



Riociguat: Mode of Action and Clinical Development in Pulmonary Hypertension

Hossein-Ardeschir Ghofrani, MD; Marc Humbert, MD; David Langleben, MD; Ralph Schermuly, MD; Johannes-Peter Stasch, PhD; Martin R. Wilkins, MD; and James R. Klingler, MD

Pulmonary arterial hypertension (PAH) and chronic thromboembolic pulmonary hypertension (CTEPH) are progressive and debilitating diseases characterized by gradual obstruction of the pulmonary vasculature, leading to elevated pulmonary artery pressure (PAP) and increased pulmonary vascular resistance (PVR). If untreated, they can result in death due to right-sided heart failure. Riociguat is a novel soluble guanylate cyclase (sGC) stimulator that is approved for the treatment of PAH and CTEPH. We describe in detail the role of the nitric oxide-sGC-cyclic guanosine monophosphate (cGMP) signaling pathway in the pathogenesis of PAH and CTEPH and the mode of action of riociguat. We also review the preclinical data associated with the development of riociguat, along with the efficacy and safety data of riociguat from initial clinical trials and pivotal phase III randomized clinical trials in PAH and CTEPH. CHEST 2017; 151(2):468-480

KEY WORDS: chronic thromboembolic pulmonary hypertension; pulmonary arterial hypertension; sGC stimulator

According to the current World Health Organization (WHO) classification, five categories of pulmonary hypertension (PH) exist: pulmonary arterial hypertension

(PAH; group 1), PH resulting from left-sided heart disease (group 2), PH due to hypoxia or disorders of the respiratory system, or both (group 3), chronic thromboembolic

ABBREVIATIONS: 6MWD = 6-minute walking distance; ADMA = asymmetric dimethylarginine; AE = adverse event; BH4 = tetrahydrobiopterin; BMPR-2 = bone morphogenetic protein receptor-2; cGMP = cyclic guanosine monophosphate; CHEST-1 and -2 = Chronic Thromboembolic Pulmonary Hypertension sGC-Stimulator Trial-1 and -2; CO = cardiac output; CTEPH = chronic thromboembolic pulmonary hypertension; eNOS = endothelial nitric oxide synthase; EQ-5D = EuroQol 5 Dimensions questionnaire; ERA = endothelin receptor antagonist; FC = functional class; HFpEF = heart failure with preserved ejection fraction; HR = heart rate; ILD = interstitial lung disease; LPH = Living with Pulmonary Hypertension questionnaire; LS = least squares; mPAP = mean pulmonary artery pressure; NADPH = nicotinamide adenine dinucleotide phosphate; NO = nitric oxide; NOS = nitric oxide synthase; NT-proBNP = N-terminal pro-hormone of brain natriuretic peptide; PAH = pulmonary arterial hypertension; PAP = pulmonary artery pressure; PATENT-1 and -2 = Pulmonary Arterial Hypertension sGC-Stimulator Trial-1 and -2; PDE5 = phosphodiesterase 5; PEA = pulmonary endarterectomy; PH = pulmonary hypertension; PH-sLVD = PH due to systolic left ventricular dysfunction; PKG = cGMP-dependent protein kinase; PVR = pulmonary vascular resistance; QoL = quality of life; RV = right ventricular; SAE = serious adverse event; sGC = soluble guanylate cyclase; sLVD = systolic left ventricular dysfunction; SvO₂ = mixed venous oxygen saturation; SVR = systemic vascular resistance; WHO = World Health Organization

AFFILIATIONS: From the University of Giessen and Marburg Lung Center (Drs Ghofrani and Schermuly), Giessen, Germany and the

German Center for Lung Research (DZL); Department of Medicine (Drs Ghofrani and Wilkins), Imperial College London, London, England; Assistance Publique-Hôpitaux de Paris (Dr Humbert), Service de Pneumologie, DHU Thorax Innovation, Hôpital Bicêtre and Université Paris-Sud, Laboratoire d'Excellence en Recherche sur le Médicament et Innovation Thérapeutique and INSERM Unité 999, Le Kremlin-Bicêtre, France; Center for Pulmonary Vascular Disease and Lady Davis Institute (Dr Langleben), Jewish General Hospital, McGill University, Montreal, Canada; Bayer Pharma AG (Dr Stasch), Wuppertal and University Halle, Institute of Pharmacy, Halle (Saale), Germany; Division of Pulmonary, Sleep, and Critical Care Medicine (Dr Klingler), Rhode Island Hospital, Alpert Medical School of Brown University, Providence, RI.

FUNDING/SUPPORT: Editorial assistance was provided by Adelphi Communications, Ltd (Bollington, England), supported by Bayer Pharma AG (Berlin, Germany).

CORRESPONDENCE TO: Hossein-Ardeschir Ghofrani, MD, Department of Internal Medicine, Medical Clinic II/V, University Hospital Giessen and Marburg, Klinikstr 33, Giessen 35392, Germany; e-mail: ardeschir.ghofrani@innere.med.uni-giessen.de

Copyright © 2016 The Authors. Published by Elsevier Inc under license from the American College of Chest Physicians. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

DOI: <http://dx.doi.org/10.1016/j.chest.2016.05.024>

pulmonary hypertension (CTEPH; group 4), and PH with unclear/multifactorial mechanisms (group 5) (Table 1).¹⁻⁴ PH is defined as a mean pulmonary artery pressure (PAP) \geq 25 mm Hg. PAH and CTEPH are progressive pulmonary hypertensive diseases that usually lead to extensive pulmonary vascular remodeling, causing a marked increase in pulmonary vascular resistance (PVR) that leads to right ventricular (RV) overload and death when untreated.^{5,6}

Endothelial dysfunction of the pulmonary vasculature plays a key role in the progression of PAH and is characterized by impaired production of vasodilators such as nitric oxide (NO) and prostacyclin, and overexpression of vasoconstrictors such as endothelin-1, leading to elevated vascular tone and promotion of vascular remodeling.⁷ These observations have underpinned three therapeutic target pathways in PAH: the prostacyclin pathway (eg, prostanoids such as epoprostenol, iloprost, and treprostinil), the endothelin pathway (eg, endothelin receptor antagonists [ERAs] such as bosentan, ambrisentan, and macitentan), and the cyclic guanosine monophosphate (cGMP) degradation pathway (eg, phosphodiesterase-5 [PDE5] inhibitors such as sildenafil and tadalafil).^{5,6,8} Despite the availability of numerous treatment options, mortality rates in patients with PAH remain high.⁹⁻¹³

Pulmonary endarterectomy (PEA) is the gold standard treatment for CTEPH, as it is potentially curative, making CTEPH the only form of PH that is curable.^{5,6,14,15} However, 24% to 37% of patients with CTEPH are

ineligible for PEA, and 17% to 35% experience persistent/recurrent PH after PEA.¹⁶⁻²² These patients are candidates for medical therapy.^{5,6} Balloon pulmonary angioplasty may also be considered in patients with CTEPH who are technically inoperable or have an unfavorable risk to benefit ratio for PEA.^{5,6}

Riociguat, a soluble guanylate cyclase (sGC) stimulator, is approved for the treatment of both PAH and inoperable CTEPH or persistent/recurrent CTEPH after PEA.^{5,6} Riociguat adds sGC stimulation as a fourth therapeutic target in PAH.⁸ We review the mode of action of riociguat, its clinical efficacy, and its safety in PAH and CTEPH.

The Role of the NO-sGC-cGMP Signaling Pathway in PH

NO is a key regulator of flow-induced vasodilatation in the lung (Fig 1).²³⁻²⁶ In the healthy lung, NO is produced by the vascular endothelium, airway, and alveolar epithelial cells and diffuses to the smooth muscle layer, where it binds to a prosthetic heme group on sGC.^{23,24} On binding with NO, sGC catalyzes the synthesis of the secondary messenger cGMP from guanosine triphosphate,²⁷ leading to activation of cGMP-dependent protein kinase (PKG), which acts through a variety of mechanisms to reduce intracellular calcium concentrations and inhibit smooth muscle cell contraction.

A reduction in endogenous NO levels has been observed in patients with PAH, CTEPH, and PH associated with COPD and interstitial lung disease (ILD).²³ Levels of

TABLE 1] Classification of Pulmonary Hypertension¹

PH Classification Group	Features
1: Pulmonary arterial hypertension	Remodeling of the pulmonary vasculature leading to thickened artery walls, increased PVR, RV overload, and right-sided heart dysfunction ² Can be idiopathic, heritable, induced by certain drugs/toxins or associated with other diseases such as connective tissue disease, HIV, portal hypertension, congenital heart disease, and schistosomiasis
2: PH due to left-sided heart disease	The most common cause of PH ³ Caused by LV systolic or diastolic dysfunction, valvular disease, or congenital heart conditions
3: PH due to lung diseases or hypoxia, or both	PH occurs commonly in COPD and ILD PH is also associated with sleep-disordered breathing, alveolar hypoventilation disorders, chronic exposure to high altitude, and developmental lung diseases
4: Chronic thromboembolic pulmonary hypertension	Characterized by the presence of organized thrombi in the pulmonary vasculature and remodeling of unaffected vessels ⁴ Can be cured surgically by pulmonary endarterectomy ¹⁵
5: PH with unclear multifactorial mechanisms	Includes hematologic disorders such as sickle cell disease, myeloproliferative disorders, metabolic disorders, and other conditions such as renal failure

ILD = interstitial lung disease; LV = left ventricular; PH = pulmonary hypertension; PVR = pulmonary vascular resistance; RV = right ventricular.

