

# Novel Treatment Pathways in Pulmonary Arterial Hypertension

Kanza N Qaiser, MD; Adriano R Tonelli, MD, MSc

CLEVELAND CLINIC, CLEVELAND, OHIO

**ABSTRACT:** Pulmonary arterial hypertension (PAH) is a severe and progressive vascular disease characterized by pulmonary vascular remodeling, proliferation, and inflammation. Despite the availability of effective treatments, PAH may culminate in right ventricular failure and death. Currently approved medications act through three well-characterized pathways: the nitric oxide, endothelin, and prostacyclin pathways. Ongoing research efforts continue to expand our understanding of the molecular pathogenesis of this complex and multifactorial disease. Based on recent discoveries in the pathobiology of PAH, several new treatments are being developed and tested with the goal of modifying the disease process and ultimately improving the long-term prognosis.

## INTRODUCTION

Pulmonary arterial hypertension (PAH) is a progressive disease characterized by pulmonary arterial vasoconstriction, inflammation, thrombosis, vascular proliferation, and remodeling, and it is likely to culminate in right ventricular (RV) failure and death.<sup>1,2</sup> Despite striking advances in treatment, PAH mortality continues to be high. In fact, according to the Registry to Evaluate Early and Long-term PAH Disease Management (REVEAL registry), the 7-year survival rate is only 49%.<sup>3</sup> Currently available treatments act on three major pathways (nitric oxide, endothelin and prostacyclin),<sup>4</sup> and all US Food and Drug Administration (FDA)-approved medications are predominantly vasodilators<sup>1,5,6</sup> with limited effects on the disease process.<sup>7</sup> Disease-modifying agents that can alter the course of the disease are certainly needed to improve outcomes. Continuous progress in the understanding of the pathophysiology of PAH offers exciting opportunities for the development of new therapeutic targets, with the central objective of modifying the disease process and ultimately improving long-term survival. This review highlights existing treatments and their pathways for PAH and discusses novel therapies currently in development.

## CURRENTLY APPROVED THERAPIES

There are ten unique molecules currently approved by the FDA to treat PAH.<sup>6</sup> In addition, calcium channel blockers are approved specifically for the subgroup of PAH patients with positive acute vasoreactivity during right heart catheterization.<sup>7,8</sup> It should be noted that there are no randomized controlled trials (RCTs) evaluating the role and efficacy of calcium channel blockers in treating PAH, and many of the recommendations are based on clinician experience and expert consensus.<sup>9</sup> The ten FDA-approved drugs are grouped into five categories—phosphodiesterase type 5 inhibitors, soluble guanylate cyclase stimulators,

endothelin receptor antagonists, prostacyclin analogs, and prostacyclin receptor agonists—and have different mechanisms of action.

Nitric oxide (NO) is a powerful pulmonary vasodilator that activates soluble guanylate cyclase, leading to increased cyclic guanine monophosphate (cGMP) levels and resulting in smooth muscle relaxation.<sup>5</sup> In PAH, endothelial dysfunction triggers reduced levels of endothelial NO synthase and increased levels of phosphodiesterase 5 (PDE5), leading to lower NO production and increased cGMP degradation.<sup>7</sup> Phosphodiesterase type 5 inhibitors (eg, sildenafil, tadalafil) and soluble guanylate cyclase stimulators (eg, riociguat) exert their effects via the NO-cGMP pathway.<sup>5,7</sup>

Endothelin (ET), a potent vasoconstrictor produced primarily by endothelial cells, is upregulated in PAH.<sup>5,7</sup> It mediates vasoconstriction via its effects on the ET-A and ET-B receptors, which are targeted by the endothelin receptor antagonists (ambrisentan, bosentan, and macitentan).<sup>5</sup>

Prostacyclin (PGI<sub>2</sub>) is produced by endothelial cells and has vasodilatory, anti-inflammatory, antithrombotic, and antiproliferative properties through its action on PGI<sub>2</sub> receptors, leading to increased cyclic adenosine monophosphate (cAMP) levels.<sup>5</sup> Patients with PAH have a marked reduction in PGI<sub>2</sub> synthase, with lower plasma levels and urinary excretion of PGI<sub>2</sub> metabolites,<sup>10</sup> and decreased expression of PGI<sub>2</sub> receptors in the lungs.<sup>11</sup> Prostacyclin analogs (eg, eprostenol, treprostinil, and iloprost) and PGI<sub>2</sub> receptor agonists (eg, selexipag) act on the prostacyclin pathway.<sup>7</sup>

