Cardiac Pharmacology

A Pharmacokinetic and Pharmacodynamic Study of Intravenous Levosimendan in Healthy Chinese Volunteers and Ethnic Comparisons

Kai-Min Chu, Oliver Yoa-Pu Hu and Jun-Ting Liou

Background: The objective of this study was to assess the pharmacokinetics and pharmacodynamics of intravenous levosimendan and the metabolites (OR1855 and OR1896) in healthy Chinese male subjects and to post hoc compare with Caucasian subjects.

Methods: One single 2 mg dose of levosimendan was infused intravenously over 10 minutes to each of 14 healthy male subjects. Plasma levosimendan was analyzed by high performance liquid chromatography. Pharmacodynamics was evaluated using echocardiography.

Results: The Cmax (peak concentration) and AUC (area under the curve from time 0 to infinity) of levosimendan of Chinese subjects were significantly higher than for Caucasian subjects as 256.1 ± 37.8 (mean ± SD) vs. 142.1 ± 17.5 ng/mL and 207.5 ± 35.2 vs. 117.0 ± 17.0 hr·ng/mL, respectively. The clearance of Chinese subjects was significantly lower than Caucasian subjects at 9.9 ± 2.3 L/hr, respectively. The elimination half-life of Chinese subjects was significantly longer than for Caucasian subjects (1.18 ± 0.18 vs. 0.76 ± 0.10 hr, respectively). Chinese subjects eliminated levosimendan significantly slower than Caucasian subjects, leading to a higher exposure of levosimendan in Chinese subjects. However, this higher exposure did not significantly change the most pharmacodynamic properties of levosimendan except for ejection fraction (EF). The EF increased 12.2 ± 11.4% in Chinese subjects 20 min after the end of intravenous infusion, which was significantly lower than Caucasian subjects with EF increased by 22.7 ± 7.0%.

Conclusions: The intravenous levosimendan in healthy Chinese volunteers was safe, and well-tolerated with significant inotropic effect. The clearance of levosimendan of Chinese subjects was significantly lower and elimination half-life longer than Caucasian subjects.

Key Words: Chinese • Ethnic comparison • Levosimendan • Pharmacodynamics • Pharmacokinetics • Volunteer

INTRODUCTION

Positive inotropic agents have been used to treat patients with acute congestive heart failure (CHF) and have demonstrated short-term hemodynamic support and symptomatic relief. Clinical trials with positive inotropic agents in patients with CHF have corroborated that these agents provide an improvement in the symptoms; however, they may also increase the mortality rate.1,2

Dobutamine is the agent most frequently used to treat hemodynamic disturbances resulting from episodes of a worsening heart failure. Side effects of dobutamine observed in clinical trials include tachphy-
laxis, an increasing heart rate, cardiac arrhythmias and increasing incidences of myocardial ischemia. Some of these side effects have also been observed with adenosine monophosphate-dependent inotropes (phosphodiesterase inhibitors) such as amrinone, milrinone, or enoximone. Due to lack of positive long-term patient outcomes following treatment, the overall benefits of such agents are questionable.

Levosimendan is an intravenously administered calcium-sensitizing agent indicated for the short-term (24 hour) treatment of decompensated chronic heart failure. In vitro and in vivo studies have demonstrated that levosimendan increases myocardial contractility, lowers cardiac filling pressures, and dilates peripheral and coronary blood vessels.

The main mechanism of action of levosimendan is calcium sensitization of the contractile protein in cardiac muscle. The effect is obtained by calcium-dependent binding of the drug to cardiac troponin C. It increases the contractile force of cardiomyocytes. Due to calcium dependency, levosimendan does not impair relaxation. Additionally, levosimendan opens the ATP-sensitive potassium channels in both vascular and cardiac tissues, thereby producing vasodilatory and anti-ischemic effects.

The pharmacokinetics (PK) of levosimendan are linear with plasma concentrations increasing in a dose-proportional manner following a single bolus intravenous infusion of doses ranging from 0.2 to 5.0 mg in healthy volunteers and patients with CHF. Levosimendan is rapidly distributed to tissues and 95-98% of the drug in plasma is bound to plasma proteins, mainly albumin. The parent drug has a half-life (t½) of approximately one-hour with steady-state concentrations reached within four hours of initiating a continuous infusion, without a preceding loading dose.