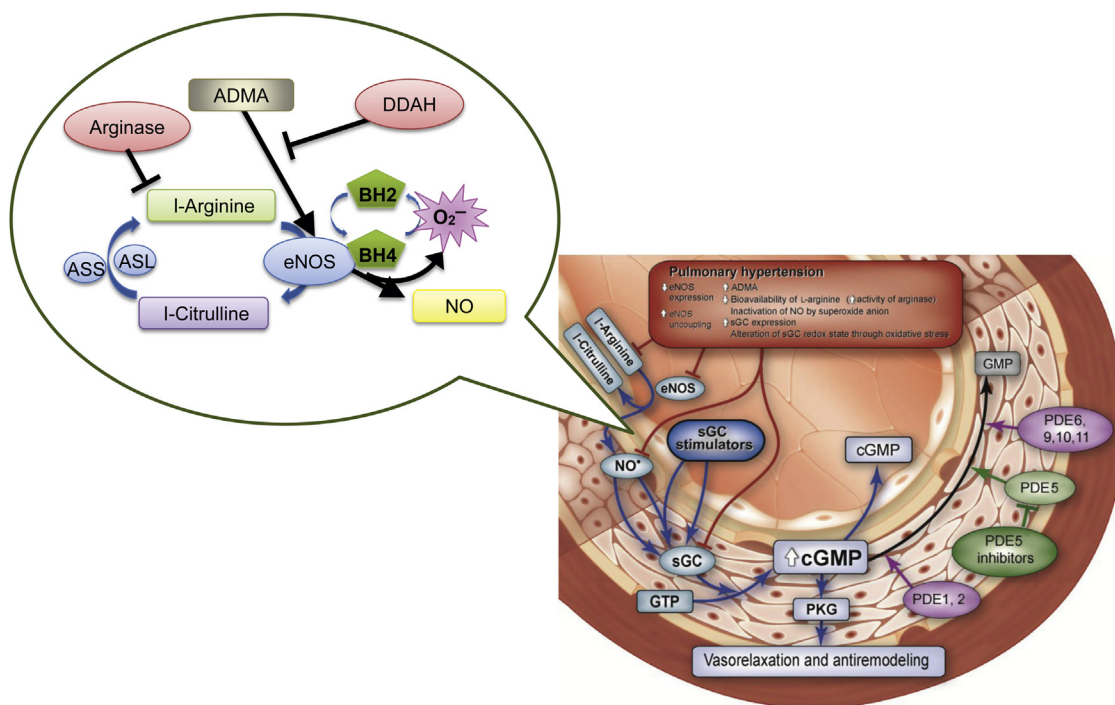


Figure 1 – The NO-sGC-cGMP signaling pathway. The primary route of NO synthesis is the oxidation of the amino group of L-arginine, a multiple-step chemical reaction that is catalyzed by a family of enzymes known as NO synthases (NOSs). NOSs facilitate the oxidation of the L-arginine amino group with the assistance of several cofactors, notably BH4 and NADPH. NO synthesis can be impaired through decreased availability of L-arginine, NOS or NOS cofactors, or increased availability of NOS inhibitors such as ADMA. NO synthesis can also be impaired by eNOS overactivity, as depletion of cofactors results in incomplete oxidation of the L-arginine amino group, leading to uncoupling of NO synthesis and the generation of reactive oxygen species (see text for details). ADMA = asymmetric dimethylarginine; ASL = argininosuccinate lysase; ASS = argininosuccinate synthase; BH2 = dihydrobiopterin; BH4 = tetrahydrobiopterin; cGMP = cyclic guanosine monophosphate; DDAH = dimethylarginine dimethylaminohydrolase; eNOS = endothelial nitric oxide synthase; GMP = guanosine monophosphate; GTP = guanosine triphosphate; NADPH = nicotinamide adenine dinucleotide phosphate; NO = nitric oxide; O_2^- = superoxide anion; PDE = phosphodiesterase; PKG = cGMP-dependent protein kinase; sGC = soluble guanylate cyclase. Adapted with permission from Stasch JP, Pacher P, Evgenov OV. Soluble guanylate cyclase as an emerging therapeutic target in cardiopulmonary disease. *Circulation*. 2011;123(20):2263-2273.²⁶ Promotional and commercial use of the material in print, digital or mobile device format is prohibited without permission from the publisher Wolters Kluwer Health. Please contact healthpermissions@wolterskluwer.com for further information.

exhaled NO have also been shown to be reduced in patients with idiopathic PAH and PAH associated with scleroderma.^{28,29} NO levels are also reduced in conditions of oxidative stress,³⁰ biomarkers of which are increased in the lungs of patients with idiopathic PAH, PH associated with left-sided heart disease, and hypoxia-induced PH.³¹⁻³³ The primary route of NO synthesis is the oxidation of the amino group of L-arginine, a multiple-step chemical reaction that is catalyzed by NO synthase (NOS) with the assistance of several cofactors, such as tetrahydrobiopterin (BH4) and nicotinamide adenine dinucleotide phosphate (NADPH) (Fig 1). The NOS family consists of endothelial and neuronal NOSs that are constitutively expressed, and inducible NOS that is capable of being stimulated by a variety of environmental signals. NO synthesis can be impaired by decreased availability of L-arginine, NOS, or NOS cofactors. All three abnormalities have been reported in PAH and CTEPH and are associated with disease progression and poor outcomes.^{23,34-36} Decreased

L-arginine levels can occur from increased activity of arginase, an enzyme that metabolizes L-arginine to ornithine. Alternatively, elevated levels of asymmetric dimethylarginine (ADMA), which competes with L-arginine for NOS binding sites but does not activate the enzyme, can impair NO synthesis.³⁷

There is also evidence that endothelial NOS (eNOS) expression and activity are dysregulated in PAH. Studies have shown both decreased eNOS expression in small pulmonary arteries and increased eNOS expression in the plexiform lesions of patients with PAH.^{38,39} Mice with genetically engineered suppression of caveolin, an endothelial membrane protein that binds eNOS and suppresses its activity, have increased eNOS activity, and PH develops.⁴⁰ This seemingly paradoxical finding may be explained by the observation that NO synthesis may be impaired by eNOS overactivity. This occurs because of depletion of the cofactors (eg, BH4) that act as electron donors and are necessary for complete

oxidation of the L-arginine amino group. In the absence of adequate cofactors, or in the presence of substrate deficiency, NO synthesis by eNOS becomes uncoupled, leading to decreased synthesis of NO and increased production of reactive oxygen species, which contribute to vascular pathology.²⁵

Changes in NO-sGC-cGMP signaling have been implicated in the downstream effects of endothelial dysfunction on the pulmonary vasculature, including pulmonary artery smooth muscle cell proliferation, vasoconstriction, platelet aggregation and fibrosis, leukocyte recruitment, inflammation, maladaptive cardiac hypertrophy, and pulmonary vascular remodeling.^{23-25,41,42}

There is increasing interest in the role of genetic factors in PH. Mutations in genes coding for bone morphogenetic protein receptor-2 (BMPR-2) are the main cause of heritable PAH.⁴³ cGMP-activated PKG is a component of the NO signaling pathway downstream of cGMP and plays an important role in BMPR-2 signaling. Mice with low or absent PKG isotype I exhibit impaired BMP signaling, with decreased contractile gene expression and abnormal vascular remodeling.⁴⁴ A mutation in the gene encoding caveolin-1 has also been associated with an increased risk of PAH developing.⁴⁵

Targeting sGC in PH: Mode of Action of Riociguat

Previous attempts to target the NO-sGC-cGMP pathway have focused on preventing cGMP degradation by inhibiting PDE5, one of the primary enzymes responsible for cGMP degradation in the lung (Fig 1). The PDE5 inhibitors sildenafil and tadalafil have been shown to improve 6-minute walking distance (6MWD), WHO functional class (WHO FC), and pulmonary hemodynamics in treatment-naïve patients with PAH.^{5,6,46,47} Sildenafil has also been shown to increase 6MWD, reduce PVR, and delay time to clinical worsening in patients who are already being treated with intravenous epoprostenol.⁴⁸ Similarly, tadalafil has been shown to increase functional capacity and delay clinical worsening when added to background ERA therapy.⁴⁹

Registry and long-term extension study data have shown that a proportion of patients with PAH may not respond to PDE5 inhibitors alone.^{11,50-52} It is possible that in some of these patients, lack of response is due in part to insufficient pulmonary cGMP production. In mice with decreased cGMP synthesis due to disrupted expression of natriuretic peptide receptor A, sildenafil

was ineffective at preventing hypoxic PH.⁵³ Similarly, decreased NO synthesis may restrict the effectiveness of PDE5 inhibitors by reducing the pulmonary vascular cGMP levels.^{23,26,54,55} Results from the Ambrisentan and Tadalafil in Patients with Pulmonary Arterial Hypertension (AMBITION) trial and Study With an Endothelin Receptor Antagonist in Pulmonary Arterial Hypertension to Improve Clinical Outcome (SERAPHIN) trials have suggested that patients receiving PDE5 inhibitor monotherapy may demonstrate insufficient response to treatment compared with patients receiving combination therapy, suggesting that further optimization of the NO-sGC-cGMP pathway may be necessary.^{56,57}

