CLINICAL STUDY

Association Between Dose and Plasma Concentration of Bisoprolol in Patients with Heart Failure (CVI ARO 6)

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Summary

Although bisoprolol is used widely to treat patients with heart failure (HF), little information is available regarding the association between the dose of bisoprolol administered and the bisoprolol plasma concentration (Bis-PC) in real-world clinical practice.

This was a single-center, observational study in 114 patients with HF receiving once-daily bisoprolol. After determination of trough Bis-PC, the relationship between the dose of bisoprolol and Bis-PC was analyzed. In a multiple linear regression model, the dose of bisoprolol and estimated creatinine clearance (reciprocal number) were identified as independent predictors. HF severity and hepatic function were not associated with Bis-PC.

Bis-PC was increased by renal dysfunction, which explained most of the discrepancy between the dose of bisoprolol administered and Bis-PC.

(Int Heart J 2020; 61: 748-754)

Key words: Bisoprolol plasma concentration, Renal function, Pharmacokinetics

Beta-blockers are widely used to treat hypertension, angina pectoris, and cardiac arrhythmia. Bisoprolol is a beta-1 selective beta-blocker, which has various beneficial effects in patients with heart failure (HF). The CIBIS-II study showed significant reductions of morbidity and mortality in HF with reduced ejection fraction by treatment with bisoprolol.1)

Bisoprolol shows small intra- and interindividual variations in plasma concentration in healthy people.2) Moreover, a report of population pharmacokinetics of bisoprolol in patients with HF from Europe has indicated that neither renal function nor liver function affected bisoprolol clearance.3) However, several reports of population pharmacokinetics of bisoprolol from Asia demonstrated that various patient background characteristics, including renal function and HF, affect bisoprolol clearance.4-6) Recently, we reported that high plasma concentrations of bisoprolol (Bis-PC), especially in elderly patients, patients with preserved ejection fraction (HFpEF), and patients with renal dysfunction, worsens HF.7) Notably, in one third of the population treated with a mean dose of 2.8 mg, trough Bis-PC reached a high level that was almost equivalent to that in healthy volunteers with a dose of 10 mg (11 ng/mL).7) Given the discrepancy between the dose and plasma concentration of bisoprolol in patients with HF,8) we analyzed the factors influencing Bis-PC other than the administration dose.

Methods

Study participants and ethical issues: We conducted a single-center observational study among Japanese patients with HF treated with bisoprolol (UMIN Clinical Trials Registry: UMIN000023353). This study was supported by the Cardiovascular Institute Academic Research Organization (CVI ARO), which was established in 2014 to encourage operations of investigator-initiated clinical investigations in Japan.8-17)

This study was performed in accordance with the ethical norms based on the Declaration of Helsinki (revised in 2013) and Ethical Guidelines for Medical and Health Research Involving Human Subjects (Public Notice of the Ministry of Education, Culture, Sports, Science and Technology, and the Ministry of Health, Labor, and Welfare, Japan, issued in 2017). Written informed consent was obtained from all participants. The Institutional Review Board of the Cardiovascular Institute reviewed and accepted the study protocol.

Patients with HF receiving bisoprolol treatment who

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Received for publication January 28, 2020. Revised and accepted March 25, 2020.

Released in advance online on J-STAGE July 18, 2020.

doi: 10.1536/ihj.20-052

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visited the Cardiovascular Institute were eligible for inclusion in the present study. The inclusion criteria were: (1) age ≥ 20 years; (2) receiving bisoprolol once daily; (3) receiving bisoprolol stably for more than one week; and (4) diagnosed with HF. In the present study, HF diagnosis was determined in patients with any one of the following conditions: (1) left ventricular ejection fraction (LVEF) < 50%; (2) LVEF ≥ 50% with E/e’ ≥ 15; (3) using diuretics (loop diuretics, mineral corticoid receptor antagonist, or vasopressin-2 receptor antagonist) to relieve HF symptoms; or (4) positive for a history of HF admission. The exclusion criteria were: (1) within seven days after onset of cardiac emergencies, such as acute decompensated HF, myocardial infarction, or cardiac surgery; (2) receiving bisoprolol twice daily; (3) dialysis; (4) hyperthyroidism; (5) any contraindications described on the product label; and (6) patients deemed by the researchers to be ineligible for the study (i.e., judged not to have sufficient ability to understand the study protocol due to dementia, intellectual disturbance, and/or psychiatric/psychosomatic disorders). A total of 128 consecutive patients were enrolled between May 1, 2016, and April 30, 2017. Among them, 14 patients were excluded from the analysis due to the following reasons: (1) one patient withdrew informed consent; (2) three patients could not complete blood sampling within a specified period due to unexpected hospital transfer; and (3) 10 patients with extremely high Bis-PC probably due to erroneous peak sampling. The remaining 114 patients were included in the present study.

