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EDITORIAL

Learning to pair therapies and the expanding matrix for pulmonary arterial hypertension: is more better?

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Study design for treatment of a rare disease with few therapeutic options is straightforward as ''something is likely better than nothing''. With the surge of therapeutic options for pulmonary arterial hypertension (PAH) since the late 1990s, trials have been designed based on limited data without the luxury of patient numbers. As community physicians can offer combination therapy without evidence of benefit, clinical trialists are challenged to accrue patients to studies to prove what some envision is intuitively obvious, that combining active agents is likely to be better than treatment with a single agent. PAH is a debilitating, progressive condition with diverse causes, and for a disease with no therapeutic cure, targeting multiple pathways concurrently is appealing. However, without prior evidence for safety and activity, it is entirely possible that some combinations might be more harmful than helpful.

PAH is an angioproliferative vasculopathy resulting from abnormal endothelial and smooth muscle cell interactions, which leads to a progressive narrowing of the pulmonary arteries and their branches, resulting in right-heart failure and death. At the cellular level, PAH is characterised by vasoconstriction, endothelial dysfunction and smooth muscle cell proliferation of the pulmonary arterioles, a process known as vascular remodelling, and *in situ* thrombosis of the small pulmonary arteries [1, 2]. Proliferating endothelial cells obliterate medium-sized pre-capillary arteries [3, 4], thereby forming the characteristic ''plexiform'' lesions. At the molecular level, the balance of vasculature regulation shifts towards factors that produce vasoconstriction and smooth muscle cell proliferation. Animal models and culture of cells from patients with PAH have demonstrated dysregulation of specific mediators of various signalling pathways, including: prostacyclin [5–7]; nitric oxide (NO); and endothelin [8–10]. Angiopoietins [11, 12], autoantibodies [13], and proinflammatory cytokines and chemokines have also been implicated [14]. In addition, endothelial cells form tumourlets or plexiform lesions [2, 4] that obliterate the medium-sized vessel walls. These lesions express angiogenic factors, vascular endothelial growth factor (VEGF) and VEGF receptor (VEGFR)-2 [15], and these endothelial cells expand in a monoclonal pattern [16]. Due to these properties, PAH is now thought of as a process of dysregulated angiogenesis with some features shared with neoplastic processes.

There are three major pathways involved in the development and progression of PAH, and each one is a target for the treatment of the disease. Prostacyclin induces relaxation of vascular smooth muscle by stimulating the production of cyclic adenosine monophosphate [17] and inhibiting the growth of smooth muscle cells [17] and platelet aggregation [18]. Current mainstays of therapy for PAH include prostacyclin supplementation with epoprostenol, treprostinil or iloprost [19–23]. The NO pathway reduces intracellular calcium by stimulating cyclic guanosine monophosphate (cGMP) synthesis by the smooth muscle cell, leading to pulmonary vasodilatation [24]. As the phosphodiesterase (PDE)-5 enzyme degrades cGMP and the pulmonary vasculature has a higher concentration of the PDE-5 enzyme than most vascular beds, another strategy is to increase cGMP indirectly by inhibiting PDE-5 with agents such as sildenafil [25]. The third pathway, the endothelin (ET) system, produces vasoconstriction and vascular proliferation by ET-1 binding to two receptors, ET_A and ET_B [26]. The nonselective inhibitor of ET_A and ET_B, bosentan [27], and the selective ET_A inhibitors, sitaxsentan [28] and ambrisentan [29], have both demonstrated benefit for some PAH patients.

Currently, Food and Drug Administration-approved therapies for PAH, such as prostacyclins (epoprostenol, treprostinil and iloprost), [19–23], ET receptor blockers (bosentan) [27] and phosphodiesterase inhibitors (sildenafil) [25], improve functional class (symptoms) and improve exercise capacity as assessed by the 6-min walk distance (6MWD). Additional objective evidence of therapeutic benefit varies; few improve right ventricular function on echocardiography, and all have minimal change in haemodynamic measurements at cardiac catheterisation. Only epoprostenol has provided survival benefit, with the 5-yr survival remaining at 50% [30, 31] without demonstrable reversal of the vasculopathy. Recently approved monotherapy is not as successful as we had hoped. Of patients treated with bosentan (56%) [32] or iloprost (71%) [33], >50% require change or addition in therapy or lung transplantation after 2 yrs of therapy. In addition, >40% required transition to *i.v.* prostanoid therapy (42% iloprost, 45% bosentan). With this information, it is not unexpected that physicians use combination therapy in the hope of improving outcomes.