To this end, recent efforts have centered on identifying pharmacologic agents that enhance sGC-induced cGMP synthesis directly.^{50,58-60} sGC stimulators such as riociguat have a dual mode of action that enhances sGC response to endogenous NO and directly stimulates sGC independent of NO binding. In vitro studies have shown that riociguat increases the activity of sGC by up to 73-fold and acts in synergy with NO to increase sGC activity up to 112-fold.⁵⁴ Thus, these agents circumvent the need for endogenous NO production for their mode of action, increasing cGMP levels through a mechanism that is different from that of PDE5 inhibitors (Fig 2).^{23,26,54,61}

Riociguat: Preclinical Data

The vasodilatory effects of riociguat have been demonstrated in several preclinical models of hypertension. Riociguat relaxed isolated saphenous artery rings from normal and nitrate-resistant rabbits and also normalized blood pressure and improved survival in high- and low-renin rat models of hypertension.⁶² Moreover, in a Dahl salt-sensitive rat model of malignant hypertension, riociguat attenuated systemic hypertension, systolic dysfunction, fibrotic tissue remodeling, and degenerative changes in myocardium and renal cortex.⁶³

The beneficial effects on other pathologic processes associated with PH have also been demonstrated in several preclinical studies. Administration of riociguat partially reversed PH, RV hypertrophy, and remodeling of lung vascular tissue in hypoxic mouse and monocrotaline-injected rat models.⁵⁴ Similarly, riociguat attenuated bleomycin-induced PH and RV hypertrophy in mice, and to a greater extent than sildenafil. Additionally, amelioration of pulmonary inflammation and fibrosis was observed with riociguat treatment in the same mouse

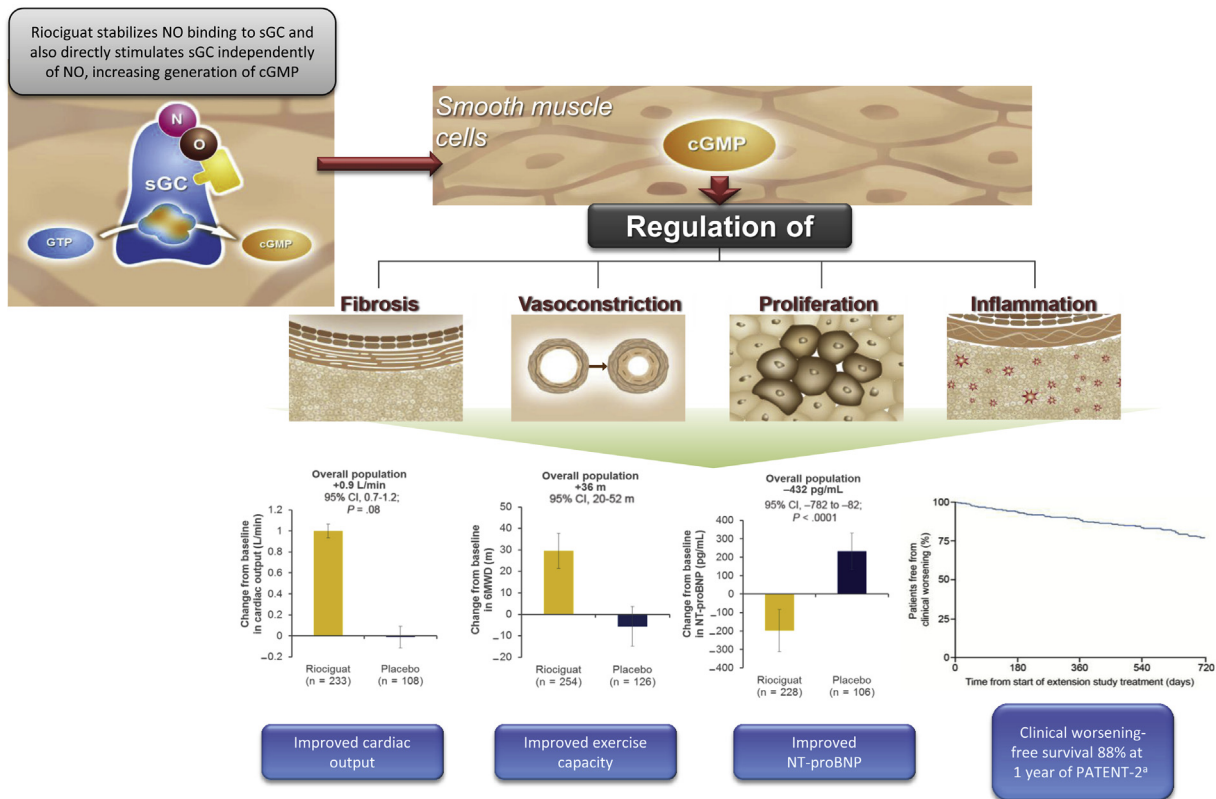


Figure 2 – Summary of the mode of action and effects of riociguat. NT-proBNP = N-terminal pro-brain natriuretic peptide. Data are from Pulmonary Arterial Hypertension sGC-Stimulator Trial (PATENT)-1 and -2. See Figure 1 legend for expansion of other abbreviations. ^aReproduced with permission from Rubin LJ, Galie N, Grimminger F, et al. Riociguat for the treatment of pulmonary arterial hypertension: a long-term extension study (PATENT-2). *Eur Respir J*. 2015;45(5):1303-1313.⁶¹

model.⁶⁴ Furthermore, riociguat has been shown to prevent the development of PH, RV hypertrophy, and vascular remodeling compared with control subjects in a cigarette-smoke-induced mouse model of COPD and PH.⁶⁵ In a rat model of angioproliferative PAH induced by hypoxia and the vascular endothelial growth factor receptor antagonist SU5416, riociguat treatment increased the open to occluded artery ratio, decreased the neointima to media ratio, reduced RV systolic pressure, and increased cardiac output (CO). Moreover, there was a greater decrease in RV hypertrophy and an increase in RV function than observed following treatment with sildenafil.⁶⁶ The antifibrotic effects of riociguat have also been demonstrated in three separate mouse models of fibrosis. Riociguat reduced skin thickening, myofibroblast differentiation, and collagen accumulation in the tight skin (TSK-1) and bleomycin-induced models of skin fibrosis in a dose-dependent manner, and in a sclerodermatous chronic graft-vs-host disease model of systemic sclerosis, riociguat ameliorated GI tract fibrosis to a greater extent than sildenafil.⁶⁷ The preclinical results observed with riociguat led to it becoming the first sGC stimulator to enter clinical development.⁵⁹

Clinical Efficacy of Riociguat in PAH and CTEPH

Riociguat has been evaluated in a number of phase II studies and two phase III studies that enrolled patients with PAH (Pulmonary Arterial Hypertension sGC-Stimulator Trial [PATENT-1]) and inoperable CTEPH or persistent/recurrent CTEPH after PEA (Chronic Thromboembolic Pulmonary Hypertension sGC-Stimulator Trial-1 [CHEST-1]) (Table 2).^{61,68-78}

Phase II Studies of Riociguat

In a single-dose, proof-of-concept study in 19 patients with PH (including patients with PAH, CTEPH, and PH with mild to moderate ILD), riociguat reduced mean PAP (mPAP) and PVR, and increased cardiac index. Riociguat had no effect on gas exchange or ventilation-perfusion matching and was well tolerated.⁶⁸ In an open-label, 12-week, phase II study that included 33 patients with PAH and 42 patients with CTEPH, riociguat significantly improved 6MWD, cardiac index, mPAP, PVR, and WHO FC at week 12 compared with baseline.⁶⁹ The results from the long-term extension to this study show that long-term riociguat treatment

