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Effect of treatment with epoprostenol and endothelin receptor antagonists on the development of thyrotoxicosis in patients with pulmonary arterial hypertension

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Abstract. Thyroid disease is known to be associated with pulmonary arterial hypertension (PAH). We investigated the prevalence of thyroid disease in patients with idiopathic PAH (IPAH) or heritable PAH (HPAH), and the factors affecting the pathogenesis of thyroid disease. We retrospectively evaluated 59 patients with IPAH or HPAH who had been diagnosed with PAH before the age of 20 years. Thyrotoxicosis was detected in 12 of the 59 patients (6 patients with Graves' disease, 3 with hashitoxicosis, and 3 with silent thyroiditis) after the start of PAH treatment. The proportion of patients who received epoprostenol in the thyrotoxicosis group was significantly higher than that in the euthyroid group (12/12 vs. 27/47, $p=0.015$). In the 39 patients treated with epoprostenol, the proportion of patients who received combination therapy with epoprostenol and an endothelin receptor antagonist (ERA) in the thyrotoxicosis group was significantly lower than that in the euthyroid group (5/12 vs. 23/27, $p=0.016$). Logistic regression analysis revealed that thyrotoxicosis development was significantly associated with administration of epoprostenol (odds ratio [OR] 8.22, 95% confidence interval [CI] 1.26–53.74, $p=0.028$) and non-administration of ERA (OR 5.33, 95% CI 1.29–22.06, $p=0.021$). The prevalence of thyrotoxicosis was high in patients with IPAH or HPAH. The onset of thyrotoxicosis might be promoted by epoprostenol and inhibited by ERA.

Key words: Pulmonary arterial hypertension, Thyrotoxicosis, Autoimmune thyroid disease, Prostacyclin analogue, Endothelin Receptor Antagonist

PULMONARY ARTERIAL HYPERTENSION (PAH) is a rare and progressive disease associated with a poor prognosis. Obstructions and stenoses of the small pulmonary arterioles result in an increase in pulmonary artery pressure and severe right heart failure. PAH is classified into four groups according to the underlying etiology [1]: idiopathic PAH (IPAH), heritable PAH (HPAH), drug- or toxin-induced PAH, and secondary PAH associated with other disorders such as connective tissue diseases (associated PAH or APAH). Mutations in the genes for bone morphogenetic protein receptor 2 (*BMPR2*), activin receptor-like kinase 1 (*ALK1*), endoglin, *SMAD9*, caveolin-1 (*CAV-1*), *KCNK3*, and *NOTCH1* have been reported to cause sporadic IPAH and HPAH [2].

Thyroid disease is associated with PAH [3–7]. The causes of this association could be that autoimmunity contributes to the pathogenesis of PAH [8] and thyroid disease, and an intravenous prostacyclin analogue epoprostenol, which is a therapeutic agent used for PAH, may trigger thyroid disease [4, 7]. In a previous study, we reported a high prevalence of autoimmune thyroid disease in 16 children and adolescents with IPAH or HPAH [6]. In the present study, we investigated the prevalence of thyroid disease in patients with IPAH or HPAH, and evaluated the factors involved in the pathogenesis of thyroid disease.

Materials and Methods

Participants

We retrospectively studied 59 patients with IPAH or HPAH (45 with IPAH and 14 with HPAH) who had been diagnosed with PAH before the age of 20 years, including 16 patients from a previously reported study [6]. They had been treated at Toho University Omori Medical Center between April 1998 and May 2016.

PAH was diagnosed during right heart catheterization when the mean pulmonary arterial pressure was higher than 25 mmHg and the pulmonary capillary wedge pressure at rest was lower than 15 mmHg.

The median age at the time of diagnosis of PAH was 10.1 years (range, 1.9–19.6 years), and the median disease duration was 7.6 years (range, 0.4–19.4 years). The disease duration was defined as the interval between PAH diagnosis and thyroid dysfunction diagnosis in patients with thyroid dysfunction, or between PAH diagnosis and the most recent clinical visit in patients without thyroid dysfunction. Of the 59 patients, 39 received a continuous intravenous infusion of epoprostenol, a prostacyclin analogue, alone or combined with an endothelin receptor antagonist (ERA) and/or a phosphodiesterase-5 inhibitor (PDE5i). Epoprostenol was administered with a portable infusion pump through a tunneled central venous catheter. The remaining 20 patients were treated with two or all three kinds of the following oral drugs: ERA, PDE5i and a prostacyclin analogue other than epoprostenol. Nine patients died and the 4 patients received lung transplantations.

