REGULAR ARTICLE

The Future of PAH Treatment

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Pulmonary arterial hypertension (PAH) is a devastating disease mediated by vasoconstriction and vascular remodeling of the pulmonary vasculature. Current therapies target the imbalance of vasoconstrictors and vasorelaxants in 3 pathways: nitric oxide, prostacyclin, and endothelin. While these have extended lifespans for PAH patients, significant morbidity and mortality remains. Notably, the progress in PAH therapy for over a decade has utilized these same 3 pathways. Fortunately, several new treatment options utilizing different mechanisms are emerging and will be reviewed here.

INTRODUCTION

Pulmonary arterial hypertension (PAH) is a result of complex pathologic processes culminating in a progressive, incurable disease characterized by elevated pulmonary vascular resistance and right ventricular (RV) dysfunction. Elevated pulmonary afterload derives from both increased vasoconstrictive tone and deranged vascular remodeling that has been likened to a pseudomalignant phenotype. Significant efforts have been made to understand the underlying pathophysiologic processes in the quest for treatment options. Currently approved therapies target 3 pathways—nitric oxide, prostacyclin, and endothelin—as patients with PAH have chronic upregulation of vasoconstrictors such as endothelin and chronic deficiency in vasodilators such as nitric oxide and prostacyclins. However, significant pulmonary vascular disease remains, reflected clinically with an improved but persistently high mortality, particularly in those with high-risk disease.[1] Fortunately, additional pathways involved in the disease have been elucidated and are now candidates for targeted intervention. Many emerging pharmaceuticals target pulmonary vascular fixed remodeling mediated by imbalanced pro-proliferative and antiapoptotic pathways (Figure 1). These potential treatments, together with current therapies, may provide synergistic effects to improve outcomes for our PAH patients. At the same time, ideally we will continue to advance our understanding of precision-based treatments in PAH and move toward the "right drug(s)" for

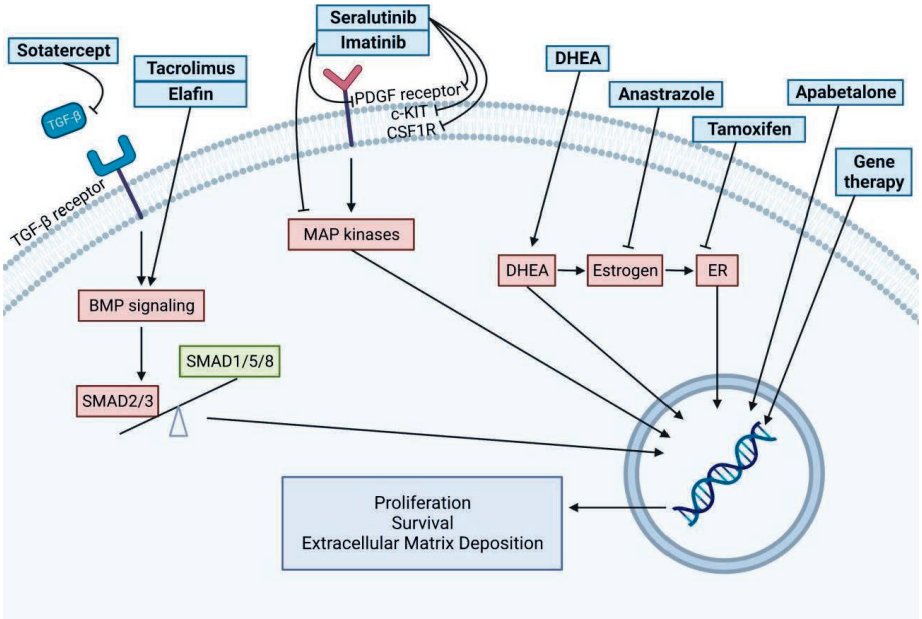

Figure 1: Mechanisms of action of emerging PAH therapies.

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Table 1. Summary of Selected New Potential Drugs in the Treatment of PAH