TABLE 2] Overview of Main Clinical Studies with Riociguat

Study Design	Patients	Riociguat Dose	Key Results	Reference
Phase II proof-of-concept study	PAH (n = 12) CTEPH (n = 6) PH-ILD (n = 1)	Single 2.5-mg or 1-mg dose	Significantly reduced mPAP and PVR and increased cardiac index No effect on gas exchange or ventilation-perfusion matching	Grimminger et al, 2009 ⁶⁸
Multicenter, open-label, uncontrolled, phase II study	PAH (n = 33) and CTEPH (n = 42)	1.0-2.5 mg tid for 12 wk	Improved median 6MWD (PAH: +57 m; CTEPH: +55 m; $P < .0001$) Increased cardiac index (0.43 mm Hg; $P < .0001$), and reduced mPAP (-4.5 mm Hg; $P < .0001$), PVR (-215 dyn·s·cm ⁻⁵ ; $P < .0001$), and SVR (-441 dyn·s·cm ⁻⁵ ; $P < .0001$) compared with baseline	Ghofrani et al, 2010 ⁶⁹ (NCT00454558)
Multicenter, open-label, uncontrolled, phase II, long-term extension study	PAH (n = 27) and CTEPH (n = 41)	1.0-2.5 mg tid	Long-term treatment was well tolerated and had a good safety profile Sustained improvements in 6MWD for up to a further 45 mo (PAH: +80 m; CTEPH: +47 m, respectively)	Hoeper et al, 2015 ⁷⁰ (NCT00454558)
Randomized, double-blind, placebo-controlled, phase II, interaction study (PATENT PLUS)	Patients with PAH receiving sildenafil (n = 18)	1.0-2.5 mg tid for 12 wk	No effect of sildenafil plus riociguat on changes in BP vs sildenafil plus placebo No evidence of a favorable clinical effect with combination therapy High rate of discontinuation due to hypotension in long-term extension	Galiè et al, 2015 ⁷¹ (NCT01179334)
Randomized, double-blind, placebo-controlled, parallel-group, multicenter, phase IIb study (LEPHT)	PH-sLVD (n = 201)	0.5-2.0 mg tid for 16 wk	No significant change in mPAP (primary end point) Cardiac index (+0.4 L·min ⁻¹ ·m ⁻² ; $P = .0001$) and SVI (+5.2 mL·m ⁻² ; $P = .0018$) were significantly increased with riociguat 2 mg PVR (-46.6 dyn·s·cm ⁻⁵ ; $P = .03$) and SVR (-293.3 dyn·s·cm ⁻⁵ ; $P = .0002$) were significantly decreased with riociguat 2 mg No significant changes in HR or systolic BP MLHF score was significantly improved ($P = .0002$)	Bonderman et al, 2013 ⁷² (NCT01065454)
Randomized, double-blind, placebo-controlled, single-dose, phase IIa study (DILATE)	PH associated with HFpEF (n = 39)	Single 0.5-2.0 mg dose	No significant change in peak decrease in mPAP (primary end point) Riociguat 2 mg significantly increased SV (+9 mL; $P = .04$) and cardiac index (+0.4 L/min/m ² ; $P = .001$) and decreased SVR (-247 dyn·s·cm ⁻⁵ ; $P < .05$) and systolic BP (-12 mm Hg; $P = .03$) No significant change in HR, PCWP, TPG, or PVR Riociguat significantly decreased RVED area (-5.6 cm ² ; $P = .04$)	Bonderman et al, 2014 ⁷³ (NCT01172756)
Phase II proof-of-concept study	PH-COPD (n = 23)	Single 2.5-mg or 1-mg dose	Decreased mPAP (1.0 mg: -3.6 mm Hg; 2.5 mg: -4.8 mm Hg), PVR (1.0 mg: -58.3 dyn·s·cm ⁻⁵ ;	Ghofrani et al, 2015 ⁷⁴ (NCT00640315)

(Continued)

TABLE 2] (Continued)

Study Design	Patients	Riociguat Dose	Key Results	Reference
Multicenter, open-label, uncontrolled, phase II study	PH-ILD (n = 22)	1.0-2.5 mg tid for 12 wk plus 12-mo extension	<p>2.5 mg: $-123.8 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$, SVR (1.0 mg: $-440 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$; 2.5 mg: $-468 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$), systolic BP (1.0 mg: -26.3 mm Hg; 2.5 mg: $-22.2 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$), and diastolic BP (1.0 mg: -13.5 mm Hg; 2.5 mg: $-11.3 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$)</p> <p>Slight improvements in lung function No change in gas exchange</p> <p>CO (+1.2 L/min) and SvO₂ (+2%) increased, PVR ($-120 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$) and SVR decreased ($-821 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$), and mPAP remained unchanged compared with baseline</p> <p>6MWD increased by 25 m after 12 wk; further improvements during follow-up</p>	Hoeper et al, 2012 ⁷⁵ (NCT00694850)
Multicenter, double-blind, randomized, placebo-controlled, phase III study (CHEST-1)	CTEPH (n = 261)	1.0-2.5 mg tid for 16 wk	<p>Significant improvements in 6MWD vs placebo at wk 16 (+46 m; $P < .001$)</p> <p>Significant improvements in PVR ($-246 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$; $P < .001$), CO (+0.9 L/min; $P < .001$), and mPAP (-5 mm Hg; $P < .001$)</p> <p>Significant improvements in NT-proBNP ($P < .001$) and WHO FC ($P = .003$)</p> <p>Nominally significant^a improvements in Borg dyspnea score and QoL</p>	Ghofrani et al, 2013 ⁷⁶ (NCT00855465)
Multicenter, double-blind, randomized, placebo-controlled, phase III study (CHEST-2)	CTEPH (n = 237)	1.0-2.5 mg tid	<p>Long-term treatment was well tolerated and had a good safety profile</p> <p>Improvements in 6MWD sustained for 1 y (+51 m; n = 172)</p> <p>Improvements in WHO FC also sustained for 1 y</p>	Simonneau et al, 2015 ⁷⁷ (NCT00910429)
Multicenter, double-blind, randomized, placebo-controlled, phase III study (PATENT-1)	PAH (n = 443)	1.0-2.5 mg tid for 12 wk	<p>Significant improvements in 6MWD vs placebo at wk 12 (+36 m; $P < .001$)</p> <p>Significant improvements in PVR ($-226 \text{ dyn}\cdot\text{s}\cdot\text{cm}^{-5}$; $P < .001$), CO (+0.9 L/min; $P < .001$), and mPAP (-4 mm Hg; $P < .001$)</p> <p>Significant improvements in NT-proBNP ($P < .0001$), WHO FC ($P = .003$), time to clinical worsening ($P = .005$), and Borg dyspnea score ($P = .002$)</p> <p>Nominally significant^a improvements in QoL</p>	Ghofrani et al, 2013 ⁷⁸ (NCT00810693)

(Continued)

TABLE 2] (Continued)

Study Design	Patients	Riociguat Dose	Key Results	Reference
Multicenter, double-blind, randomized, placebo-controlled, phase III study (PATENT-2)	PAH (n = 396)	1.0-2.5 mg tid	Long-term treatment was well tolerated and had a good safety profile Improvements in 6MWD sustained for 1 y (+51 m; n = 327) Improvements in WHO FC also sustained for 1 y	Rubin et al, 2015 ⁶¹ (NCT00863681)

6MWD = 6-min walking distance; CHEST = Chronic Thromboembolic Pulmonary Hypertension sGC-Stimulator Trial; CO = cardiac output; CTEPH = chronic thromboembolic pulmonary hypertension; DILATE = Acute Hemodynamic Effects of Riociguat in Patients With Pulmonary Hypertension Associated With Diastolic Heart Failure; HFpEF = heart failure with preserved ejection fraction; HR = heart rate; LEPHT = A Study to Test the Effects of Riociguat in Patients With Pulmonary Hypertension Associated With Left Ventricular Systolic Dysfunction; MLHF = Minnesota Living with Heart Failure survey; mPAP = mean pulmonary artery pressure; NT-proBNP = N-terminal prohormone of brain natriuretic peptide; PAH = pulmonary arterial hypertension; PATENT = Pulmonary Arterial Hypertension sGC-Stimulator Trial; PCWP = pulmonary capillary wedge pressure; QoL = quality of life; RVED = right ventricular end-diastolic; sLVD = systolic left ventricular dysfunction; SV = stroke volume; SVI = systemic vascular index; SvO₂ = mixed venous oxygen saturation; SVR = systemic vascular resistance; TPG = transpulmonary pressure gradient; WHO FC = World Health Organization functional class. See Table 1 legend for expansion of other abbreviations.

^aBecause of the hierarchical testing procedure in CHEST-1 and PATENT-1.^{76,78}

(median treatment duration, 77 months) was well tolerated, and the improvements in 6MWD and WHO FC were sustained for at least 48 months in those patients who continued on riociguat therapy.⁷⁰

Pulmonary Arterial Hypertension sGC-Stimulator Trial-1 Study

The phase III, randomized, placebo-controlled Pulmonary Arterial Hypertension sGC-Stimulator Trial-1 (PATENT-1) study investigated the safety and efficacy of 12 weeks of riociguat therapy (up to 2.5 mg tid) in 443 patients with symptomatic PAH. The primary end point of PATENT-1, a change in 6MWD at week 12, was significantly greater in the riociguat group receiving a maximum of 2.5 mg compared with the placebo group (least squares [LS] mean difference +36 m; 95% CI, 20-52; $P < .001$).⁷⁸ Riociguat also improved a range of clinically relevant secondary end points, including PVR ($-226 \text{ dyn} \cdot \text{s} \cdot \text{cm}^{-5}$; 95% CI, 20-52; $P < .001$), N-terminal prohormone of brain natriuretic peptide (NT-proBNP) (-432 pg/mL ; 95% CI, -782 to -82 ; $P < .001$), WHO FC ($P = .003$), time to clinical worsening ($P = .005$), and Borg dyspnea score ($P = .002$). Several other hemodynamic parameters including CO ($+0.9 \text{ L/min}$; 95% CI, 0.7-1.2; $P < .001$), mPAP (-5 mm Hg ; 95% CI, -6 to -2 ; $P < .001$), and mixed venous oxygen saturation (SvO₂) ($+5\%$; 95% CI, 3-7; $P < .001$) were also improved with riociguat vs placebo in PATENT-1.⁷⁸ There was also evidence that the beneficial effects of riociguat were reflected in patient quality of life (QoL). Measures of health-related QoL were improved in PATENT-1 (EuroQol 5 Dimensions [EQ-5D] questionnaire score, $+0.06$; 95% CI, 0.01-0.11; $P = .07$ [minimally important difference, 0.074]⁷⁹; Living with Pulmonary

Hypertension questionnaire [LPH] score, -6 ; 95% CI, -10 to -3 ; $P = .02$ [minimally important difference, 3 points for subscales, 7 points for total score]⁸⁰), with the change in LPH score reaching only nominal significance considering the hierarchical testing procedure.^{76,78} The effects of riociguat in PATENT-1 were similar in treatment-naïve patients and in patients receiving riociguat added to a background therapy of ERAs or non-intravenous prostanoids.⁷⁸

In the PATENT-2 long-term extension study,⁶¹ after 12 weeks, mean 6MWD \pm SD had changed by $+52 \pm 61 \text{ m}$ in the former group receiving a maximum of 2.5 mg riociguat ($n = 218$) and by $+52 \pm 67 \text{ m}$ in the former placebo group following transition to riociguat ($n = 103$). At the same 12-week time point in PATENT-2, patients formerly receiving placebo showed a decrease in NT-proBNP levels similar to those of patients formerly receiving a maximum of 2.5 mg riociguat.