Due to a lack of head-to-head comparison studies, the relative advantages of these treatment options is not yet known.<sup>12</sup> However, abundant data demonstrate superiority of combination-therapy regimens over monotherapy in either an

upfront<sup>13</sup> or sequential<sup>14-16</sup> combination approach. Current proceedings in pulmonary hypertension (PH) based their treatment recommendations on the severity of the disease,<sup>17</sup> with PAH patients stratified as low, intermediate, or high risk based on the 1-year mortality of < 5%, 5% to 10%, and > 10%, respectively.<sup>18</sup> There are several risk-assessment tools to determine this risk, including the recently updated REVEAL 2.0,<sup>19</sup> COMPERA (Comparative, Prospective Registry of Newly Initiated Therapies for Pulmonary Hypertension),<sup>20</sup> and French Pulmonary Hypertension Registry methods.<sup>21</sup> The latest recommendations for PH include initial dual oral combination therapy for the majority of patients with PAH, with the addition of a third agent when a low-risk status is not achieved.<sup>17</sup>

Several meta-analyses have explored the effect of therapies on PAH. A meta-analysis of 26 trials with a total of 3,519 patients reported an all-cause mortality risk reduction of 39% regardless of the class of therapy used.<sup>22</sup> Another meta-analysis of 17 RCTs with 4,095 total patients compared sequential combination therapy with monotherapy and reported a 35% reduced risk of clinical worsening with combination therapy; however, this effect did not translate to a significant improvement in mortality (14% reduction;  $P = .09$ ).<sup>23</sup>

Two studies of PAH that were recently presented in meetings are worth mentioning. The TRITON trial (Efficacy and Safety of Initial Triple Versus Initial Dual Oral Combination Therapy in Patients With Newly Diagnosed Pulmonary Arterial Hypertension)—which compared triple therapy with tadalafil, macitentan, and selexipag versus double therapy with tadalafil and macitentan—showed no statistical differences in the primary end point (change in pulmonary vascular resistance, or PVR) and multiple secondary end points (6-minute walk distance [6MWD], NT pro-brain natriuretic

peptide, and no worsening in functional class) between the two groups.<sup>24</sup> The Riociguat trial (rEplacing PDE-5i Therapy evaluated Against Continued PDE-5i thErapy) tested whether switching treatment to riociguat would be effective in intermediate-risk PAH patients who did not achieve adequate clinical response on stable doses of PDE5 inhibitors.<sup>25</sup> A total of 41% of patients transitioned to riociguat achieved the primary end point of satisfactory clinical response in the absence of clinical worsening compared with 20% of the group on PDE5 inhibitors (OR 2.8; 95% CI, 1.5–5.1;  $P = .0007$ ).<sup>26</sup>

#### ONGOING STUDIES ON TRADITIONAL PAH TREATMENT PATHWAYS

UNISUS (Outcome Study Assessing a 75 Milligrams Dose of Macitentan in Patients With Pulmonary Arterial Hypertension) is a phase 3 multicenter, randomized, double-blind trial testing the superiority of high-dose macitentan (75 mg/day) compared with the FDA-approved dose of 10 mg/day in patients with PAH.<sup>27</sup> The primary outcome of this study is time to first clinical morbidity (unplanned PAH-related hospitalization, disease progression, or worsening functional class) or mortality.<sup>27</sup> Another study involving the same ET pathway, this one a phase 1 trial of getagozumab, is currently underway.<sup>28</sup> Getagozumab, a long-lasting humanized monoclonal antibody that targets the receptor ET-A, has been shown to lower pulmonary artery pressures and curtail pulmonary artery and right ventricular remodeling when administered parenterally in animal models.<sup>29</sup>

Ralinepag is a novel oral selective PGI<sub>2</sub> receptor agonist that has been shown to reduce PVR in patients with PAH who are on mono or dual combination background therapy.<sup>30</sup> ADVANCE OUTCOMES (A Study Evaluating the Efficacy and Safety of Ralinepag to Improve Treatment Outcomes in PAH Patients) is a phase 3 RCT currently underway to evaluate the effectiveness of

ralinepag when added to approved PAH therapies.<sup>31</sup> The primary end point is time-to-clinical-events, defined as mortality, PAH-related unplanned hospitalization, disease progression, unsatisfactory treatment response, and need for intravenous PGI<sub>2</sub> therapy.<sup>31</sup> Additional trials (NCT04084678)<sup>32</sup> are planned to evaluate ralinepag's effects on exercise capacity and 6MWD.