Two metabolites (OR1855 and OR1896) of levosimendan have been identified in human plasma following intravenous infusion. Only the acetylated metabolite OR1896 is pharmacologically active, and only about 5% of the levosimendan dose is metabolized to OR1896. It is formed and eliminated slowly, with an elimination half-life of approximately 80 hours. Following a continuous 24-hour infusion of levosimendan (0.1 to 0.4 mcg/kg/min), peak concentrations of these active metabolites are reached within one to four days (mean of 2 days), and are detectable for at least 10 days.

The efficacy and safety of levosimendan have been investigated in several studies. Levosimendan has been shown to increase cardiac output and ejection fraction in healthy subjects. In addition, levosimendan decreased pulmonary capillary wedge pressure and peripheral vascular resistance, and increased stroke volume and cardiac output dose-dependently in patients with heart failure. Use of the recommended dose of levosimendan (0.05-0.2 µg/kg/min) produced a modest increase in heart rate, which was long-lasting.

In general, the safety profile was related to the pharmacology of levosimendan. The most frequent adverse drug reactions were headache and hypotension (both about 6%). Other less common adverse drug reactions included nausea and dizziness (both about 2%), atrial fibrillation and ventricular tachycardia (both about 1%). Adverse events related to arrhythmias and myocardial ischemia were significantly less frequently reported with levosimendan than with dopamine.

All of the clinical data thus far have been collected in Caucasian subjects. It is considered important to assess the pharmacokinetic and pharmacodynamic effects associated with the intravenous administration of levosimendan in Chinese subjects. This study was the first levosimendan study in the Chinese population. The objective of this study was to assess the pharmacokinetics, pharmacodynamics and safety effects of intravenous levosimendan and the metabolites (OR1855 and OR1896) in healthy Chinese male subjects.

**MATERIALS AND METHODS**

**Study design and plan**

This was a Phase 1, single-dose, open-label, and single-center study. In vitro protein binding assay and distribution kinetics assay were also conducted.

Fourteen healthy male Chinese subjects were enrolled in this study. All subjects were determined to be in good health on the basis of history, physical examination, urinalysis, blood chemistry, chest roentgenogram and electrocardiogram. None of the subjects had a history of cardiovascular, renal, hepatic, gastrointestinal, respiratory, hematological, metabolic or other diseases that could affect the absorption, distribution, meta-
bolism, or excretion of the study drug. None had a history of drug or alcohol abuse, donation or loss of 550 ml or more blood within 8 weeks prior to study, or use of tobacco or nicotine-containing products within the 6-month period prior to our study. The protocol was approved by the Human Subject Institutional Review Board. The study was carried out in accordance with those parameters as established by the Declaration of Helsinki. Written informed consent was obtained after the purpose, procedures, and risks of the study had been explained to the volunteers. The volunteers were instructed to avoid any other drugs, including alcohol, caffeine or nicotine for two weeks before and during the course of the study.

On study day 1, all subjects received, in a fasting state (at least 10 hours fasting), a single 2 mg dose of intravenous infusion of levosimendan, over 10 minutes. Blood samples for levosimendan, OR1855 and OR1896 concentration assay

Blood samples (6 mL) from each patient were obtained from an antecubital vein in the contralateral arm (the other was for infusion) for levosimendan; OR1855 and OR1896 concentration assay were drawn into pre-cooled plastic tubes containing ethylenediaminetetraacetic acid (EDTA) as an anticoagulant. The blood samples were collected at -15 min, 0 min (end of infusion), 5 min, 10 min, 20 min, 30 min, 1 hr, 2 hr, 4 hr, 6 hr, 8 hr, 12 hr, 24 hr, 32 hr, 48 hr, 60 hr, 72 hr, 120 hr, 168 hr and 240 hr following dosing.

Immediately after collection, the blood samples were inverted several times to ensure good mixing of the blood and anticoagulant. Blood samples were kept in ice and the plasma was separated within 10 minutes by centrifugation in a centrifuge for 10 minutes at approximately 3300 rpm. At least 2 mL of plasma was transferred into one pre-cooled polypropylene test tube. The plasma samples were frozen immediately at -70°C until shipped to Abbott Laboratories.

In vitro protein binding assay

Blood samples (6 mL from each and 12 mL in total) for protein binding assay were collected at 0 min and 10 min after infusion stopped for each subject. The blood samples were drawn into pre-cooled plastic tubes containing sodium heparin as an anticoagulant. The subsequent procedures were the same as noted in the above two paragraphs.