In PATENT-2, improvements in 6MWD, NT-proBNP, WHO FC, Borg dyspnea score, and QoL seen during PATENT-1 were sustained for at least 2 years; at 2 years (mean treatment duration 135 weeks) in the overall population, mean 6MWD \pm SD had increased from PATENT-1 baseline by $+47 \pm 85 \text{ m}$ ($n = 296$), and WHO FC had improved/stabilized/worsened compared with baseline in 33%/58%/9% ($n = 306$).^{61,81} The estimated survival was 93% at 2 years.^{61,81}

Chronic Thromboembolic Pulmonary Hypertension sGC-Stimulator Trials

CHEST-1 was a phase III study of riociguat in 261 patients with inoperable CTEPH or persistent/recurrent CTEPH after PEA.⁷⁶ After 16 weeks' treatment with riociguat

(up to 2.5 mg tid), the primary end point of 6MWD increased by 46 m vs placebo (LS mean difference, 95% CI, 25-67; $P < .001$). Riociguat-treated patients also showed improvements across a range of clinically relevant secondary end points, including PVR ($-246 \text{ dyn} \cdot \text{s} \cdot \text{cm}^{-5}$; 95% CI, -303 to -190 ; $P < .001$), NT-proBNP (-444 pg/mL ; 95% CI, -843 to -45 ; $P < .001$), and WHO FC ($P = .03$). The effects of riociguat were consistent, although somewhat stronger, in patients with inoperable CTEPH compared with those with persistent/recurrent CTEPH after PEA.⁷⁶ Riociguat is the first pharmacologic therapy that has been shown to significantly improve both 6MWD and PVR in patients with inoperable CTEPH and in patients with recurrent/persistent CTEPH after PEA in a large randomized controlled trial.

In addition to PVR, other hemodynamic effects of riociguat in the CHEST-1 study were comparable with its effects in the PATENT-1 study; CO ($+0.9 \text{ L/min}$; 95% CI, 0.6-1.1; $P < .001$), mPAP (-5 mm Hg ; 95% CI, -7 to -3 ; $P < .001$), and SvO₂ ($+4\%$; 95% CI, 1-6; $P = .001$) were all improved without significant changes in heart rate.^{76,78} Nominally significant improvements in health-related QoL were also evident in the CHEST-1 study (EQ-5D score, $+0.13$; 95% CI, 0.06-0.21; $P < .001$), suggesting that the improvements in exercise capacity, hemodynamics, NT-proBNP levels, and WHO FC were translated into patient QoL benefits.

In the CHEST-2 long-term extension study,⁷⁷ mean 6MWD \pm SD had increased by $+61 \pm 59 \text{ m}$ after 12 weeks in the former riociguat group ($n = 145$) and by $+51 \pm 64 \text{ m}$ in the former placebo group following transition to riociguat ($n = 75$). Patients formerly receiving placebo showed a decrease in NT-proBNP levels similar to that in patients formerly receiving riociguat at the same time point.

In CHEST-2, improvements in 6MWD and WHO FC seen during CHEST-1 were sustained for at least 2 years; at 2 years (mean treatment duration, 127 weeks) in the overall population 6MWD \pm SD had increased from CHEST-1 baseline by $+50 \pm 68 \text{ m}$ ($n = 162$) and WHO FC was improved/stabilized/worsened in 39%/58%/3% of the overall population ($n = 170$).^{77,82} Estimated survival was 93% at 2 years.⁸²

Clinical Safety of Riociguat in PAH and CTEPH

Overall, riociguat was well tolerated in the clinical studies.^{68,69,72,75,77,78} The most common adverse events (AEs) were headache, dizziness, indigestion, peripheral edema, nausea, diarrhea, and vomiting.

In the 12-week PATENT-1 study, eight patients (3%) receiving riociguat in doses up to 2.5 mg tid withdrew because of AEs, whereas 33 of the patients (8%) who entered PATENT-2 withdrew because of AEs by the March 2013 cutoff (mean treatment duration, 95 weeks).^{78,81} In CHEST-1, four patients (2%) receiving riociguat withdrew because of AEs, whereas seven of the 237 patients (3%) who entered CHEST-2 had withdrawn because of AEs at the March 2013 cutoff (mean treatment duration, 83 weeks).^{76,77}

There is an increased likelihood of bleeding in patients with PH, particularly among patients receiving anticoagulation therapy.⁸³ The total rate of bleeding was not increased with riociguat vs placebo in the CHEST-1 and PATENT-1 studies.^{76,78} In the randomized riociguat studies, respiratory tract bleeding was reported in 2% of patients treated with riociguat and in 1% of patients treated with placebo. Hemoptysis (reported as a serious adverse event [SAE]) was observed in three riociguat-treated patients (2%) in CHEST-1 and in two riociguat-treated patients (1%) in PATENT-1. In CHEST-2 at 1 year ($n = 237$; mean treatment duration, 83 weeks), a further three patients (1%) experienced significant hemoptysis, and one patient ($< 1\%$) experienced pulmonary hemorrhage.⁷⁷ At the same time point in PATENT-2, hemoptysis and pulmonary hemorrhage were reported as SAEs in seven (2%) and three (1%) patients, respectively (overall population $n = 396$; mean treatment duration, 95 weeks).⁶¹ The mechanism by which riociguat could influence the incidence of respiratory tract bleeding is unclear, although bronchial artery vasodilation might contribute.

In PATENT-1, SAEs of syncope occurred in three patients (1%) in the group receiving a maximum of 2.5 mg, all of which were considered study drug related, and in five patients (4%) in the placebo group, one of which was considered study drug related (1%). In CHEST-1, the SAE of syncope was experienced by four patients (2%) in the riociguat group, three cases of which (2%) were considered study drug related, and by three patients (3%) in the placebo group, one case of which (1%) was considered study drug related. At 1 year of both PATENT-2 and CHEST-2, 2% of patients had experienced study-drug-related syncope.^{61,77} Renal failure was rare in the CHEST or PATENT studies.^{61,76-78}

Riociguat: Indication, Dosing, and Clinical Pharmacology

Riociguat is indicated for the management of PAH and inoperable CTEPH or persistent/recurrent CTEPH

after PEA in adult patients.^{5,6,84,85} To avoid the risk of hypotension from the vasodilatory effect of riociguat on the systemic circulation, the oral dose is adjusted for the individual patient, guided by the signs and symptoms of hypotension. The usual starting dose is 1 mg tid for the first 2 weeks, and the dose is increased by 0.5 mg tid at 2-week intervals to a maximum of 2.5 mg tid based on tolerability (systolic blood pressure \geq 95 mm Hg and no signs or symptoms of hypotension). Dose reduction should be considered any time the drug is not tolerated. During phase III studies, 75% of patients with PAH and 77% of those with CTEPH tolerated the maximum dose of 2.5 mg tid.^{76,78}

Oral riociguat is rapidly absorbed, with maximum plasma concentrations occurring after about 1 to 1 1/2 hours and a terminal elimination half-life of approximately 12 hours in patients with PAH.^{84,85} The absolute bioavailability of riociguat is approximately 94%, and food intake has no clinically relevant effect on pharmacokinetics.^{84,85} Pharmacokinetic studies indicate that riociguat has no effect on platelet aggregation at clinically relevant concentrations^{86,87} and has no clinically relevant drug to drug interactions with warfarin.⁸⁶ Riociguat can be used in combination with ERAs or prostanoids, or both, as seen in the PATENT study.^{61,78}

Concerns over pharmacodynamic interactions between riociguat and PDE5 inhibitors were supported by the results of the PATENT PLUS study.⁷¹ In this 12-week phase II study, coadministration of riociguat and sildenafil resulted in changes in blood pressure similar to those of sildenafil plus placebo. However, in the long-term extension study, a high rate of discontinuation was observed because of hypotension, and there were three deaths (18%) (not considered study drug related by the investigator). The potentially unfavorable safety signals with sildenafil plus riociguat, and no evidence of a positive benefit to risk ratio, mean that concomitant administration of riociguat with PDE5 inhibitors is contraindicated, as is coadministration of riociguat with nitrates or NO donors.^{84,85}