Several other treatment options and delivery methods have recently been tested or have already launched in the US market. A recent phase 3 study demonstrated the safety and tolerability of switching from oral to intravenous selexipag as bridging therapy in PAH patients who were temporarily unable to take the oral medication.<sup>33</sup> Remunity, a novel subcutaneous continuous infusion pump from United Therapeutics that delivers treprostinil to patients with PAH, received final FDA clearance in 2020 and launched earlier this year.<sup>34</sup> The pump has a compact size and uses prefilled, disposable cassettes.<sup>34</sup> Medtronic has developed and tested a fully implantable system that delivers treprostinil without an external pump or tubing.<sup>35,36</sup> This system, which has shown to save time and increase independence and quality of life, is awaiting final FDA approval.<sup>37</sup>

Newer delivery systems for inhaled treprostinil are currently under development. These include dry powder inhaled forms of treprostinil (LIQ861 by Liquidia and Tyvaso DPI, and Treprostinil Technosphere® by United Therapeutics and Mannkind) that would increase portability and convenience compared to the currently available portable inhalation system (TYVASO®).

#### NOVEL TREATMENT PATHWAYS

Increased understanding of PAH and its molecular pathogenesis has uncovered a number of promising pathways for disease-modifying therapeutic targets (Figure 1).

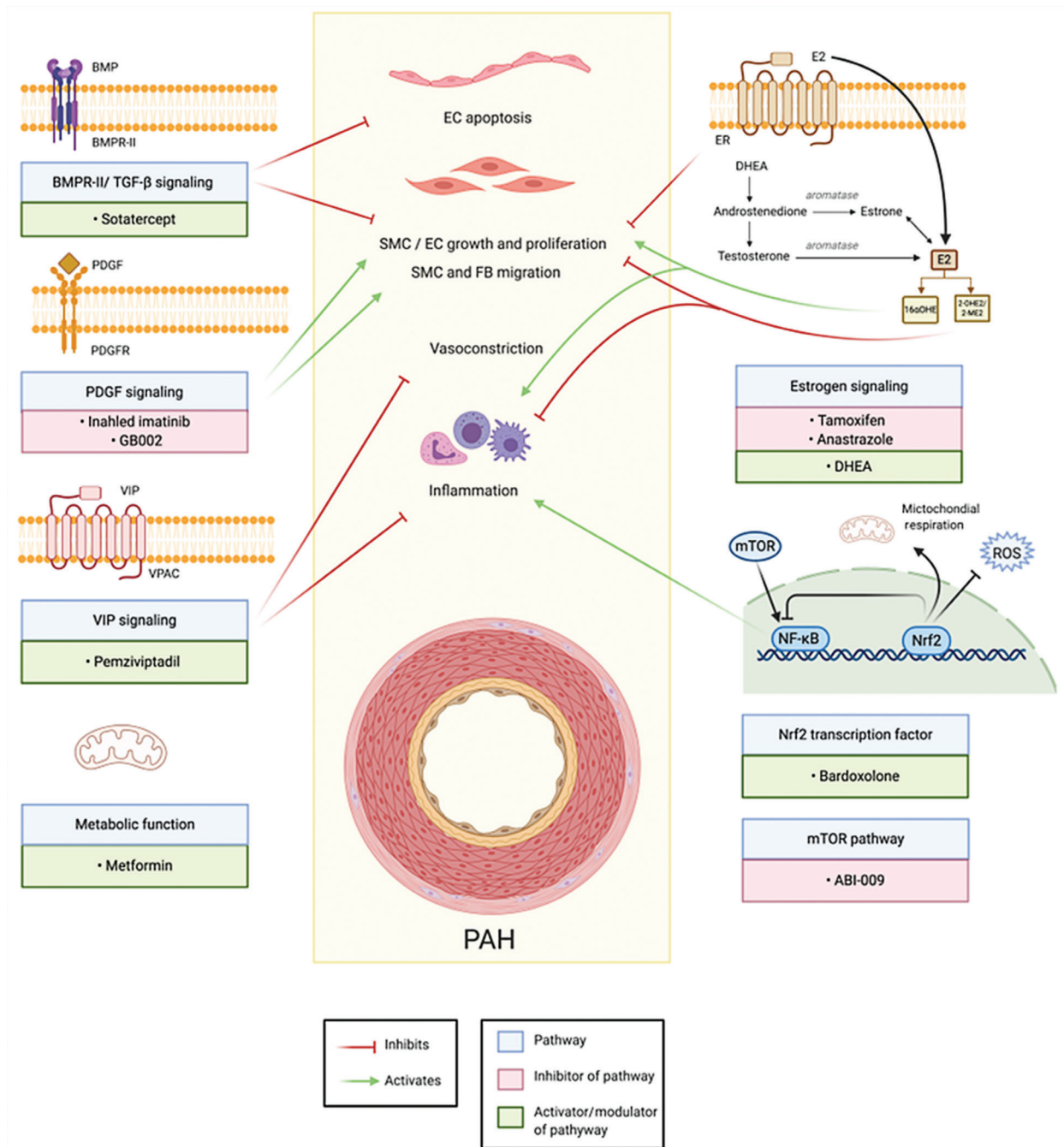


Figure 1.

Promising therapeutic pathways in pulmonary arterial hypertension (PAH). The interplay of several pathways and cellular processes contributes to the development of PAH by various mechanisms, including vasoconstriction, inflammation, dysregulated endothelial and smooth muscle cell growth, proliferation, migration, and apoptosis. This figure represents the novel treatment pathways discussed in the article, along with the medications that inhibit, activate, or modulate these pathways. It does not represent an exhaustive list. BMP: bone morphogenetic protein; BMPR-II: BMP receptor type 2; TGF-β: transforming growth factor-β; PDGF: platelet-derived growth factor; PDGFR: PDGF receptor; VIP: vasoactive intestinal peptide; VPAC: vasoactive intestinal peptide receptor; EC: endothelial cells; SMC: smooth muscle cells; FB: fibroblasts; PAH: pulmonary arterial hypertension; E2: estradiol; ER: estrogen receptor; 16αOHE: 16α-hydroxyoestrone; 2-OHE2: 2-hydroxyoestradiol; 2-ME2: 2-methoxyoestradiol; DHEA: dehydroepiandrosterone; ROS: reactive oxygen species; Nrf2: nuclear factor erythroid 2-related factor 2; NF-κB: nuclear factor kappa-light-chain-enhancer of activated B cells; mTOR: mammalian target of rapamycin. Created with BioRender.com

