



Update on Pediatric Pulmonary Arterial Hypertension – Differences and Similarities to Adult Disease –

Tsutomu Saji, MD

Children and adults with pulmonary arterial hypertension (PAH) have similarities and differences in their background characteristics, hemodynamics, and clinical manifestations. Regarding genetic background, mutations in *BMPR2*-related pathways seem to be pivotal; however, it is likely that other modifier genes and bioactive mediators have roles in the various forms of PAH in children and adults. In pediatric PAH, there are no clear sex differences in incidence, age at onset, disease severity, or prognosis but, as compared with adults, syncope incidence, pulmonary vascular resistance, and mean pulmonary artery pressure are higher, and vasoreactivity to acute drug testing is more frequent, among children. Nevertheless, the pharmacokinetic effects of 3 major pulmonary vasodilators appear to be similar in children and adults with PAH. This review focuses on the specific pathophysiologic features of PAH in children. (*Circ J* 2013; **77**: 2639–2650)

Key Words: Children; Genes; Prostaglandins; Pulmonary hypertension

In children, pulmonary arterial hypertension (PAH) is usually a severe, refractory, and life-long disease. However, pediatric and adult PAH differ somewhat in etiology, background characteristics, sex predominance, symptoms at onset, response to medical treatment, and prognosis. Here, I review the available evidence and outline current perspectives on the clinical management of pediatric PAH.

Epidemiology

The incidence of pediatric idiopathic PAH (IPAH) in Japan was first surveyed in 1996 by the Japanese Society of Pediatric Cardiology and Cardiac Surgery (JSPCCS).¹ There were 219 PAH cases in 103 centers among children aged 0–15 years: 131 patients (88%) were 1 year or older and 18 (12%) were younger than 1 year. During a 6-year period 136 patients were evaluated. The male:female ratio was 51:72 (1:1.4). There were 2 periods of peak onset: before the age of 1 year and at approximately 12 years of age. Higher incidences were also seen in the age groups 4–7 years and 9–12 years. However, the large proportions of children who undergo school physical examinations at these times might explain the high percentages of cases (26%) in these age groups.

Disease onset was concentrated in the age groups 4–7 (34.5% of cases) and 9–12 years (41.2% of cases); mean (SD) age at onset was 8 years 2 months (3 years 8 months) (n=115). Mean age at first visit to a medical center to seek treatment was 9 years 1 month (4 years 0 months) (n=121), which indicates that the mean interval between disease onset and first medical visit

was 1 year 8 months (2 years 10 months) (n=112). This suggests that disease progression was quite slow in many patients.

In 7 cases, the patient (6.2%) had family members with PAH: 7 males and 7 females, a ratio of 1:1. This ratio differed from that seen in sporadic cases. In 4 cases the patient had siblings with IPAH: 3 (2 girls and 1 boy) had an older brother with IPAH. In another family, an older sister and a younger brother both had IPAH. There was also 1 case of a mother and son having IPAH. The incidence was approximately 1–2 per 1 million children under the age of 15 years.¹ During 2005–2011, the annual JSPCCS survey identified 131 children with IPAH.

The annual incidence of IPAH was reported to be approximately 26 out of 17,800,000 (1.49 per 1 million) Japanese children younger than 15 years,² which is similar to that reported by Barst.³

Clinical Manifestations at Onset

According to the Japanese nationwide survey, initial symptoms of pediatric PAH included fatigue and lassitude (35 cases, 28%), shortness of breath (33 cases, 27%), fainting (21 cases, 17%), chest pain (6 cases, 5%), difficulty breathing (5 cases, 4%), cyanosis (4 cases, 3%), abnormalities on cardiac testing (25 cases, 20%), and abnormalities on chest radiographs (7 cases, 6%) (n=123 cases). The mean (\pm SD) New York Heart Association (NYHA) functional class (FC) for all those responding (n=110) was 2.2 \pm 0.9 at first presentation; 24 patients (22%) were classified as NYHA-FC I and 50 (45%) as NYHA-FC II; thus, 74 patients (67%) had less severe disease. However, 27 patients (25%) were classified as NYHA-

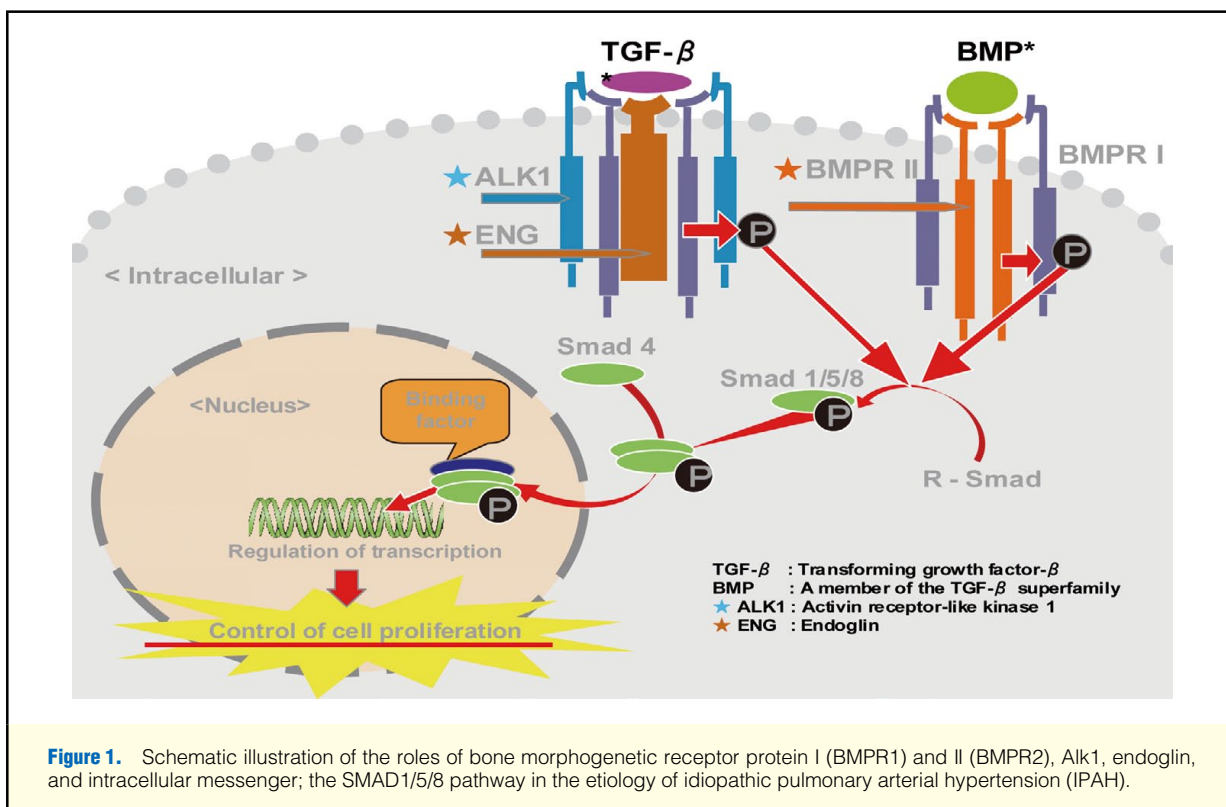
Received September 26, 2013; accepted September 26, 2013; released online October 22, 2013

Division of Pediatric Cardiology & Cardiac Surgery, Omori Hospital Medical Center, Toho University, Tokyo, Japan

Mailing address: Division of Pediatric Cardiology & Cardiac Surgery, Omori Hospital Medical Center, Toho University, 6-11-1 Omori-Nishi, Ota-Ku, Tokyo 143-8541, Japan. E-mail: saji34ben@med.toho-u.ac.jp

ISSN-1346-9843 doi:10.1253/circj.CJ-13-1180

All rights are reserved to the Japanese Circulation Society. For permissions, please e-mail: cj@j-circ.or.jp



FC III, for which registration on a lung transplant waiting list is recommended, and 9 patients (8%) were classified as NYHA-FC IV.

In subsequent generations of a pedigree, PAH presents at a younger age and with increased severity, which has been referred to as “genetic anticipation”.⁴ PAH usually appears at a younger age in each successive generation. This phenomenon is controversial but is typically observed in diseases with trinucleotide repeat expansion.⁵

As for associated PAH (APAH), collagen vascular disease-related PAH is rare in children, but a report from China found PAH in 5 of 8 (62.5%) pediatric patients with connective tissue disease (CTD), 2 of 4 (50%) with antiphospholipid antibody syndrome, 4 of 14 (28.6%) with systemic vasculitis, 3 of 28 (10.9%) with dermatomyositis, and 17 of 223 (7.6%) with systemic lupus erythematosus.⁶ Antinuclear antibody positivity has not been found in children with PAH but was observed in 4 of 16 (25%) mothers of children with IPAH.⁷

Sex Disparity

Among adults, the incidence of IPAH is considerably higher among women; the reported female:male ratio ranges from 1.7:1 to 3.5:1. In contrast, a study found that the incidence of IPAH was approximately equal between boys and girls.⁸ Another study reported a female:male incidence ratio of 1.5:1, and moreover, the female:male incidence ratio did not significantly differ between younger and older children.³

Although among adults the incidence of PAH is higher in women, numerous recent experimental studies have demonstrated better outcomes in female animals, worsening of the disease after ovariectomy, and a strong protective effect of estrogen, a phenomenon known as the “estrogen paradox” of

PAH.⁹ The hypothesis that hormonal and endocrinologic activity explains the nearly equal incidence of PAH in boys and girls remains to be studied.

