The pharmacokinetics of sildenafil may be affected by intestinal absorption rate in children admitted to the intensive care unit

Yukino OYA, Daisuke WATAHIKI, Mitsuki MATSUNAGA, Keiichi HIRONO, Fukiko ICHIDA, Masaya AOKI, Naoki YOSHIMURA, and Masato TAGUCHI *

Graduate School of Medicine and Pharmaceutical Sciences, University of Toyama,

*Address correspondence to Masato Taguchi, Ph.D.
2630 Sugitani, Toyama 930-0194, Japan
E-mail: taguchi@pha.u-toyama.ac.jp
Abstract

This study was performed for a better understanding of the pharmacokinetics of sildenafil (SIL) and N-desmethyl sildenafil (DMS) in 13 children treated in the intensive care unit (ICU). Blood samples were taken periodically after the first oral administration of SIL (0.5 mg/kg). Plasma concentrations were analyzed by LC-MS/MS. Of the 13 patients, apparent peaks in the plasma concentration of SIL were observed in four patients, with the other nine patients showing reduced or delayed drug absorption of SIL. The maximum plasma concentrations of SIL after administration varied in range from 7.8 to 101.0 ng/mL. The parent drug-to-metabolite (SIL/DMS) ratios of the nine patients with reduced or delayed drug absorption of SIL were relatively lower than those in the four patients with rapid absorption of the drug. These observations suggested that the inter-individual variability of intestinal absorption and/or first-pass extraction of SIL was involved in the pharmacokinetic variability of the drug. Next, we evaluated the impact of changes in the gastrointestinal absorption rate on the pharmacokinetics of the drug. That is, SIL (2.5 mg/body) was administered at two different rates in the duodenum of rats. When SIL was administered for 10 min, the $C_{max}$ and bioavailability were $3.46 \pm 1.65 \mu g/mL$ and $23.2 \pm 11.1\%$, respectively. When SIL was administered for 60 min, the $C_{max}$ and bioavailability were $0.990 \pm 0.352 \mu g/mL$ and $9.91 \pm 3.79\%$, respectively. These findings suggest that the drug absorption rate was at least partly responsible for the pharmacokinetic variability of SIL in the ICU children.

Key words: sildenafil; children; intensive care unit; gastrointestinal motility; bioavailability
Introduction

Sildenafil (SIL), a selective phosphodiesterase-5 (PDE-5) inhibitor, causes relaxation of vascular smooth-muscle cells without affecting systemic pressure\(^1\) and acts as an adjunctive or alternative pulmonary vasodilator in the treatment of pulmonary hypertension.\(^2,3\) Matamis et al.\(^2\) demonstrated that the combination SIL and inhaled nitric oxide (iNO) decreased pulmonary vascular pressure and resistance more than either treatment alone.\(^2\) Namachivayam et al.\(^3\) reported that a single dose of SIL effectively prevented the rebound of pulmonary hypertension in infants and children after withdrawal of iNO and reduced the subsequent duration of mechanical ventilation. Indeed, patients administered SIL had a mean pulmonary pressure of 35.1 ± 3.3 mm Hg at baseline and 35.8 ± 14.8 mm Hg 1 h after stopping iNO, whereas patients administered with a placebo, had a mean pulmonary pressure of 31.0 ± 9.0 mm Hg at baseline and 45.2 ± 20.4 mm Hg 1 h after stopping iNO.\(^3\) Therefore, SIL is effective as an adjunctive and alternative treatment of iNO in patients with pulmonary hypertension.

Ahsman et al.\(^4\) previously investigated the pharmacokinetics of repeated administration of SIL in 11 neonates ranged in postnatal days from 2 to 121 days with pulmonary hypertension. The patients had undergone extracorporeal membrane oxygenation (ECMO) and received SIL (0.5 mg/kg) three or four times daily in the intensive care unit (ICU). Because desmethyl-sildenafil (DMS), metabolite of SIL, possesses activity itself (50 % as potent as SIL\(^5\)), the plasma concentration area under the curve (AUC) over 24 h was evaluated as AUC\(_{24h}(SIL+DMS)\) which was calculated by combination AUC of SIL and 50 % of that of DMS. As a result, the AUC\(_{24h}(SIL+DMS)\) values in individual patients ranged from 625 to 13579 ng·h/mL.\(^4\) In the study of
Ahsman et al.⁴), however, the mechanisms responsible for the great pharmacokinetic variability of SIL in the neonatal patients was unclear.