Drug	Mechanism	Trial	Phase	Primary Outcome	Status
Sotatercept	TGF-β ligand trap, BMP signal potentiation	STELLAR	Phase III	Change in 6MWD at 24 weeks	Completed
		SOTERIA	Phase III	Adverse events, detectable anti-drug antibodies, abnormal hematology or chemistry laboratory results, abnormal weight, abnormal blood pressure, ECG, abnormal urinalysis up to 200 weeks	Recruiting
		ZENITH	Phase III	Time to first confirmed morbidity or mortality up to 46 months	Recruiting
		HYPERION	Phase III	Time to clinical worsening	Recruiting
Elafin	Elastase inhibitor, BMP signaling potentiation	*Planned*			
Tacrolimus (FK506)	Calcineurin inhibitor, BMP signaling potentiation	*Planned*			
Imatinib	Tyrosine kinase inhibitor	PIPAH (NCT04416750)	Phase II	Change in PVR at 24 weeks	Recruiting
		IMPAHCT (NCT05036135)	Phase IIb/III	2b: Change in PVR at 24 weeks 3: Change in 6MWD at 24 weeks	Recruiting
Seralutinib (GB002)	Tyrosine kinase inhibitor BMP signaling potentiation	TORREY (NCT04456998)	Phase II	Change in PVR at 24 weeks	Completed
Apabetalone	BET protein inhibitor	APPROACH-2 (NCT04915300)	Phase II	Change in PVR at 24 weeks	Not yet recruiting
Tamoxifen	Estrogen receptor	T3PAH	Phase II	Change in TAPSE at 24 weeks	Recruiting
	inhibitor	(NCT03528902)			
Anastrazole	Aromatase inhibitor	PHANTOM (NCT03229499)	Phase II	Change in 6MWD at 6 months	Completed
DHEA	Steroid hormone precursor	EDIPHY (NCT03648385)	Phase II	Change in RV longitudinal strain on CMRI at 18, 40 weeks	Recruiting
Gene therapy		*Preclinical stage*			
eNOS enhanced progenitor cell transplant	eNOS enhancement	SAPPHIRE (NCT03001414)	Phase II/III	Change in 6MWD at 6 months	Recruiting
Microbiome transfer	Modulating systemic inflammation	NCT04884971	Phase I	Adverse effects and compliance	Recruiting

Abbreviations: TGF indicates transforming growth factor; BMP, bone morphogenetic protein; 6MWD, 6-minute walk distance; ECG, electrocardiogram; PVR, pulmonary vascular resistance; BET, bromodomain and extraterminal motif; TAPSE, tricuspid annular plane systolic excursion; DHEA, dehydroepiandrosterone, RV, right ventricle; CMRI, cardiac magnetic resonance imaging; eNOS, endothelial nitric oxide synthase.

the "right patients" to minimize costly and burdensome regimens and maximize outcomes and quality of life for our patients. It is equally important to validate surrogate endpoints that reflect patient-centered outcomes in PAH trials alongside the discovery of novel mechanistic targets. Readily available PAH risk scores have been suggested as surrogate outcomes in randomized controlled trials of PAH but further validation is required (data unpublished). Identification of alternative validated surrogates is warranted as the efficacy of these emerging potential therapeutics is studied. We will review some of the most promising novel pharmaceuticals currently on the horizon and in clinical trials (Table 1).

BONE MORPHOGENETIC PROTEIN SIGNALING MODULATORS

Disruptions in signaling of the transforming growth factor–β (TGF-β) superfamily contributes significantly to the dysregulated vascular proliferation of PAH. Germline mutations specifically in the TGF receptor of bone morphogenetic protein receptor type 2 (*BMPR2*) and its downstream signalers are the most common genetic cause of heritable PAH,[2] with BMPR2 itself playing a critical gatekeeping role.[3] Bone morphogenetic protein signaling and function is also decreased in nonheritable PAH.[4,5] Downstream of BMPR2, evidence

suggests an imbalance of SMAD signaling with underactive antiproliferative SMAD 1/5/8 signaling and overactive pro-proliferative SMAD 2/3.[6]

Sotatercept

The recombinant fusion protein sotatercept targets this deranged signaling by preferentially inhibiting the pro-proliferative SMAD2/3 pathway. Sotatercept has been previously studied in conditions characterized by TGF-β signaling dysregulation, including multiple myeloma and myelodysplastic syndrome.[7,8] The recent phase II PULSAR trial evaluated sotatercept's efficacy in PAH patients on stable background therapy, at doses of 0.3 mg/kg and 0.7 mg/kg for 24 weeks.[9] The least-squares mean difference of change in pulmonary vascular resistance (PVR) in the sotatercept 0.3-mg/kg group as compared with the placebo group was −145.8 dyn·sec·cm^{-5} (95% confidence interval [CI]: −241.0 to −50.6 dyn·sec·cm^{-5}, $P = .003$), while that for the sotatercept 0.7-mg/kg group compared with placebo group was −239.5 dyn·sec·cm^{-5} (95% CI: −329.3 to 149.7 dyn·sec·cm^{-5}; $P < .001$). Secondary endpoints included improved 6-minute walk distance (6MWD) with least-squares mean difference for the 0.3-mg/kg sotatercept participants compared to placebo participants of 29.4 m (95% CI: 3.8 to 55.0 m) and that of the 0.7-mg/kg group compared to placebo at 21.4 m (95% CI: −2.8 to 45.7 m). Additionally, N-terminal pro-B-type natriuretic peptide (NT-proBNP) decreased with least-squares mean difference between the sotatercept 0.3-mg/kg group and placebo group of −931.5 pg/mL (95% CI: −1353.2 to −509.7 pg/mL) and of the sotatercept 0.7-mg/kg group, −651.0 pg/mL (95% CI: −1043.3 to −258.7 pg/mL).