Genetic and Cellular Characteristics

Since 1999, a number of mutations in the bone morphogenetic protein receptor (BMPR)/SMAD pathway have been reported (Figure 1). Familial PAH (FPAH) is transmitted as an autosomal-dominant trait that exhibits genetic anticipation but also markedly reduced penetrance ($\approx 20\%$). The genetic backgrounds of patients with pediatric PAH have been analyzed by my group for almost 2 decades. Mutations in *BMPR2* and the activin receptor-like kinase 1 gene (*ALK1*) were studied in 21 PAH probands younger than 16 years of age. In all 4 familial aggregates of PAH, 3 *ALK1* and 1 *BMPR2* mutation were identified. Among 17 probands aged between 4 and 14 years with idiopathic PAH, we identified 2 *ALK1* mutations (2/17: 11.8%) and 3 *BMPR2* mutations (3/17: 17.6%; 5 mutations in total: 5/17: 29.4%). The probands with the *ALK1* mutation developed PAH, as did the probands with the *BMPR2* mutation. Hence, it has been suggested that *ALK1* has as important a role as *BMPR2* in the etiology of PAH. Furthermore, asymptomatic carriers with the *ALK1* mutation within the serine-threonine kinase domain are at risk of developing PAH and hereditary hemorrhagic telangiectasia, so careful follow-up is recommended for such patients.¹⁰

Associations between clinical characteristics and gene mutations and age at onset younger than 16 years were analyzed by Chida et al in 54 patients with IPAH or hereditary PAH (HPAH).¹¹ Overall 5-year survival was 76%, and 18 *BMPR2* mutation carriers and 7 *ALK1* mutation carriers were detected among the 54 patients. The 5-year survival was lower in carri-

Table 1. Transforming Growth Factor (TGF)- β and Bone Morphogenetic Protein Receptor (BMPR)-Related Mutations Reported in Pediatric IPAH, HPAH, and CHD-PAH

Study	No. of patients	Age	Type	Positive (n, %)
Harrison et al 2003 ¹⁴	11	Adults	IPAH+HHT	8 (73%)
Roberts et al 2004 ¹⁵	106	66 children 40 adults	CHD-PAH	6 (3 AVSD; 3 Complex) ^{****}
Gruning et al 2004 ¹⁶	13	Children	IPAH	0 (0%)
Harrison et al 2005 ¹⁷	18	Children	IPAH/APAH	4 mutations (22%) (2 BMPR, 1 ALK1, 1 Endoglin)
Austin & Lloyd 2007 ¹⁸				
Fujiwara et al 2008 ^{*10}	21	Children	IPAH/HPAH	4 (3 ALK1, 1 BMPR2) (100%) in HPAH 5 (3 BMPR, 2 ALK1) (29.4%) in 17 IPAH
Shintani et al 2009 ^{**12}	23	Children	IPAH/HPAH	1 SMAD8 (4.4%)
Rosenzweig et al 2009 ¹⁹	78	Children	IPAH/FPAH	7/13 FPAH (54%); 1/65 IPAH (1.5%)
Chida et al 2012 ^{***11,13}	72	Children	IPAH/HPAH	18 BMPR2 (24%) 7 ALK1 (9.5%) 5 BMPR2 mutation carrier (6.8%) 1 SMAD8 (1.4%) 2 BMPR1b (2.8%)

*; **, ***Reports from same group. ****Complex CHD (1 ASD/PDA, 1 ASD/PDA/PAPVR, 1 APW/VSD). APW, aortopulmonary window; ASD, atrial septal defect; CHD, congenital heart disease; CHD-PAH, congenital heart disease-related pulmonary arterial hypertension; HHT, hereditary hemorrhagic telangiectasia; HPAH, hereditary pulmonary arterial hypertension; IPAH, idiopathic pulmonary arterial hypertension; PAH, pulmonary arterial hypertension; PAPVR, partial anomalous pulmonary venous return; PDA, patent ductus arteriosus; VSD, ventricular septal defect.

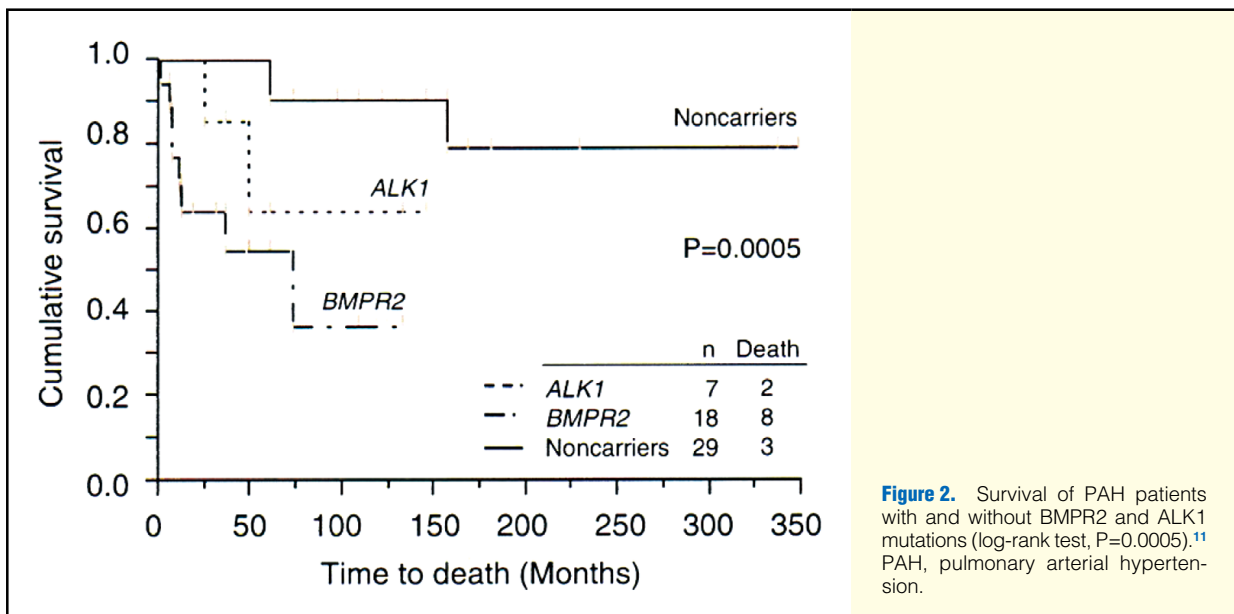


Figure 2. Survival of PAH patients with and without BMPR2 and ALK1 mutations (log-rank test, P=0.0005).¹¹ PAH, pulmonary arterial hypertension.

ers vs. noncarriers of *BMPR2* mutation (55% vs. 90%, respectively; hazard ratio, 12.54; P=0.0003). It was also worse in carriers vs. noncarriers of *ALK1* mutation, but not significantly so (5-year survival rate 64%; hazard ratio 5.14, P=0.1205). Patients with childhood IPAH or HPAH and *BMPR2* mutation may have the worst clinical outcomes. Outcomes were worse among *ALK1* mutation carriers than among noncarriers.

Mutations in *SMAD8* have been also reported in HPAH and IPAH.^{12,13} However, almost 30% of HPAH cases and 60–90% of IPAH cases have no *SMAD8* mutations, which suggests that there other, unidentified genes associated with HPAH and IPAH.¹² Chida et al¹³ screened for mutations in *endoglin*,

SMAD1, *SMAD2*, *SMAD3*, *SMAD4*, *SMAD5*, *SMAD6*, *SMAD7*, and the BMPRr type 1A (*BMPRIA*) and type 1B (*BMPR1B*) genes in 43 IPAH patients who had no mutations in *BMPR2*, *ALK1*, or *SMAD8*. Two missense mutations (c.479 G>A S160N, c.1176 C>A F392L) in *BMPR1B* were both found in 2 patients with IPAH.

IPAH and FPAH have developed in patients with hereditary hemorrhagic telangiectasia associated with mutations in *ALK1*.¹⁴ Genetic mutations reported in pediatric PAH are summarized in **Table 1**.

In considering medical treatment, patients with *BMPR2* mutations are less likely than those without such mutations to re-

spond to acute vasodilator testing (4% vs. 33%, respectively; $P < 0.003$; $n = 147$) and appear to have more severe disease at diagnosis. Patients with *BMPR2* mutations had lower mixed venous saturation ($57 \pm 9\%$ vs. $62 \pm 10\%$; $P < 0.05$) and cardiac index (CI; 2.0 ± 1.1 vs. 2.4 ± 1.5 L/min; $P < 0.05$) than those without such mutations. Patients with IPAH or FPAH appear unlikely to benefit from calcium-channel blockade.¹⁹

Cumulative survival was longer among patients with *ALK-1* mutation than among those with *BMPR2* mutation (Figure 2).