In the present issue of the *European Respiratory Journal* (*ERJ*), HOEPER *et al.* [34] report the results of their multicentre,

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randomised, open-label, stepwise approach to assessing the safety and efficacy of inhaled iloprost added to oral bosentan therapy. HOEPER *et al.* [34] and the *ERJ* should be commended for realising the importance of reporting this ''negative'' trial, as it raises important issues in PAH treatment and trial design. This 12-week study (the Combination Therapy of Bosentan and Aerosolized Iloprost in Idiopathic Pulmonary Arterial Hypertension (COMBI) trial) randomised stable World Health Organization functional class III idiopathic PAH patients currently on bosentan (125 mg *b.i.d.*), to either inhaled iloprost 5 μg six times daily or no additional therapy. The investigators powered this study with the assumption that the addition of iloprost to bosentan would yield the same magnitude of improvement as iloprost monotherapy compared with placebo: an increase in the 6MWD of 45 ± 75 m [21]. With this assumption, the study required a total of 72 patients and included an interim safety/efficacy analysis at the study midpoint. According to the interim analysis (36 completed and 40 enrolled patients), a larger study could not demonstrate a statistically significant improvement in 6MWD for combination therapy; consequently, the investigators terminated the study.

Concerning and intriguing, three subjects in the combination arm of this trial had significant clinical deterioration with a marked decline in all objective measures. Although the study failed to meet its planned end-point, it raises two important and interconnected questions: 1) Why was the trial underpowered? and 2) Does the deterioration of three subjects in this study represent a true signal of harm or is it noise? HUMBERT *et al.* [35] completed a similar trial with upfront initiation of bosentan and epoprostenol, with three patient deaths during or shortly after patients were exposed to this combination. The study by HUMBERT *et al.* [35] was powered based on the magnitude of expected haemodynamic changes on cardiac catheterisation at 16 weeks, determined for prostacyclin monotherapy when compared with placebo. Similar to HUMBERT *et al.* [35], HOEPER *et al.* [34] overestimated the improvement for their power calculation, but if they had expected less, the sample size would have been prohibitive. With little data on the mechanistic basis for combination strategies and the range of potential responses, estimating improvement over a short time period can be precise only with luck. These trials were underpowered to meet the predetermined clinical end-points, unless the arbitrarily determined doses and schedules for the combinations had additive or better benefit for all subjects receiving them.

It is unclear if when combined with ET receptor blockers, prostacyclin causes harm in some patients and improvement in others. Without obtaining more definitive outcome measures, such as catheterisation, or longer trial duration with frequent visits to assess 6MWD and cardiopulmonary exercise measures, it is difficult to determine if the changes are within the normal variability of the 6MWD itself, if the effects may have improved over time, or if they are due to negative effects of the combination therapy. Importantly, we have not learned the reasons for the deterioration of three subjects in both the studies by HOEPER *et al.* [34] and HUMBERT *et al.* [35]. Is the 6MWD as the primary end-point responsible for some of this confusion? Maybe it is time to develop new end-points for PAH trials such as more discriminative exercise measures, PAH-specific quality-of-life measures, measures of right ventricular function, and new biomarkers to help us answer these questions [36]. In addition, we need to redefine progress in PAH prognosis and not only continue comparisons to the National Institute of Health registry historical control [37].

Despite the limitations cited by the investigators, such as short observation time, open-label design and small sample size, the results are similar to the double-blind trial of similar design, the STEP trial (iloprost inhalation solution safety and pilot efficacy trial in combination with bosentan for evaluation in PAH; V.V. McLaughlin, University of Michigan, Ann Arbor, MI, USA; unpublished data), with neither improving exercise capacity. Patients in the COMBI trial appeared more ill based on haemodynamics and 6MWD compared with the STEP trial; this may be why the combination in the STEP trial improved functional class and decreased time to clinical worsening. To further complicate the matter, the investigators previously reported improvement in 6MWD and cardiopulmonary exercise measures with the addition of bosentan to patients stable on either inhaled iloprost or beraprost, the converse approach to their present study [38]. When they evaluated therapy using a ''real-life'' approach of goal-oriented therapy using combinations in clinical practice, this approach appeared successful compared with historical controls [39]. From these studies, the community is left with unclear data, except that this combination is not curative.