Multiple phase 3 trials of sotatercept in patients with PAH with varying disease duration and risk profiles are currently ongoing. Early results from STELLAR, a randomized, multi-center study of sotatercept in patients with PAH with World Health Organization (WHO) functional class (FC) II or III on background PAH therapies, were recently reported as meeting the study's primary endpoint of improvement in 6MWD, along with 8 of 9 secondary endpoints including time to clinical worsening, a multicomponent improvement from baseline, maintenance or improvement in WHO FC, change from baseline PVR, change from baseline in NT-proBNP, maintenance or improvement to low risk score, change from baseline in physical impacts, and change from baseline cardiopulmonary symptoms.[10,11] Additional active phase III trials of sotatercept include SOTERIA (NCT04796337),[12] evaluating long-term efficacy and safety; ZENITH (NCT04896008),[13] investigating efficacy in advanced WHO FC III or IV patients on maximally tolerated background therapy; and HYPERION (NCT04896008)[14], studying the drug's efficacy in incident PAH patients.

Elafin

Increased elastase activity has been demonstrated in the pulmonary arteries of experimental PAH models with degradation of elastin, a structural protein that contributes to pulmonary vascular integrity and elastance. Elastin degradation is associated with pulmonary artery smooth muscle cell (PASMC) proliferation.[15] Elafin is a naturally occurring elastase inhibitor with additional antimicrobial and anti-inflammatory properties. Treating pulmonary artery endothelial cells from PAH patients with elafin led to an increase in BMP signaling and a reduction in neointimal formation in cultured pulmonary artery endothelial cells.[16] In the Sugen-hypoxia rat model of PAH, elafin reversed pulmonary vascular occlusive changes and normalized RV pressure.[16,17] These data suggest that elafin may augment BMPR2 signaling in PAH as well as increase expression of apelin, a target of BMPR2 signaling. A small phase I trial (NCT03522935) in healthy patients treated with elafin is complete with plans for a phase 2 proof-of-concept study[18].

Tacrolimus

Tacrolimus, a calcineurin inhibitor used routinely for immunosuppression in transplant patients, also activates BMPR2 signaling.[19] In animal models, low-dose tacrolimus increased BMPR2 signaling in pulmonary artery endothelial cells and reversed pulmonary vascular remodeling in a murine BMPR2 knockout.[19] A small phase 2a trial of tacrolimus at 3 different target levels in PAH patients on background therapy showed no improvement in 6MWD or RV function, but may be efficacious in select patients.[20] Nonetheless, a larger phase II study is being planned to determine the efficacy of tacrolimus more definitively in PAH.

TYROSINE KINASE PATHWAY

Aberrant proliferation of pulmonary vascular smooth muscle cells has been in part attributed to growth factors such as platelet-derived growth factor (PDGF), a potent PASMC mitogen[21] that is increased in PAH patients.[22] PDGF receptors (PDGFRs) belong to a family of tyrosine kinase receptors, and preclinical data have demonstrated that tyrosine kinase inhibitors both attenuate pulmonary vascular remodeling through PDGFR inhibition but also directly relax the pulmonary vasculature.[23] As such, several tyrosine kinase inhibitors are now under clinical investigation for PAH.

Imatinib

Imatinib, a Bcr-Abl inhibitor originally developed to treat chronic myeloid leukemia, also inhibits PDGF. Imatinib potently inhibited PDGF-dependent PASMC proliferation, with near full reversal of pulmonary hypertension in the monocrotaline and hypoxic rat model.[24] Further in vitro data demonstrated that imatinib exerted proapoptotic effects in PDGF-stimulated PASMCs from idiopathic PAH patients.[25] A phase II study of imatinib versus placebo found improvements in PVR and cardiac output specifically in patients with more significant hemodynamic impairment (PVR ≥ 1000 dyn·sec·cm^{-5}), suggesting a role as add-on therapy for a subset of advanced PAH patients.[26] The compelling preclinical data and phase II trial ultimately led to evaluation of imatinib in the IMPRES trial, which enrolled subjects on at least dual background PAH therapies. At 24 weeks, the mean placebo-corrected treatment effect on 6MWD was 32 m (95% CI: 12 to 52 m; $P = .002$) and PVR decreased by 379

