The Clinical Efficacy of Endothelin Receptor Antagonists in Patients with Pulmonary Arterial Hypertension
Comparison Between Each Generation

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Summary
Therapeutic strategies for pulmonary arterial hypertension (PAH) have made remarkable progress over the last two decades. Currently, 3 types of drugs can be used to treat PAH; prostacyclins, phosphodiesterase 5 inhibitors, and endothelin receptor antagonists (ERA). In Japan, the first generation ERA bosentan was reimbursed in 2005, following which the 2nd generation ERAs ambrisentan and macitentan were reimbursed in 2009 and 2015, respectively. The efficacy of each ERA on hemodynamics in PAH patients remains to be elucidated. The aims of this study were to evaluate the hemodynamic effects of ERAs and compare these effects among each generation of ERAs.

We retrospectively examined the clinical parameters of 42 PAH patients who were prescribed an ERA (15 bosentan, 12 ambrisentan, and 15 macitentan) and who underwent a hemodynamic examination before and after ERA introduction at our institution from January 2007 to July 2019.

In a total of 42 patients, mean pulmonary arterial pressure (mPAP) and pulmonary vascular resistance (PVR) were significantly decreased and cardiac index was significantly increased after ERA introduction (P < 0.001) and the World Health Organization-Functional class (WHO-Fc) was significantly improved after ERA introduction (P = 0.005). Next, in a comparison between 1st and 2nd generation ERAs, 2nd generation ERAs were found to have brought about greater improvements in hemodynamic parameters (mPAP and PVR. P < 0.01), heart rate, brain natriuretic peptide, arterial oxygen saturation, and mixed venous oxygen saturation than the 1st generation ERA bosentan.

We conclude that all ERAs could successfully improve the hemodynamics of PAH patients and that the newer generation ERAs, ambrisentan and macitentan, seemed to be preferable to bosentan.

Key words: Bosentan, Ambrisentan, Macitentan, Hemodynamics

 Pulmonary hypertension used to have a poor prognosis and few effective treatments were available. Remarkable progress has been made in the treatment of pulmonary arterial hypertension (PAH) in the last two decades. Currently available therapeutic agents for PAH exert their efficacy in the pulmonary vascular endothelium and media by modifying mainly 3 pathways; the prostacyclin-adenylate cyclase-cyclic adenosine monophosphate pathway, nitric monoxide (NO)-guanylate cyclase-cyclic guanosine monophosphate pathway, and endothelin (ET) pathway. Prostacyclin and nitric oxide were discovered to be relaxing factors produced in the endothelium in 1976 and 1987, respectively,1,2 ET was first reported to be an endothelium-derived potent vasoconstrictor by Yanagisawa, et al in 1988.3 ET family peptides are 21-residue peptides including ET-1, ET-2, and ET-3. Among the ETs, ET-1 is the most potent vasoconstrictive subtype and deeply related to the pathophysiology of PAH.4 Indeed, circulating ET levels were found to be elevated5 and local pulmonary production of ET-1 was reported to be elevated in PAH patients.6 There are two subtypes of ET receptors (ETα receptor and ETβ receptor) in mammals. The ETα receptor is involved in vasoconstriction, cell growth, and inflammation, while the ETβ receptor is involved in vasodilation, increasing sodium excretion in the kidney, and inhibition of cell growth and inflammation.7,8 ET-1 affects both ETα and ETβ receptors on smooth muscle cells (SMC) in the pulmonary vasculature, leading to...
SMC constriction and proliferation, whereas ET-1 might promote vasodilation of the pulmonary vasculature via ET<sub>A</sub> receptors on vascular endothelial cells through NO synthesis. However, the effect derived from endothelial ET<sub>A</sub> receptors is considered to be minute, and ET receptor blockade is an important therapeutic target for decreasing pulmonary arterial pressure in PAH patients. The first ET receptor antagonist (ERA) bosentan is a dual ET<sub>A</sub> and ET<sub>B</sub> antagonist, whereas ET-1 might promote vasodilation of the pulmonary vasculature via ET<sub>B</sub> receptors.