Methods

Serum free T3 (fT3), free T4 (fT4), and TSH levels were examined at the time of PAH diagnosis, and subsequently, at intervals of 6–12 months, or when right heart failure worsened. Serum levels of antithyroid antibodies

(thyroid peroxidase antibody: TPOAb; thyroglobulin antibody: TgAb; and TSH receptor antibody: TRAb) were evaluated when thyroid dysfunction was detected.

The Mann-Whitney U test, chi-squared test, and logistic regression analysis were used for statistical analysis. A *p*-value <0.05 was considered significant.

The study procedure was approved by the Ethics Committee of Toho University Medical Center, and conducted according to the Ethical Guidelines for Clinical Studies outlined by the Ministry of Health, Labour and Welfare in Japan. The ethics committee approved an opt-out approach instead of obtaining informed consent from each patient.

Results

Thyroid dysfunction was not detected at the time of PAH diagnosis in all patients. Of the 59 patients, 12 (20.3 %) developed thyrotoxicosis after the start of PAH treatment: 6 developed Graves' disease, 3 developed hashitoxicosis, and 3 developed silent thyroiditis. Thyrotoxicosis was diagnosed according to the criteria listed in Table 1. None of the patients developed hypothyroidism due to Hashimoto's disease.

The 59 patients were divided into two groups on the basis of the presence or absence of thyrotoxicosis: a thyrotoxicosis group (*n*=12) and a euthyroid group (*n*=47) (Table 2). There were no significant differences

Table 1 Criteria for diagnosis of thyrotoxicosis

	Graves' disease	Hashitoxicosis	Silent thyroiditis
Serum FT3 and/or FT4 level	Increase	Increase	Increase
Serum TSH level	Decrease	Decrease	Decrease
TRAb	+	–	–
TgAb and/or TPOAb	+ or –	+	–

FT3, free T3; FT4, free T4; TRAb, thyroid-stimulating hormone receptor antibody; TgAb, thyroglobulin antibody; TPOAb, thyroid peroxidase antibody; +, presence; –, absence.

Table 2 Comparison of clinical characteristics of the thyrotoxicosis and euthyroid groups

Clinical characteristics	Thyrotoxicosis group (<i>n</i> =12)	Euthyroid group (<i>n</i> =47)	<i>p</i> value
Male patients/Female patients	5/7	25/22	0.697
Mutation of <i>BMPT2</i> gene +/–	2/8	5/10	0.785
PAH disease duration in years, median (range)	8.3 (3.1–13.6)	7.5 (0.4–19.4)	0.970
PAH therapy, number of patients (%)			
Epoprostenol	12 (100)	27 (57.4)	0.015
Endothelin receptor antagonist	5 (41.7)	36 (76.6)	0.046
Phosphodiesterase-5 inhibitor	10 (83.3)	45 (95.7)	0.377

Disease duration was defined as the interval between the diagnosis of PAH and the diagnosis of thyroid dysfunction for the thyrotoxicosis group, and between the diagnosis of PAH and the most recent clinical visit for the euthyroid group. PAH, pulmonary arterial hypertension; BMPT2, bone morphogenetic protein receptor 2; +, presence; –, absence.

between the two groups in patient sex, the frequency of *BMP2* mutation-positive patients, or disease duration. All 12 patients in the thyrotoxicosis group were treated with epoprostenol. The proportion of patients who received epoprostenol in the thyrotoxicosis group was significantly higher than that in the euthyroid group (12/12 vs. 27/47, respectively; $p=0.015$).

The 39 patients treated with epoprostenol were divided into two groups on the basis of the presence or absence of thyrotoxicosis: a thyrotoxicosis group ($n=12$) and an epo-euthyroid group ($n=27$) (Table 3). There were no significant differences between the two groups in patient sex or the frequency of *BMP2* mutation-positive patients. In regard to epoprostenol, there were no significant differences between the two groups in the duration of epoprostenol treatment, the maximum epoprostenol dose, or the cumulative epoprostenol dose. The proportion of patients who received combination therapy with epoprostenol and ERA in the thyrotoxicosis group was significantly lower than that in the epo-euthyroid group (5/12 vs. 23/27, respectively; $p=0.016$). The median duration of epoprostenol treatment without ERA in the thyrotoxicosis group (5.1 years) was longer than that in the epo-euthyroid group (2.5 years), but this difference was not significant ($p=0.248$).