Data collection and bisoprolol assay: After obtaining informed consent, plasma samples were collected once before drug intake (22-26 hours after the last drug intake) to measure trough Bis-PC.

For each patient, in addition to age, gender, height, weight, and blood pressure, we evaluated cardiovascular status by electrocardiography, chest X-ray, blood laboratory findings, and echocardiogram at baseline. Body mass index (BMI) was calculated as weight in kilograms divided by height in meters squared. The estimated creatinine clearance (eCCR) was calculated using the demographic source information according to the reported formulas.

The amount of bisoprolol in the sample was determined by liquid chromatography-tandem mass spectrometry (LC-MS/MS) (KAC Co, Ltd., Saitama, Japan). Briefly, a 100-μL aliquot of the sample was treated by precipitating protein with 500 μL of acetonitrile containing metoprolol. The supernatant (500 μL) was aspirated off, evaporated to dryness, and dissolved in 250 μL of acetonitrile-100 mM ammonium formate (19:1 v/v). An aliquot of 3 μL of the supernatant was injected into an LC-MS/MS system (ACQUITY UPLC BEH HILIC column, 10 cm × 2.1 mm i.d., 1.7 μm particle size; Waters Corporation, Milford, MA). Samples were chromatographed in gradient mode (eluent A, 5 mM/L ammonium formate; eluent B, acetonitrile-100 mM ammonium formate, 19:1 v/v). The initial content of eluent B was 95%, decreasing linearly to 40% by 4.2-4.4 minutes with a 7.0-minute hold at 40%. Multiple-reaction monitoring transitions were performed at m/z 326.40 → 115.9 for bisoprolol and m/z 268.22 → 115.8 for metoprolol under positive electrospay ionization conditions.

Statistical analysis: All analyses were performed using SPSS version 19.0 (IBM Corp, Armonk, NY). In all analyses, P < 0.05 was taken to indicate statistical significance.

Categorical and consecutive data are presented as number (%) and mean ± standard deviation, respectively. The prediction model for Bis-PC using bisoprolol doses and other covariables was determined using simple and multiple linear regression analyses. In the multiple linear regression model, dose of bisoprolol was forcedly entered and then other covariables that were significantly associated with Bis-PC in univariate models were entered by the stepwise method. The predictive accuracy of the predicted trough Bis-PC for the observed one was assessed by the coefficient of determinants. Each parameter’s weight in the final multiple linear regression model was assessed by the relative contribution.

Results

Patient characteristics: Table I summarizes the baseline clinical characteristics of the study population. The subjects consisted of 71 male and 43 female Japanese patients aged 40-94 years old, with a mean BMI of 23.6 kg/m². The severity of HF was relatively low with 69 and 44 patients corresponding to New York Heart Association (NYHA) classification 1 and 2, respectively.

Dose of bisoprolol and trough Bis-PC: The average dose of bisoprolol administered was 1.94 mg and the average trough Bis-PC was 4.56 ng/mL. The correlation between dose of bisoprolol and Bis-PC is shown in Figure 1. The regression coefficient (β) of the dose of bisoprolol for Bis-PC was 1.912 (95% CI 1.481-2.342). Although Bis-PC was significantly related to the dose, it was widely scattered. Even at the same dose, the highest was several times greater than the lowest.

Prediction model for trough Bis-PC: The results of simple and multiple linear regression analyses are shown in Tables II, III, respectively. In the multiple linear regression model, the dose of bisoprolol (β, 2.307; 95%CI, 1.959-2.655) and 1/eCCR (β, 190.582; 95%CI, 146.476-234.688) were identified as independent explanatory covariates for Bis-PC. Based on the multiple linear regression model, the prediction model for trough Bis-PC was developed as follows:

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\text{Predicted trough Bis-PC (ng/mL)} = -3.292 + 2.307 \times [\text{dose of bisoprolol (mg)}] + 190.582 \times [1/eCCR (minute/mL)]
\] (Table III)

Figure 2 shows the correlation between the observed trough Bis-PC and the predicted trough Bis-PC. The regression coefficient \(R^2\) was 0.644 (95% CI, 0.495-0.687). In the prediction model, the relative contribution of bisoprolol dose was 81.9%, whereas that of 1/eCCR was 18.1%, indicating eCCR predicted 18% of the variance of Bis-PC in the prediction model.

