α₁-Adrenoceptor-dependent vascular hypertrophy and remodeling in murine hypoxic pulmonary hypertension

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Faber JE, Szymeczek CL, Cotecchia S, Thomas SA, Tanoue A, Tsujimoto G, Zhang H. α₁-Adrenoceptor-dependent vascular hypertrophy and remodeling in murine hypoxic pulmonary hypertension. Am J Physiol Heart Circ Physiol 292: H2316–H2323, 2007. First published January 12, 2007; doi:10.1152/ajpheart.00792.2006—Excessive proliferation of vascular wall cells underlies the development of elevated vascular resistance in hypoxic pulmonary hypertension (PH), but the responsible mechanisms remain unclear. Growth-promoting effects of catecholamines may contribute. Hypoxemia causes sympathetic excitation, and prolonged stimulation of α₁-adrenoceptors (α₁-ARs) induces hypertrophy and hyperplasia of arterial smooth muscle cells and adventitial fibroblasts. Catecholamine trophic actions in arteries are enhanced when other conditions favoring growth or remodeling are present, e.g., injury or altered shear stress, in isolated pulmonary arteries from rats with hypoxic PH. The present study examined the hypothesis that catecholamines contribute to pulmonary vascular remodeling in vivo in hypoxic PH. Mice genetically deficient in norepinephrine and epinephrine production [dopamine β-hydroxylase−− (DBH−−)] or α₁-ARs were examined for alterations in PH, cardiac hypertrophy, and vascular remodeling after 21 days exposure to normobaric 0.1 inspired oxygen fraction (FiO₂). A decrease in the lumen area and an increase in the wall thickness of arteries were strongly inhibited in knockout mice (order of extent of inhibition: DBH−− = α₁D-AR−− > α₁B-AR−− > α₁H-AR−− mice). Distal muscularization of small arteries was also reduced (DBH−− > α₁D-AR−− > α₁H-AR−− mice). Despite these reductions, increases in right ventricular pressure and hypertrophy were not attenuated in DBH−− and α₁AR−− mice. However, hematocrit increased more in these mice, possibly as a consequence of impaired cardiovascular activation that occurs during reduction of FiO₂. In contrast, in α₁D-AR−− mice, where hematocrit increased the same as in wild-type mice, right ventricular pressure was reduced. These data suggest that catecholamine stimulation of α₁B- and α₁D-ARs contributes significantly to vascular remodeling in hypoxic PH.

vascular smooth muscle; adrenergic receptors; hypoxia

OBSTRUCTIVE, RESTRICTIVE, and hypoventilatory pulmonary diseases are the most common causes of secondary pulmonary arterial hypertension (PH), wherein chronic alveolar hypoxia and hypoxemia are considered the main pathogenic disturbances (reviewed in Refs. 14, 17, 28, 29, 33). In contrast, to the less common primary PH of familial or idiopathic origin, secondary PH is also caused by a chronic increase in pulmonary vascular shear stress, vascular inflammation, and prolonged exposure to high altitude (>2,500 m). PH is characterized by an increase in pulmonary vascular tone, combined with a hypertrophy of pulmonary arteries and muscularization of small arterioles, the walls of which are normally either partially or completely devoid of vascular smooth muscle cells (VSMCs) (17, 29, 33). This adverse remodeling is caused by a proliferation of VSMCs and adventitial fibroblasts, matrix accumulation, and acquisition of new mural cells, which result in lumen narrowing and decreased compliance. Evidence suggests that these effects are brought about by endothelial dysfunction and an imbalance in factors that stimulate and inhibit growth of vascular wall cells (4, 14, 17, 28, 29, 33). However, many of these mechanisms are not well understood. Remodeling increases pulmonary vascular resistance (PVR), leading to PH and right ventricular hypertrophy that can progress to right heart failure and death. Current therapies have improved the clinical course, but responses vary, and no therapy reverses the disease. Thus further understanding of mechanisms is needed to develop additional treatments.