In Japan, epoprostenol, ERA, and PDE5i became available in 1999, 2005, and 2008, respectively. Therefore, the therapeutic strategy for PAH has changed, and it has become possible for PAH patients to receive a combination therapy after 2005. The proportion of patients who received epoprostenol treatment in the patients diagnosed with PAH before 2004 was significantly higher than that in the patients diagnosed with PAH after 2005 (27/29 vs. 12/30, respectively; $p<0.001$). The proportion of patients who developed thyrotoxicosis in the patients diagnosed with PAH before 2004 was significantly higher than that in the patients diagnosed with PAH after 2005 (10/29 vs. 2/30, respectively; $p=0.020$). The 27 patients who had been diagnosed with PAH before 2004 and received epoprostenol treatment were divided into two groups on the basis of the presence or absence of thyrotoxicosis: a thyrotoxicosis before 2004 group ($n=10$) and an epo-euthyroid before 2004 group ($n=17$) (Table 4). Although the significant differences were not found, the median duration of epoprostenol treatment was short and the median cumulative dose of epoprostenol was small in the thyrotoxicosis before 2004 group in comparison with the epo-euthyroid before 2004 group. On the other hand, the proportion of patients who received combination therapy with epoprostenol and ERA in

Table 3 Comparison of clinical characteristics of the thyrotoxicosis and epo-euthyroid groups in the 39 patients treated with epoprostenol

Clinical characteristics	Thyrotoxicosis group ($n=12$)	Epo-euthyroid ($n=27$)	p value
Age at the diagnosis of thyroid dysfunction or at the most recent clinical visit (years)	17.0 (12.5–28.1)	17.5 (9.0–33.6)	0.808
Male patients/Female patients	5/7	13/14	0.979
Mutation of <i>BMP2</i> gene +/–	2/8	4/9	0.917
Age at the time of diagnosis of PAH (years)	10.7 (4.1–14.5)	8.2 (2.5–17.6)	0.330
Disease duration of PAH (years)	8.3 (3.1–13.6)	9.9 (0.4–19.4)	0.563
Epoprostenol treatment			
Age at the time of start of epoprostenol treatment (years)	12.0 (5.4–18.8)	9.9 (3.3–17.6)	0.605
Duration of epoprostenol treatment (years)	6.8 (1.5–13.3)	6.4 (0.3–16.6)	0.819
Maximum dose of epoprostenol (ng/kg/min)	29.5 (13.0–49.0)	24.0 (4.5–65.0)	0.808
Cumulative dose of epoprostenol (mg/kg)	70.9 (8.9–179.6)	72.3 (0.4–287.0)	0.930
Combination therapy			
Combination therapy with ERA, number of patients (%)	5 (41.7)	23 (84.2)	0.016
Combination therapy with PDE5i, number of patients (%)	10 (83.3)	27 (100.0)	0.164
Duration of epoprostenol treatment without ERA (years)	5.1 (0–10.5)	2.5 (0–11.3)	0.248
Duration of epoprostenol treatment without PDE5i (years)	2.2 (0–7.1)	0.9 (0–8.4)	0.927
Duration of isolated epoprostenol treatment (years)	2.2 (0–7.1)	0.9 (0–5.2)	0.879

Data are shown median value (range) unless otherwise indicated. Disease duration was defined as the interval between the diagnosis of PAH and the diagnosis of thyroid dysfunction for the thyrotoxicosis group, and between the diagnosis of PAH and the most recent clinical visit for the epo-euthyroid group. PAH, pulmonary arterial hypertension; BMP2, bone morphogenetic protein receptor 2; +, presence; –, absence; ERA, endothelin receptor antagonist; PDE5i, phosphodiesterase-5 inhibitor.