Analysis of plasma samples for levosimendan, OR1855 and OR1896 concentrations

Plasma concentrations of levosimendan, OR1855 and OR1896 were measured by using liquid chromatography tandem mass spectrometry at the Department of Drug Analysis of Abbott Laboratories.

Pharmacokinetic variables

Values for the pharmacokinetic parameters of levosimendan, OR1855 and OR1896, including the maximum observed plasma concentration (C_max), the time to C_max (peak time, T_max), the terminal phase elimination rate constant (β), terminal phase elimination half-life (t_1/2), and the area under the plasma concentration-time curve (AUC) from time 0 to the time of the last measurable concentration (AUC_0) and from time 0 to infinite time (AUC_∞), were determined using noncompartmental methods. Clearance (CL) and volume of distribution (V_d) for levosimendan were determined using noncompartmental methods. The WinNonlin version 5.0.1 of Pharsight Corp. was used for PK parameters calculation.

In addition, plasma protein binding and redistribution phenomenon were also determined. The plasma protein binding (P_b) was calculated using: P_b = (1 - C_f/C_t) × 100%, where C_f: concentration of the top layer after centrifugation, and C_t: initial plasma concentration before centrifuge.

Pharmacodynamic variables

Left atrium diameter (LA), left ventricle end-diastolic diameter (LVEDD), LV end-systolic diameter (LVESD), heart rate (HR), pre-ejection period (PEP), LV ejection time (LVET), systolic time interval (STI), mean circumferential fiber shortening (MVCf), ejection fraction (EF), cardiac output (CO), cardiac index (CI), aortic valve peak velocity (AOPV) and mitral valve peak velocity (MVPV) were measured and calculated.

Safety variables

The following safety evaluations were performed during the study: adverse event monitoring and vital signs, physical examination, electrocardiography (ECG) and laboratory test assessments.
Study procedure

A complete medical history, including alcohol, tobacco and nicotine-containing product use histories was taken when blood samples (6 mL for each and 12 mL in total) for protein binding assay were collected at 0 min and 10 min after infusion was stopped for each subject. The medical history was updated on study day 1, and the medical history obtained at screening served as the baseline for clinical assessment. Medication (prescription or over-the-counter, including vitamins and herbal supplements) use from 4 weeks prior to study drug administration through the end of the study was also recorded. A physical examination was performed at screening. A brief physical examination was performed on the study day 1 (-60 min), day 3 (48 ± 0.5 hr), day 4 (72 ± 0.5 hr), day 6 (120 ± 0.5 hr), day 8 (168 ± 0.5 hr) and day 11 (240 ± 0.5 hr) or upon subject discontinuation. A symptom-directed physical examination was performed when necessary. The physical examination performed at screening served as the baseline physical examination for clinical assessment. Any significant physical examination findings after dosing were recorded as adverse events.

Patient body weight was measured at screening (baseline for clinical assessment), -60 min on the study day 1, and day 11 (240 ± 0.5 hr) or upon subject discontinuation in the study.

Body temperature (oral), blood pressure and pulse were measured at screening, on the study day 1 (-60 min, -10 min, 0 min, 5 min, 10 min, 20 min, 30 min, 1 hr, 2 hr, 4 hr, 6 hr, 8 hr, 12 ± 0.5 hr), day 2 (24 ± 0.5 hr, 32 ± 0.5 hr), day 3 (48 ± 0.5 hr, 60 ± 0.5 hr), day 4 (72 ± 0.5 hr), day 6 (120 ± 0.5 hr), day 8 (168 ± 0.5 hr), and day 11 (240 ± 0.5 hr) or upon early discontinuation.

Blood pressure and pulse rate were measured after the subject had been lying down for at least 3 minutes. The measurements were completed within 5 minutes prior to draw of PK blood sample.

The vital signs measurements just prior to dosing on study day 1 served as the baseline measurements for clinical assessment.

A 12-lead resting ECG was obtained at screening, on the study day 1 (-60 min, 10 min, 1 hr, and 6 hr), day 3 (48 ± 0.5 hr) and day 11 (240 ± 0.5 hr) or upon early discontinuation. The ECG measurements at -60 min on the study day 1 served as the baseline measurements for clinical assessment.