To identify the genes responsible for triggering PAH, we analyzed factors with an important role in IPAH by comparing discrepant or controversial expression patterns in a murine model with those previously published for human IPAH. We used a mouse model that, after repeated intratracheal injection of *Stachybotrys chartarum* (a ubiquitous, nonpathogenic fungus), induced muscularization of the pulmonary artery, leading to hypertension. Microarray assays with ontology and pathway analyses were performed. For some pathways the expression patterns in our model and in IPAH were the same, including BMP signaling with downregulation of *BMPR2*, *ALK1*, and *endoglin*. However, both Wnt/planar cell polarity (PCP) signaling and its downstream *Rho/ROCK* signaling were activated in human IPAH but not in our model. Activation of Wnt/PCP signaling, in upstream positions of the pathway, was found only in lung tissue from a patient with endstage IPAH and may have essential roles in the pathogenesis of the disease.²⁰ Mutations in *CAVI* (caveolin-1) were reported to be associated with PAH, in rare cases.²¹

Pathobiology

Although previously reported,²¹ herpesvirus (HHV)-8 infection has not been associated with IPAH in Japanese patients with PAH. All examined samples contained plexiform lesions around pulmonary arterial vessels, but immunohistochemistry did not detect HHV-8-encoded latency-associated nuclear antigen. HHV-8 DNA could not be amplified by polymerase chain reaction for the HHV-8-encoded K1 and KS330 (233 bp) genes in any sample.²²

Regarding Ca^{2+} and K^{+} channelopathy, uncontrolled, open-label, prospective observational studies^{23,24} have reported improved hemodynamics and survival with long-term administration of calcium-channel blockers in cases of pediatric IPAH/HPAH. An association of the novel gene *KCNK3* with familial PAH and IPAH was recently reported. Mutations in this gene reduced the potassium-channel current, which was successfully reversed by pharmacologic treatment.²⁵

Myeloid-derived suppressor cells (MDSCs) are increased in inflammatory and autoimmune disorders, and immature myeloid cells are present in the lungs of humans and animals with PAH. Among children with PAH, the level of MDSCs in girls was twice that in boys. Circulating activated MDSCs are significantly elevated in children with PAH as compared with control subjects. MDSCs might thus have important immunomodulatory roles in PAH pathogenesis.²⁶

Impaired endothelial homeostasis is fundamental to PAH pathogenesis. The number of circulating endothelial progenitor cells (EPCs) is diminished in adults with PAH and Eisenmenger syndrome. Sildenafil treatment is a pharmacologic means of increasing circulating EPCs. In addition, treprostinil increases the number and angiogenic potential of EPCs in children with PAH.²⁷

Chronic inflammation is an important component of fibroproliferative changes in PAH vasculopathy. Fibrocytes contribute to tissue remodeling in the setting of chronic inflamma-

tion and in animal models of PAH. No differences in fibrocytes have been observed among people with idiopathic vs. secondary PAH. Fibrocyte levels correlate with mean pulmonary arterial pressure (mPAP) and age. Differences have not been found in plasma levels of CC chemokine ligand 2 (CCL2) or CXC chemokine ligand 12 (CXCL12) which can mobilize fibrocytes from bone marrow. Circulating fibrocytes are significantly elevated in PAH.²⁸

Biomarkers in Children

Circulating levels of endothelin-1 (ET-1) are elevated in children with IPAH/HPAH and are probably related to the degree of hypoxic pulmonary vasoconstriction.²⁹ ET-1 is also elevated in congenital heart disease (CHD)-related PAH.³⁰ The plasma concentration of adrenomedullin (ADM), a potent vasodilating endogenous neurohumoral mediator, was reported to be useful in assessing the severity of pediatric IPAH. Levels of the neurohumoral mediators atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP), ET-1, and ADM positively correlated with NYHA-FC and significantly negatively correlated with exercise capacity. Stepwise regression analysis revealed that the BNP:ANP ratio was the most powerful independent factor of total pulmonary resistance ($r = 0.85$, $P = 0.0071$) and CI ($r = 0.84$, $P = 0.009$). ADM significantly correlated with BNP ($r = 0.53$, $P = 0.03$), ET-1 ($r = 0.66$, $P = 0.006$), and BNP:ANP ($r = 0.73$, $P = 0.0009$). After long-term intravenous prostacyclin (PGI₂) treatment, both ANP and BNP were again elevated at 3 months after discharge, regardless of improvement in NYHA-FC or exercise capacity. In contrast, ADM decreased from 3.0 ± 2.2 fmol/ml at baseline to 1.7 ± 0.7 fmol/ml at 1 month to 1.6 ± 0.5 fmol/ml at 3 months. In pediatric patients with PAH, a change in ADM concentration at 3 months was significantly associated with the ADM concentration at the start of PGI₂ therapy and change in mPAP ($r = 0.97$, $P = 0.0041$).³¹

In pediatric PAH, both the BNP and N-terminal proBNP (NT-proBNP) concentration strongly correlate with and predict changes in clinical variables and hemodynamics. In a cross-sectional analysis, echocardiographic and exercise data were better correlated with NT-proBNP than with BNP. NT-proBNP also showed less within-patient variability over time. Therefore, NT-proBNP is additional information that is useful in predicting these clinical variables. BNP and NT-proBNP correlate with mPAP/mean systemic arterial pressure (SAP) ratio ($r = 0.40$, $P < 0.01$; $r = 0.45$, $P < 0.01$; respectively), mean right atrial pressure ($r = 0.48$, $P < 0.01$; $r = 0.48$, $P < 0.01$), and tricuspid regurgitation velocity ($r = 0.36$, $P < 0.01$; $r = 0.41$, $P < 0.01$). In a longitudinal study, BNP and NT-proBNP were associated with the 6-min walk distance (6MWD), mPAP, mPAP/mSAP ratio, mean right atrial pressure, pulmonary vascular resistance (PVR) index, and tricuspid regurgitation velocity.³²

Pathology

Pathologic studies showed that younger patients with IPAH have greater pulmonary vascular medial hypertrophy but less intimal fibrosis and fewer plexiform lesions. Medial hypertrophy is severe in patients younger than 15 years of age, and usually the only change seen in young infants.³³ The pathology and cellular composition of plexiform lesions in children younger than 3 years of age differ from findings in older children and adults, in that young children have a more prominent smooth muscle component.³⁴ In addition, young children will often have marked muscularization of arterioles.³⁵ Medial thickening of the pulmonary arterioles usually develops earlier in children, whereas intimal thickening occurs earlier in adults.

Vasoconstriction appears to be more pronounced in children.³⁶ Plexiform lesions can be sparse and difficult to find in collagen vascular disease-related PAH. The pathology of PAH in portal hypertension is indistinguishable from that in IPAH and FPAH. Irreversible PAH in children with high-risk CHD is strongly associated with impaired endothelial cell apoptosis and antiapoptotic signaling from perivascular inflammatory cells.³⁷

Cardiac Catheterization and Acute Vasoreactivity

As compared with adults, children with IPAH have a higher CI (4.3 ± 3.9 vs. 2.1 ± 0.7 L · min⁻¹ · m⁻²), heart rate (121 ± 29 vs. 83 ± 13 beats/min), PVR (28.1 ± 18.1 vs. 15.4 ± 8.2 units), mPAP (70 ± 15 vs. 55 ± 16 mmHg), and PVR:SVR ratio (1.01 vs. 0.61), lower SAP (75 ± 15 vs. 91 ± 12 mmHg), and a better response to drug testing.³ In a recent study comparing hemodynamic parameters in children and adults, the children were better able than adults to withstand an increased right heart workload. As compared with adults, children with IPAH/HPAH had a higher mPAP/SAP ratio (0.83 vs. 0.62 mmHg, respectively) and a higher PVR index:SVR index ratio (0.83 vs. 0.64 U/m², respectively) at diagnosis.³⁸ Cardiac output (CO) is low in children, so PVR is higher than in adults. Pulmonary vasoreactivity during active vasodilatation and vasoconstriction is more obvious in younger children than in adults. The response rate to acute vasoreactivity testing appears to be higher before the age of 6 years, but decreases markedly with increasing age. PH crisis occurs more often in infants and younger children.²³

The frequency of a positive response to drug testing was reported to be highest among the youngest children (86% for age <1 year, 47% for age 1–7 years, and 27% for age >7 years at time of diagnosis), using the following criteria for response to drug testing: (1) 20% or greater decrease in mPAP and (2) no change or an increase in CI.³⁹ Children treated with long-term epoprostenol improved regardless of response to short-term vasodilator challenge. However, another study found no relationship between the acute and long-term effects of epoprostenol in adults (ie, absence of a short-term, acute response did not preclude long-term efficacy).⁴⁰

Acute pulmonary vasodilator testing appears essential in selecting the initial therapy for children with PAH. One study⁴¹ reported initial experience with inhaled treprostinil in children with PAH and evaluated and compared hemodynamic changes with those after inhaled nitric oxide treatment. The median total treprostinil dose was 1.53 µg/kg (range 0.71–2.89 µg/kg). Inhaled nitric oxide and inhaled treprostinil significantly decreased mPAP and the PVR index as compared with baseline. Inhaled treprostinil was effectively delivered and well tolerated and may be useful as an acute vasodilative test.

Treatment

General Support

Although it is not known if diuretics, digitalis, and anticoagulation therapy are clearly beneficial for pediatric PAH, these treatments may be useful in supporting cardiopulmonary performance and preventing right heart failure and hypercoagulable state-induced cardiac and pulmonary thrombosis.⁴²

Calcium-Channel Blockers

A subgroup of pediatric IPAH patients appears to have a higher rate of acute pulmonary vasoreactivity (defined as >20% mPAP, no decrease in CO, and a decrease in PVR:SVR); children younger than age 8 years had a 40–70% rate of acute reactivity,²³ but acute vasoreactivity resolved in a study with a

longer follow-up period.²⁴

Approximately 80% of children with severe PAH do not respond to acute drug testing, in which case nifedipine (2–5 mg/kg/day), diltiazem (3–5 mg/kg/day), and amlodipine (2.5–10 mg/kg/day) are recommended; verapamil is contraindicated in PAH.⁴² None of 22 children with acute vasoreactivity who were treated with a high-dose calcium-channel blocker as monotherapy or in combination therapy died within 5 years of diagnosis.^{43,44}

PGI₂

Children with IPAH and APAH typically have an imbalance in biosynthesis of thromboxane A₂ and PGI₂.^{44,45}

Oral PGI₂ (Beraprost) The oral prostacyclin analog, beraprost (BPS), was first marketed in Japan in 1992 and was the first available orally active PGI₂. BPS is approved in Japan and Korea. It has beneficial effects for patients in NYHA-FC I or II, and the effects persist from several to 9 months.⁴⁶ BPS reaches peak concentration after 30 min and has an elimination half-life of 35–40 min.^{46,47} However, the effect of treatment is not sustained at 9 or 12 months. When change in pulmonary vascular reactivity was defined as a decrease in mPAP or PVR greater than 20% from baseline, the response rate for 100% O₂ (10 min), inhaled nitric oxide 40 ppm (10 min), and oral BPS (1 µg/kg; 30 min) during cardiac catheterization was 84%, 72.7%, and 64%, respectively.⁴⁸ The pulmonary vasodilating effects of BPS are similar to those of inhaled nitric oxide, and BPS can be used as an acute vasodilating agent during cardiac catheterization.⁴⁹ Long-acting oral BPS (beraprost LA) is currently approved for PAH in adults. In Japan, BPS has been used for 2 decades and is usually combined with a phosphodiesterase type 5 inhibitor (PDE5-I) and/or an endothelin-receptor antagonist (ERA) as initial treatment. Combination therapy with oral BPS LA and inhaled aerosolized iloprost is an alternative therapy for CHD-PAH. Treated patients have improved exercise capacity and right ventricular systolic pressure without worsening of oxygen saturation.