Chronic alveolar hypoxia is thought to cause metabolic and/or hemodynamic injury of the vascular wall, leading to activation of trophic pathways (3, 4, 14, 17, 28, 29, 33). Besides growth factor receptor tyrosine kinases, e.g., BMP receptor 2, angiotensin/Tie2, PDGFβ receptor, and heparin-binding-EGF/EGF receptor (ErbB1), several G protein-coupled receptor ligands with trophic activity have also been implicated in PH, including serotonin and its transporter and endothelin-1 (7, 17, 21, 22, 26, 27, 33). Catecholamines have also been proposed to contribute to PH (12, 29, 31, 32). Stimulation of certain α₁-adrenoceptor (α₁-AR) subtypes by norepinephrine (NE) induces growth of VSMCs and adventitial fibroblasts (discussed below). The concentration of NE in venous (i.e., pulmonary arterial) blood is normally ~40% higher than in arterial blood, owing to diffusion from innervated vessels and secretion from the adrenal gland, together with hepatic and pulmonary metabolism (19). In experimental studies and in patients with primary and secondary PH, venous NE levels are further increased by chemoreflex-dependent sympathoexcitation (18, 31, 35). Hypoxia directly increases expression of both tyrosine hydroxylase, the rate-limiting enzyme in catecholamine synthesis (31), and certain vascular α₁-ARs (9). Appetite suppressants, cocaine, and liver disease,
which elevate plasma catecholamines, increase the incidence of PH (31, 32). The pulmonary artery has the highest contractile sensitivity to NE (up to 200-fold greater) among 12 different rabbit vessels studied in vitro (1). Whether this extends to the trophic activity of NE is unknown.

NE induces α1-AR-dependent proliferation, hypertrophy, and migration of VSMCs and adventitial fibroblasts studied in cell culture and, in aorta and carotid arteries, studied in organ culture (13, 40, 41) and in vivo (10, 36). α2- and β-ARs do not appear to contribute to this growth response. NE-mediated growth is signaled by reactive oxygen species-dependent trans-activation of EGFRs (2, 42), a pathway shared in part by endothelin-1 receptor stimulation (26). Furthermore, adrenergic growth is strongly augmented in systemic arteries after either balloon injury or altered shear stress, contributing significantly to the attendant hypertrophic changes (10, 11, 40, 43), as well as to growth and remodeling of collateral arteries in ischemia (5). For example, local or systemic blockade of specific α1-AR subtypes or catecholamine synthesis by pharmacological or genetic approaches markedly reduced neointimal growth, adventitial fibrosis, and lumen loss after carotid injury (10, 11, 43). Of note, nonsubtype-specific α1-AR antagonists reduce wall hypertrophy, resistance, and pressure in the pulmonary circulation of patients with PH (reviewed in Ref. 31). However, the basis for these beneficial effects has not been examined, presumably because the hypotension caused by these agents has discouraged their use in patients with PH.

These findings suggest that NE may contribute to adverse structural remodeling in PH. In support of this hypothesis, we recently found that NE stimulates α1-AR-dependent growth of the rat pulmonary artery maintained in organ culture (12). Furthermore, this effect was significantly augmented in pulmonary arteries obtained from animals with hypoxic PH. The purpose of the present study was to examine, in vivo, whether NE contributes to pulmonary hypertensive remodeling. Remodeling was studied in small arteries and arterioles that account for most of the elevated resistance in hypoxic PH (29, 33). PH was induced in mice with genetic defects in catecholamine signaling by chronic exposure to reduced inspired oxygen (0.1 FIO2). This model is frequently used to investigate pathophysiological mechanisms and new therapies.