dyn·sec·cm^{-5} (95% CI: −502 to −255 dyn·sec·cm^{-5}; P < .001, between-group difference); however, WHO FC, time to clinical worsening, and mortality did not differ between the groups.[27] Serious adverse effects were noted in the imatinib group compared to placebo (44% versus 30%), particularly subdural hematomas, which in addition to significant dropout due to intolerances tempered enthusiasm for imatinib. At present, the ongoing phase II PIPAH trial (NCT04416750)[28] aims to identify the highest tolerated dose of oral imatinib, assess efficacy as measured by PVR reduction, and identify the patients most likely to respond via analyses of plasma proteins and genes, particularly those encoding PDGF. A phase III study of an additional oral formulation of imatinib with enteric coating meant to mitigate gastrointestinal side effects is also planned.[29] Concurrently, IMPAHCT (NCT05036135) is a phase IIb/III trial of dry powder inhaled form of imatinib, which will identify the optimal dose and examine effects of inhaled imatinib on 6MWD and PVR at 24 weeks.[30]

Seralutinib

Seralutinib, which was specifically developed to target several tyrosine kinase inhibitors implicated in PAH pathogenesis including PDGFRα/β, colony stimulating factor 1 receptor, and c-KIT while also increasing BMPR2, has reversed pulmonary vascular remodeling and improved hemodynamics in 2 preclinical pulmonary hypertension models.[31] The phase II TORREY trial of seralutinib delivered by dry powder inhaler was recently completed with early reports of modest improvements in the primary endpoint of PVR at 24 weeks (14.3% placebo-corrected improvement; P = .03) with the secondary endpoint of 6MWD favoring seralutinib as well.[32,33] Findings were more striking in subgroup analyses of more symptomatic WHO FC III patients for the seralutinib arm versus placebo (21% reduction in PVR, P = .04; 37-m improvement in 6MWD, P = .048), and in patients with intermediate- or high-risk REVEAL 2.0 scores (23% reduction in PVR, P = .01; 22-m increase in 6MWD, P = .25) for the seralutinib arm versus placebo.

BROMODOMAIN PROTEINS

Bromodomains (BRDs) are epigenetic drivers of the BRD and extraterminal motif protein family that regulate gene transcription. BRD and extraterminal motif inhibitors may also exert favorable effects on the myocardium and decreased hospitalizations in patients with left heart disease following acute coronary syndrome.[34] BRD4 specifically can inhibit apoptosis, promote hyperproliferation, and stimulate a switch into a proinflammatory phenotype and as such has been implicated in cancer.[35-37] BRD inhibitors have thus been identified as treatment options for cancers.[38] Given the cancer-like proliferation of PASMCs in PAH, it is not surprising that BRD4 has also been identified as a contributor to the proliferation in PAH, with significant upregulation detected in human pulmonary artery tissue.[35]. Accordingly, inhibition of BRD4 reverses pulmonary vascular remodeling and improves hemodynamics in preclinical pulmonary hypertension models.[35] Similar findings were observed in the Phase I APPROACH-p trial of the BRD4 inhibitor apabetalone, with decreased PVR (−140 dyn·s·cm^{-5}; 95% CI: −200 to −79 dyn·sec·cm^{-5}) noted in 7 PAH patients treated for 16 weeks.[39] Improvements in cardiac output (+0.73 L/min; 95% CI: −0.22 to +1.68 L/min) and stroke volume (+8 mL; 95% CI: −4 to +20 mL) were also noted with apabetalone. The larger phase II APPROACH-2 trial (NCT04915300) will confirm or refute these findings.[40]