To date, we have reported on the pharmacokinetics of tadalafil, another PDE-5 inhibitor, in a pediatric population, and demonstrated the interindividual variability of the pharmacokinetics of the drug.⁵) That is, post dose concentrations of tadalafil varied 9-fold (from 128 to 1135 ng/mL) in 23 children.⁶) In addition, we compared enzymes involved in the metabolism of both SIL and tadalafil and found that tadalafil was metabolized by cytochrome P450 (CYP)3A4, but not by CYP3A5 and CYP3A7.⁷) On the other hand, SIL was metabolized by CYP3A4, CYP3A5, and to a lesser extent by CYP3A7, and the intrinsic clearance ($CL_{int}$) values of the N-demethylation for CYP3A4, 3A5, and 3A7 were 1.108 μL/min/pmol P450, 1.086 μL/min/pmol P450, and 0.093 μL/min/pmol P450, respectively.⁷) Considering reports that CYP3A7 and CYP3A4 are expressed specially in human fetal and adult livers, respectively⁸), the clinical impact of the developmental changes in drug metabolizing enzymes on the pharmacokinetics of SIL may be more complex than that of tadalafil.

In this study, we evaluated the pharmacokinetic variability of SIL in pediatric Japanese postoperative cardiovascular ICU patients, including not only neonates but also young infants. In addition, we revealed a potential mechanism of the pharmacokinetic variability of SIL using an animal model.
Materials & methods

Materials
SIL was purchased from Toronto Research Chemicals (North York, ON, Canada). All other chemicals and solvents were the highest purity available.

Patients and study protocol
We recruited 13 Japanese pediatric patients (five boys and eight girls) treated in the ICU and their demographic data are summarized in Table 1. The patients ranged in postnatal days from 12 to 1150 days (mean: 385 days) and in body weight from 3.03 to 10.7 kg (mean: 6.67 kg). All patients were postoperative for congenital heart disease and on a mechanical ventilator and recovered to good condition for weaning (Table 1). No critically ill patients with severe symptoms was included. SIL citrate (Revatio®; Pfizer Inc.) was administered at a dose of 0.5 mg/kg four times a day after cardiovascular surgery at Toyama University Hospital. The dose is confirmed by Japan Circulation Society 2018 Guideline. Ten of the 13 patients had been co-administered 2-3 drugs, for example phenobarbital, chloral hydrate, midazolam, rocuronium bromide, and digoxin (Supplemental Table 1). At the first oral administration of SIL, 1 mL blood samples were taken from arterial line pre-dose and at 30, 60, 90, 120, 150, 180, and 360 min post-dose, and stored at -30°C until analysis. The patients and/or parents gave written informed consent to participate in this study, which was approved by the ethics committee of the University of Toyama (EG22-1).

Animals and experimental protocol
Male Wistar rats aged 8 weeks old (200-250 g) were used for animal experiments.
The rats were housed in a temperature and humidity-controlled room with free access to water and standard rat chow, and fasted overnight for 12-24 h prior to experiments. All animal experiments were performed in accordance with The Guidelines for Experiments of University of Toyama (A2017PHA-18).