MATERIALS AND METHODS

Three strains of 4–6-mo-old male mice constructed on the Sv129 × C57BL/6 background, as detailed elsewhere (see Ref. 11 for references), were used: dopamine β-hydroxylase knockout (DBH−/−) mice and heterozygote “control” littermates (DBH+−/); DBH−/− have normal levels of plasma and tissue catecholamines; see Ref. 11 for references); α1B-AR−/− mice and wild-type littermates (α1B-AR−/−); and α1D-AR−/− mice and wild-type littermates (α1D-AR−/−). Body weights for the different groups are given in Figs. 1, 4, and 5. Controls and knockouts for each gene were from at least two litters. To control for genetic variation among the knockout models, which were generated in different laboratories, wild-types for each strain were studied in normoxia and hypoxia. To induce PH, mice were maintained in a normobaric 0.1 FIO2-elevated nitrogen environment for 21 days, except for two 15-min exposures to normoxia to allow for a change of bedding and replenishment of food, water, and absorbents for CO2 and water vapor. Procedures were conducted in accordance with National Institutes of Health (NIH) guidelines, and the protocols were approved by the Institutional Animal Care and Use Committee of the University of North Carolina at Chapel Hill.

After 21 days of normoxia or hypoxia, animals were anesthetized with ketamine and acepromazine (150 and 1.50 mg/kg im). Right ventricular systolic pressure in the closed-chest condition was recorded using subxiphisternal cardiac puncture with a 25-gauge needle connected through a stiff-walled catheter filled with heparinized saline to a pressure transducer. The chest was then opened, blood was collected for determination of hematocrit, the trachea was cannulated, and the lungs were inflated to 23 cmH2O with phosphate-buffered solution (PBS). The pulmonary artery trunk was cannulated through the right ventricle, and the circulation was perfused for 10 min at 75 and 100 cmH2O for normoxic and hypoxic groups, respectively, across the opened left atrium with heparinized PBS at 4°C containing 100 μmol/l of sodium nitroprusside (prepared fresh and maintained in darkness at 4°C until use). This protocol produces maximal dilation (see RESULTS). After perfusion, the pulmonary vasculature was immediately perfused at the same pressures for 20 min with paraformaldehyde at 4°C. Catheters were clamped, and the heart-lung was post-fixed for 24 h in paraformaldehyde at 4°C. Hearts were dissected for determination of wet weight of the right ventricle (RV) and the left ventricle plus septum (LV + S).

The mid thoracic portion of the left lung lobe was blocked from hilum to the periphery and embedded in paraffin. Lung blocks were processed by interrupted sectioning at 8-mm thickness every 64 mm and mounted to 10 sections (1 at each interruption) per slide. Multiple slides were constructed of sequentially adjacent sections to the first slide and were stained for morphometry (hematoxylin and eosin or Masson’s trichrome), elastin (Verhoff van Giesson), and α-smooth muscle actin (α-SMA). Detection of α-SMA was with mouse antihuman α-SMA antibody, 1:100 (Dako, Carpinteria, CA), biotinylated anti-mouse antibody, 1:200 (Vectastain ABC, Burlingame, VT), and diaminobenzidine (DAB React, Sigma, St. Louis, MO). Sections stained for α-SMA were lightly counterstained with 0.1% Light Green SF Yellowish. For determination of the lumen area, wall thickness, and distal muscularization, in each section, 4–6 elastin-positive vessels that ranged from ~25–100 mm lumen diameter and that were associated with intra-acinar airways (terminal and respiratory bronchioles, alveolar ducts, and alveoli) were imaged and subjected to digital morphometry (Scion Image Software, NIH), and an average was obtained for each animal. For determination of distal muscularization, intra-acinar arterioles were counted as fully muscularized (>75% of wall circumference α-SMA positive), partially muscularized (<75% of wall circumference α-SMA positive), or nonmuscularized (29).

Areas were determined as follows: lumen area = (lumen circumference)2/4π, and wall area = area between the lumen and outer vessel circumference defined by the outer edge of the dense collagen-containing adventitial layer. Wall thickness was calculated as (outer vessel circumference/2π) − (circumference of lumen/2π). Thicknesses were obtained because wall areas change secondary to changes in vessel circumference, thus increased area may not equate with hypertrophy of the vessel wall. All histological analysis was conducted by an investigator blinded to animal strain.