Table 4 Comparison of clinical characteristics of the thyrotoxicosis before 2004 and epo-euthyroid before 2004 groups in the 27 patients treated with epoprostenol, who had been diagnosed with PAH before 2004

Clinical characteristics	Thyrotoxicosis before 2004 group (n=10)	Epo-euthyroid before 2004 group (n=17)	<i>p</i> value
Age at the diagnosis of thyrotoxicosis or at the most recent clinical visit (years)	17.5 (12.5–28.1)	19.7 (11.5–33.6)	0.581
Male patients/Female patients	6/4	8/9	0.802
Mutation of <i>BMPR2</i> gene +/–	2/7	3/6	1.000
Age at the time of diagnosis of PAH (years)	11.1 (4.1–14.5)	8.1 (2.9–17.6)	0.366
Disease duration of PAH (years)	8.7 (3.1–13.6)	12.8 (2.7–19.4)	0.027
Epoprostenol treatment			
Age at the time of start of epoprostenol treatment (years)	12.0 (5.4–18.8)	9.4 (3.3–17.6)	0.482
Duration of epoprostenol treatment (years)	6.8 (1.5–13.3)	9.7 (1.2–16.6)	0.079
Maximum dose of epoprostenol (ng/kg/min)	26.4 (13.0–44.0)	24.0 (9.3–65.0)	1.000
Cumulative dose of epoprostenol (mg/kg)	64.6 (8.9–179.6)	84.2 (17.8–287.0)	0.130
Combination therapy			
Combination therapy with ERA, number of patients (%)	3 (30.0)	13 (76.5)	0.049
Combination therapy with PDE5i, number of patients (%)	8 (80.0)	17 (100.0)	0.248
Duration of epoprostenol treatment without ERA (years)	6.1 (0–10.5)	5.2 (0–11.3)	0.393
Duration of epoprostenol treatment without PDE5i (years)	2.5 (0–7.1)	2.3 (0–8.4)	0.802
Duration of isolated epoprostenol treatment (years)	2.5 (0–7.1)	1.8 (0–5.2)	0.725

Data are shown median value (range) unless otherwise indicated. Disease duration was defined as the interval between the diagnosis of PAH and the diagnosis of thyroid dysfunction for the thyrotoxicosis before 2004 group, and between the diagnosis of PAH and the most recent clinical visit for the epo-euthyroid before 2004 group; *BMPR2*, bone morphogenetic protein receptor 2; +, presence; –, absence; ERA, endothelin receptor antagonist; PDE5i, phosphodiesterase-5 inhibitor.

the thyrotoxicosis before 2004 group was significantly lower than that in the epo-euthyroid before 2004 group (3/10 vs. 13/17, respectively; $p=0.049$).

Logistic regression analysis revealed that the development of thyrotoxicosis was significantly associated with administration of epoprostenol (odds ratio [OR] 8.22, 95% confidence interval [CI] 1.26–53.74, $p=0.028$) and non-administration of ERA (OR 5.33, 95% CI 1.29–22.06, $p=0.021$).

Clinical characteristics, examination findings, diagnoses, and outcomes of 12 patients with thyrotoxicosis are shown in Table 5. The median age at the time of diagnosis of thyrotoxicosis was 17.0 years (range, 12.5–28.1 years). The reasons for the evaluation of thyroid function at the time of diagnosis of thyrotoxicosis were worsening of right heart failure in 5 of 12 patients, who were all diagnosed with Graves' disease, and routine check-up in the remaining 7 patients. Thyroid enlargement was observed on thyroid ultrasonography in all patients except for Patient no. 2. Of the 6 patients with Graves' disease, 4 showed increased blood flow in the thyroid gland on ultrasonography. Patient no. 1 developed agranulocytosis (neutrophil count, 396/ μ L) 2 months after the start of methimazole, which was replaced with potassium iodide. She had

received potassium iodide treatment for 9.4 years, and underwent thyroidectomy after amelioration of right heart failure. Patient no. 9 underwent thyroidectomy for a massive goiter 3.4 years after the development of Graves' disease. Patient no. 5, 6, and 7 died because of worsening of right heart failure after the development of Graves' disease. Several thyrotoxicosis recurrences occurred in all patients with hashitoxicosis. In contrast, all patients with silent thyroiditis experienced only one episode of thyrotoxicosis. The ERA treatment was started after the onset of thyrotoxicosis in 6 of 7 patients who had not received ERA treatment before the onset of thyrotoxicosis.