The body temperature of the rats was maintained with heating lamps. The femoral artery was cannulated with a polyethylene tube (SP-31, Natsume Seisakusho, Tokyo, Japan) for blood sampling. For the intra-duodenum administration study (2.5 mg/body), a catheter with a 20 G needle was inserted into the duodenum and the intestines were restored to their former location. Thereafter, SIL was administered at two different rates; at a slow infusion rate of 0.75 mL/h for 60 min and at a fast infusion rate of 4.5 mL/h for 10 min. Blood samples were withdrawn pre-dose and at 5, 10, 20, 30, 40, 50, 60, 70, 80, 90, 120, and 180 min after the slow infusion, and pre-dose and at 1, 5, 10, 15, 20, 25, 30, 45, 60, 90, 120, and 150 min after the fast infusion. For the intravenous administration study, the jugular vein was cannulated with a polyethylene tube (SP-31) for intravenous infusion of SIL (2.5 mg/body). SIL solution was infused via a catheter using a constant rate infusion pump. Arterial blood samples for measurement of SIL concentration were obtained pre-dose and at 1, 5, 15, 30, 31, 35, 40, 60, 90, and 120 min after initiation of 30-min intravenous infusion.

SIL assay

The solid-phase extraction (SPE) was performed by the previously validated method for the sample preparation of human plasma. That is, fifty-five µL of human plasma sample was mixed with 990 µL of distilled water and 55 µL of methanol containing 2 mg/mL SIL-d3 as an internal standard. The 1 mL sample was instilled in
OASIS HLB® 1cc Vac Cartridge (Waters, MA, USA) at 1 mL/min. The column was washed with 1 mL of distilled water then 1 mL of 5% methanol. SIL was eluted with 2 mL of acetonitrile at 1 mL/min, and the solvent was evaporated to dryness with a ThermoSavant SPD1010 SpeedVac® System at 45°C for 45 min. The resulting residue was reconstituted with 200 µL of mobile phase.

In the case of rat plasma, the liquid-liquid extraction was performed because SPE cartridges are too expensive to handle many animal samples and because plasma levels of SIL in rats were sufficiently high to measure. That is, 50 µL of plasma sample was mixed with 1 mL of glycine buffer (pH 10.6, 0.1M, saturated with sodium chloride) and 5 mL of tert-butylmethyl ether and centrifuged at 3500 rpm for 5 min at 4 °C. Four mL of the supernatant was taken and tert-butylmethyl ether was evaporated to dryness with SpeedVac at 45 °C for 40 min. The residues were reconstituted with 150 µL mobile phase containing 500 ng/mL caffeine as an internal standard. The linearly range was from 0.01 to 100 µg/mL. The recovery of SIL was 86.1 ± 9.81 % and the quantification limit for SIL was 5 ng/mL. The inter day coefficient of variation was 16.2 % at 10 ng/mL, 6.97 % at 50 ng/mL and 3.47 % at 500 ng/mL.

**LC-MS/MS analysis**

Concentrations of SIL and DMS were determined by the LC-MS/MS method. The injection volume was 25 µL. The mobile phase was 10 mM ammonium formate (with 0.1% formic acid) and acetonitrile (40:60). The flow rate was 300 µL/min, and the column temperature was 40°C. LC-MS/MS analysis was carried out in a Thermo Fisher Accela™ LC system (Thermo Fisher Scientific) coupled to an LTQ-Orbitrap XL™ ETD system (Thermo Fisher Scientific). A COSMOSIL® PACKED column
2.5C18-MS-II (2.0 × 50 mm) was used for the chromatographic separation of SIL. The cone voltage was 35 V and the collision energy was 30 eV. Mass spectra were recorded by electrospray ionization in ESI positive mode. The detector was operated in selected reaction monitoring mode using the transitions of SIL at m/z 475.5→283.4, DMS at m/z 461.5→283.4, SIL-d3 at m/z 478.6→283.4, and caffeine at m/z 195.2→138.1. The peak areas were calculated using Qualbrowser® software (Thermo Fisher Scientific). The limits of quantification for SIL and DMS were 1.0 ng/mL and 1.0 ng/mL, respectively.