Data are given as means ± SE for n number of animals, unless stated otherwise. P values (significance, <0.05) were obtained from paired or unpaired t-tests or ANOVA, followed by Bonferroni protection where appropriate.

RESULTS

Vascular remodeling is inhibited in mice lacking catecholamine synthesis. In this study, the pulmonary circulation was dilated with 100 μmol/l nitroprusside in PBS, followed by perfusion fixation. We have found in other studies that the rate of perfusion with this solution was not different from the rate obtained with a combination of 4 mmol/l adenosine and 0.1 mmol/l papavarine in PBS, suggesting that a maximum relax-
ation of smooth muscle was obtained. To test this assumption, we determined whether perfusion of the lungs with zero-calcium PBS containing 100 μmol/l EGTA caused additional dilation beyond nitroprusside perfusion. Three-to-four-month-old male C57BL/6 mice maintained a normoxic state or, under 0.1 FiO2 for 3 wk, were perfused, as described in MATERIALS AND METHODS, for three sequential 5-min intervals, first with PBS, then with 100 μmol/l nitroprusside, and finally with zero-calcium EGTA. PVR (PVR = pulmonary artery pressure/pulsat flow) during PBS perfusion in hypoxic mice (n = 8) was 89 ± 17% higher than in normoxic controls (n = 8; P = 0.003). During nitroprusside perfusion, PVR changed in normoxic mice by -3.9 ± 3.2% (not significant) and in hypoxic mice by -11.4 ± 3.5% (P = 0.18), reflecting greater smooth muscle tone in hypoxic mice. During perfusion with zero-calcium EGTA, PVR changed in normoxic mice by 0 ± 3.3% and in hypoxic mice by +3.3 ± 3.3% (both not significant). These data demonstrate that nitroprusside perfusion, as done herein, produced maximal dilation of the pulmonary vasculature before fixation for subsequent histomorphometry.

To determine whether catecholamines contribute to PH and vascular remodeling, mice lacking DBH that is required for NE and epinephrine synthesis were studied. After exposure for 21 days to 0.1 FiO2, hematocrit and right ventricular hypertrophy (RV/LV + S) increased more in DBH−/− than in wild-type mice; body weight declined similarly, and right ventricular systolic pressure increased similarly (Fig. 1). Thus PH and right ventricular hypertrophy were not diminished in DBH−/− mice. However, the increase in wall thickness (+65%) and decrease in lumen area (−16%) of distal airway-associated arterioles induced by hypoxia (Fig. 2) were both reduced (+10% and −3%, respectively) in DBH−/− mice (Fig. 2). Circumferences were not significantly different among any of the groups (n = 25–34 mice/group)—wild-type: DBH-normoxia (NX) = 307 ± 9, DBH-hypoxia (HX) = 306 ± 6, α1B-NX = 290 ± 13, α1B-HX = 301 ± 18, α1D-NX = 304 ± 10, and α1D-HX = 304 ± 9; and knockout: DBH-NX = 298 ± 6, DBH-HX = 299 ± 3, α1B-NX = 313 ± 12, α1B-HX = 326 ± 13, α1D-NX = 318 ± 8, and α1D-HX = 311 ± 5. Distal muscularization of 25–100 mm arterioles in controls was also reduced in DBH−/− mice (Figs. 3). We also determined the number of alveoli and elastin-positive vessels (25–100 mm diameter) associated with distal airways in adjacent histological sections stained for elastin and with trichrome, respectively (four 40X fields quantified in each animal). The vessel-to-alveoli ratios in normoxic and hypoxic control mice (0.046 ± 0.006 and 0.048 ± 0.006) were similar in DBH−/− mice (0.038 ± 0.005 and 0.055 ± 0.005).