Positioning of Riociguat in PAH and CTEPH Therapy

The efficacy data obtained in patients with PAH in the PATENT-1 and PATENT-2 studies led to the recommendation of riociguat as a first-line treatment as well as an add-on to background bosentan therapy in the 2015 European Society of Cardiology/European Respiratory Society PH treatment guidelines. Additionally,

riociguat is the only pharmacotherapy recommended in the 2015 guidelines for the treatment of patients with symptomatic CTEPH that is inoperable or persistent/recurrent after surgery.^{5,6} The use of riociguat as first-line therapy in PAH gives the option of sequential combination therapy with an ERA or prostanoid to potentially augment the benefit experienced by patients. In PATENT-1, it was demonstrated that riociguat provided additional efficacy to that achieved with ongoing ERA therapy. Previous studies adding a PDE5 inhibitor to ERA therapy have either not met their primary end point or not shown additional benefit.⁸⁸⁻⁹⁰

In PATENT-1, riociguat showed efficacy in both the primary end point and several secondary end points in patients who were treatment-naïve with respect to PAH-specific drugs.^{61,78,81} Furthermore, there was a uniform response pattern between treatment-naïve and pretreated patients, which was not seen in the Pulmonary Arterial Hypertension and Response to Tadalafil (PHIRST) trial of tadalafil in PAH.⁴⁷ In addition, the proportion of patients in PATENT-1 fulfilling several responder criteria^{91,92} reflects the clinically meaningful improvements achieved in patients receiving riociguat and suggests that riociguat has a prognostically relevant impact on PAH. Riociguat has also been shown to be effective in patients with PAH associated with congenital heart disease⁹³ and in patients with PAH associated with connective tissue disease.⁹⁴

The approval of riociguat for the treatment of PAH adds to a growing armamentarium of medications that have recently been approved for this indication. Although few data are available regarding the comparative efficacy of riociguat vs other medications or which patients are more likely to respond to sGC stimulators vs other drug classes, recent studies suggest that patients with PAH do better when agents from two or more classes of drugs are combined.⁵⁶ Although several choices are available for drugs that target the endothelin or prostacyclin pathways, those that target NO-cGMP have been limited to PDE5 inhibitors. The development of riociguat now provides clinicians with another option to augment NO-cGMP signaling. A significant proportion of patients with PAH fail to reach or maintain treatment goals with PDE5 inhibitors. Because of its mode of action, riociguat could potentially overcome these issues and provide a possible replacement drug for patients not responding to PDE5 inhibitor therapy. The ongoing open-label Riociguat Clinical Effects Studied in Patients With Insufficient Treatment Response to Inadequate Response to Phosphodiesterase-5 inhibitor (RESPITE) study (NCT02007629) is investigating whether it is safe,

feasible, and beneficial to transition from PDE5 inhibitor therapy to riociguat in patients with PAH and an inadequate response to PDE5 inhibitors or PDE5 inhibitors in combination with an ERA.⁹⁵ In addition, the different side-effect profile of riociguat makes it a potential candidate for transition in patients who experience side effects with PDE5 inhibitors.^{46,47,51,52}

Clinical Development of Riociguat for Other Forms of PH

Endothelial dysfunction and deficits in NO-sGC-cGMP signaling are involved in the progression of several subtypes of PH. To this end, a number of phase II studies with riociguat have been undertaken in other forms of PH, including PH associated with systolic LV dysfunction,^{72,96} PH associated with heart failure with preserved ejection fraction,⁹⁶ PH associated with ILD,⁷⁵ PH associated with COPD (Table 2),⁷⁴ and PH associated with idiopathic interstitial pneumonia.⁹⁷

Conclusions

Inadequate signaling of the NO-cGMP pathway may play an important role in the pathogenesis of PAH. sGC stimulators such as riociguat offer a novel approach to enhancing NO-mediated cGMP synthesis for the treatment of PH (Fig 2). Findings from the phase III CHEST and PATENT studies indicate that riociguat significantly improves exercise capacity and a range of secondary end points, including pulmonary hemodynamics, NT-proBNP levels, and WHO FC, in patients with inoperable CTEPH and persistent/recurrent CTEPH after PEA and in PAH. Riociguat is effective in treatment-naïve patients with PAH and in those who are receiving ERAs or prostanoids, or both, and is the only approved medical therapy for inoperable or persistent/recurrent CTEPH.

Stimulating the NO-sGC-cGMP pathway is a promising approach for the treatment of PAH and CTEPH. Further studies are needed to identify which patients are most likely to respond favorably to sGC stimulation and to determine if patients who do not respond to PDE5 inhibitors or other medical therapies for PAH may respond to sGC stimulation.

Acknowledgments

Financial/nonfinancial disclosures: The authors have reported to CHEST the following: H-A. G. reports grants from Actelion, Bayer HealthCare Pharmaceuticals, Ergonex, and Pfizer and personal fees from Actelion, Bayer HealthCare Pharmaceuticals, Ergonex, Gilead, GSK, Merck, Novartis, and Pfizer. M. H. reports personal fees from

Actelion, Bayer HealthCare Pharmaceuticals, GSK, Novartis, and Pfizer. D. L. reports grants, personal fees, and nonfinancial support from Actelion, Bayer HealthCare Pharmaceuticals, Gilead, GSK, and Ikaria. R. S. reports grants from Actelion, Bayer HealthCare Pharmaceuticals, Gilead, and Pfizer and personal fees from Bayer HealthCare Pharmaceuticals and Ergonex. J-P. S. is an employee of Bayer Pharma AG, Wuppertal, Germany. M. R. W. reports personal fees and nonfinancial support from Bayer HealthCare Pharmaceuticals. J. R. K. reports grants from Actelion, Bayer HealthCare Pharmaceuticals, Gilead, Ikaria, Lung Biotechnology, NIH-NHLBI, Pfizer, and United Therapeutics.

Role of sponsors: The sponsor had no role in the design of the study, the collection and analysis of the data, or the preparation of the manuscript.

References

1. Simonneau G, Gatzoulis MA, Adatia I, et al. Updated clinical classification of pulmonary hypertension. *J Am Coll Cardiol*. 2013;62(25 suppl):D34-D41.
2. Gaine S. Pulmonary hypertension. *JAMA*. 2000;284(24):3160-3168.
3. Vachery JL, Adir Y, Barbera JA, et al. Pulmonary hypertension due to left heart diseases. *J Am Coll Cardiol*. 2013;62(25 suppl):D100-D108.
4. Simonneau G, Robbins IM, Beghetti M, et al. Updated clinical classification of pulmonary hypertension. *J Am Coll Cardiol*. 2009; 54(1 suppl):S43-S54.
5. Galie N, Humbert M, Vachery JL, et al. 2015 ESC/ERS Guidelines for the diagnosis and treatment of pulmonary hypertension: the Joint Task Force for the Diagnosis and Treatment of Pulmonary Hypertension of the European Society of Cardiology (ESC) and the European Respiratory Society (ERS): Endorsed by: Association for European Paediatric and Congenital Cardiology (AEPC), International Society for Heart and Lung Transplantation (ISHLT). *Eur Heart J*. 2016;37(1):67-119.
6. Galie N, Humbert M, Vachery JL, et al. 2015 ESC/ERS Guidelines for the diagnosis and treatment of pulmonary hypertension: the Joint Task Force for the Diagnosis and Treatment of Pulmonary Hypertension of the European Society of Cardiology (ESC) and the European Respiratory Society (ERS): Endorsed by: Association for European Paediatric and Congenital Cardiology (AEPC), International Society for Heart and Lung Transplantation (ISHLT). *Eur Respir J*. 2015;46(4):903-975.
7. McLaughlin VV, McGoon MD. Pulmonary arterial hypertension. *Circulation*. 2006;114(13):1417-1431.
8. Humbert M, Ghofrani HA. The molecular targets of approved treatments for pulmonary arterial hypertension. *Thorax*. 2016;71(1):73-83.
9. Benza RL, Miller DP, Barst RJ, et al. An evaluation of long-term survival from time of diagnosis in pulmonary arterial hypertension from REVEAL. *Chest*. 2012;142(2):448-456.
10. Humbert M, Sitbon O, Chaouat A, et al. Survival in patients with idiopathic, familial, and anorexigen-associated pulmonary arterial hypertension in the modern management era. *Circulation*. 2010;122(2):156-163.
11. Humbert M, Sitbon O, Yaici A, et al. Survival in incident and prevalent cohorts of patients with pulmonary arterial hypertension. *Eur Respir J*. 2010;36(3):549-555.
12. Benza RL, Miller DP, Barst RJ, et al. An evaluation of long-term survival from time of diagnosis in pulmonary arterial hypertension from the REVEAL Registry. *Chest*. 2012;142(2):448-456.
13. Farber HW, Miller DP, Poms AD, et al. Five-year outcomes of patients enrolled in the REVEAL Registry. *Chest*. 2015;148(4):1043-1054.
14. Jenkins D. PEA: a potentially curative treatment option for patients with CTEPH. *Eur Respir Rev*. 2014;24(136):263-271.
15. Kim NH, Delcroix M, Jenkins DP, et al. Chronic thromboembolic pulmonary hypertension. *J Am Coll Cardiol*. 2013;62(25 suppl): D92-D99.
16. Bonderman D, Martitschnig AM, Moertl D, Lang IM. Pulmonary hypertension in chronic heart failure. *Int J Clin Pract Suppl*. 2009;(161):4-10.