Table 1. Baseline Characteristics of PAH Patients

|                      | Total ERA (n = 42) | 1st generation ERA (n = 15) | 2nd generation ERA (n = 27) | P value |
|----------------------|-------------------|-----------------------------|-----------------------------|---------|
| Age (years)          | 50.8 ± 15.4       | 52.9 ± 15.9                 | 49.6 ± 15.3                 | 0.51    |
| Sex (female:male)    | 29:13             | 10:5                        | 19:8                        | 0.54    |
| Heart rate (beats/minute) | 73.4 ± 13.5   | 74.7 ± 13.1                 | 72.7 ± 14.7                 | 0.67    |
| Hb (g/dL)            | 12.9 (11.9, 14.5) | 12.5 (11.7, 14.5)           | 13.3 (12.5, 14.5)           | 0.38    |
| BNP (pg/mL)          | 71.0 (24.8, 127.0)| 59.5 (28.8, 250.5)          | 72.5 (23.1, 120.0)          | 0.55    |
| mean BP (mmHg)       | 85.8 ± 11.9       | 85.1 ± 13.4                 | 86.3 ± 11.2                 | 0.76    |
| mean RAP (mmHg)      | 5.0 (4.0, 7.0)    | 5.0 (3.0, 7.0)              | 5.0 (4.0, 7.0)              | 0.95    |
| mean PAWP (mmHg)     | 8.3 ± 2.9         | 7.7 ± 2.7                   | 8.7 ± 3.0                   | 0.28    |
| mean PAP (mmHg)      | 38.0 (32.8, 45.0)| 36.0 (31.0, 43.0)           | 39.0 (34.0, 45.0)           | 0.45    |
| CI (L/minute/m²)     | 2.6 (2.4, 3.0)    | 2.6 (2.4, 3.1)              | 2.6 (2.5, 3.0)              | 0.72    |
| PVR (dyne-second/cm<sup>5</sup>) | 573.4 (399.5, 687.0) | 628.0 (400.0, 789.0) | 553.8 (398.0, 663.0) | 0.65    |
| SvO₂ (%)             | 94.9 (92.6, 96.5) | 96.0 (91.6, 96.5)           | 94.9 (92.6, 95.9)           | 0.82    |
| Type of PAH           |                   |                             |                             | 0.03*   |
| I/HPAH               | 12                | 2                           | 10                          |         |
| CTD-PAH              | 19                | 11                          | 8                           |         |
| Others               | 11                | 2                           | 9                           |         |
| WHO-Fc               |                   |                             |                             | 0.78    |
| I                    | 3                 | 1                           | 2                           |         |
| II                   | 20                | 6                           | 14                          |         |
| III                  | 19                | 8                           | 11                          |         |
| IV                   | 0                 | 0                           | 0                           |         |
| Baseline PAH medication |                 |                             |                             |         |
| Number               | 0.9 ± 0.7         | 1.1 ± 0.6                   | 0.9 ± 0.8                   | 0.34    |
| Naive                | 13 (31%)          | 2 (15%)                     | 11 (41%)                    |         |
| Monotherapy          | 19 (45%)          | 10 (67%)                    | 9 (33%)                     |         |
| Dual combination     | 10 (24%)          | 3 (20%)                     | 7 (26%)                     |         |
| Beraprost            | 15 (36%)          | 10 (67%)                    | 5 (19%)                     |         |
| Parenteral prostanoids | 3 (7%)          | 0 (0%)                      | 3 (11%)                     |         |
| PDE5-I               | 14 (33%)          | 4 (27%)                     | 10 (37%)                    |         |
| Riociguat             | 5 (12%)           | 1 (7%)                      | 4 (15%)                     |         |
| CCB                  | 1 (2%)            | 1 (7%)                      | 0 (0%)                      |         |

BNP indicates brain natriuretic peptide; BP, blood pressure; CCB, calcium channel blocker; CI, cardiac index; CTD, connective tissue disease; ERA, endothelin receptor antagonist; Hb, hemoglobin; I/HPAH, idiopathic/heritable pulmonary arterial hypertension; PAP, pulmonary arterial pressure; PAWP, pulmonary arterial wedge pressure; PDE5-I, phosphodiesterase 5 inhibitor; PVR, pulmonary vascular resistance; RAP, right atrial pressure; SaO₂, arterial oxygen saturation; and SvO₂, mixed venous oxygen saturation. *Statistically significant between 1st generation ERA group and 2nd generation ERA group.