**Pharmacokinetic analysis in rats**

The AUC after intravenous and intra-duodenum infusion in rats were calculated using the linear trapezoidal rule and extrapolated to infinity by dividing the last measurable SIL concentration by the elimination rate constant (k_e). The apparent clearance (CL/F) value expressed by the systemic clearance (CL) and bioavailability (F) following the intra-duodenum administration were calculated from Dose/AUC. The F values following intra-duodenum injection were calculated by dividing AUC following intra-duodenum injection (AUC_{i.d.}) by AUC following intravenous infusion (AUC_{i.v.}).

**Statistical analysis**

The significance of the differences between the two groups was evaluated using the Student’s t-test if the variance of the group was similar, or Welch’s t-test if the variance of the group was not similar. p<0.05 was considered to be significant. Data are expressed as the mean ± S.D.
Results

Figure 1A shows the time course of plasma concentrations of SIL administered through a nasogastric tube in individual patients. The dose of SIL was set at 0.5 mg/kg. Of the 13 patients, rapid absorption of SIL was observed in four patients (#3, #4, #8 and #10). On the other hand, the other nine patients (#1, #2, #5-7, #9, #11-13) showed reduced or delayed SIL absorption. Overall, the $C_{\text{max}}$ of SIL within 6 h after administration ranged widely from 7.8 to 101.0 ng/mL between patients (Fig. 1A). We measured the plasma concentrations of DMS as it is an active metabolite of SIL (Fig. 1B). Of the 13 patients, the $C_{\text{max}}$ of DMS was observed in six patients (#4, #6-#8, #11 and #12). On the other hand, seven patients (#1-3, #5, #9, #10, #13) showed a slow but constant production of DMS, and the $C_{\text{max}}$ of DMS within 6 h after administration ranged widely from 9.4 to 76.9 ng/mL between patients (Fig.1B). These findings suggested that gastrointestinal absorption and/or first-pass extract of SIL was markedly varied among patients.

To clarify the mechanisms responsible for the variability of drug absorption and/or the first-pass effect, the parent drug-to-metabolite (SIL/DMS) ratio was evaluated in individual patients. (Fig. 2). The SIL/DMS ratio in the nine patients with reduced or delayed absorption of SIL (open circle) were significantly lower than those in the four patients with rapid absorption of the drug (closed circle) (Fig. 2). Summary of comparisons between the two patient groups with and without rapid absorption of SIL were listed in Table 2. The mean postnatal age in the patients with rapid absorption of SIL was numerically but not statistically smaller than that in the patients with reduced or delayed absorption of SIL. The SIL/DMS ratio corresponding to Fig. 2 in patients who showed apparent peaks of SIL (4.08 ± 4.85) was significantly greater than that in
patients who showed no apparent peaks (1.82 ± 1.94) (Table 2). In addition, it was noteworthy that four patients (#6, #7, #11, #12) showed an apparent increase in the plasma DMS concentration even though the absorption of SIL was considerably reduced or delayed (Fig. 1A and B). These findings suggested that the first-pass hepatic extraction of SIL was increased by the prolonged and sustained intestinal absorption of the drug under ICU conditions.

To evaluate the effects of change in the intestinal absorption rate on the bioavailability of SIL, we conducted an *in vivo* pharmacokinetic study in which SIL solution (3.33 mg/mL) was directly administered into the rat duodenum at different infusion rates. That is, the time course of the plasma concentration of SIL in rats infused with 4.5 mL/h for 10 min was compared with that of those infused with 0.75 mL/h for 60 min, which mimics prolonged and sustained intestinal absorption of the drug. As shown in Fig. 3A, when SIL was infused for 60 min, the mean plasma concentration was considerably decreased compared with that for 10 min. That is, when SIL was administered to the duodenum for 10 min, the $C_{\text{max}}$ and bioavailability were 3.46 ± 1.65 µg/mL and 23.2 ± 11.1%, respectively (Table 3). When SIL was administered to the duodenum for 60 min, the $C_{\text{max}}$ and bioavailability were 0.99 ± 0.35 µg/mL and 9.91 ± 3.79%, respectively (Table 3). These results suggested that altered bioavailability of SIL was at least partly involved in the mechanism of the pharmacokinetic variability.
Discussion