The mortality rate in the thyrotoxicosis group (4/12, 33.3 %) was higher than that in the euthyroid group (5/47, 10.6 %) and in the epo-euthyroid group (4/27, 14.8 %), but this difference was not significant ($p=0.131$ and $p=0.372$, respectively). Since the patients who had been diagnosed with PAH before 2004 and received epoprostenol treatment were followed up for a long term, the prognosis of them was studied. The ratio of the lung transplantation and/or death in the thyrotoxicosis before 2004 group (2/10, 20.0 %) was similar with that in the epo-euthyroid before 2004 group (3/17, 17.6 %) ($p=1.000$). There was no significant difference in

Table 5-a Clinical characteristics of 12 patients with pulmonary arterial hypertension and thyrotoxicosis

Patient No	Sex	PAH subtype	Genetic mutation	Family history of autoimmune thyroid disease	At the diagnosis of thyrotoxicosis						Combination therapy with ERA / PDE5i
					Age (years)	Reason for the evaluation of thyroid function	Disease duration of PAH (years)	Duration of epoprostenol treatment (years)	Maximum dose of epoprostenol (ng/kg/min)	Cumulative dose of epoprostenol (mg/kg)	
1	F	IPAH	<i>NOTCH1</i>	—	12.5	Worsening of RHF	7.7	7.1	43.7	103.8	— / —
2	M	IPAH	—	NA	12.8	Routine check-up	6.2	5.8	23.0	55.4	— / +
3	F	IPAH	—	+	15.5	Routine check-up	3.1	2.3	13.0	12.0	— / —
4	F	IPAH	—	—	15.9	Worsening of RHF	11.8	7.3	32.0	93.5	— / +
5	F	IPAH	—	+	15.9	Worsening of RHF	4.5	4.3	29.0	27.3	— / +
6	F	IPAH	NA	+	17.1	Worsening of RHF	8.8	8.7	30.0	114.5	+ / +
7	F	IPAH	—	NA	16.9	Worsening of RHF	7.6	4.5	49.0	68.0	+ / +
8	M	HPAH	<i>BMPR2</i>	—	19.0	Routine check-up	7.2	6.4	15.0	45.4	— / +
9	M	IPAH	—	—	20.2	Routine check-up	9.6	1.5	23.7	8.9	+ / +
10	M	IPAH	NA	NA	21.1	Routine check-up	10.5	10.3	17.0	73.8	— / +
11	M	HPAH	<i>BMPR2</i>	NA	23.0	Routine check-up	10.8	10.6	44.0	179.6	+ / +
12	F	HPAH	<i>ALK1</i>	+	28.1	Routine check-up	13.6	13.3	30.0	159.5	+ / +

PAH, pulmonary arterial hypertension; IPAH, idiopathic pulmonary arterial hypertension; HPAH, heritable pulmonary arterial hypertension; BMPR2, bone morphogenetic protein receptor 2; ALK1, activin receptor-like kinase 1; RHF, right heart failure; ERA, endothelin receptor antagonist; PDE5i, phosphodiesterase-5 inhibitor; NA, not available; +, presence; —, absence.

Table 5-b Examination findings, diagnoses and treatment of 12 patients with pulmonary arterial hypertension and thyrotoxicosis at the diagnosis of thyrotoxicosis

Patient No	TSH (μIU/mL)	Free T4 (ng/dL)	Free T3 (pg/mL)	TRAb (%)	TRAb (IU/mL)	TgAb (IU/mL)	TPOAb (IU/mL)	BNP (pg/mL)	Thyroid ultrasound Enlargement / Blood flow		Diagnosis	Treatment
1	0.01	>7.77	16.84	86.7	NA	NA	NA	338.2	+	Increase	Graves' disease	MMI
2	0.01	1.66	4.31	6.1	NA	<0.3	<0.3	99.2	—	No increase	Silent thyroiditis	Observation
3	0.22	1.68	5.03	NA	NA	53.68	26.91	14.1	+	No increase	Hashitoxicosis	Observation
4	<0.01	3.38	10.08	18.9	NA	15.6	242.0	329.6	+	Increase	Graves' disease	MMI
5	0.01	3.44	7.46	77.5	NA	<0.3	<0.3	635.4	+	No increase	Graves' disease	MMI β-blocker
6	<0.01	3.42	7.08	NA	4.32	12.55	10.94	581.3	+	(mild) Increase	Graves' disease	MMI
7	0.01	2.51	5.05	NA	3.47	24.84	9.06	474.4	+	No increase	Graves' disease	MMI
8	<0.01	3.09	9.94	8.3	NA	<0.3	<0.3	28.9	+	(mild) No increase	Silent thyroiditis	Observation
9	<0.01	1.66	10.47	NA	5.12	15.87	5.63	32.2	+	Increase	Graves' disease	MMI
10	0.01	4.17	13.91	NA	<0.3	47.69	6.67	7.8	+	(mild) No increase	Hashitoxicosis	β-blocker
11	0.01	2.03	5.93	NA	0.519	637.9	236.2	29.5	+	No increase	Hashitoxicosis	Observation
12	<0.01	1.87	5.58	NA	<0.3	NA	NA	77.4	+	No increase	Silent thyroiditis	β-blocker