We retrospectively reviewed the medical records of 45 PAH patients who were prescribed an ERA (16 bosentan, 13 ambrisentan, and 16 macitentan) and underwent a
right heart catheterization (RHC) before and after ERA introduction at The University of Tokyo from January 2007 to July 2019. The type of ERA and the other medications for PAH treatment between the two RHCs remained unchanged. The etiology of PAH included idiopathic or heritable PAH, connective tissue disease-associated PAH (CTD-PAH), portopulmonary PH, congenital heart disease-associated PAH (CHD-PAH), and drug/toxin related PAH. The diagnosis of PAH was made based on a mean pulmonary arterial pressure (mPAP) ≥ 25 mmHg and mean pulmonary arterial wedge pressure ≤ 15 mmHg on RHC. Three unrepaired CHD-PAH patients were excluded from the 45 patients. After these exclusions, 42 patients were included in this study.

For laboratory data, blood samples were assessed using standard laboratory methods at the central laboratory of The University of Tokyo Hospital. Hemodynamic data were evaluated with a Swan-Ganz catheter within 1 week before the ERA introduction. The study protocol conformed to the tenets of the Declaration of Helsinki, and the Institutional Review Board of The University of Tokyo reviewed and approved the protocol (2650).

**Statistical analysis:** Data are presented as the mean ± standard deviation or as the median (inter-quartile range). We performed statistical analyses using IBM SPSS Statistics version 26 (IBM Inc., Armonk, NY, USA). A P value < 0.05 was considered statistically significant. We compared parameters in all study patients or each group before and after ERA introduction using the paired t-test or Wilcoxon signed-rank test. Furthermore, we evaluated the differences between two groups using Student’s (or Welch’s) t-test or the Mann-Whitney U-test for continuous variables and the chi-square test for categorical variables.

**Results**

The study population consisted of 29 female patients (69%) and 13 male patients (31%). The mean age at ERA introduction was 51 ± 15 years. Table 1 shows the baseline characteristics of the patients. There was no significant difference in age, sex, laboratory data, values of hemodynamic examination with Swan-Ganz catheterization, WHO-Fc, or the number of PAH medications at
Figure 2. Changes in WHO-Fc between ERA therapy. A: Total ERA (n = 42), B: 1st generation ERA (n = 15), and C: 2nd generation ERA (n = 27). *statistically significant with Wilcoxon signed-rank test.

ERA introduction between the two ERA groups. The etiology of PAH was significantly different between the two groups; the 1st generation ERA-treated group had a higher proportion of CTD-PAH patients (73%). The interval between right heart catheterization before and after the introduction of the ERA was 133.0 (89.3, 187.5) days, and there was no significant difference between the two groups (135.0 (84.0, 189.0) days vs 126.0 (91.0, 173.0) days, P = 0.79).

First, we evaluated the changes in clinical parameters after ERA introduction in all study patients. In the 42 patients, heart rate, hemoglobin, and SaO2 were significantly decreased after ERA introduction. Among hemodynamic parameters in RHC, mPAP and pulmonary vascular resistance (PVR) were significantly decreased and cardiac index (CI) was significantly increased after ERA introduction (P < 0.001 in mPAP, PVR and CI; Table II and Figure 1). On the other hand, mean right atrial pressure (mRAP) was unexpectedly increased after ERA introduction (P = 0.04, Table II and Figure 1). WHO-Fc was significantly improved after ERA introduction (P = 0.005; Figure 2). Indeed, these trends in hemodynamic parameters (mPAP, CI, PVR and mRAP) were also observed in each ERA group after the patients were divided into two groups, however, the significant changes before and after ERA were observed more robustly in the 2nd ERA group (Table II and Figure 3).