### *Modulation of Transforming Growth Factor-B/Bone Morphogenetic Protein Receptor 2 Signaling*

Bone morphogenetic proteins (BMP) are part of the transforming growth factor- $\beta$  (TGF- $\beta$ ) superfamily of cytokines.<sup>38</sup> Heterozygous mutations with loss of function in bone morphogenetic protein receptor 2 gene (*BMPR2*) lead to an unopposed TGF- $\beta$  signal.<sup>4</sup> The *BMPR2* gene has the most mutations associated with PAH, found in up to 80% of patients with heritable PAH and 11% to 40% of those with idiopathic PAH.<sup>39</sup> The lifetime penetrance of *BMPR2* mutations in patients with PAH is 14% to 42%,<sup>40</sup> supporting the concomitant role of environmental and other genetic factors in the disease pathogenesis.<sup>39</sup> *BMPR2* signaling is important in regulating vascular cell apoptosis and maintaining the integrity of the pulmonary artery endothelium.<sup>38</sup> Suppression of *BMPR2* signaling leads to maladaptive endothelial inflammatory response, culminating in vascular cell proliferation and remodeling.<sup>38,39</sup> Interestingly, the pulmonary vascular *BMPR2* expression was found to be reduced in PAH patients, even in the absence of *BMPR2* mutations.<sup>41</sup> Therefore, balancing the opposing effect of *BMPR2* and TGF- $\beta$  signaling is a promising novel approach to treat PAH patients.

Sotatercept is a first-in-class medication that modulates the TGF- $\beta$  and *BMPR2* signaling pathways.<sup>42</sup> It is a fusion protein that works as a selective ligand trap for members of the TGF- $\beta$  superfamily (such as activin A and B and growth differentiation factor 11), thereby suppressing TGF- $\beta$  and enhancing *BMPR2* signaling.<sup>42</sup> The results of PULSAR, a phase 2 double-blind placebo-controlled trial evaluating the efficacy of sotatercept in functional class II-III PAH patients, were recently presented in an abstract form.<sup>43</sup> Patients were randomized to receive placebo or sotatercept (0.3 or 0.7 mg/kg) subcutaneously every 21 days in addition to standard PAH therapy.<sup>43</sup> At week 24, sotatercept significantly reduced PVR compared with placebo.<sup>43</sup> Furthermore, statistically significant improvements were also noted in secondary end points with sotatercept, such as 6MWD.<sup>43</sup> A phase 3 trial is planned to confirm these exciting results.