TRAb, thyroid-stimulating hormone receptor antibody; TgAb, thyroglobulin antibody; TPOAb, thyroid peroxidase antibody; BNP, brain natriuretic peptide; NA, not available; MMI, methimazole; +, presence; —, absence. Reference values: TSH, 0.32–4.12 μIU/mL; Free T4, 1.01–1.67 ng/dL; Free T3, 2.26–4.15 pg/mL; TRAb, <10 %, TRAb, <2.0 IU/mL; TgAb, <28 IU/mL; TPOAb, <16 IU/mL. BNP, <18.4 pg/mL.

Table 5-c Outcome of 12 patients with pulmonary arterial hypertension and thyrotoxicosis after the diagnosis of thyrotoxicosis

Patient No	Diagnosis	Duration of observation after the diagnosis of thyrotoxicosis	Outcome of thyrotoxicosis	Final outcome
1	Graves' disease	10.1	Thyroidectomy	Clinically stable
2	Silent thyroiditis	7.3	Remission	Death
3	Hashitoxicosis	11.0	Three recurrences	Clinically stable
4	Graves' disease	7.7	One recurrence	Clinically stable
5	Graves' disease	3.7	Euthyroid after the cessation of MMI	Death after the lung transplantation
6	Graves' disease	0.5	TRAb-negative conversion Cessation of MMI Euthyroid sick syndrome due to severe heart failure	Death
7	Graves' disease	1.9	Unknown because of changing hospital	Death
8	Silent thyroiditis	8.3	Remission	Clinically stable
9	Graves' disease	3.6	Thyroidectomy	Clinically stable
10	Hashitoxicosis	3.1	Two recurrences	Clinically stable
11	Hashitoxicosis	3.9	One recurrence	Clinically stable
12	Silent thyroiditis	2.2	Remission	Clinically stable

MMI, methimazole; TRAb, thyroid-stimulating hormone receptor antibody.

the lung transplantation-free survival period after the start of epoprostenol treatment between the thyrotoxicosis before 2004 group (median, 13.6 years; range, 7.9–17.2 years) and the epo-euthyroid before 2004 group (median, 12.0 years; range, 1.2–16.6 years) ($p=0.422$).

Discussion

The prevalence of thyrotoxicosis in Japanese adults has been reported to be approximately 0.6 % [9]. Therefore, the prevalence of thyrotoxicosis in patients with IPAH or HPAH in the present study (20.3 %) was significantly higher than that in the general Japanese population ($p<0.001$). All patients with thyrotoxicosis received epoprostenol treatment. Some studies have suggested that epoprostenol treatment is associated with the development of non-autoimmune thyrotoxicosis [5] and autoimmune thyrotoxicosis [3, 4, 6, 7]. Prostacyclin stimulates adenylate cyclase activity and increases cyclic adenosine monophosphate (cAMP) synthesis in thyroid follicular cells *via* a TSH-independent system. Therefore, prostacyclin may induce non-autoimmune thyrotoxicosis [10, 11].

Three kinds of drugs are used for PAH treatment: prostacyclin analogues, ERA, and PDE5i. Prostacyclin increases cAMP synthesis by stimulating adenylate cyclase activity [12], ERA blocks endothelin activity, and PDE5i increases cyclic guanosine monophosphate (cGMP) levels by inhibiting the degradation of cGMP. The vasodilating and antiproliferative effects of these three drugs in pulmonary blood vessels are useful for PAH treatment. In addition to these effects, these three drugs also act on the immune system. The T-helper 17 (Th17)–interleukin (IL)-17 axis plays important roles in the pathogenesis of autoimmune disease. Prostacyclin accelerates the differentiation of naïve T cells into Th17 cells and enhances Th17 cell function [13, 14], endothelin increases the production of IL-17 from Th17 cells [15, 16], and PDE5 accelerates the differentiation of naïve T cells into Th17 cells [17]. Thus, prostacyclin analogues may accelerate the development of autoimmune diseases, whereas inhibitors of endothelin and PDE5 may prevent the development of autoimmune diseases.