Intravenous PGI₂ (Epoprostenol) There are few data on the long-term effects of continuous intravenous epoprostenol for children with IPAH. However, treatment with intravenous epoprostenol is at least as effective in children as in adults with respect to increasing survival, improving hemodynamics, and relieving symptoms. Without coadministration of oral PDE5-I or ERA, survival at 1, 3, 5, and 10 years was 94%, 88%, 81%, and 61%, respectively, in a 2004 study.²⁴

Nakayama et al⁵⁰ reviewed the records of 31 IPAH patients aged 18 years or younger who had begun epoprostenol treatment between January 1999 and June 2004. During a mean follow-up of 3.4 years, the rate of those who survived or did not undergo lung transplantation among the 27 patients who received intravenous epoprostenol was 100% at 1 year, 96.3% at 2 years, and 79.4% at 3 years. Among 82% of the survivors, World Health Organization functional class (WHO-FC) changed from III or IV to II, as indicated by improvement in plasma BNP and 6MWD during follow-up. In most patients, mPAP and PVR:SVR remained high, although CI had improved to normal range at 1 year after initiation of epoprostenol. Therefore, oral sildenafil was given as additional therapy to 16 patients who subsequently presented with sustained severe PAH.

Continuous intravenous epoprostenol improves survival and exercise tolerance in childhood IPAH, although improvement in PVR is insufficient despite long-term therapy. Therefore, the addition of oral vasodilators such as sildenafil or bosentan is recommended as add-on therapy to intravenous epoprostenol. The indications for continuous intravenous epoprostenol

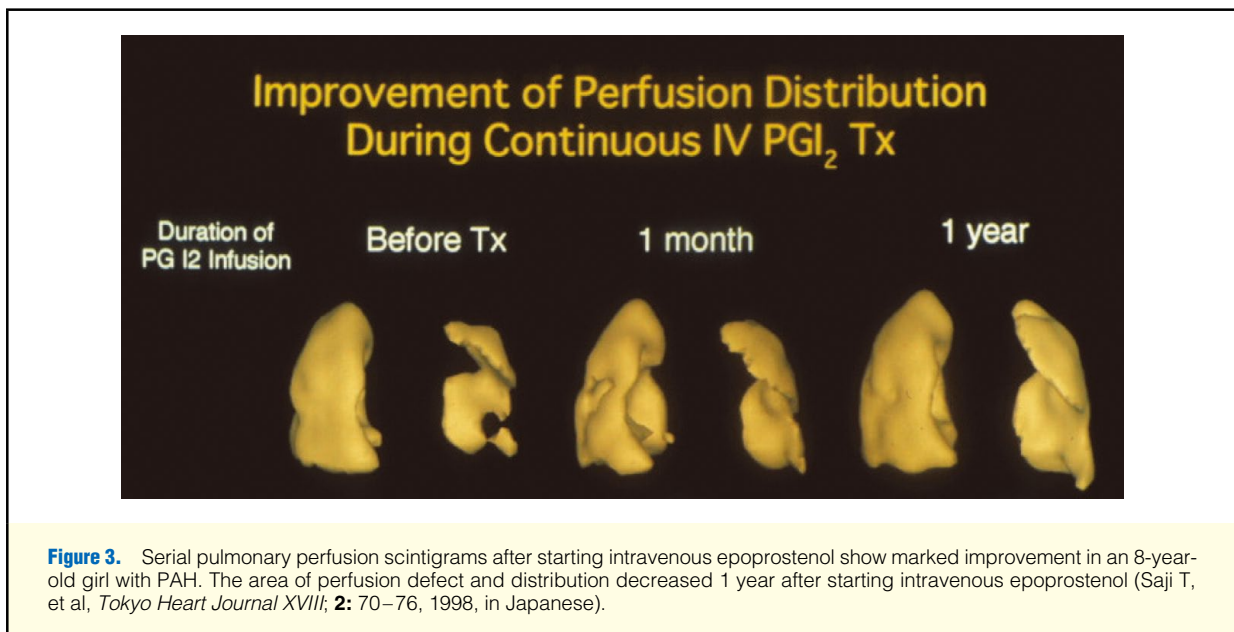


Figure 3. Serial pulmonary perfusion scintigrams after starting intravenous epoprostenol show marked improvement in an 8-year-old girl with PAH. The area of perfusion defect and distribution decreased 1 year after starting intravenous epoprostenol (Saji T, et al, *Tokyo Heart Journal XVIII*; **2**: 70–76, 1998, in Japanese).

are the same as those for adults, but the optimal dose may vary.^{23,42} Subcutaneous treprostinil injection may cause intolerable local pain when given to children. Inhaled iloprost has some efficacy.⁵¹ Currently, subcutaneous, inhaled, and oral PGI₂ are not yet approved for treatment of children in Japan.

In patients with PAH, lung perfusion scintigraphy usually shows abnormal defects, namely, a segmental pattern or non-segmental, mottled pattern. After starting intravenous epoprostenol infusion, perfusion defects markedly improve, probably because of an increase in pulmonary vascular beds (**Figure 3**).

Inhaled PGI₂ (Treprostinil) A study by Krishnan et al⁵² described the safety and efficacy of inhaled treprostinil in children with PAH. Patients must receive 3–9 breaths (6 μg/breath) of inhaled treprostinil 4–6 times per day. In their retrospective analysis, data from 29 children treated with inhaled treprostinil for at least 6 weeks were analyzed. Inhaled treprostinil was discontinued in 4 patients because of cough and bronchospasm (n=3) and PAH progression (n=1). Mild adverse effects, including cough (n=9) and sore throat (n=6), did not require discontinuation of therapy. WHO-FC improved in 19 children and was unchanged in 10; exercise capacity (6MWD) improved during follow-up from 455.7±71.5 to 498±70 m (P=0.01), and peak oxygen consumption increased from 25.5±10.2 to 27.4±10 ml·kg⁻¹·min⁻¹ (P=0.04). Inhaled treprostinil was associated with improved exercise capacity and WHO-FC when added to background targeted therapy for PAH in children and had an acceptable safety profile. In another study of children treated with inhaled iloprost, WHO-FC improved in 35%, remained unchanged in 50%, and decreased in 15%.⁵¹

During intravenous PGI₂ therapy, a high prevalence of autoimmune thyroid disease (AITD) has been reported in adult patients with PAH. Abnormal thyroid function was reviewed in children with IPAH who had been treated with intravenous epoprostenol infusion.⁵³ Thyroid function and antithyroid antibody levels were examined regularly at 6- to 12-month intervals. In total, 7 of 16 patients (44%) had AITD: 2 developed Graves' disease, 2 developed silent thyroiditis, and 3 were euthyroid but had positive tests for antithyroid antibodies.

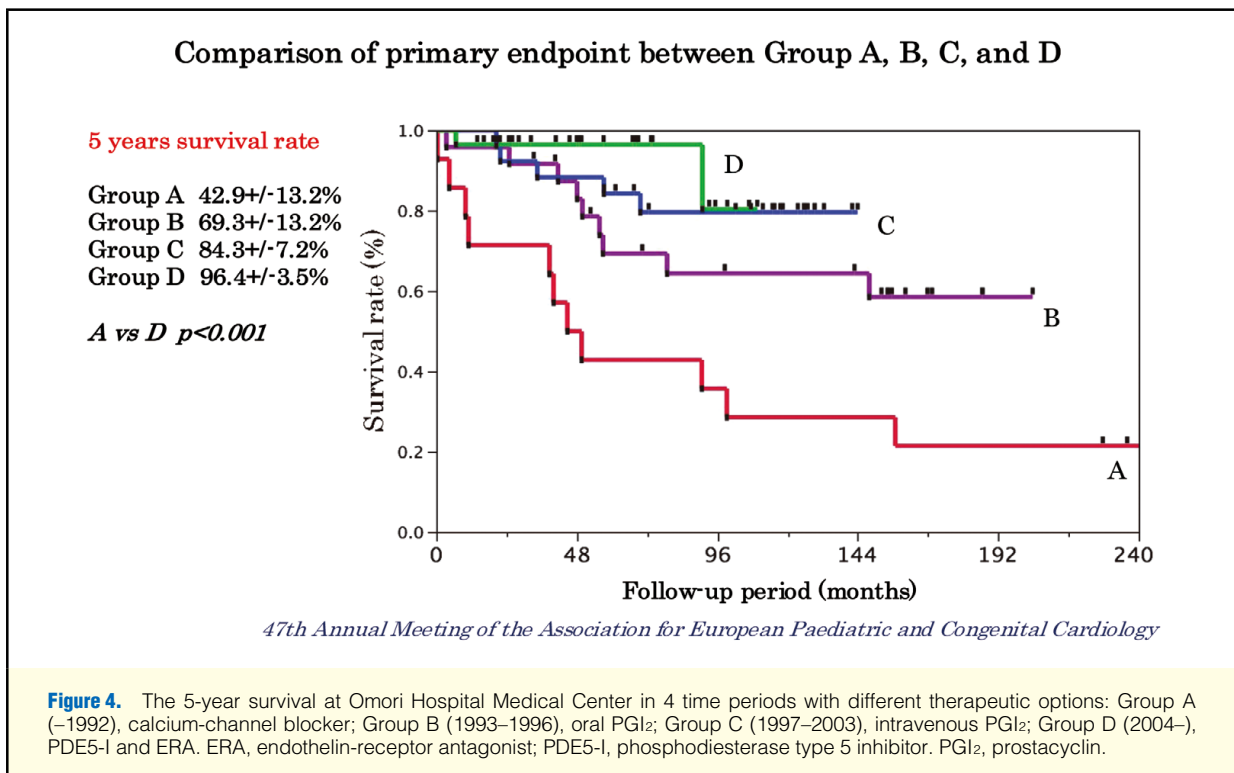
Duration after PAH onset and duration of intravenous epoprostenol treatment were significantly longer in patients with AITD (7.6±2.1 and 7.4±2.3 years, respectively) than in patients without AITD (5.0±1.1 and 4.8±1.2 years, respectively; P<0.01 and P<0.05). The prevalence of AITD was high in children and adolescents with IPAH, so regular evaluation of thyroid function is important in detecting deterioration of heart failure or AITD.⁵³