Figure 3 displays a simulation graph which plots the predicted Bis-PC according to eCCR separated by bisoprolol dose. According to the simulation graph, we can see that the decline of eCCR from 100 minute/mL to 50 minute/mL or from 50 minute/mL to 30 minute/mL both
Consecutive values are given as mean ± SD. Bis-PC indicates bisoprolol plasma concentration; NYHA, New York Heart Association; HFpEF, heart failure with preserved ejection fraction; history of ISTIA, history of ischemic stroke or transient ischemic attack; eGFR, estimated glomerular filtration rate; eCCr, estimated creatinine clearance; and AST, aspartate aminotransferase.

lead to increase of approximately 2 ng/mL in trough Bis-PC, which were almost equivalent to an increase in bisoprolol dose of 1.25 mg.

### Discussion

**Main findings:** The present study results indicate that the dose of bisoprolol and 1/eCCr were independent explanatory factors for trough Bis-PC in Japanese patients with HF, where eCCr explained the discrepancy between dose and plasma concentration of bisoprolol. We developed a prediction model for trough Bis-PC, which could explain two thirds of the observed Bis-PC (R² = 0.644).

**Prediction model for trough Bis-PC:** The present study results indicated that Bis-PC increased with decreasing eCCr and higher bisoprolol dose. A previous study of the pharmacokinetics of bisoprolol showed that Bis-PC was significantly higher in patients with kidney or liver diseases than in healthy volunteers.™ While bisoprolol is eliminated by both renal and non-renal metabolic pathways, the renal pathway accounts for the majority, with excretion of 50% of the dose.™ Horikiri, et al. reported that hepatic clearance of bisoprolol was around 30% of the total clearance.™ In the present study, no patient had severe hepatic dysfunction, but almost half had chronic kidney disease. Thus, the impact of the hepatic function on the Bis-PC may have been relatively small compared to that of renal function. Bisoprolol is known to be metabolized in the liver mainly via CYP3A4, and CYP enzyme activity is differentially affected by the presence of liver disease along with the Child-Pugh score.™ The measurement of total bilirubin or prothrombin activity value may be needed in patients with severe hepatic dysfunction, and further attempts of genetic screening for CYP3A4 is desired for optimal model.

Age and body weight, which generally affect the pharmacokinetics of various drugs, were not included in the final predictive model for Bis-PC. This may have been because CCr, estimated by the Cockcroft Gault equation, involved the effects of age and body weight.

In the previous reports of population pharmacokinetic analysis (PPK) analysis, the importance of the dose of bisoprolol and the estimated renal function on bisoprolol pharmacokinetics was concluded (of note, these study mostly included patients with NYHA class II, like our study). Our methodology was not based on such pharmacological analysis, but our results were generally in line with the previous PPK analyses.

**Clinical implications:** In HF patients, the clearance of drugs is generally diminished via decreased blood flow to various organs, including the kidneys and the liver. Although the severity of HF in our patients was relatively low (NYHA class ≤ 2), clearance of bisoprolol may have been decreased, resulting in the large discrepancy between the dose and Bis-PC.

According to the prediction model for trough Bis-PC in our data, decreases in renal function from normal (eCCr 100 minutes/mL) to moderate dysfunction (eCCr 50 minutes/mL), or from moderate to severe dysfunction (eCCr 30 minutes/mL), both lead to increases of approximately 2 ng/mL in trough Bis-PC, which were almost
Figure 1. Relationship between dose of bisoprolol and Bis-PC. Bis-PC indicates bisoprolol plasma concentration.