### *Inhibition of Platelet-Derived Growth Factor Signaling*

The platelet-derived growth factor (PDGF) signaling pathway is overexpressed in the pulmonary vasculature of patients with PAH, with upregulation of PDGF-A, PDGF-B, and their receptors (PDGFR- $\alpha$  and - $\beta$ ).<sup>44</sup> An increase in PDGF activity leads to endothelial dysregulation and anomalous proliferation of smooth muscle cells (SMCs).<sup>4</sup> Since PDGF acts through transmembrane tyrosine kinase receptors, tyrosine kinase inhibitors such as imatinib have

been repurposed as a therapeutic option in PAH.<sup>5</sup> IMPRES (Imatinib [QTI571] in Pulmonary Arterial Hypertension)<sup>45</sup> was a phase 3 RCT investigating the role of oral imatinib in the treatment of severe PAH (PVR  $\geq$  10 Wood units) in patients receiving two or more PAH-specific background therapies.<sup>46</sup> After 24-week follow-up, oral imatinib significantly improved exercise capacity, measured by the 6MWD, and reduced PVR.<sup>46</sup> Despite these benefits, further investigation of imatinib for PAH was halted due to unexpectedly high rates of discontinuations (33%) and serious adverse events (44%), including subdural hematoma in patients who were also receiving anticoagulation.<sup>46</sup> Inhaled imatinib is expected to reduce the overall dose by delivering the medication directly to the lungs and, in doing so, mitigate the adverse events observed with oral imatinib.<sup>47</sup> A phase 1 clinical trial testing inhaled imatinib in patients with PAH is expected to start in the near future.

A novel PDGF receptor- $\alpha/\beta$  inhibitor (GB002) given by inhalation improved cardiopulmonary hemodynamics by reducing RV systolic pressure and mean pulmonary artery pressure, reduced pulmonary arteriole muscularization and inflammatory cytokines, and restored *BMPR2* signaling.<sup>48</sup> A phase 1B trial is currently underway,<sup>49</sup> with plans for a phase 2 trial in the near future.<sup>50</sup>

### *Improved Mitochondrial and Metabolic Function*

The “metabolic theory” of PAH, also known as the Warburg Effect, is based on the observation that pulmonary vascular cells preferentially switch their metabolism from mitochondrial oxidative phosphorylation to glycolysis for ATP production.<sup>51</sup> This metabolic change contributes to abnormal cell proliferation and resistance to apoptosis, mimicking the cellular processes seen in neoplasia.<sup>51</sup> Changes in aerobic respiration affect reactive oxygen species (ROS) levels, which in turn modulate vascular tone.<sup>51</sup> Activation of the transcription factor nuclear factor erythroid 2-related factor 2 (Nrf2) suppresses the effects of the proinflammatory factor nuclear factor kappa-light-chain-enhancer of activated B cells (NF- $\kappa$ B) and has been shown to improve metabolic dysfunction by promoting mitochondrial respiration, reducing ROS levels, and decreasing inflammation.<sup>51,52</sup> In addition, NF- $\kappa$ B has been shown to facilitate autophagy in PAH in preclinical models.<sup>53</sup>

Bardoxolone methyl is an oral Nrf2 transcription factor activator that targets multiple cell types involved in the pathogenesis of PAH, including SMCs, endothelial cells, and macrophages.<sup>52</sup> Bardoxolone also increases endothelial NO bioavailability and reduces vascular remodeling.<sup>52</sup> The phase 2 study LARIAT (Bardoxolone Methyl Evaluation in Patients With Pulmonary Hypertension)<sup>52</sup> showed

improvement in placebo-corrected 6MWD at 16 weeks in PAH patients treated with bardoxolone in addition to background therapy.<sup>54</sup> However, the phase 3 RANGER (Extended Access Program to Assess Long-term Safety of Bardoxolone Methyl in Patients With Pulmonary Hypertension RANGER)<sup>55</sup> and CATALYST trials (Bardoxolone Methyl in Patients With Connective Tissue Disease-associated Pulmonary Arterial Hypertension)<sup>56</sup> were prematurely halted over concerns about COVID-19 exposure in this vulnerable patient population; this was further compounded by interim data suggesting that the study was “unlikely” to meet its primary efficacy end point.