Phosphodiesterase 5 Inhibitors

Sildenafil Add-on of a PDE5-I or ERA reduced dosing increases of intravenous epoprostenol in children.⁵⁰ Some evidence indicates that PDE5-Is are effective in treating PAH. A phase III multicenter open-label clinical trial of sildenafil in Japan examined data from 21 adult and pediatric patients with PAH.⁵⁴ At week 12, 6MWD increased by 84.2 m from baseline. Mean PAP, PVR, Borg dyspnea score, and plasma BNP also improved. WHO-FC was improved or unchanged in most patients. The 7 patients among whom sildenafil pharmacokinetics were examined had relatively large interindividual variations in maximum and minimum concentrations, area under the plasma concentration-time curve from 0 to 8 h, and average plasma concentration at steady state. Sildenafil 1 mg/kg/day was found to be effective and safe for pediatric IPAH.⁵⁴

During sildenafil therapy in another study, 6MWD increased from 278±114 to 443±107 m at 6 months (P=0.02) and to 432±156 m at 12 months (P=0.005). A plateau was reached between 6 and 12 months (P=0.48). Mean PAP decreased from a median of 60 mmHg (range, 50–105) to 50 mmHg (range, 38–84) (P=0.014). Median PVR decreased from 15 (range, 9–42) to 12 Wood units/m² (range, 5–29) (P=0.024).⁵⁵

A recent study of 3 sildenafil dosages (20, 40, and 80 mg, t.i.d.) for pediatric PAH (STARTS-1) showed improvements in peak oxygen uptake, WHO-FC, and hemodynamics; the moderate and high doses had greater efficacy. An extension study (STARTS-2) found that the moderate sildenafil dosage was optimal (10 mg, t.i.d. for ≥8–20 kg of body weight; 20 mg, t.i.d., for >20–45 kg; 40 mg, t.i.d., for >45 kg), as the high dose level was associated with increased mortality.⁵⁶



Tadalafil A retrospective study⁵⁷ investigated the safety, tolerability, and effects of tadalafil as initial therapy or after transition from sildenafil among 33 children with PAH; 29 were switched from sildenafil to tadalafil. The main reason for the change from sildenafil was the convenience of once-daily dosing for tadalafil. Average dose was $3.4 \pm 1.1 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ for sildenafil and $1.0 \pm 0.4 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{day}^{-1}$ for tadalafil. Among 14 of the 29 patients who underwent repeat catheterization, statistically significant improvements were observed after transition from sildenafil to tadalafil in mPAP (53.2 ± 18.3 vs. $47.4 \pm 13.7 \text{ mmHg}$; $P < 0.05$) and PVR index (12.2 ± 7.0 vs. 10.6 ± 7.2 Woods units/m²; $P < 0.05$). Among the 4 patients treated with tadalafil as initial therapy, clinical improvement was noted, and the side effect profile was similar to that of the patients who had transitioned from sildenafil to tadalafil, including headache, nausea, myalgia, nasal congestion, flushing, and allergic reaction. Two patients discontinued tadalafil because of migraine or allergic reaction. One patient receiving sildenafil had no breakthrough syncope after transition to tadalafil. Tadalafil can be safely used for pediatric patients with PAH, but without significant advantages over sildenafil.⁵⁷ Duration of adverse and main effects was longer than those observed for sildenafil because of the longer half-life of tadalafil.

Endothelin-Receptor Antagonists

Bosentan The pharmacokinetic profile of bosentan is similar between pediatric patients with PAH and healthy adults, and treatment with bosentan results in hemodynamic improvement, which suggest that adult dosing regimens may be appropriate in treating pediatric patients. Patients weighing 10–20 kg, >20–40 kg, and >40 kg received a single dose of 31.25, 62.5, and 125 mg, respectively, on day 1, followed by 4 weeks of treatment at the initial dose.⁵⁸ The pharmacokinetic

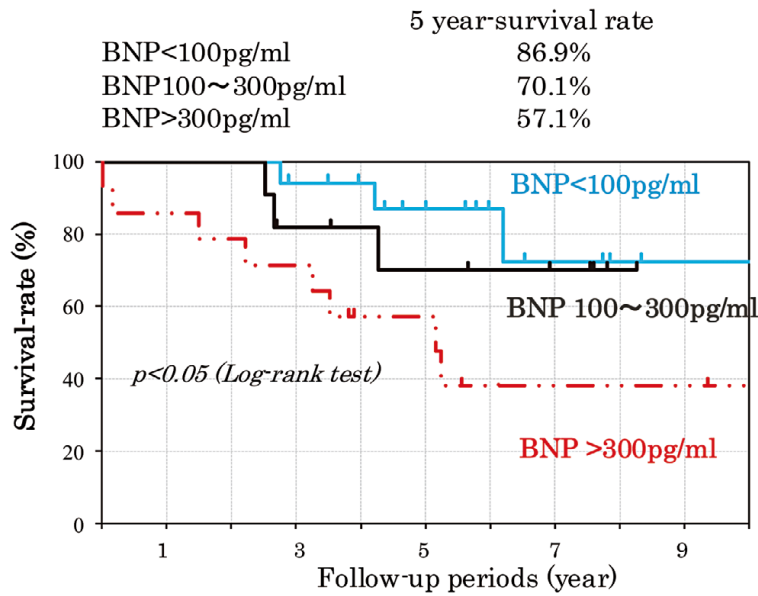
profile of bosentan is not altered by heart failure or coadministration of sildenafil.⁵⁹ Hepatic dysfunction in children is less frequent than in adults.

Ambrisentan The clinical safety and pharmacokinetics of ambrisentan were studied in 38 children with PAH, 15 of whom had switched from bosentan to ambrisentan.⁶⁰ The remaining 23 children were treated with ambrisentan as add-on therapy because of disease progression. In both transition and add-on cases, mPAP significantly improved (transition cases: 55 ± 18 vs. $45 \pm 20 \text{ mmHg}$ [$n=13$], $P=0.04$; add-on cases: 52 ± 17 vs. $45 \pm 19 \text{ mmHg}$ [$n=13$]; $P=0.03$, during follow-up). WHO-FC improved in 31% of patients during the follow-up period (median 20, range, 4–44 months). In total, 5 patients (13%) discontinued ambrisentan because of severe headache, lack of clinical efficacy, or near syncope and 10 patients (26%) had adverse effects associated with ambrisentan treatment, including nasal congestion, headache, and flushing. No patients developed hepatic dysfunction, and there were no deaths after initiation of ambrisentan. Pharmacokinetics were evaluated in 16 children treated with ambrisentan 2.5–10.0 mg. Mean peak plasma concentration was $738 \pm 452 \text{ ng/ml}$, mean time to peak plasma concentration was $3.2 \pm 2.1 \text{ h}$, and mean area under the curve for the plasma concentration was $6,657 \pm 4,246 \text{ ng} \cdot \text{h}^{-1} \cdot \text{ml}^{-1}$.

Rehabilitation

Cardiopulmonary rehabilitation was reported effective in a study of 26 IPAH patients (age, 5–37 years) with severe heart failure.⁶¹ Average duration of cardiopulmonary rehabilitation was 6.7 weeks, and the regimen consisted of breathing exercises, training of upper extremity muscles, gait training, bicycle ergometer training, and treadmill walking for 30–60 min/day, 5 days a week. There was no deterioration in CTR, ANP, or BNP level, tricuspid regurgitation, right ventricular Tei index,

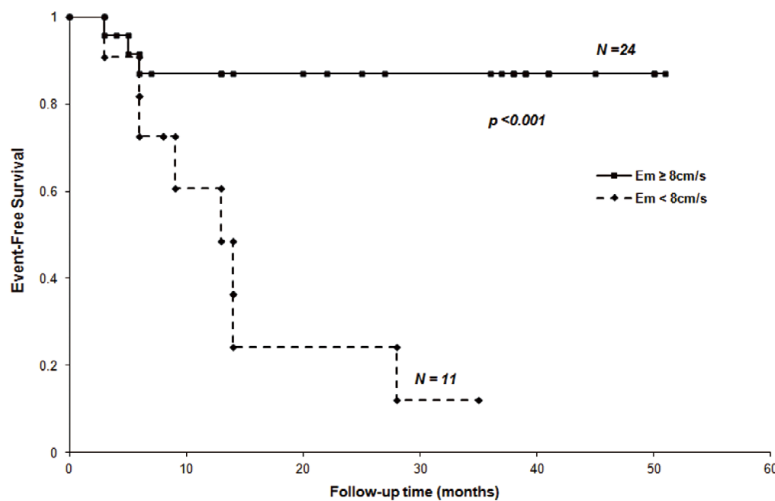
BNP level at baseline and survival rate



47th Annual Meeting of the Association for European Paediatric and Congenital Cardiology

Figure 5. Brain natriuretic peptide (BNP) and survival. The 5-year survival was worse among patients with BNP >300 pg/ml at treatment start than among those with BNP <100 pg/ml.