Increased functionality of the Th17–IL-17 axis has also been reported in autoimmune thyroid diseases. Circulating Th17 lymphocyte levels increase in patients with intractable Graves' disease [18], and levels of circulating Th17 lymphocytes and serum IL-17

increase in patients with newly diagnosed Graves' disease [19] and Hashimoto's disease [20]. Moreover, IL-17 has been reported to contribute to the development of autoimmune thyroid diseases in mouse models of both Graves' disease [21] and in Hashimoto's disease [22]. Silent thyroiditis is also caused by an autoimmune mechanism similar to that involved in the development of Graves' disease and Hashimoto's disease. The present study results suggest that the development of thyrotoxicosis is promoted by intravenous epoprostenol administration and inhibited by ERA treatment. This may reflect the facilitatory effect of epoprostenol and inhibitory effect of ERA on the function of the Th17–IL-17 axis.

In the present study, the patients treated with oral prostacyclin analogue did not develop thyroid dysfunction. Oral prostacyclin analogue is not recommended as a PAH therapeutic agent because currently available evidence indicates that its efficacy in PAH is limited [12]. Therefore, the effect of oral prostacyclin analogues on the immune system may also be limited. In contrast, since almost all patients (55 out of 59 patients) were treated with PDE5i, the prevalence of thyrotoxicosis did not differ in the presence and absence of PDE5i treatment.

A mutation in *BMPR2* is reported to be one of the causes of sporadic IPAH and HPAH. An association has previously been reported between thyroid disease and mutations in *BMPR2* [23]. However, in the present study, no differences were found in the frequency of *BMPR2* mutation-positive patients between the thyrotoxicosis group and the euthyroid, thyrotoxicosis, and epo-euthyroid groups.

The prevalence of family history of autoimmune thyroid disease in 12 patients with thyrotoxicosis was high (4/12, 33 %). It is unknown whether the high frequency of family history of autoimmune thyroid disease is characteristic of the patients with IPAH or HPAH or not, because the family history of autoimmune thyroid disease was not checked in the patients without thyroid dysfunction. Recent studies have indicated that the promotion of Th17 cell function is involved in the pathogenesis of IPAH and HPAH [24, 25]. Therefore, the pathogenesis of IPAH and HPAH itself may predispose the patients to the development of autoimmune diseases. However, other than autoimmune thyroid disease no other autoimmune diseases were detected in the patients with PAH in the present study. Only a few reports have described the comorbidity of IPAH

or HPAH with autoimmune diseases other than autoimmune thyroid disease: two IPAH patients developed polyglandular autoimmune syndromes [26, 27] and one IPAH patient developed autoimmune hepatitis [28] after the start of PAH treatment. The reason for the low incidence of IPAH or HPAH complicated with autoimmune diseases other than autoimmune thyroid disease is unclear.

No significant differences in mortality rate were found between the thyrotoxicosis group and the euthyroid, thyrotoxicosis, and epo-euthyroid groups. However, the 3 patients with Graves' disease died because of worsening of right heart failure after the development of thyrotoxicosis. Therefore, it is very important to routinely monitor thyroid function, especially immediately after worsening of right heart failure, in order to avoid the progression of PAH, as previously reported [7].

In conclusion, we found a high prevalence of thyrotoxicosis in patients with IPAH or HPAH. Our results indicate that the onset of thyrotoxicosis is promoted by intravenous prostacyclin analogue treatment and inhibited by endothelin receptor antagonists. Further studies are needed to evaluate the relationship between the drugs used for PAH treatment and the immune system, especially Th17–IL17 axis function. It is important to regularly evaluate thyroid function in patients with PAH, particularly those receiving epoprostenol without ERA.

Discosure

Saji T is affiliated with the endowed chair supported by Actelion Pharmaceuticals Japan. Satoh M, Aso K and Nakayama T declare that there is no potential conflict of interest associated with this research.

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