These data suggest that, in hypoxic DBH−/− mice, the further increase in polycythemia and associated viscosity, together with a potential increase in pulmonary vascular tone (see DISCUSSION), prevented a decrease in pulmonary resistance, pulmonary arterial pressure, and right ventricular hypertrophy that was favored by the reduction in structural remodeling. The accentuated polycythemia may be a consequence of greater hypoxemia, due to a loss of catecholamine stimulation of the heart and vasculature during hypoxia, which is especially strong during the first several days of hypoxia (see DISCUSSION).

Vascular remodeling is inhibited in mice deficient in α1B- and α1D-ARs. We next examined the involvement of α1-AR subtypes in the catecholamine contribution to hypoxic pulmonary vascular remodeling. We also sought to confirm the above results in a genetic model involving global deficiency in adrenergic catecholamines. Therefore, mice lacking either α1B- or α1D-ARs were examined. α1A-Adrenoceptors null mice were not studied because expression of this receptor was not detected in the mouse pulmonary vasculature (30). Right ventricular pressure and hypertrophy were similar in α1B-AR−/− mice and wild-type controls during hypoxia. Like DBH−/− mice (Fig. 1), α1B-AR−/− evidenced a greater increase in hematocrit and a similar decrease in body weight (Fig. 4). An increase in arteriolar wall thickness (+97%) and a decrease in lumen area (−22%) in wild-type mice were attenuated in α1B-AR-deficient mice (+51% and −8%, respectively; Fig. 2). Distal muscularization showed less inhibition in α1B-AR−/− mice than in DBH−/− or α1D-AR−/− mice (Fig. 3). These data suggest that stimulation of α1B-ARs contributes to vascular remodeling in hypoxic PH. However, like that observed in the

![Fig. 1](http://ajpheart.physiology.org/10.220336e)
Fig. 2. Hypoxia-induced reduction in lumen area and increase in wall thickness of pulmonary arterioles in wild-type mice are reduced in mice deficient in catecholamine synthesis (DBH<sup>−/−</sup>) or α<sub>1</sub>-adrenoceptor subtypes (α<sub>1B</sub> and α<sub>1D</sub>). Top: histological sections of peripheral lung lobe showing small arterioles in wild-type mice for DBH exposed to normoxia (A and C) and after 21 days 0.1 Fio<sub>2</sub> (B and D) stained with Masson’s trichrome (A and B) and anti-α-smooth muscle actin (α-SMA) antibody (C and D). Note wall thickening and lumen narrowing (B and D) and acquisition of anti-α-SMA staining (D) over nearly 3 quarters of the wall circumference after exposure to 21 days of 0.1 Fio<sub>2</sub>. Middle and bottom: histomorphometry was determined on 25–100-μm-diameter intra-acinar arterioles in normoxic mice (white bars) or after 21 days exposure to 0.1 Fio<sub>2</sub> (black bars). Lumen area normalized to outer circumference squared for graphical scaling purposes (see RESULTS for circumference values, which did not differ among the 12 groups). Numbers in bars denote percent increases and decreases relative to normoxic mice. ***P < 0.001 vs. normoxia for same strain; ####P < 0.001 vs. corresponding hypoxic wild-type strain.
muscularization was also partially inhibited in mice deficient in DBH (order of extent of inhibition: DBH<sup>-/-</sup> = α<sub>1D</sub>-AR<sup>-/-</sup> > α<sub>1B</sub>-AR<sup>-/-</sup>). Distal muscularization of small arterioles was also inhibited (DBH<sup>-/-</sup> > α<sub>1D</sub>-AR<sup>-/-</sup> > α<sub>1B</sub>-AR<sup>-/-</sup> mice). Increases in right ventricular pressure and hypertrophy were not reduced in DBH<sup>-/-</sup> and α<sub>1B</sub>-AR<sup>-/-</sup> mice; however, hematocrit increased more in these mice. In contrast, hematocrit increased the same in α<sub>1D</sub>-AR<sup>-/-</sup> and wild-type mice, and right ventricular pressure increased by a significantly smaller amount relative to pressure during normoxia in α<sub>1D</sub>-AR<sup>-/-</sup> mice. These data suggest that catecholamine stimulation of α<sub>1</sub>-ARs contributes significantly to vascular remodeling in hypoxic PH.