SEX HORMONES

Estrogen

Despite early identification of female sex as a major risk factor for PAH[41,42] and subsequent intense investigation into sex hormones and their contribution to PAH pathobiology, the exact role of estrogen remains incompletely defined as reviewed more thoroughly in other works.[43–46] Briefly, it is clear that despite female predominance, once PAH is established estrogen provides protective effects on RV function,[47,48] allowing female patients better prognoses.[33,34,41,42,47,49] However, the role of estrogen signaling and metabolism in the pulmonary vasculature itself is complex. Beneficial effects including vasodilation and angiogenesis are noted in some animal studies,[50,51] but other studies report that estrogen promoted destructive vascular remodeling.[52,53] Clinical evidence supports a deleterious relationship with higher circulating estrogen noted in PAH patients, including men.[54] Furthermore, in animal models of PAH, inhibiting estrogen receptors with tamoxifen or inhibiting conversion of androgens to estrogen with anastrazole reversed PAH[55,56] A small phase II clinical trial showed that anastrazole decreased circulating estrogen by 40% and increased 6MWD.[57] This launched the PHANTOM trial (NCT03229499) now underway examining the effects of anastrazole in postmenopausal women and men with PAH.[58] Whether inhibition of estrogen receptors with tamoxifen may benefit PAH patients is also being evaluated with the single-center Phase II T3PAH trial (NCT03528902).[59]

Dehydroepiandrosterone

The precursor to both estrogen and androgens, dehydroepiandrosterone (DHEA) prevented and treated PAH and RV dysfunction in animal models.[60] Clinically, lower DHEA is associated with higher risk of PAH in men[46] and increased risk and severity in women.[61] The consistent data suggesting benefit of DHEA in PAH may be explained by DHEA-mediated enhanced endothelial nitric oxide synthesis or through direct cardioprotective effects. The single-center crossover trial EDIPHY (NCT03648385) is currently testing DHEA efficacy in PAH patients by measuring RV longitudinal strain.[62]

EXPLORATORY THERAPIES

Beyond typical pharmacologic options, other novel approaches are currently under investigation for this complex and morbid disease. As specific gene mutations are implicated in heritable PAH and account for 6% to 10% of all PAH, gene therapy provides an attractive approach to directly correct aberrant genes and restore balance between proliferation and apoptosis. Preclinical pulmonary hypertension models have proven amenable with improvements in

pulmonary vascular remodeling via viral transfection endotracheally and intravenously.[63–65] Work in experimental models is ongoing to determine ideal and effective gene therapy delivery methods.

Stem or progenitor cell therapy may offer similar direct restoration of pulmonary vasculature homeostasis. With the abnormal endothelial dysfunction and hyperproliferation of PASMCs, regenerative cell treatment could interfere and restore vasculature architecture. Endothelial progenitor cells (EPCs) appear protective in PH animal models, including specifically with BMPR2-augmented EPCs, which improved mean pulmonary artery pressures and RV hypertrophy in monocrotaline-induced models.[66] Small pilot randomized controlled trials have demonstrated safety and efficacy of stem cell therapy in humans and a 2019 meta-analysis of 16 small clinical trials with stem cell therapy in PAH patients revealed that despite heterogeneity in findings, weighted-means differences indicated improvements in RV systolic pressure, mean pulmonary artery pressure, and mean RV pressure with P values all $< .001$ in patients treated with stem cells.[67] The PHACeT study in 2015 reported that when treated with 3 doses of enhanced endothelial nitric oxide synthase EPCs, PAH patient demonstrated improved hemodynamics in the short term with good tolerance; however, findings were not sustained at 3 and 6 months,[68] despite prior EPC data showing sustained hemodynamic and exertional effects at 3 months.[69] A recent landmark report described the use of human umbilical cord mesenchymal stems cells to treat a child with heritable PAH and hereditary hemorrhagic telangiectasia which improved clinical parameters at 6 months.[70] Currently the phase II SAPPHIRE study (NCT03001414) is recruiting and aims to assess safety and efficacy of monthly administration of autologous EPCs transfected with human endothelial nitric oxide synthase in severe PAH patients.[71]

Finally, investigation into the microbiome may elucidate novel mechanisms and therapeutic targets in PAH. Compared to controls, PAH patient microbiomes demonstrated decreased alpha diversity with distinct signatures even from unaffected family members, and enrichment of bacteria associated with the proinflammatory metabolite trimethylamine oxide.[72,73] Species associated with trimethylamine oxide were increased as was serum trimethylamine oxide in high-risk PAH patients, whereas species associated with anti-inflammatory metabolites were reduced. Guided by this data, a Phase I trial (NCT04884971) is currently evaluating the safety of microbiome transplant in PAH patients.[74]

CONCLUSION

Despite significant progress in PAH therapeutics over the last 2 decades, innovative treatments are needed to ameliorate morbidity and mortality in this progressive deadly disease. Beyond our current arsenal of treatments, BMP signaling, tyrosine kinase signaling, BRD proteins, sex hormones, and other more novel approaches such as gene therapy targeting pulmonary vascular remodeling are in varied stages of development. With continued scientific rigor used to explore new signaling pathways and mechanisms, we are one step closer to halting, if not reversing, this devastating disease.

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