An alternative study design, such as a crossover study, might have yielded more informative results. Learning trials, studies aimed at investigating therapeutic mechanism and differences in individual response, enable the investigator to discriminate variation that is related to differences based on treatment and disease classification, a true signal, from noise, variation based on measurement variability and individual patient differences [40]. With combination therapy, one cannot assume additive response to therapy based on dose alone. Variability in pharmacokinetics and pharmacodynamics for each individual, and as yet poorly understood variability in disease status, confound our ability to interpret small studies of combination therapy *versus* monotherapy. Hence, our traditional ''confirming'' trial designs [40] might be ill-suited to developing methods of appropriate patient selection and identification of treatment combinations that will benefit our patients. We need to initiate smaller scale mechanistic studies before embarking on confirmatory trials set out to prove superiority. These data highlight the need to improve trial design, end-points and measures of prognosis to answer the important questions and expedite development of better treatment regimens for our patients.

REFERENCES

1 Wagenvoort CA, Mulder PG. Thrombotic lesions in primary plexogenic arteriopathy. Similar pathogenesis or complication? *Chest* 1993; 103: 844–849.
2 Wagenvoort C, Wagenvoort N. Pathology of Pulmonary Hypertension. 2nd Edn. New York, John Wiley and Sons, 1977.
3 Cool CD, Stewart JS, Weahera P, *et al.* Three-dimensional reconstruction of pulmonary arteries in plexiform