Next, we also compared these parameter changes during ERA introduction between the 1st generation ERA group and 2nd ERA group. Patients treated with a 2nd generation ERA had a greater improvement of hemodynamic parameters (median mPAP change; −5.0 mmHg (−11.0, −3.0) versus −2.0 mmHg (−3.0, 1.0), P = 0.003, median PVR change; −199.0 dyne · s/cm5 (−257.5, −109.5) versus −94.0 dyne · s/cm5 (−193.0, 2.0), P = 0.007) than patients treated with the 1st generation ERA bosentan, as well as HR (mean HR change; −4.9 ± 7.4 bpm versus +0.1 ± 6.7 bpm, P = 0.04), BNP (median BNP change; −13.1 pg/mL (−56.9, 4.1) versus +14.6 pg/dL (−7.2, 186.9), P = 0.005), SaO2 (median SaO2 change; +0.1 % (−1.6, 1.5) versus −2.2 % (−4.1, −0.6), P = 0.001) and SvO2 (mean SvO2 change; +1.7 ± 4.3 % versus −2.3 ± 6.4 %, P = 0.02) (Table II and Figure 3). The WHO-Fc was significantly improved in the 2nd generation ERA-treated group but not in the 1st generation ERA-treated group (P = 0.002 and P = 1.0, respectively; Figure 2).

Discussion

Our study demonstrated that ERA therapy could improve the hemodynamics parameters and WHO-Fc in PAH patients. Although there was no significant difference in the baseline severity of PAH between these 2 ERA treatment groups, the magnitude of the improvement of these parameters was larger in patients treated with 2nd generation ERAs than in patients treated with bosentan. These results suggested that 2nd generation ERAs had a stronger vasodilator effect than bosentan in the short-to-medium treatment term (median treatment duration was about 4 months). According to the literature reviews on the effects of ERA on the hemodynamics of PAH patients, all 3 ERAs were reported to improve hemodynamic parameters. Firstly, for bosentan, one meta-analysis including 10 RCTs showed that bosentan reduced mPAP by 5.7 mmHg, increased CI by 0.4 L/minute/m², and reduced PVR by 305.1 dyne · s/cm5 as compared with placebo. Secondly, for ambrisentan, the result of the ATHENA-1 trial which evaluated the safety and efficacy of sequential add-on therapy of ambrisentan to 33 PAH patients who remained symptomatic despite background phosphodiesterase 5-inhibitor (PDE5-I) monotherapy, indicated that ambrisentan achieved significant improvements in mPAP, CI, and PVR, the mean changes of which from baseline were −5.4 mmHg, +0.58 L/minute/m², and −249 dyne · s/cm5, respectively, after 6 months. Thirdly, for macitentan, the results of the SERAPHIN hemodynamic substudy, which evaluated the hemodynamic efficacy of macitentan as compared with placebo in PAH patients, demonstrated that macitentan 10 mg achieved significant improvement of mPAP, CI, and PVR in 57 PAH patients after 6 months, the mean changes of which from baseline were −5.3 mmHg, +0.3 L/minute/m², and −274 dyne · s/cm², respec-
Figure 3. Box plots of hemodynamic parameters before and after ERA treatment in each ERA therapy group (1st generation \( n = 15 \) and 2nd generation \( n = 27 \)). CI indicates cardiac index; mPAP, mean pulmonary arterial pressure; mRAP, mean right atrial pressure; and PVR, pulmonary vascular resistance. *Statistically significant with Wilcoxon signed-rank test.