An appropriately qualified cardiologist at the study site read all ECGs. The reader wrote on the ECG tracing his/her global interpretation as either “normal ECG” or “abnormal ECG” and then signs and dates the ECG. The QT interval measurement (corrected by Fridericia formula, QTcF) was documented only if the reader selects the “prolonged QT” box.

Echocardiography recording and analysis

Echocardiographic recordings were obtained on study day 1 (-60 min, -10 min, 0 min, 5 min, 10 min, 20 min, 30 min, 1 hr, 2 hr, 4 hr, 6 hr, 4 hr) or upon early discontinuation.

Cardiac function indices were evaluated by a Hewlett Packard SONOS ultrasound system with phased-array Doppler (Model 5500). M-mode, two-dimensional (2D) echocardiographic images and pulsed-wave Doppler velocity signals were recorded on computer hard disc and magneto-optical (MO) disc for a later retrieval and analysis. The subjects were examined in a slightly left-lateral decubitus position during quiet respiration. Left parasternal long-axis, left parasternal short-axis, apical 4 chamber, apical 5-chamber views were recorded. Doppler tracings of aortic flow were obtained using the apical 5-chamber view, with the sample-volume placed just beyond the valve leaflets within the proximal aortic root. Slight adjustments in transducer angulations or sample-volume position were at times required to maximize the audio and graphic quality of the Doppler signal. The aortic valve opening on parasternal long axis view, velocities of mitral and aortic flow were recorded over several cardiac cycles at a monitor sweep speed of 100 mm/sec. AOPV and MVPV were measured. M-mode echocardiographic measurements followed the standard method as recommended by the American Society of Echocardiography. LVEDD and LVESD were measured in parasternal long-axis view at the level between the papillary muscle and mitral leaflet tips. LA was measured at the aortic valve level. Left ventricle volume (V) was determined by the diameter (D) using Teichholz’s equation: V = [7.0/(2.4 + D)] × D. Stroke volume (SV) equals LVEDV-LVESV; EF equals SV/LVEDV, cardiac output equals SV × HR, and CI equals SV × HR/body surface area. The PEP was the time duration measured from the onset of ventricular depolarization (onset of QRS complex on simultaneous electrocardiogram) to the opening of the
aortic valve. LVET was measured from the opening to the closure of the aortic valve. All measurements were made with the on board computer. Systolic time interval was calculated by the equation PEP/LVET.

**Ethnicity comparison**

The pharmacokinetic and pharmacodynamic parameters were tested among study result and previous Caucasian male results by analysis of variance (ANOVA) for any ethnic difference. P < 0.05 was considered significant.

**RESULTS**

A total of 14 Chinese male subjects in the age range of 25.0 ± 2.8 years old, weight of 66.8 ± 5.2 kg, and height of 173.5 ± 3.5 cm had completed the study.

**Assessment of pharmacokinetics**

Table 1 shows the pharmacokinetic variables in this study. Figure 1 shows the mean concentration-time curve of levosimendan after intravenous infusion of 2 mg of levosimendan.

The pharmacokinetic parameters were calculated using non-compartment model and actual time vs. concentration of levosimendan, OR1855 and OR1896. For levosimendan, $C_{\text{max}}, T_{\text{max}}, \beta, t_{1/2}, AUC_t, AUC_{\infty}, C_L,$ and $V_d$ were 256.1 ± 37.8 ng/mL, 0.02 ± 0.06 hr, 0.60 ± 0.07 1/hr, 1.18 ± 0.18 hr, 206.6 ± 35.0 hr-ng/mL, 207.5 ± 35.2 hr-ng/mL, 9.89 ± 1.78 L/hr and 9.83 ± 1.41L, respectively.

For OR1855 and OR1896, there was not enough concentration data to estimate the pharmacokinetic parameters in most subjects. Pharmacokinetic parameters of OR1855 could be estimated for only one subject and OR1896 could be estimated for only two subjects. For these subjects only $C_{\text{max}}, T_{\text{max}}$ and $AUC_t$ could be estimated.

**Plasma protein binding of levosimendan**

The plasma protein binding of levosimendan was 99.15 ± 0.28%.