17. Condliffe R, Kiely DG, Gibbs JS, et al. Improved outcomes in medically and surgically treated chronic thromboembolic pulmonary hypertension. *Am J Respir Crit Care Med.* 2008;177(10):1122-1127.
18. Condliffe R, Kiely DG, Gibbs JS, et al. Prognostic and aetiological factors in chronic thromboembolic pulmonary hypertension. *Eur Respir J.* 2009;33(2):332-338.
19. Freed DH, Thomson BM, Berman M, et al. Survival after pulmonary thromboendarterectomy: effect of residual pulmonary hypertension. *J Thorac Cardiovasc Surg.* 2011;141(2):383-387.
20. Hurdman J, Condliffe R, Elliot CA, et al. ASPIRE registry: assessing the Spectrum of Pulmonary hypertension Identified at a REferral centre. *Eur Respir J.* 2012;39(4):945-955.
21. Mayer E, Jenkins D, Lindner J, et al. Surgical management and outcome of patients with chronic thromboembolic pulmonary hypertension: results from an international prospective registry. *J Thorac Cardiovasc Surg.* 2011;141(3):702-710.
22. Pepke-Zaba J, Delcroix M, Lang I, et al. Chronic thromboembolic pulmonary hypertension (CTEPH): results from an international prospective registry. *Circulation.* 2011;124(18):1973-1981.
23. Stasch JP, Evgenov OV. Soluble guanylate cyclase stimulators in pulmonary hypertension. *Handb Exp Pharmacol.* 2013;218:279-313.
24. Schmidt HH, Walter U. NO at work. *Cell.* 1994;78(6):919-925.
25. Klinger JR, Abman SH, Gladwin MT. Nitric oxide deficiency and endothelial dysfunction in pulmonary arterial hypertension. *Am J Respir Crit Care Med.* 2013;188(6):639-646.
26. Stasch JP, Pacher P, Evgenov OV. Soluble guanylate cyclase as an emerging therapeutic target in cardiopulmonary disease. *Circulation.* 2011;123(20):2263-2273.
27. Derbyshire ER, Marletta MA. Structure and regulation of soluble guanylate cyclase. *Annu Rev Biochem.* 2012;81:533-559.
28. Kaneko FT, Arroliga AC, Dweik RA, et al. Biochemical reaction products of nitric oxide as quantitative markers of primary pulmonary hypertension. *Am J Respir Crit Care Med.* 1998;158(3):917-923.
29. Kharitonov SA, Cailles JB, Black CM, du Bois RM, Barnes PJ. Decreased nitric oxide in the exhaled air of patients with systemic sclerosis with pulmonary hypertension. *Thorax.* 1997;52(12):1051-1055.
30. Laursen JB, Somers M, Kurz S, et al. Endothelial regulation of vasomotion in apoE-deficient mice: implications for interactions between peroxynitrite and tetrahydrobiopterin. *Circulation.* 2001;103(9):1282-1288.
31. Bowers R, Cool C, Murphy RC, et al. Oxidative stress in severe pulmonary hypertension. *Am J Respir Crit Care Med.* 2004;169(6):764-769.
32. Cracowski JL, Cracowski C, Bessard G, et al. Increased lipid peroxidation in patients with pulmonary hypertension. *Am J Respir Crit Care Med.* 2001;164(6):1038-1042.
33. Hoshikawa Y, Ono S, Suzuki S, et al. Generation of oxidative stress contributes to the development of pulmonary hypertension induced by hypoxia. *J Appl Physiol (1985).* 2001;90(4):1299-1306.
34. Kielstein JT, Bode-Boger SM, Hesse G, et al. Asymmetrical dimethylarginine in idiopathic pulmonary arterial hypertension. *Arterioscler Thromb Vasc Biol.* 2005;25(7):1414-1418.
35. Skoro-Sajer N, Mittermayer F, Panzenboeck A, et al. Asymmetric dimethylarginine is increased in chronic thromboembolic pulmonary hypertension. *Am J Respir Crit Care Med.* 2007;176(11):1154-1160.
36. Xu W, Kaneko FT, Zheng S, et al. Increased arginase II and decreased NO synthesis in endothelial cells of patients with pulmonary arterial hypertension. *FASEB J.* 2004;18(14):1746-1748.
37. Pullamsetti S, Kiss L, Ghofrani HA, et al. Increased levels and reduced catabolism of asymmetric and symmetric dimethylarginines in pulmonary hypertension. *FASEB J.* 2005;19(9):1175-1177.
38. Giaid A, Saleh D. Reduced expression of endothelial nitric oxide synthase in the lungs of patients with pulmonary hypertension. *N Engl J Med.* 1995;333(4):214-221.
39. Berger RM, Geiger R, Hess J, Bogers AJ, Mooi WJ. Altered arterial expression patterns of inducible and endothelial nitric oxide synthase in pulmonary plexogenic arteriopathy caused by congenital heart disease. *Am J Respir Crit Care Med.* 2001;163(6):1493-1499.
40. Zhao YY, Zhao YD, Mirza MK, et al. Persistent eNOS activation secondary to caveolin-1 deficiency induces pulmonary hypertension in mice and humans through PKG nitration. *J Clin Invest.* 2009;119(7):2009-2018.
41. Schermuly RT, Ghofrani HA, Wilkins MR, Grimminger F. Mechanisms of disease: pulmonary arterial hypertension. *Nat Rev Cardiol.* 2011;8(8):443-455.
42. Wilkins MR. Pulmonary hypertension: the science behind the disease spectrum. *Eur Respir Rev.* 2012;21(123):19-26.
43. Ehret GB, Munroe PB, Rice KM, et al. Genetic variants in novel pathways influence blood pressure and cardiovascular disease risk. *Nature.* 2011;478(7367):103-109.
44. Schwappacher R, Kilic A, Kojonazarov B, et al. A molecular mechanism for therapeutic effects of cGMP-elevating agents in pulmonary arterial hypertension. *J Biol Chem.* 2013;288(23):16557-16566.
45. Austin ED, Ma L, LeDuc C, et al. Whole exome sequencing to identify a novel gene (caveolin-1) associated with human pulmonary arterial hypertension. *Circ Cardiovasc Genet.* 2012;5(3):336-343.
46. Galiè N, Ghofrani HA, Torbicki A, et al. Sildenafil citrate therapy for pulmonary arterial hypertension. *N Engl J Med.* 2005;353(20):2148-2157.
47. Galiè N, Brundage BH, Ghofrani HA, et al. Tadalafil therapy for pulmonary arterial hypertension. *Circulation.* 2009;119(22):2894-2903.
48. Simonneau G, Rubin LJ, Galiè N, et al. Addition of sildenafil to long-term intravenous epoprostenol therapy in patients with pulmonary arterial hypertension: a randomized trial. *Ann Intern Med.* 2008;149(8):521-530.
49. Zhuang Y, Jiang B, Gao H, Zhao W. Randomized study of adding tadalafil to existing ambrisentan in pulmonary arterial hypertension. *Hypertens Res.* 2014;37(6):507-512.
50. Lundberg JO, Gladwin MT, Weitzberg E. Strategies to increase nitric oxide signalling in cardiovascular disease. *Nat Rev Drug Discov.* 2015;14(9):623-641.
51. Oudiz RJ, Brundage BH, Galie N, et al. Tadalafil for the treatment of pulmonary arterial hypertension: a double-blind 52-week uncontrolled extension study. *J Am Coll Cardiol.* 2012;60(8):768-774.
52. Rubin LJ, Badesch DB, Fleming TR, et al. Long-term treatment with sildenafil citrate in pulmonary arterial hypertension: SUPER-2. *Chest.* 2011;140(5):1274-1283.
53. Zhao L, Mason NA, Strange JW, Walker H, Wilkins MR. Beneficial effects of phosphodiesterase 5 inhibition in pulmonary hypertension are influenced by natriuretic peptide activity. *Circulation.* 2003;107(2):234-237.
54. Schermuly RT, Stasch JP, Pullamsetti SS, et al. Expression and function of soluble guanylate cyclase in pulmonary arterial hypertension. *Eur Respir J.* 2008;32(4):881-891.
55. Stasch JP, Hobbs AJ. NO-independent, haem-dependent soluble guanylate cyclase stimulators. *Handb Exp Pharmacol.* 2009;(191):277-308.
56. Galie N, Barbera JA, Frost AE, et al. Initial use of ambrisentan plus tadalafil in pulmonary arterial hypertension. *N Engl J Med.* 2015;373(9):834-844.
57. Pulido T, Adzerikho I, Channick RN, et al. Macitentan and morbidity and mortality in pulmonary arterial hypertension. *N Engl J Med.* 2013;369(9):809-818.
58. Evgenov OV, Pacher P, Schmidt PM, et al. NO-independent stimulators and activators of soluble guanylate cyclase: discovery and therapeutic potential. *Nat Rev Drug Discov.* 2006;5(9):755-768.
59. Follmann M, Griebenow N, Hahn MG, et al. The chemistry and biology of soluble guanylate cyclase stimulators and activators. *Angew Chem Int Ed Engl.* 2013;52(36):9442-9462.
60. Lim SL, Lam CS, Segers VF, Brutsaert DL, De Keulenaer GW. Cardiac endothelium-myocyte interaction: clinical opportunities for