Tricuspid E' velocity by tissue Doppler imaging at baseline and survival rate



47th Annual Meeting of the Association for European Paediatric and Congenital Cardiology

Figure 6. Event-free survival and tricuspid Em velocity on tissue Doppler imaging at baseline, and survival rate. Cumulative event-free survival was significantly lower when tricuspid Em was 8 cm/s or less (log-rank test, *P* < 0.001).⁶⁵

Table 2. Specific Characteristics of Pediatric PAH as Compared with Adult PAH

Epidemiology	Same prevalence: 1–2 million per year; less connective tissue disease-PAH, less CTEPH, less portal PAH, less ANA positivity
Sex ratio	Reduced female predominance (adults, 3.8–4.0:1; children, 1.0–1.8:1)
Genetics	Same incidence of mutation; BMPR2/Alk1, serotonin transporter; ALK1(HHT) develops with increasing age
Pathology	Less fibrosis, fewer plexiform lesions, more medial hypertrophy, more reversible lesions
Hemodynamics	Better acute response, higher PVR index, higher Pp:Ps ratio, higher CI; higher rate of complications from cardiac catheterization ^{68,69}
Response to acute vasodilating test	Greater AVT response (11–40% responders in children vs. 6–27% in adults); same response to chronic treatment
Clinical manifestations	Earlier diagnosis due to greater physical exercise, less severe NYHA-FC at diagnosis, more PAH crisis, more syncopal episodes, more sudden cardiac death, less heart failure, less edema, lower BNP and NT-proBNP
Treatment response	Same PGI ₂ effect, NO production, ET-1, vWF, factor VIII, and TxA ₂ values. Same incidence of adverse events; slightly higher optimal dose of PGI ₂ ; poor compliance with inhaled iloprost; Same response to SC/inhaled/intravenous PGI ₂ analog
Survival, prognosis	Same or slightly better prognosis and survival

ANA, antinuclear antibody; AVT, acute vasodilatation; BNP, brain natriuretic peptide; CI, cardiac index; CTEPH, chronic thromboembolic pulmonary hypertension; HHT, hereditary hemorrhagic telangiectasia; NO, nitric oxide; NT-proBNP, N-terminal pro-brain natriuretic peptide; PAH, pulmonary arterial hypertension; PVR, pulmonary vascular resistance; Pp:Ps, pulmonary-to-systemic pressure ratio; SC, subcutaneous; TxA₂, thromboxane A₂; vWF, von Willebrand factor.

or pulse oxymetry saturation. The results also showed decreased heart rate at rest ($P=0.007$), and improved NYHA-FC ($P=0.010$), lower extremity strength ($P<0.001$), ambulatory ability ($P<0.001$), Barthel index ($P<0.001$), and 6MWD ($P=0.001$).

Long-Term Survival and Prognosis

In the 1980s, survival in a group of children (average age, 7.4 years) with IPAH was relatively short: 37% at 1 year and 12% at 2.5 years, which was lower than reported in a nationwide prospective study published in 1993.⁶² Another study found that mean survival was 8.7 years in children aged 1–15 years.⁶³

The prognosis for children with IPAH has markedly improved during the past 3 decades, through the use of PGI₂, PDE5-I, and ERA. My group analyzed 92 consecutive patients with IPAH (47 females, 45 males) who had been enrolled since 1978.⁶⁴ Age at onset was 11.7 ± 8.4 years, and median follow-up was 70 months (range, 0.4 months to 28 years). To assess the correlation between available therapeutic options and prognosis, patients were divided into 4 subgroups depending on the era of vasodilator therapy: group A (–1993), B (1994–1999), C (2000–2003), and D (2004–). Patients started receiving oral PGI₂ in 1994, intravenous PGI₂ in 2000, sildenafil in 2004, bosentan in 2005, tadalafil in 2010, and ambrisentan in 2011. In May 2013, the 5-year survival of groups A–D was $42.9\pm 13.2\%$, $69.3\pm 13.2\%$, $84.3\pm 7.2\%$, and $96.4\pm 3.5\%$, respectively (group A vs. group D; $P<0.0001$; **Figure 4**); 64 patients (69.6%) were alive, including 7 (7.6%) who had received lung transplants, and 28 (30.4%) had died, including 1 death after lung transplantation. Mean actuarial survival at 3, 5, and 10 years was $88.8\pm 11\%$, $74.1\pm 26\%$, and $59.7\pm 40\%$, respectively. Their survival rate was significantly better than that for patients with cardiomegaly of 50% or more (Kaplan-Meier curve with log-rank test; $P=0.02$). Results were excellent when advanced treatment was started for patients with a BNP <100 or <200 and NYHA-FC I or II, for those identified during school health screening, and for those without mutations in *BMPR2* or *ALK1* (**Figure 5**).

Tissue Doppler imaging is reported to be quite useful in assessing disease severity and prognosis in children with IPAH. Takatsuki et al evaluated tissue Doppler imaging velocities

(systolic myocardial velocity, early diastolic myocardial relaxation velocity [Em], and late diastolic myocardial velocity associated with atrial contraction), BNP, NYHA-FC, and hemodynamics in 51 children (mean age, 11.6 years) with IPAH.⁶⁵ Em, the ratio of Em to late diastolic myocardial velocity associated with atrial contraction ratio, and systolic myocardial velocity at the mitral annulus, septum, and tricuspid annulus in IPAH were significantly lower than in controls. Tricuspid Em was significantly inversely correlated with plasma BNP ($r=-0.60$, $P<0.001$), right ventricular end-diastolic pressure ($r=-0.79$, $P<0.001$), and mPAP ($r=-0.67$, $P<0.001$). Tricuspid Em significantly differed between patients in NYHA-FC II and those in NYHA-FC III or IV (mean, 11.9 ± 4.2 cm/s vs. 8.2 ± 3.6 cm/s, respectively, $P=0.002$). Cumulative event-free survival was significantly lower when tricuspid Em was 8 cm/s or less (log-rank test, $P<0.001$). Tricuspid Em velocity correlated with disease severity (NYHA-FC) and may be a useful prognostic marker in children with IPAH (**Figure 6**).

Barst et al⁴³ investigated survival in 216 children (age <18 years) with PAH. Survival at 5 years after diagnosis was $74\pm 6\%$ overall, and there was no significant difference between IPAH/FPAH ($75\pm 7\%$) and PAH associated with CHD ($71\pm 13\%$). High PVR index and FPAH were associated with decreased survival. Factors associated with increased survival included acute vasoreactivity and lower BNP. In our study of 92 consecutive pediatric patients with IPAH (47 females, 45 males),⁶⁴ there was no female predominance in incidence or sex difference in age at diagnosis, variety of symptoms, disease severity, response to treatment, or prognosis. The 5-year survival was $96.4\pm 3.5\%$ in a subgroup of patients who started combination therapy with pulmonary vasodilators after 2003.

Despite encouraging data from recent trials of pulmonary vasodilators, there may be no cure for PAH, and lung transplantation is often the only effective treatment.

Adverse Events

Among patients aged 0–18 years, 9,329 adverse events were reported in 588 patients (6.3%).

As part of the USFDA postmarketing surveillance, side effects and adverse events associated with PAH therapy have been documented in children. For bosentan, epoprostenol, and

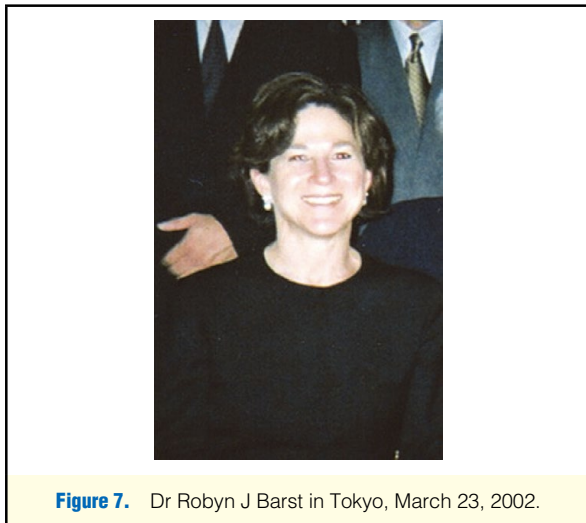


Figure 7. Dr Robyn J Barst in Tokyo, March 23, 2002.

sildenafil, adverse events occurred in at least 5% of patients, and the events were judged to be associated with the targeted PAH medication in children.⁶⁶ The 3 most frequent adverse events were liver dysfunction (62%), cardiac failure (11%), and syncope (8.6%) for bosentan; pulmonary hemorrhage (13.1%), cardiac failure (9.7%), and hemoptysis (8%) for epoprostenol; and cardiac failure (12.4%), hypotension (11.2%), and dyspnea (10.1%) for sildenafil. Serious adverse events not previously reported in the literature (eg, pulmonary hemorrhage, hemoptysis, pneumonia, cardiac arrest, hypoxia) were also reported.

In a small open-label study,⁶⁷ so-called “upfront triple therapy” with bosentan, sildenafil, and intravenous epoprostenol had much better effects on hemodynamics and clinical outcomes than did monotherapy or dual therapy among adults with severe disease. Combination therapy has been recommended for children in NYHA-FC III treated with monotherapy.

We need to collect additional data to develop promising therapies. Improving treatment for pediatric patients will require additional controlled clinical trials and data on the pharmacokinetics of innovative drugs in order to determine the best options with respect to risk-benefit ratio and cost-effectiveness.