The primary purpose of the present study was to gain a better understanding of the pharmacokinetics of SIL and DMS in children admitted to the ICU. The plasma concentration of SIL and DMS varies in the clinical setting, and some patients showed rapid absorption of SIL whereas others showed reduced or delayed SIL absorption (Fig.1). In addition, there were patients that showed an apparent increase in the plasma concentration of DMS even though the absorption of SIL was reduced or delayed (Fig.1). To demonstrate our hypothesis that the bioavailability of SIL was decreased by prolonged and sustained absorption of the drug, SIL solution was directly administered into the rat duodenum at two different infusion rates (Fig.3). The bioavailability of SIL following an infusion at 4.5 mL/min for 10 min and at 0.75 mL/min for 60 min was 23.2% and 9.91%, respectively, although total system plasma clearance was almost identical irrespective of the infusion rate (Table 3). These findings indicated that the bioavailability of SIL was largely dependent on the intestinal absorption rate of the drug.

The bioavailability in humans after oral SIL administration has been reported to be 38-41%. To investigate the cause of this incomplete bioavailability of SIL, Osman et al. evaluated the intestinal transport parameters of SIL citrate in rabbits using an in situ intestinal perfusion technique. The intestinal absorption of SIL differed among the four gastrointestinal tract segments with the highest absorption occurring in the jejunoileum, followed by the duodenum, the ascending colon, and finally the rectum. They demonstrated that the absorption of SIL occurs mainly in the jejunoileum and suggested that slowing the gastric emptying rate may slow down SIL absorption. We previously demonstrated that the accelerated intestinal absorption
associated with acute renal failure resulted in an increased bioavailability of propranolol in rats.\textsuperscript{14) That is, after intra-intestinal injection of propranolol, the mean blood concentration in acute renal failure rats was significantly higher than that in control rats. However, when propranolol was infused into the portal vein, the hepatic extraction did not differ between the rats with acute renal failure and control rats.\textsuperscript{14)}

Several studies have indicated that critically ill patients, particularly postoperative cardiovascular patients in the ICU, are prone to have decreased intestinal motility.\textsuperscript{15-18)} The determinants of gastrointestinal complications in cardiac surgery were the use of anesthetic drugs\textsuperscript{15)}, mechanical ventilation\textsuperscript{16)}, and increased activity in the sympathetic nerve.\textsuperscript{17)} In addition, Morisawa et al.\textsuperscript{19)} reported that decreased intestinal motility in patients in the ICU after cardiovascular surgery can be corrected by passive exercise of the lower limbs and trunk (PELT). That is, the integral value of bowel sounds after 5 min at rest was $63.1 \pm 41.3\text{ mVsec}$ and that 5 min after PELT was $115.0 \pm 57.8\text{ mVsec}$, suggesting that the resting state had a considerable suppressive effect on the gastrointestinal motility.\textsuperscript{19)} These reports strongly support our hypothesis that the first-pass hepatic extraction of SIL was increased by the prolonged and sustained intestinal absorption of the drug under ICU conditions.

The large pharmacokinetic variability of SIL observed in the study by Ahsman et al.\textsuperscript{4)} was fairly in line with the observations of our present study. However, interestingly, the plasma concentrations of SIL in the present study were much lower than those reported by Ahsman et al.\textsuperscript{4)} There are three possible reasons for this large difference in the plasma concentration between these two studies. First, the activity of metabolic enzymes varies with age, which may affect the pharmacokinetics of SIL. We previously reported that SIL N-demethylation, which is the main metabolic pathway,
is catalyzed by CYP3A4, CYP3A5, and to a lesser extent by CYP3A7. As CYP3A7 is more active than CYP3A4 in the fetal period, and CYP3A7 is replaced by CYP3A4 during the early neonate period, hepatic clearance increased rapidly after birth. The patients ages ranged from postnatal days 2 to 121 (average 33.5 days) in the report by Ahsman et al., and ranged from postnatal days 12 to 1150 (average 385 days) in the present study. Second, the pharmacokinetics of SIL may vary depending on the experimental procedure or the severity of the patients. That is, SIL administration route via a nasogastric tube and blood sampling using arterial line were consistent in the two studies, but patients of the Ahsman study were on ECMO and those of present study were on standard respiratory support (non-ECMO). Third, cultural differences in medication use may affect an individual’s exposure and/or response to drugs. The study by Ahsman et al. included patients that received fluconazole (three of 11 patients) which is a potent inhibitor of CYP3A activity. In contrast, our study included patients that received phenobarbital (eight of 13 patients), which is an inducer of hepatic CYP3A activity in humans (Supplemental Table 1).