Table II. Changes in Parameters Before and After ERA Treatment

|                  | Total ERA \( n = 42 \) | 1st generation ERA \( n = 15 \) | 2nd generation ERA \( n = 27 \) | 1st versus 2nd |
|------------------|-------------------------|---------------------------------|---------------------------------|-----------------|
|                  | \( \Delta \) | \( P \) value | \( \Delta \) | \( P \) value | \( \Delta \) | \( P \) value | \( \Delta \) | \( P \) value |
| Interval (days)  | 133.0 (89.3, 187.5) | 0.0001* | 135.0 (84.0, 189.0) | 0.09 | 126.0 (91.0, 173.0) | 0.79 |
| Heart rate (bpm) | -3.1 ± 7.5 | 0.02* | 0.1 ± 6.7 | 0.94 | -4.9 ± 7.4 | 0.002* |
| Hb (g/dL)        | -0.8 (-1.6, 0.0) | < 0.001* | -0.8 (-1.4, 0.0) | 0.02* | -0.7 (-2.0, 0.0) | 0.002* |
| BNP (pg/mL)      | -4.1 (-46.6, 22.5) | 0.6 | 14.6 (-7.2, 186.9) | 0.07 | -13.1 (-56.9, 4.1) | 0.02* |
| mean BP (mmHg)   | -4.0 ± 13.2 | 0.06 | 0.1 ± 15.4 | 0.98 | -6.5 ± 11.2 | 0.008* |
| mean RAP (mmHg)  | 1.0 (-1.0, 2.0) | 0.04* | 1.0 (-1.0, 2.0) | 0.32 | 1.0 (-1.0, 2.0) | 0.06 |
| mean PAWP (mmHg) | 0.8 ± 3.5 | 0.16 | 0.7 ± 2.2 | 0.23 | 0.8 ± 4.0 | 0.32 |
| mean PAP (mmHg)  | -4.0 (-9.0, 1.0) | < 0.001* | -2.0 (-3.0, 1.0) | 0.11 | -5.0 (-11.0, -3.0) | < 0.001* |
| CI (L/minute/m²) | 0.4 (0.1, 0.6) | < 0.001* | 0.3 (0.4) | 0.1 | 0.4 (0.3, 0.8) | < 0.001* |
| PVR (dyne·s/cm⁵) | -142.5 (-228.0, -84.5) | < 0.001* | -94.0 (-193.0, 2.0) | 0.06 | -199.0 (-257.5, -109.5) | < 0.001* |
| SaO₂ (%)         | -0.5 (-2.5, 0.6) | 0.05* | -2.2 (-4.1, -0.6) | 0.002* | 0.1 (-1.6, 1.5) | 0.83 |
| SvO₂ (%)         | 0.3 ± 5.4 | 0.74 | -2.3 ± 6.4 | 0.18 | 1.7 ± 4.3 | 0.05* |

BNP indicates brain natriuretic peptide; BP, blood pressure; CI, cardiac index; ERA, endothelin receptor antagonist; Hb, hemoglobin; PAP, pulmonary arterial pressure; PAWP, pulmonary arterial wedge pressure; PVR, pulmonary vascular resistance; RAP, right atrial pressure; SaO₂, arterial oxygen saturation; and SvO₂, mixed venous oxygen saturation. *Statistically significant in comparison between two matched groups. †Statistically significant in comparison between 1st generation ERA group and 2nd generation ERA group.