**Assessment of pharmacodynamics**

The pharmacodynamic parameters included systolic

<table>
<thead>
<tr>
<th>Pharmacokinetic parameters</th>
<th>Levosimendan (n = 14)</th>
<th>OR1855 (n = 1)</th>
<th>OR1896 (n = 2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>$C_{\text{max}}$, ng/mL</td>
<td>Mean (SD) 256.1 (37.8)</td>
<td>0.6 (-)</td>
<td>1.1 (0.01)</td>
</tr>
<tr>
<td>Median</td>
<td>248.2</td>
<td>0.6</td>
<td>1.1</td>
</tr>
<tr>
<td>$T_{\text{max}}$, hr</td>
<td>Mean (SD) 0.02 (0.06)</td>
<td>60 (-)</td>
<td>72 (0)</td>
</tr>
<tr>
<td>Median</td>
<td>0</td>
<td>60</td>
<td>72</td>
</tr>
<tr>
<td>$\beta$, 1/hr</td>
<td>Mean (SD) 0.60 (0.07)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median</td>
<td>0.61</td>
<td></td>
<td></td>
</tr>
<tr>
<td>$t_{1/2}$, hr</td>
<td>Mean (SD) 1.18 (0.18)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median</td>
<td>1.13</td>
<td></td>
<td></td>
</tr>
<tr>
<td>$AUC_{t}$, hr-ng/mL</td>
<td>Mean (SD) 206.6 (35.0)</td>
<td>18.7 (-)</td>
<td>83.6 (0.2)</td>
</tr>
<tr>
<td>Median</td>
<td>207.9</td>
<td>18.7</td>
<td>83.6</td>
</tr>
<tr>
<td>$AUC_{\infty}$, hr-ng/mL</td>
<td>Mean (SD) 207.5 (35.2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median</td>
<td>208.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>$C_L$, L/hr</td>
<td>Mean (SD) 9.89 (1.78)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median</td>
<td>9.58</td>
<td></td>
<td></td>
</tr>
<tr>
<td>$V_d$, L</td>
<td>Mean (SD) 9.83 (1.41)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median</td>
<td>9.84</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Figure 1. The mean ± SD of plasma concentration-time profile of levosimendan of 14 subjects after IV infusion of 2 mg of levosimendan.
and diastolic blood pressure, heart rate, and the following echocardiography parameters: LA, PEP, LVET, PEP/LVET, HR, LVEDD, LVESD, EF, CO, CI, AOPV, MVPV, and MVcf.

The changes in the pharmacodynamic parameters versus time profile of subjects at different time points before and after IV infusion of 2 mg of levosimendan are shown in Figures 2-5. For most of the parameters, the maximum hemodynamic effect was seen within 10 or 20 minutes after the end of infusion.

Comparisons of pharmacokinetic parameters between Chinese and Caucasian subjects

Table 2 shows the comparisons of pharmacokinetic parameters of levosimendan between Chinese and Caucasian subjects.

The $C_{\text{max}}$, $AUC_{t}$, and $AUC_{\infty}$ of levosimendan of Chinese subjects were significantly higher than in the Caucasian subjects. Mean ± SD of weight un-normalized and normalized CL of levosimendan of Chinese and Caucasian subjects were 9.9 ± 1.8 vs. 17.4 ± 2.7 L/hr and 0.149 ± 0.025 vs. 0.246 ± 0.028 L/hr/kg. It was found that CL of levosimendan of Chinese subjects were significantly lower than Caucasian subjects irrespective of whether CL was normalized by weight or not.

The elimination half-life of levosimendan of Chinese subjects was significantly longer than Caucasian subjects (1.18 ± 0.18 vs. 0.76 ± 0.10 hr).

The results suggest that Chinese subjects eliminated levosimendan significantly slower than Caucasian subjects leading to higher exposure of levosimendan in Chinese subjects as compared to Caucasian subjects.
Comparisons of pharmacodynamic parameters between Chinese and Caucasian subjects

Table 3 shows the comparisons of pharmacodynamic parameters after IV infusion of 2 mg of levosimendan between Chinese and Caucasian subjects.

The percentage changes of pharmacodynamic parameters of Chinese subjects after IV infusion of 2 mg of levosimendan were similar to those of Caucasian subjects except for EF. The EF increased up to 12.2%/177 11.4% in Chinese subjects after the end of intravenous infusion, which was significantly lower than in Caucasian subjects where EF increased up to 22.7%/177 7.0% (p = 0.0282).

