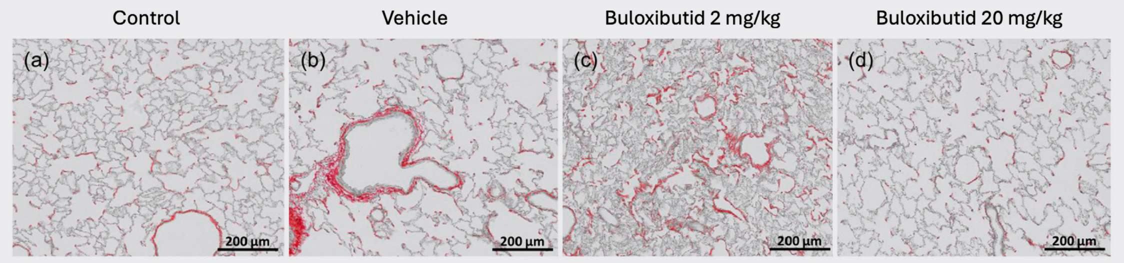


Figure 6. Correlation between non-obliterated/open vessels and pulmonary collagen content.

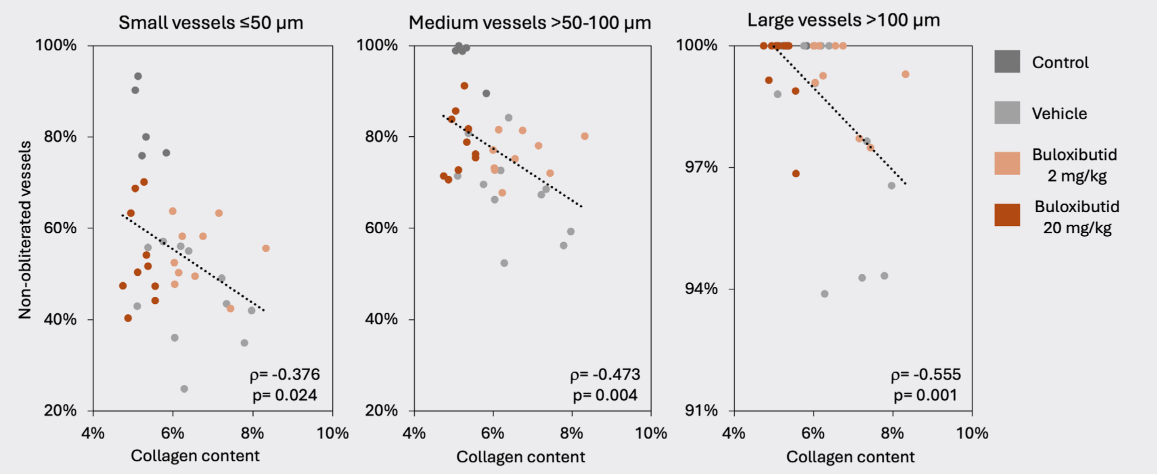
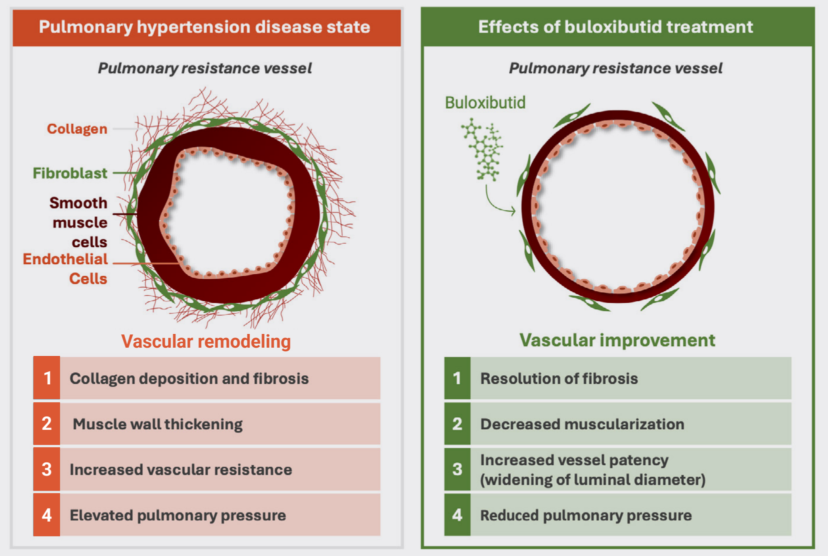


Figure 7. Summary of effects of buloxibutid in pulmonary hypertension and fibrosis.



Conclusions

- The results support that buloxibutid has disease-modifying potential with regard to both the vasculopathy and the pulmonary fibrosis in interstitial lung disease.
- Buloxibutid reduced pulmonary vascular remodeling, increased resistance vessel patency/openness and reduced right ventricular hypertrophy.
- Pulmonary hemodynamics improved without systemic hypotension, supporting a pulmonary-selective effect of buloxibutid.
- Vascular improvements correlated with antifibrotic effects, reflecting a shared pro-fibrotic upstream pathobiology targeted in concert by buloxibutid through AT2R activation.

Methods

Male Sprague–Dawley rats receive a single dose of Sugen 5416 in DMSO (20 mg/kg, s.c.) followed by 21 days of hypoxia (FiO_2 = 10%). After disease establishment, rats received oral buloxibutid (2 or 20 mg/kg twice daily) or vehicle from days 21–55 under normoxia. Controls received DMSO s.c. and normoxia for the full study duration. There were 5 animals in the control group and 10-11 animals per treatment group.

Echocardiography (days 0, 21, 56) was used to assess pulmonary artery flow and cardiac output. On day 56, invasive catheterization measured right ventricular and pulmonary artery pressures and systemic hemodynamics.

Lungs were harvested at sacrifice. Lung sections were stained with picrosirius red for collagen content. Automated morphometric analysis quantified vascular remodeling and collagen content. Right ventricular hypertrophy was assessed by Fulton's index.

Vessels were divided into Small (≤50 µm), Medium (<50-100 µm) and Large (>100 µm). Pulmonary arteries 100–300 µm in diameter contribute the most to total pulmonary vascular resistance.

Group comparisons employed Mann–Whitney or Kruskal–Wallis tests. For correlation between collagen content in the lungs and proportion of non-obliterated vessels, correlation coefficients (rho) and p-values were calculated for all groups combined. Significance was defined as p < 0.05.

References

[1] Ganslandt et al., Am J Respir Crit Care Med 09: A1055, 2024.
[2] Tornling et al., Int J Mol Sci 24: 7478, 2023.

vicore pharma