Conclusions

Many advances have been made in understanding the genetic background and clinical management of pediatric PAH. As more well-designed clinical trials of innovative drugs are conducted and the extent of research collaborations increases, prognosis and quality of life may improve. Dr Robyn Barst, a well-known expert on PAH in children and adults, maintained that PAH in children appears to have more similarities than differences with PAH in adults (Table 2).³⁸ If so, we may be able to extrapolate from guidelines and criteria for adult PAH patients in devising pediatric clinical algorithms.

Acknowledgments

This review article was written after consulting the numerous published works of experts and my personal collaborators, including Dr Rumiko Matsuoka, Dr Hisato Yagi, Dr Michiko Furutani, Professor Toshio Nakanishi (Heart Institute of Japan, Tokyo Women’s Medical College, Tokyo, Japan), Dr Maya Fujiwara, Dr Tomotaka Nakayama, Dr Shinichi Takatsuki, and Dr Hiroyuki Matsuura (Department of Pediatrics, Toho University Omori Medical Center, Tokyo, Japan). I thank them all for

their ongoing collaboration in basic and clinical research. I also thank Professors Dunbar Ivy (Children’s Hospital Colorado) and Timothy Feltes (Nationwide Children’s Hospital, in Columbus, Ohio) for their scientific advice and critical comments on my work in the field of PAH, Chiaki Goto for her administrative assistance, and David Kipler for reviewing the language of the manuscript.

In Memoriam

I would like to dedicate this work to the memory of Robyn J Barst, MD (Figure 7), FAHA, Professor Emeritus of Columbia University and a distinguished member of the Council on Cardiovascular Disease in the Young, who passed away in April 2013 after a long illness. She devoted her life to the health and happiness of children with PAH.

Dr Barst first visited Japan, with her daughter, in 2002 and presented information that offered new hope for managing PAH. I would like to express my personal appreciation to Robyn, who was a great lover of the healthy food, nature, and culture of Japan, and a gracious colleague to all pediatric cardiologists in this country. She shared her immense knowledge regarding the reliability and safety of newer drugs that offered possibilities to address the challenges of refractory disease and patients with poor prognoses. I will always remember her e-mail response to a question of mine in 2009 regarding the off-label use of a drug for PAH (which was found, 3 year later, to increase the risk of PAH). With great foresight, she urged caution in light of the possible risks.

The drug data is also interesting but again very preliminary and I would not use it off-label in any patients at this time. We did not see any adverse effects on cardiac function in 6 months RCT trial of the drug, but that does not preclude cardiac toxicity long-term and very few patients continued on drug past the 6 months due to our concerns at the time. Thus the long-term open-label extension of trial will be very important.

Disclosures

None.

References

- Saji T, Momma K, Shibata T, Kondo C, Matsuda A, Yasui H, et al. National survey of idiopathic pulmonary arterial hypertension. *Jpn J Pediatr Cardiol Card Surg* 2000; **16**: 230–237.
- Ichida F, Saji T, Kajino H, Ogawa S, Nakanishi T, et al. Annual report of the national survey of cardiovascular rare diseases. *Jpn J Pediatr Cardiol Card Surg* 2010; **26**: 348–350.
- Barst RJ. Primary pulmonary hypertension in children: Primary pulmonary hypertension. In: Rubin LJ, Rich S, editors. Lung biology in health and disease. New York: Marcel Dekker, 1997; **99**: 179–225.
- Lloyd JE, Atkinson JB, Pietra GG, Virmani R, Newman JH. Heterogeneity of pathologic lesions in familial primary pulmonary hypertension. *Am Rev Respir Dis* 1988; **138**: 952–957.
- Lloyd JE, Butler MG, Foroud TM, Conneally PM, Phillips JA 3rd, Newman JH. Genetic anticipation and abnormal gender ratio at birth in familial primary pulmonary hypertension. *Am J Respir Crit Care Med* 1995; **152**: 93–97.
- Xing Y, Song HM, Wu XY, He YY, Wei M. Clinical analysis of pulmonary arterial hypertension secondary to connective tissue disease in children. *Zhonghua Er Ke Za Zhi* 2008; **46**: 822–826 [in Chinese].
- Barst RJ, Flaster ER, Menon A, Fotino M, Morse JH. Evidence for the association of unexplained pulmonary hypertension in children with the major histocompatibility complex. *Circulation* 1992; **85**: 249–258.
- Moride Y, Abenham L, Xu J. Epidemiology of pulmonary hypertension in primary pulmonary hypertension. In: Rubin LJ, Rich S, editors. Primary pulmonary hypertension. New York: Marcel Dekker, 1997; 163–178.
- Umar S, Ravinovitch M, Eghbali M. Estrogen paradox in pulmonary hypertension. *Am J Respir Crit Care Med* 2012; **186**: 125–131.
- Fujiwara M, Yagi H, Matsuoka R, Akimoto K, Furutani M, Imamura S, et al. Implications of mutations of activin receptor-like kinase 1 gene (ALK1) in addition to bone morphogenetic protein receptor II gene (BMP2) in children with pulmonary arterial hypertension. *Circ J* 2008; **72**: 127–133.
- Chida A, Shintani M, Yagi H, Fujiwara M, Kojima Y, Sato H, et al. Outcomes of childhood pulmonary arterial hypertension in BMPR2 and ALK1 mutation carriers. *Am J Cardiol* 2012; **110**: 586–593.
- Shintani M, Yagi H, Nakayama T, Saji T, Matsuoka R. A new nonsense mutation of SMAD8 associated with pulmonary arterial hypertension. *J Med Genet* 2009; **46**: 331–337.
- Chida A, Shintani M, Nakayama T, Furutani Y, Hayama E, Inai K,