Safety evaluation

Throughout the study period, 2 subjects were reported to have experienced 3 adverse events (chest tightness, cough rhino rhea nasal stiffness and tachycardia). No subject experienced any serious adverse event and no subject discontinued due to any adverse event. In terms of these three reported adverse events, one was probably not related to the study drug; the other two, in the investigator’s opinion, were also not related to the study drug.

DISCUSSION

In our study subjects, plasma concentration of levosimendan was higher with lower clearance and longer elimination half-life than in Caucasian subjects. However, the inotropic index, as EF, was less increased than in Caucasian subjects. The higher protein binding in our subjects might explain this. In our study, the percentage of protein binding of plasma levosimendan is 99.15%/177 0.28%, higher than the previously reported 95-98%. 10-12 When the fraction unbound is lower, less free drug is distributed into red blood cells and tissue, and also less free drug is available for both renal excretion and...
hepatic biotransformation, which might be demonstrated by a decreased clearance and a longer half-life.\textsuperscript{24,25} Less tissue distribution might also indicate less target organ distribution and less pharmacodynamic effect as shown by less increased EF. There are important clinical considerations when using a drug with very high (> 95%) protein binding rate.\textsuperscript{26} First of all, the patient's plasma protein concentration should be considered. If a patient has a low plasma protein concentration, then for any given dose of drug the concentration of free bioactive drug might be higher than anticipated. A number of diseases such as cirrhosis of liver, nephrotic syndrome, malnutrition, age, trauma, and related circumstances affect the plasma protein concentration. In nephrotic syndrome, excessive renal loss of plasma protein and accumulation of waste metabolites such as urea and uric acid, as well as an accumulation of drug metabolites, may also alter protein binding of drugs.\textsuperscript{27} Second, drug-drug and drug-metabolite interactions are also important in a high protein binding drug with a narrow therapeutic index such as warfarin.\textsuperscript{28} In this case one drug might displace a second bound drug from the protein, causing a sudden increase in pharmacologic response due to an increase in free drug concentration. Third, the increase of plasma concentration of a drug might not necessarily mean the increase of the pharmacodynamic effect. In a condition with no change of the protein binding, higher plasma concentration will have higher free drug concentration and the following greater pharmacodynamic effect according to the dose-concentration-response curve. However, displacement of drug from plasma protein or target tissue is completely different.\textsuperscript{29} If a free drug is increased due to drug-interaction with the displacement by a second drug from the plasma protein-binding site, than the tissue distribution will be increased and the pharmacodynamic effect will be increased. The increased free drug will also increase the elimination and total plasma concentration will transiently be increased and returned to the baseline level before drug-interaction. This has been shown in the drug interaction with warfarin.\textsuperscript{28,30-32} If a free drug increases due to low plasma protein with less binding site, the change in pharmacodynamic effect will be the same as in the condition of displacement from plasma protein binding site. On the other hand, if a drug-interaction acts at the tissue-binding site with displacement of drug from tissue to plasma, than the higher plasma concentration means lower tissue concentration and thus will have smaller pharmacodynamic effect. This effect has been demonstrated in digoxin, a cardiac glycoside, which binds strongly to the myocardial tissue of the heart. If patients who are on digoxin take a quinidine, steady-state digoxin plasma levels increase, possibly due to displacement of the tissue-bound digoxin.\textsuperscript{33-35}

Therefore, when applying our study result in determining the infusion rate and duration, subsequent studies based only on plasma concentration are probably not appropriate. Careful monitoring of the therapeutic effect is also important.

**CONCLUSIONS**

The administration of intravenous levosimendan into healthy Chinese volunteers was safe, well-tolerated and had a significant inotropic effect. The clearance of levosimendan from Chinese subjects was significantly lower, and the elimination of half-life longer than in Caucasian subjects.

**ACKNOWLEDGMENTS**

Authors thank Abbott Pharmaceuticals for supporting this study, providing the raw data of previously unpublished studies and plasma sample analysis. The protocol was initiated by Abbott Pharmaceuticals and revised by authors. The conduction, pharmacokinetic analysis and manuscript preparation were not supported.

**REFERENCES**

1. McMurray JJ, Adamopoulos S, Anker SD, et al. ESC guidelines for the diagnosis and treatment of acute and chronic heart failure 2012: The Task Force for the Diagnosis and Treatment of Acute and Chronic Heart Failure 2012 of the European Society of Cardiology. Developed in collaboration with the Heart Failure Association (HFA) of the ESC. *Eur Heart J* 2012;33:1787-847.