... and mean treatment effects of which as compared with placebo were −6.4 mmHg, +0.63 L/minute/m², and −416 dyne · s/cm², respectively. Judging from these results, all 3 ERAs seem to have comparable efficacy on the hemodynamics of PAH patients, however, we must pay attention to the fact that these trials did not directly compare the effectiveness among each ERA. One single-center study compared the safety and clinical efficacy between bosentan and ambrisentan in 20 stable PAH patients who were prescribed oral 5 mg ambrisentan daily. Eight of the 20 patients switched to bosentan (started with 62.5 mg b.i.d and increased to 125 mg b.i.d. after 4 weeks) and the
remaining 12 patients continued ambrisentan treatment and all patients have been followed up for 2 years. The results of this study indicated that there were no significant differences in the WHO-Fc and echocardiographic parameters of systolic PAP, TAPSE (tricuspid annular plane systolic excursion), and left ventricular eccentricity index between the two groups. Although there is no large RCT evaluating the hemodynamic efficacy of switching ERAs. Although most of the clinical background values in our study were similar between the two ERA groups, the subclassifications of PAH were different between them, more specifically, the proportion of CTD-PAH was higher in the 1st generation ERA group than in the 2nd generation ERA group. CTD-PAH may be associated with heart and lung disorders and may have diminished the effect of PAH treatment, which might have adversely affected our results in the 1st generation ERA group. In addition, 7 of 15 patients (47%) in the 1st generation ERA group started ERA treatment before September 2010, while 26 of 27 (96%) patients in the 2nd generation ERA group started ERA treatment after September 2010, which may have biased the treatment results. The number of PAH drugs at ERA initiation was similar between the two groups, however, looking into the breakdown, there were some differences in the baseline PAH treatment between the two groups; 2 (13%) of the 1st ERA group and 11 (41%) of the 2nd ERA group were naive for PAH treatment, 10 (67%) of the 1st and 9 (33%) of the 2nd ERA group were treated with monotherapy, and 3 (20%) of the 1st and 7 (26%) of the 2nd ERA group were treated with dual combination therapy. These variations in the background PAH treatment might have had some effects on the therapeutic effect of ERA due to the drug interaction. There are several reports regarding the combination of PAH therapeutics. Although combination therapy with an ERA and PDE-5-I was a standard therapeutic regimen, specific combinations of bosentan with a PDE5-I might not be valid due to the pharmacological interaction in which bosentan decreases the maximum plasma concentration (C_{max}) and the area under the plasma concentration versus time curve over a dosing interval (AUC) of PDE5-Is. These drug interactions were caused by inducing cytochrome P450 (CYP) 3A4 and CYP2C9 with bosentan which is the potent ligand of the nuclear pregnane X receptor (PXR). Indeed, the chronic use of bosentan attenuated the effect of sildenafil, while on the other hand, sildenafil was reported to increase the C_{max} and AUC of bosentan. Also, Hatano, et al demonstrated that there was no significant difference in the hemodynamic effects of sildenafil with or without bosentan in PAH patients, although bosentan decreased both the C_{max} and AUC_{0-6h} of sildenafil and its active metabolite desmethylsildenafil. Another RCT evaluated the clinical benefit of sildenafil as compared with placebo in PAH patients treated with a stable dosage of bosentan (62.5 mg or 125 mg, b.i.d.) and demonstrated that sildenafil provided no additional benefit versus placebo on 6 MWT at 12 weeks when added to the bosentan monotherapy. Compared with bosentan, ambrisentan does not strongly induce CYP and therefore does not reduce the blood concentrations of PDE5-Is. With respect to the pharmacokinetic interactions between ERAs and PDE5-Is, one RCT demonstrated that the combination of PDE5-I with macitentan or ambrisentan should be preferable to a combination with bosentan due to maintaining the targeted plasma PDE-5I concentration. And in the AMBITION (Ambrisentan and Tadalafil in Patients with Pulmonary Arterial Hypertension) trial, initial combination therapy with ambrisentan and tadalafil resulted in a significant risk reduction in clinical worsening as compared with ambrisentan or tadalafil monotherapy.

Study limitations: There are several limitations in our study. First, it was a single-center retrospective study, therefore, we have to be cautious when generalizing the results from this study to all PAH patients. As patients were enrolled from 2007 to 2019, the disease background has changed over time, and the number of available drugs has increased over time, resulting in changes in treatment strategies. Second, the study population was small and the statistical analysis may not have adequate power for the comparisons, even when significance was indicated. Despite these issues, this study could evaluate and directly compare the effects of ERA on the hemodynamics of PAH patients in a real-world clinical setting, which is worthy of reporting.

Conclusion

We analyzed and compared the hemodynamic effect of each ERA in PAH patients. All ERAs could successfully improve the hemodynamics of PAH patients and the newer generation ERAs, ambrisentan and macitentan, seemed to be preferable to bosentan.

Disclosure

Conflicts of interest: None.

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