Table II. Simple Linear Regression Model for Bis-PC

| Model Variable          | Standardized β | B (95% confidence interval)       | P-value  | R²  |
|-------------------------|----------------|----------------------------------|----------|-----|
| Age (year)              | 0.325          | 0.090 (0.041-0.139)              | < 0.001  | 0.106|
| Female                  | 0.272          | 1.957 (0.658-3.255)              | 0.003    | 0.074|
| Bisoprolol dose (mg)    | 0.639          | 1.912 (1.481-2.342)              | < 0.001  | 0.409|
| 1/eCCr (minutes/mL)     | 0.300          | 113.703 (45.980-181.425)         | < 0.001  | 0.090|
| Albumin (g/dL)          | −0.180         | −1.701 (−3.460-0.057)            | 0.058    | 0.032|
| AST (IU/L)              | −0.140         | −0.044 (–0.105-0.018)            | 0.161    | 0.020|
| Hemoglobin (g/dL)       | −0.269         | −0.592 (−0.989–−0.195)           | 0.004    | 0.072|
| Brain natriuretic peptide (pg/dL) | 0.071 | 0.001 (−0.002-0.004) | 0.453    | 0.005|

Bis-PC indicates bisoprolol plasma concentration; eCCr, estimated creatinine clearance; and AST, aspartate aminotransferase.

Table III. Multiple Linear Regression Model for Bis-PC

| Model Variable          | Standardized β | B (95% confidence interval)       | P-value  | R²  |
|-------------------------|----------------|----------------------------------|----------|-----|
| Bisoprolol dose (mg)    | 0.771          | 2.307 (1.959-2.655)              | < 0.001  | 0.644|
| 1/eCCr (min/mL)         | 0.503          | 190.582 (146.476-234.688)        | < 0.001  | 0.644|

In multiple regression model, bisoprolol dose was forsiely entered, and then other variables significantly associated with Bis-PC in univariate analysis were entered using the stepwise method. Bis-PC indicates bisoprolol plasma concentration; and eCCr, estimated creatinine clearance.

equivalent to an increase in bisoprolol dose of 1.25 mg. Caution should be exercised to prevent HF patients from unexpected exposure to high Bis-PC, especially those with renal dysfunction, the number of whom is increasing in clinical practice with the aging of the population.

Limitations: Our study had several limitations. First, our
Figure 2. Relationship between observed and predicted Bis-PC. Predicted trough Bis-PC = −3.292 + 2.307 \times \text{[the dose of bisoprolol (mg)]} + 190.582 \times \text{[1/eCCr (minute/mL)]} \quad \text{Bis-PC indicates bisoprolol plasma concentration; and eCCr, estimated creatinine clearance.}

Figure 3. A simulation graph of estimated Bis-PC according to eCCr separated by bisoprolol dose. According to the simulation graph, we can see that the decline of eCCr from 100 minute/mL to 50 minute/mL or from 50 minute/mL to 30 minute/mL both lead to increase of approximately 2 ng/mL in trough Bis-PC, which were almost equivalent to an increase in bisoprolol dose of 1.25 mg. Bis-PC indicates bisoprolol plasma concentration; and eCCr, estimated creatinine clearance.
data involved a relatively small number of patients from a single cardiovascular hospital, which may have led to underestimation of the relationships between clinical factors and Bis-PC due to a lack of statistical power. Second, as Bis-PC varies according to the timing of blood sampling, our data, based on patient-reported intake time, may have somehow overestimated or underestimated Bis-PC in each patient. However, we believe the difference would be minimal because of the nature of trough sampling, where the variance by sampling time is small. Third, our results and prediction model cannot be extrapolated easily to patients with severe HF and hepatic congestion, requiring hospitalization repeatedly, because of the relatively low risk profiles in our study population. It means that unintended increase of Bis-PC may be more enhanced in patients with severe HF and hepatic congestion, and therefore further investigations in patients with high risk profiles would be necessary.

Conclusions
In this single-center study, we developed a model for prediction of trough Bis-PC in Japanese HF patients. The discrepancy between dose of bisoprolol and Bis-PC could mainly be explained by renal dysfunction.

Acknowledgment
We thank Mr. Hidefumi Kasai at Department of Clinical Pharmacokinetics and Pharmacodynamics, Keio University School of Medicine, for giving important intellectual advice in making the prediction model for trough Bis-PC.

Disclosure
Conflicts of interest: Dr. Suzuki received research funding from Daiichi Sankyo and Mitsubishi Tanabe Pharm. Dr. Yamashita has received research funding and/or lecture fees from Daiichi Sankyo, Bayer Yakuhin, Bristol-Myers Squibb, Pfizer, Nippon Boehringer Ingelheim, Eisai, Mitsubishi Tanabe Pharm, Ono Pharmaceutical, and Toa Eiyo.

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