pulmonary hypertension using cell-specific markers. *Am J Pathol* 1999; 155: 411–419.
4 Tuder R, Groves B, Badesch D, Voelkel N. Exuberant endothelial cell growth and elements of inflammation are present in plexiform lesions of pulmonary hypertension. *Am J Pathol* 1994; 144: 275–285.
5 Geraci MW, Gao B, Shepherd DC, *et al.* Pulmonary prostacyclin synthase overexpression in transgenic mice protects against development of hypoxic pulmonary hypertension. *J Clin Invest* 1999; 103: 1509–1515.
6 Nagaya N, Yokoyama C, Kyotani S, *et al.* Gene transfer of human prostacyclin synthase ameliorates monocrotaline-induced pulmonary hypertension in rats. *Circulation* 2000; 102: 2005–2010.
7 Tuder R, Cool C, Geraci M, *et al.* Prostacyclin synthase expression is decreased in lungs from patients with severe pulmonary hypertension. *Am J Respir Crit Care Med* 1999; 159: 1925–1932.
8 Giaid A. Nitric oxide and endothelin-1 in pulmonary hypertension. *Chest* 1998; 114: Suppl. 3, 208S–212S.
9 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: 214–221.
10 Giaid A, Yanagisawa M, Langleben D, *et al.* Expression of endothelin-1 in the lungs of patients with pulmonary hypertension. *N Engl J Med* 1993; 328: 1732–1739.
11 Kido M, Du L, Sullivan CC, Deutsch R, Jamieson SW, Thistlethwaite PA. Gene transfer of a TIE2 receptor antagonist prevents pulmonary hypertension in rodents. *J Thorac Cardiovasc Surg* 2005; 129: 268–276.
12 Sullivan CC, Du L, Chu D, *et al.* Induction of pulmonary hypertension by an angiopoietin 1/TIE2/serotonin pathway. *Proc Natl Acad Sci USA* 2003; 100: 12331–12336.
13 Nicolls MR, Taraseviciene-Stewart L, Rai PR, Badesch DB, Voelkel NF. Autoimmunity and pulmonary hypertension: a perspective. *Eur Respir J* 2005; 26: 1110–1118.
14 Dorfmuller P, Perros F, Balabanian K, Humbert M. Inflammation in pulmonary arterial hypertension. *Eur Respir J* 2003; 22: 358–363.
15 Tuder R, Chacon M, Alger L, *et al.* Expression of angiogenesis-related molecules in plexiform lesions in severe pulmonary hypertension: evidence for a process of disordered angiogenesis. *J Pathol* 2001; 195: 367–374.
16 Lee S, Shroyer K, Markham N, Cool C, Voelkel N, Tuder R. Monoclonal endothelial cell proliferation is present in primary but not secondary pulmonary hypertension. *J Clin Invest* 1998; 101: 927–934.
17 Clapp L, Finney P, Turcato S, Tran S, Rubin L, Tinker A. Differential effects of stable prostacyclin analogs on smooth muscle proliferation and cyclic AMP generation in human pulmonary artery. *Am J Respir Cell Mol Biol* 2002; 26: 194–201.
18 Rubin L, Mendoza J, Hood M, *et al.* Treatment of primary pulmonary hypertension with continuous intravenous prostacyclin (epoprostenol). Results of a randomized trial. *Ann Intern Med* 1990; 112: 485–491.
19 Barst R, Rubin L, Long W, *et al.* A comparison of continuous intravenous epoprostenol (prostacyclin) with conventional therapy for primary pulmonary hypertension. *N Engl J Med* 1996; 334: 296–301.
20 Gomberg-Maitland M, Tapson VF, Benza RL, *et al.* Transition from intravenous epoprostenol to intravenous treprostinil in pulmonary hypertension. *Am J Respir Crit Care Med* 2005; 172: 1586–1589.
21 Olschewski H, Simonneau G, Galie N, *et al.* Inhaled iloprost for severe pulmonary hypertension. *N Engl J Med* 2002; 347: 322–329.
22 Simonneau G, Barst RJ, Galie N, *et al.* Continuous subcutaneous infusion of treprostinil, a prostacyclin analogue, in patients with pulmonary arterial hypertension: a double-blind, randomized, placebo-controlled trial. *Am J Respir Crit Care Med* 2002; 165: 800–804.
23 Tapson VF, Gomberg-Maitland M, McLaughlin VV, *et al.* Safety and efficacy of IV treprostinil for pulmonary arterial hypertension: a prospective, multicenter, open-label, 12-week trial. *Chest* 2006; 129: 683–688.
24 Ricciardolo FL, Sterk PJ, Gaston B, Folkerts G. Nitric oxide in health and disease of the respiratory system. *Physiol Rev* 2004; 84: 731–765.
25 Galie N, Ghofrani HA, Torbicki A, *et al.* Sildenafil citrate therapy for pulmonary arterial hypertension. *N Engl J Med* 2005; 353: 2148–2157.
26 Luscher T, Barton M. Endothelins and endothelin receptor antagonists. *Circulation* 2000; 102: 2434–2440.
27 Rubin LJ, Roux S. Bosentan: a dual endothelin receptor antagonist. *Expert Opin Investig Drugs* 2002; 11: 991–1002.
28 Barst RJ, Langleben D, Badesch D, *et al.* Treatment of pulmonary arterial hypertension with the selective endothelin-A receptor antagonist sitaxsentan. *J Am Coll Cardiol* 2006; 47: 2049–2056.
29 Gallie N, Badesch D, Oudiz R, *et al.* Ambrisentan therapy for pulmonary arterial hypertension. *J Am Coll Cardiol*, 46: 529–535.
30 McLaughlin VV, Shillington A, Rich S. Survival in primary pulmonary hypertension: the impact of epoprostenol therapy. *Circulation* 2002; 106: 1477–1482.
31 Sitbon O, Humbert M, Nunes H, *et al.* Long-term intravenous epoprostenol infusion in primary pulmonary hypertension: prognostic factors and survival. *J Am Coll Cardiol* 2002; 40: 780–788.
32 Provencher S, Sitbon O, Humbert M, Cabrol S, Jais X, Simonneau G. Long-term outcome with first-line bosentan therapy in idiopathic pulmonary arterial hypertension. *Eur Heart J* 2006; 27: 589–595.
33 Opitz CF, Wensel R, Winkler J, *et al.* Clinical efficacy and survival with first-line inhaled iloprost therapy in patients with idiopathic pulmonary arterial hypertension. *Eur Heart J* 2005; 26: 1895–1902.
34 Hoeper MM, Leuchte H, Halank M, *et al.* Combining inhaled iloprost with bosentan in patients with idiopathic pulmonary arterial hypertension. *Eur Respir J* 2006; 28: 691–694.
35 Humbert M, Barst RJ, Robbins IM, *et al.* Combination of bosentan with epoprostenol in pulmonary arterial hypertension: BREATHE-2. *Eur Respir J* 2004; 24: 353–359.
36 Peacock A, Naeije R, Galie N, Reeves JT. End points in pulmonary arterial hypertension: the way forward. *Eur Respir J* 2004; 23: 947–953.
37 D'Alonzo G, Barst R, Ayres S, *et al.* Survival in patients with primary pulmonary hypertension: results from a

national prospective registry. *Ann Intern Med* 1991; 115: 343–349.

38 Hoeper MM, Taha N, Bekjarova A, Gatzke R, Spiekerkoetter E. Bosentan treatment in patients with primary pulmonary hypertension receiving nonparenteral prostanoids. *Eur Respir J* 2003; 22: 330–334.

39 Hoeper MM, Markevych I, Spiekerkoetter E, Welte T, Niedermeyer J. Goal-oriented treatment and combination therapy for pulmonary arterial hypertension. *Eur Respir J* 2005; 26: 858–863.

40 Sheiner LB. Learning *versus* confirming in clinical drug development. *Clin Pharmacol Ther* 1997; 61: 275–291.