- new heart failure therapies regardless of ejection fraction. *Eur Heart J*. 2015;36(31):2050-2060.
61. Rubin LJ, Galie N, Grimminger F, et al. Riociguat for the treatment of pulmonary arterial hypertension: a long-term extension study (PATENT-2). *Eur Respir J*. 2015;45(5):1303-1313.
 62. Sharkovska Y, Kalk P, Lawrenz B, et al. Nitric oxide-independent stimulation of soluble guanylate cyclase reduces organ damage in experimental low-renin and high-renin models. *J Hypertens*. 2010;28(8):1666-1675.
 63. Geschka S, Kretschmer A, Sharkovska Y, et al. Soluble guanylate cyclase stimulation prevents fibrotic tissue remodeling and improves survival in salt-sensitive Dahl rats. *PLoS One*. 2011;6(7):e21853.
 64. Evgenov OV, Zou L, Zhang M, et al. Stimulation of soluble guanylate cyclase attenuates bleomycin-induced pulmonary fibrosis in mice [abstract]. *Am J Respir Crit Care Med*. 2011;183:A2715.
 65. Weissmann N, Lobo B, Pichl A, et al. Stimulation of soluble guanylate cyclase prevents cigarette smoke-induced pulmonary hypertension and emphysema. *Am J Respir Crit Care Med*. 2014;189(11):1359-1373.
 66. Lang M, Kojonazarov B, Tian X, et al. The soluble guanylate cyclase stimulator riociguat ameliorates pulmonary hypertension induced by hypoxia and SU5416 in rats. *PLoS One*. 2012;7(8):e43433.
 67. Dees C, Beyer C, Distler A, et al. Stimulators of soluble guanylate cyclase (sGC) inhibit experimental skin fibrosis of different aetiologies. *Ann Rheum Dis*. 2015;74(8):1621-1625.
 68. Grimminger F, Weimann G, Frey R, et al. First acute haemodynamic study of soluble guanylate cyclase stimulator riociguat in pulmonary hypertension. *Eur Respir J*. 2009;33(4):785-792.
 69. Ghofrani HA, Hoepfer MM, Halank M, et al. Riociguat for chronic thromboembolic pulmonary hypertension and pulmonary arterial hypertension: a phase II study. *Eur Respir J*. 2010;36(4):792-799.
 70. Hoepfer M, Ghofrani H-A, Halank M, et al. Riociguat for pulmonary arterial hypertension (PAH) and chronic thromboembolic pulmonary hypertension (CTEPH): final data from a phase II long-term extension (LTE) study [abstract]. *Eur Respir J*. 2015;46(suppl 59):PA4560.
 71. Galie N, Muller K, Scalise AV, Grunig E. PATENT PLUS: a blinded, randomised and extension study of riociguat plus sildenafil in PAH. *Eur Respir J*. 2015;45(5):1314-1322.
 72. Bonderman D, Ghio S, Felix SB, et al. Riociguat for patients with pulmonary hypertension due to systolic left ventricular dysfunction: a phase IIb double-blind, randomized, placebo-controlled, dose-ranging hemodynamic study. *Circulation*. 2013;128(5):502-511.
 73. Bonderman D, Pretsch I, Steringer-Mascherbauer R, et al. Acute hemodynamic effects of riociguat in patients with pulmonary hypertension associated with diastolic heart failure (DILATE-1): a randomized, double-blind, placebo-controlled, single-dose study. *Chest*. 2014;146(5):1274-1285.
 74. Ghofrani A, Staehler G, Grünig E, et al. Acute effects of riociguat in borderline or manifest pulmonary hypertension associated with chronic obstructive pulmonary disease. *Pulm Circ*. 2015;5(2):296-304.
 75. Hoepfer MM, Halank M, Wilkens H, et al. Riociguat for interstitial lung disease and pulmonary hypertension: a pilot trial. *Eur Respir J*. 2012;41(4):853-860.
 76. Ghofrani HA, D'Armini AM, Grimminger F, et al. Riociguat for the treatment of chronic thromboembolic pulmonary hypertension. *N Engl J Med*. 2013;369(4):319-329.
 77. Simonneau G, D'Armini AM, Ghofrani HA, et al. Riociguat for the treatment of chronic thromboembolic pulmonary hypertension: a long-term extension study (CHEST-2). *Eur Respir J*. 2015;45(5):1293-1302.
 78. Ghofrani HA, Galie N, Grimminger F, et al. Riociguat for the treatment of pulmonary arterial hypertension. *N Engl J Med*. 2013;369(4):330-340.
 79. Walters SJ, Brazier JE. Comparison of the minimally important difference for two health state utility measures: EQ-5D and SF-6D. *Qual Life Res*. 2005;14(6):1523-1532.
 80. Bonner N, Abetz L, Meunier J, Sikirica M, Mathai SC. Development and validation of the living with pulmonary hypertension questionnaire in pulmonary arterial hypertension patients. *Health Qual Life Outcomes*. 2013;11:161.
 81. Rubin LJ, Galie N, Grimminger F, et al. Riociguat for the treatment of pulmonary arterial hypertension (PAH): 2-year results from the PATENT-2 long-term extension [abstract]. *Eur Respir J Suppl*. 2014;44. http://erj.ersjournals.com/content/44/Suppl_58/P1803. Accessed November 15, 2016.
 82. Simonneau G, D'Armini AM, Ghofrani HA, et al. Riociguat for the treatment of chronic thromboembolic pulmonary hypertension (CTEPH): 2-year results from the CHEST-2 long-term extension [abstract]. *Eur Respir J Suppl*. 2014;44(58):P1802.
 83. Henkens IR, Hazenoot T, Boonstra A, Huisman MV, Vonk-Noordegraaf A. Major bleeding with vitamin K antagonist anticoagulants in pulmonary hypertension. *Eur Respir J*. 2013;41(4):872-878.
 84. Bayer Pharma AG. Adempas® US prescribing information. http://labeling.bayerhealthcare.com/html/products/pi/Adempas_PL.pdf 2013. Accessed September 20, 2016.
 85. Bayer Pharma AG. Adempas® (riociguat tablets): EU summary of product characteristics. http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Product_Information/human/002737/WC500165034.pdf 2014. Accessed September 20, 2016.
 86. Frey R, Muck W, Kirschbaum N, et al. Riociguat (BAY 63-2521) and warfarin: a pharmacodynamic and pharmacokinetic interaction study. *J Clin Pharmacol*. 2011;51(7):1051-1060.
 87. Frey R, Muck W, Unger S, et al. No pharmacodynamic (PD) and pharmacokinetic (PK) interaction of riociguat (BAY 63-2521) and aspirin. *BMC Pharmacology*. 2011;11(suppl 1):25.
 88. Barst RJ, Oudiz RJ, Beardsworth A, et al. Tadalafil monotherapy and as add-on to background bosentan in patients with pulmonary arterial hypertension. *J Heart Lung Transplant*. 2011;30(6):632-643.
 89. Gruenig E, Michelakis E, Vachieri JL, et al. Acute hemodynamic effects of single-dose sildenafil when added to established bosentan therapy in patients with pulmonary arterial hypertension: results of the COMPASS-1 study. *J Clin Pharmacol*. 2009;49(11):1343-1352.
 90. Pfizer. Assess the efficacy and safety of sildenafil when added to bosentan in the treatment of pulmonary arterial hypertension. <https://clinicaltrials.gov/ct2/show/study/NCT00323297>. Accessed September 20, 2016.
 91. D'Armini AM, Ghofrani H-A, Kim NH, et al. Use of responder threshold criteria to evaluate the response to treatment in the phase III CHEST-1 study. *J Heart Lung Transplant*. 2015;34(3):348-355.
 92. Langleben D, Galie N, He J, et al. Use of clinically relevant responder threshold criteria to evaluate the response to treatment in the phase III PATENT-1 study. *J Heart Lung Transplant*. 2015;34(3):338-347.
 93. Rosenkranz S, Ghofrani HA, Beghetti M, et al. Riociguat for pulmonary arterial hypertension associated with congenital heart disease. *Heart*. 2015;101(22):1792-1799.
 94. Humbert M, Coghlan G, Denton C, et al. Efficacy and safety of riociguat in patients with pulmonary arterial hypertension associated with connective tissue disease: results from PATENT-1 and PATENT-2. In: American Thoracic Society International Conference Abstracts. A98. *Clinical Trials and Outcomes in Pulmonary Arterial Hypertension*. American Thoracic Society, 2015:A2198.
 95. Hoepfer M, Benza R, Simonneau G, et al. Rationale and study design of the RESPITE trial: riociguat clinical effects studied in pulmonary arterial hypertension (PAH) patients with insufficient treatment response to PDE-5 inhibitors (PDE-5i) [abstract]. *Eur Respir J*. 2015;46(S59):P2210.
 96. Ghio S, Bonderman D, Felix SB, et al. Left ventricular systolic dysfunction associated with pulmonary hypertension riociguat trial (LEPHT): rationale and design. *Eur J Heart Fail*. 2012;14(8):946-953.
 97. Bayer Pharma AG. Efficacy and safety of riociguat in patients with symptomatic pulmonary hypertension (PH) associated with idiopathic interstitial pneumonias (IIP) (13605RISE-IIP), 2015. <https://clinicaltrials.gov/ct2/show/NCT02138825>. Accessed September 20, 2016.