In an organ culture model that maintains wall tension, we previously observed that exposure to NE for several days exerts trophic activity on the rat pulmonary artery that is strongly augmented in arteries taken from rats after 9 days of hypoxic PH (12). This growth factor-like action of NE was blocked by an α<sub>1A</sub>-AR but not by an α<sub>1D</sub>-AR antagonist. Similar α<sub>1A</sub>-AR subtype-dependent trophic actions were found in rat carotid and aorta studied in organ culture or in vivo, where mechanical injury was also accompanied by a strong enhancement of NE-induced growth (10, 40, 43). However, in the mouse carotid, the α<sub>1A</sub>-AR is not expressed (30), and the trophic actions of NE are mediated instead by the α<sub>1B</sub>-AR (11, 43). The α<sub>1D</sub>-AR mediates constriction of the aorta and carotid in both rat and mouse (8, see Ref. 11 for references). Thus, in these large conduit arteries and also in the rat pulmonary artery (the α-AR that mediates constriction of mouse pulmonary artery has not been examined), different α<sub>1</sub>-ARs appear to signal the trophic (α<sub>1A</sub>-ARs in rat and α<sub>1B</sub>-ARs in mouse) and contractile (α<sub>1D</sub>-AR) actions of NE. The present results in the small arteries of the mouse pulmonary circulation find some similarities and differences to this conclusion. Wall hypertrophy, lumen narrowing, and distal muscularization of small arteries and arterioles in PH were strongly inhibited in mice lacking NE and epinephrine (DBH<sup>-/-</sup>), confirming augmentation of the trophic activity of NE in the presence of the conditions (here chronic reduced F<sub>O<sub>2</sub></sub> and attendant hemodynamic changes) promoting cell proliferation and remodeling of arteries. However, unlike in mouse and rat carotid and aorta, small artery remodeling in PH was partially inhibited in mice lacking either α<sub>1D</sub>-ARs or α<sub>1B</sub>-ARs. With the assumption that in vitro studies in the pulmonary artery of rat, wherein the α<sub>1</sub>-AR mediates constriction (25), predict that α<sub>1D</sub>-ARs mediate constriction of small pulmonary arteries and arterioles of rat and mouse, the current results suggest that both α<sub>1B</sub>-AR and a subtype not previously identified to signal growth, i.e., the α<sub>1D</sub>-AR, mediate trophic actions of NE in the murine pulmonary circulation.
Despite significant inhibition of pulmonary remodeling in DBH−/− and α1B-AR−/− mice, elevated right ventricular pressure and hypertrophy were not reduced. However, hypoxia-induced polycythemia increased further in these knockouts. Resistance is directly and linearly related to viscosity, which is directly related to hematocrit (6). The exponential relationship between blood viscosity and hematocrit becomes especially steep at hematocrit levels above 0.60. However, this relationship, as well as the relationship between systemic hematocrit and microvascular hematocrit, is more complex in the lung than in a system of rigid tubes where this exponential relationship and the effects of diameter on hematocrit can be readily demonstrated. Nevertheless, an accentuated increase in viscosity within the pulmonary circulation of the hypoxic DBH−/− and α1B-AR−/− mice may have opposed the reduction in resistance produced by the observed inhibition of lumen narrowing. This would favor no reduction or an increase in pulmonary artery pressure (right ventricular afterload) and right ventricular hypertrophy, which is what we found. This explanation is supported by results in the α1D-AR−/− mice. Hematocrit did not increase over wild-type during hypoxia, and inhibition of remodeling was accompanied by the expected inhibition of right ventricular pressure and hypertrophy (although the latter did not reach statistical significance). The greater polycythemia in DBH−/− and α1B-AR−/− mice during hypoxia may have resulted from greater hypoxemia and erythropoietin stimulation caused by impaired sympathetic stimulation of cardiac output during the induction and maintenance of reduced FIO2. This is because a sympathetic increase in cardiac contractility is mediated by β-ARs and α1A-ARs and α1B-ARs in the murine heart (38) but not by α1D-ARs since they are not expressed in the heart (39). Furthermore, α1-AR regulation of
contractility becomes dominant when β-ARs are downregulated by prolonged sympathoexcitation, which is favored by the chronic reduction in FIO2 in this study. Polycythemia is a determinant of the rise in pulmonary artery pressure and, hence, right ventricular hypertrophy in hypoxic PH. Normalization of hematocrit in rats, guinea pigs, and mice partially reduces PVR and pressure and right ventricular hypertrophy, whereas hypertrophy of small arteries and distal muscularization is unaffected (15, 20, 24). These findings indicate that chronically reduced inspired oxygen, rather than polycythemia, is the primary stimulus for the structural remodeling in hypoxia PH. In addition to possible effects of greater hematocrit, enhanced tone mediated by effectors of Rho kinase, endothelin, serotonin, and other contractile pathways (7, 17, 26) likely also contributes importantly to the sustained PH in the DBH−/− and α1B−/− mice. The importance of tone is underscored by the demonstration that greatly elevated hematocrit in normoxic rats produced by treating them with erythropoietin for 2 wk did not significantly increase pulmonary artery pressure, which suggests that increased hematocrit alone is insufficient to produce hypertension in the absence of remodeling or elevated vascular tone (37).