Insulin resistance may be a risk factor for patients with PAH.<sup>57</sup> Experimental studies of metformin, which improves insulin sensitivity, have shown that it enhances pulmonary vasodilation by increasing endothelial NO synthase, modulating ROS and NF- $\kappa$ B levels,<sup>51,53</sup> and inhibiting mitogen-activated protein kinase (MAPK), thereby reducing pulmonary SMC proliferation and remodeling.<sup>58,59</sup> Given these beneficial effects, metformin is currently being investigated as an adjunctive treatment modality for PAH.<sup>60</sup>

### *Enhanced Vasoactive Intestinal Peptide Signaling*

Vasoactive intestinal peptide (VIP) is a neurohormone that causes vasodilation and prevents vascular remodeling; however, it has a very short half-life.<sup>61</sup> VIP deficiency has been implicated in the development of PH in experimental models.<sup>62</sup> Pemzivipadil (PB1046), a VIP analogue that is administered once weekly by a subcutaneous injection, is currently being investigated in a phase 2 trial in PAH patients.<sup>63</sup>

### *Modulated Estrogen Signaling*

The “estrogen paradox” refers to the ambiguity between clinical studies that showed a pathogenic role of estrogen in PAH and preclinical studies that described a protective effect.<sup>64</sup> 17 $\beta$ -estradiol (E2) has been shown to cause pulmonary vascular injury leading to the development of PAH; however, it also augments RV function in established PAH.<sup>64</sup> Additionally, E2 metabolites can exert contrasting effects: 16 $\alpha$ -hydroxyoestrone (16 $\alpha$ OHE) has inflammatory and proliferative properties whereas 2-hydroxyoestradiol (2-OHE2) and 2-methoxyoestradiol (2-ME2) possess anti-inflammatory and antiproliferative properties.<sup>4</sup> Therefore, disruptions in the balance of these E2 metabolites may have a relevant role in PAH pathogenesis, in part providing a potential explanation for the inconsistencies noted between clinical and preclinical studies.

Tamoxifen is a selective estrogen receptor blocker currently being investigated in a phase 2 trial of PAH.<sup>65</sup>

Anastrozole is an aromatase inhibitor that works by inhibiting androgen conversion to estrogen.<sup>66</sup> A small phase 2 trial in patients with PAH demonstrated that anastrozole improved 6MWD compared with placebo.<sup>66</sup> A larger phase 2 trial is currently underway.<sup>67</sup> Dehydroepiandrosterone (DHEA) is an estrogen and testosterone precursor that has been shown in preclinical models to prevent and reverse PAH.<sup>68</sup> The EDIPHY trial (Effects of DHEA in Pulmonary Hypertension) is a phase 2 crossover trial investigating the effects of DHEA on RV longitudinal strain.<sup>69</sup>

### *Down-Regulated Mammalian Target of Rapamycin Pathway*

In PAH, the mammalian target of rapamycin (mTOR) pathway is upregulated, and animal models have shown that mTOR inhibitors, such as sirolimus, can prevent and reverse SMC proliferation.<sup>70</sup> ABI-009, an albumin-bound sirolimus nanoparticle, showed better penetration into lung tissue and is currently being investigated in a phase 1 trial in patients with severe PAH.<sup>71</sup>

## CONCLUSION

Despite great progress during the last 3 decades, PAH continues to be a progressive disease with high morbidity and mortality. Ongoing research efforts have led to promising new translational targets that may change the way we treat the disease in the near future. Modifying agents for PAH are needed to improve outcomes and offer hope for a cure.

## KEY POINTS

- Currently approved medications for pulmonary arterial hypertension (PAH) mainly act on three traditional pathways: the nitric oxide, endothelin, and prostacyclin pathways. These medications have greatly improved survival, and studies have shown superiority of combination therapy regimens over monotherapy.
- Several new pathways are currently being studied to develop novel medications as well as repurpose well-known medications, with the goal of modifying the course of PAH disease to further improve outcomes.
- Novel pathways currently under investigation include the transforming growth factor- $\beta$ /bone morphogenetic protein receptor 2 signaling, platelet-derived growth factor signaling, vasoactive intestinal peptide signaling, estrogen signaling, nuclear factor erythroid 2-related factor 2 transcription factor, metabolic function, and the mammalian target of rapamycin pathways.

*Corresponding Author:*

tonella@ccf.org

*Conflict of Interest Disclosure:*

The authors have completed and submitted the *Methodist DeBakey Cardiovascular Journal* Conflict of Interest Statement and none were reported.

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