- et al. Missense mutations of the BMPR1B (ALK6) gene in childhood idiopathic pulmonary arterial hypertension. *Circ J* 2012; **76**: 1501–1508.
14. Harrison RE, Flanagan JA, Sankelo M, Abdalla SA, Rowell J, Machado RD, et al. Molecular and functional analysis identifies ALK-1 as the predominant cause of pulmonary hypertension related to hereditary haemorrhagic telangiectasia. *J Med Genet* 2003; **40**: 865–871.
 15. Roberts KE, McElroy JJ, Wong WP, Yen E, Widlitz A, Barst RJ, et al. BMPR2 mutations in pulmonary arterial hypertension with congenital heart disease. *Eur Respir J* 2004; **24**: 371–374.
 16. Grünig E, Koehler R, Miltenberger-Miltenyi G, Zimmermann R, Gorenflo M, Meredes D, et al. Primary pulmonary hypertension in children may have a different genetic background than in adults. *Pediatr Res* 2004; **56**: 571–578.
 17. Harrison RE, Berger R, Haworth SG, Tulloh R, Mache CJ, Morrell NW, et al. Transforming growth factor-beta receptor mutations and pulmonary arterial hypertension in childhood. *Circulation* 2005; **111**: 435–441.
 18. Austin ED, Loyd JE. Genetics and mediators in pulmonary arterial hypertension. *Clin Chest Med* 2007; **28**: 43–57.
 19. Rosenzweig EB, Morse JH, Knowles JA, Chada KK, Khan AM, Roberts KE, et al. Clinical implications of determining BMPR2 mutation status in a large cohort of children and adults with pulmonary arterial hypertension. *J Heart Lung Transplant* 2008; **27**: 668–674.
 20. Shimodaira K, Okubo Y, Ochiai E, Nakayama H, Katano H, Wakayama M, et al. Gene expression analysis of a murine model with pulmonary vascular remodeling compared to end-stage IPAH lungs. *Respir Res* 2012; **13**: 103.
 21. Katano H, Ito K, Shibuya K, Saji T, Sato Y, Sata T. Lack of human herpesvirus 8 infection in lungs of Japanese patients with primary pulmonary hypertension. *J Infect Dis* 2005; **191**: 743–745.
 - 21'. Cool CD, Rai PR, Yeager ME, Hernandez-Saavedra D, Serls AE, Bull TM, et al. Expression of human herpesvirus 8 in primary pulmonary hypertension. *N Engl J Med* 2003; **349**: 1113–1122.
 22. Austin ED, Ma L, LeDuc C, Berman Rosenzweig E, Borczuk A, Phillips JA 3rd, et al. Whole exome sequencing to identify a novel gene (caveolin-1) associated with human pulmonary arterial hypertension. *Circ Cardiovasc Genet* 2012; **3**: 336–343.
 23. Barst RJ, Maislin G, Fishman AP. Vasodilator therapy for primary pulmonary hypertension in children. *Circulation* 1999; **99**: 1197–1208.
 24. Yung D, Widlitz AC, Rosenzweig EB, Kerstein D, Maislin G, Barst RJ. Outcome in children with idiopathic pulmonary arterial hypertension. *Circulation* 2004; **110**: 660–665.
 25. Ma L, Roman-Campos D, Austin ED, Eyries M, Sampson KS, Soubrier F, et al. A novel channelopathy in pulmonary arterial hypertension. *N Engl J Med* 2013; **369**: 351–361.
 26. Yeager ME, Nguyen CM, Belchenko DD, Colvin KL, Takatsuki S, Ivy DD, et al. Circulating myeloid-derived suppressor cells are increased and activated in pulmonary hypertension. *Chest* 2012; **141**: 944–952.
 27. Smadja DM, Mauge L, Gaussem P, d'Audigier C, Israel-Biet D, Celermajer DS, et al. Treprostinil increases the number and angiogenic potential of endothelial progenitor cells in children with pulmonary hypertension. *Angiogenesis* 2011; **14**: 17–27.
 28. Yeager ME, Nguyen CM, Belchenko DD, Colvin KL, Takatsuki S, Ivy DD, et al. Circulating fibrocytes are increased in children and young adults with pulmonary hypertension. *Eur Respir J* 2012; **39**: 104–111.
 29. Allen SW, Chatfield BA, Koppenhafer SA, Schaffer MS, Wolfe RR, Abman SH. Circulating immunoreactive endothelin-1 in children with pulmonary hypertension. Association with acute hypoxic pulmonary vasoreactivity. *Am Rev Respir Dis* 1993; **148**: 519–522.
 30. Yoshiyoshi M, Nishioka K, Nakao K, Saito Y, Matsumura M, Ueda T, et al. Plasma endothelin concentrations in patients with pulmonary hypertension associated with congenital heart defects: Evidence for increased production of endothelin in pulmonary circulation. *Circulation* 1991; **84**: 2280–2285.
 31. Nakayama T, Ishikita T, Matsuura H, Saji T. Effects of long-term treatment with prostacyclin on plasma adrenomedullin in patients with primary pulmonary hypertension. *J Cardiol* 2001; **38**: 263–271.
 32. Takatsuki S, Wagner BD, Ivy DD. B-type natriuretic peptide and amino-terminal pro-B-type natriuretic peptide in pediatric patients with pulmonary arterial hypertension. *Congenit Heart Dis* 2012; **7**: 259–267.
 33. Wagenvoort CA, Wagenvoort N. Primary pulmonary hypertension. A pathological study of the lung vessels in 156 clinically diagnosed cases. *Circulation* 1970; **42**: 1163–1184.
 34. Deuthsch GH, Davie NJ, Serls AE, Cool CD, Stenmark KR. Aberrant adventitial changes in a subset of cases with secondary pediatric pulmonary hypertension. Society for Pediatric Pathology annual meeting (poster) Washington, DC. *Lab Invest* 2003; 83.
 35. Cool CD, Deutsch G. Pulmonary arterial hypertension from a pediatric perspective. *Pediatr Dev Pathol* 2008; **11**: 169–177.
 36. Yamaki S, Wagenvoort CA. Comparison of primary plexogenic arteriopathy in adults and children: A morphometric study in 40 patients. *Br Heart J* 1985; **54**: 428–434.
 37. Lévy M, Maurey C, Celermajer DS, Vouhé PR, Danel C, Bonnet D, et al. Impaired apoptosis of pulmonary endothelial cells is associated with intimal proliferation and irreversibility of pulmonary hypertension in congenital heart disease. *J Am Coll Cardiol* 2007; **49**: 803–810.
 38. Barst RJ, Ertel SI, Beghetti M, Ivy DD. Pulmonary arterial hypertension: A comparison between children and adults. *Eur Respir J* 2011; **31**: 665–677.
 39. Barst RJ. Pharmacologically induced pulmonary vasodilatation in children and young adults with primary pulmonary hypertension. *Chest* 1986; **98**: 497–503.
 40. McLaughlin VV, Genthner DE, Panella MM, Rich S. Reduction in pulmonary vascular resistance with long-term epoprostenol (prostanol) therapy in primary pulmonary hypertension. *N Engl J Med* 1998; **338**: 273–277.
 41. Takatsuki S, Parker DK, Doran AK, Friesen RH, Ivy DD. Acute pulmonary vasodilator testing with inhaled treprostinil in children with pulmonary arterial hypertension. *Pediatr Cardiol* 2013; **34**: 1006–1012.
 42. Ivy DD, Saji B. Medical management of severe pediatric pulmonary arterial hypertension. In: Beghetti M, Barst RJ, Berger RMF, et al, editors. Pediatric pulmonary hypertension. Elsevier, 2011; 315–328.
 43. Barst RJ, McGoon MD, Elliott CG, Foreman AJ, Miller DP, Ivy DD. Survival in childhood pulmonary arterial hypertension: Insights from the registry to evaluate early and long-term pulmonary arterial hypertension disease management. *Circulation* 2012; **125**: 113–122.
 44. Adatia I, Barrow SE, Stratton PD, Miall-Allen VM, Ritter JM, Haworth SG. Thromboxane A2 and prostacyclin biosynthesis in children and adolescents with pulmonary vascular disease. *Circulation* 1993; **88**: 2117–2122.
 45. Hashiguchi R. The involvement of vasoactive prostanoids in pulmonary hypertension with congenital heart disease: The investigation of thromboxane A2 and prostaglandin I2. *Jpn J Pediatr Cardiol Card Surg* 1991; **7**: 253–260 [English abstract].
 46. Saji T, Ozawa Y, Ishikita T, Matsuura H, Matsuo N. Short-term hemodynamic effect of a new oral PG12 analog, beraprost, in primary and secondary pulmonary hypertension. *Am J Cardiol* 1996; **78**: 244–247.
 47. Galie N, Manes A, Branzi A. Effects of beraprost sodium, an oral prostacyclin analogue, in patients with pulmonary arterial hypertension: A randomized, double blind, placebo-controlled trial. *J Am Coll Cardiol* 2000; **39**: 1496–1502.
 48. Barst RJ, McGoon M, McLaughlin V, Tapson V, Rich S, Rubin L, et al. Beraprost therapy for pulmonary arterial hypertension. *J Am Coll Cardiol* 2003; **41**: 2119–2125.
 49. Durongpisitkul K, Laoprasitporn D, Layangool T, Sittiwankul R, Panamonta M, Mokrapong P, et al. Comparison of the acute pulmonary vasodilating effect of beraprost sodium and nitric oxide in congenital heart disease. *Circ J* 2005; **69**: 61–64.
 50. Nakayama T, Shimada H, Takatsuki S, Hoshida H, Ishikita T, Matsuura H, et al. Efficacy and limitations of continuous intravenous Epoprostenol therapy for idiopathic pulmonary arterial hypertension in Japanese children. *Circ J* 2007; **71**: 1785–1790.
 51. Ivy DD, Doran AK, Smith KJ, Mallory GB Jr, Beghetti M, Barst RJ, et al. Short- and long-term effects of inhaled iloprost therapy in children with pulmonary arterial hypertension. *J Am Coll Cardiol* 2008; **51**: 161–169.
 52. Krishnan U, Takatsuki S, Ivy DD, Kerstein J, Calderbank M, Coleman E, et al. Effectiveness and safety of inhaled treprostinil for the treatment of pulmonary arterial hypertension in children. *Am J Cardiol* 2012; **110**: 1704–1709.
 53. Satoh M, Aso K, Nakayama T, Naoi K, Ikehara S, Uchino Y, et al. Autoimmune thyroid disease in children and adolescents with idiopathic pulmonary arterial hypertension. *Circ J* 2010; **74**: 371–374.
 54. Satoh T, Saji T, Watanabe H, Ogawa S, Takehara K, Tanabe N, et al. A phase III, multicenter, collaborative, open-label clinical trial of sildenafil in Japanese patients with pulmonary arterial hypertension. *Circ J* 2011; **75**: 677–682.
 55. Humpl T, Reyes JT, Holtby H, Stephens D, Adatia I. Beneficial effect of oral sildenafil therapy on childhood pulmonary arterial hypertension: Twelve-month clinical trial of a single-drug, open-label, pilot study. *Circulation* 2005; **111**: 3274–3280.
 56. Barst RJ, Ivy DD, Gaitan G, Szatmari A, Rudzinski A, Garcia AE, et al. A randomized, double-blind, placebo-controlled, dose-ranging

- study of oral sildenafil citrate in treatment-naive children with pulmonary arterial hypertension. *Circulation*, 2012; **125**: 324–334.
57. Takatsuki S, Calderbank M, Ivy DD. Initial experience with tadalafil in pediatric pulmonary arterial hypertension. *Pediatr Cardiol* 2012; **33**: 683–688.
 58. Barst RJ, Ivy DD, Dingemans J, Widlitz A, Schmitt K, Doran A, et al. Pharmacokinetics, safety, and efficacy of bosentan in pediatric patients with pulmonary arterial hypertension. *Clin Pharmacol Ther* 2003; **73**: 372–382.
 59. Taguchi M, Ichida F, Hirono K, Miyawaki T, Yoshimura N, Nakamura T, et al. Pharmacokinetics of bosentan in routinely treated Japanese pediatric patients with pulmonary arterial hypertension. *Drug Metab Pharmacokinet* 2011; **26**: 280–287.
 60. Takatsuki S, Rosenzweig EB, Zuckerman W, Brady D, Calderbank M, Ivy DD. Clinical safety, pharmacokinetics, and efficacy of ambrisentan therapy in children with pulmonary arterial hypertension. *Pediatr Pulmonol* 2013; **48**: 27–34.
 61. Uchi M, Saji T, Harada T. Feasibility of cardiopulmonary rehabilitation in patients with idiopathic pulmonary arterial hypertension treated with intravenous prostacyclin infusion therapy. *J Cardiol* 2005; **46**: 183–193.
 62. Houde C, Bohn DJ, Freedom RM, Rabinovitch M. Profile of pediatric patients with pulmonary hypertension judged by responsiveness by vasodilator. *Br Heart J* 1993; **70**: 461–468.
 63. Kleinbaum DG, Kupper LL, Morgenstein H. Epidemic research: principles and quantitative. New York: Van Nostrand Reinhold, 1982.
 64. Saji T, Matsuura H, Takatsuki S, Ikehara S, Naoi K, Ozawa T. Dramatic Improvement of the prognosis of idiopathic PAH in the young during the last 3 decade: Predictive factors from a single center experience with 92 cases. *Cardiol Young* 2013; **23**(Suppl 1): S21
 65. Takatsuki S, Nakayama T, Jone PN, Wagner BD, Naoi K, Ivy DD, et al. Tissue Doppler imaging predicts adverse outcome in children with idiopathic pulmonary arterial hypertension. *J Pediatr* 2012; **161**: 1126–1131.
 66. Maxey DM, Ivy DD, Ogawa MT, Feinstein JA. Food and Drug administration (FDA) postmarket reported side effects and adverse events associated with pulmonary hypertension therapy in pediatric patients. *Pediatr Cardiol* 2013; **34**: 1628–1636.
 67. Sitbon O, Simonneau G. Optimal management of severe pulmonary arterial hypertension. *Eur Respir Rev* 2011; **20**: 254–261.