It is also noteworthy that lower arterial pressure in the knockouts versus wild-type mice does not explain the observed inhibition of pulmonary vascular remodeling. Arterial pressure is reduced by 8–15 mmHg in DBH−/− and α1B−/− mice but is unaffected in α1H-ARs mice (see Ref. 11 for references and Ref. 34). This arises from the significant dependence of systemic adrenergic vascular tone on α1D-ARs in rats and mice (16). However, remodeling was inhibited in all three knockout groups. Also, it is well known that reduction of systemic arterial pressure, which is normal in PH, does not prevent or reverse the disease in experimental animals or humans.

In conclusion, these results provide the first experimental evidence that catecholamines may contribute to hypoxic PH. They support a recent report on the pulmonary artery studied in vitro that found that adrenergic trophic sensitivity is augmented by chronic alveolar hypoxia (12). Together, they suggest that catecholamines may participate in the excessive muscularization and fibrosis of the pulmonary arterial circulation in hypoxic PH. This could result from direct trophic effects of catecholamines on VSMCs and fibroblasts in the vascular wall. The signaling pathway for NE-induced growth in these cells has thus far been defined as follows: α1-AR→NADPH-oxidase→superoxide and H2O2→HB→EGF→EGF receptor→Raf1/MEK/Erk1/2→proliferation and protein synthesis (2, 42). Activation of this trophic pathway may combine or synergize with other growth-promoting stimuli, e.g., serotonin, endothelin-1, oxidative stress, and certain receptor tyrosine kinases, that have been implicated in the pathophysiology of PH (7, 14, 17, 21, 26–29, 33). Consistent with this possibility, nonsubtype-selective α1-AR antagonists were shown some years ago to reduce vascular wall hypertrophy, resistance, and pressure in the lungs of patients with PH (31). However, before the present study, the basis for these beneficial effects had not been examined, presumably because these agents cause undesirable arterial hypotension. The recent development of subtype-selective α1-AR antagonists with minimal cardiovascular side effects for treatment of benign prostatic hyperplasia (e.g., tamsulosin) (23) and other diseases may provide an opportunity to examine their effects prospectively or retrospectively in patients with PH. However, new methods are required to achieve local inhibition of adrenergic signaling in the pulmonary circulation of animal models of PH, and the types and functions of α1-ARs in the human pulmonary circulation need to be defined.

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