From the above, there are limitations to discuss the true impact of ICU admission on the pharmacokinetics of SIL, but the comparison of our results with the previous study in non-ICU patients as close as matched in age may give at least plausible and partial information about the issue. For reference, we would like to cite the paper by Olguín et al. where the plasma concentrations of SIL was measured in non-ICU patients with pulmonary arterial hypertension. The twelve patients with mean age of 6.4 years (1.5 to 15 years) were included and received an oral administration of 1.0 mg/kg of SIL. What is noteworthy in the study of Olguín et al. is that there was a rapid process of SIL absorption with a time to maximal concentration of approximately
Study limitation

As the present study was a retrospective observational study and the number of patients was limited, concomitant drugs were not controlled. In addition, we could not evaluate gastrointestinal motility directly in young children due to ethical issues. We did not investigate the relationship between the plasma concentrations and the therapeutic effects of SIL directly, but we succeeded in weaning of iNO therapy without rebound pulmonary hypertension in all 13 patients. SIL is known to have a favorable PDE selectivity, but Wang et al.\textsuperscript{21)} reported that the inhibitor selectivity for PDE families is most likely associated with increased incidence of adverse events of PDE-5 inhibitors. That is, PDE-6 is involved in phototransduction in the retina, and transient cyanopsia, light sensitivity, blurred vision, and blindness have been shown to occur with SIL.\textsuperscript{21)} This raises another issue that lower dose of SIL may be recommended especially in pediatric patients who cannot complain of adverse effect. Further systematic studies are needed to confirm the factors responsible for the pharmacokinetic and/or pharmacodynamic variability of SIL.

In conclusion, we observed a large degree of interindividual variability in the pharmacokinetics of SIL in patients treated in the ICU. We propose that altered bioavailability of SIL was at least partly involved in the mechanism of this pharmacokinetic variability. The present study provides new insights on the proper management of pharmacotherapy in critically ill patients in the ICU.
ACKNOWLEDGMENT

This work was supported in part by JSPS KAKENHI Grant Number 18K06780.

Conflict of Interest

The authors declare no conflict of interest.

Supplementary Materials

The online version of this article contains supplementary materials.
References

1) Abrams D, Schulze-Neick I, Magee AG. Sildenafil as a selective pulmonary vasodilator in childhood primary pulmonary hypertension. *Heart*. **84**, e4 (2000).

2) Matamis D, Pampori S, Papathanasiou A, Papakonstantinou P, Tsagourias M, Galiatsou E, Koulouras V, Nakos G. Inhaled NO and sildenafil combination in cardiac surgery patients with out-of-proportion pulmonary hypertension: acute effects on postoperative gas exchange and hemodynamics. *Circ Heart Fail*. **5**, 47—53 (2012).

3) Namachivayam P, Theilen U, Butt WW, Cooper SM, Penny DJ, Shekerdemian LS. Sildenafil prevents rebound pulmonary hypertension after withdrawal of nitric oxide in children. *Am J Respir Crit Care Med*. **174**, 1042—1047 (2006).

4) Ahsman MJ, Witjes BC, Wildschut ED, Sluiter I, Vulto AG, Tibboel D, Mathot RA. Sildenafil exposure in neonates with pulmonary hypertension after administration via a nasogastric tube. *Arch Dis Child Fetal Neonate Ed*. **95**, F109—F114 (2010).

5) Mehrotra N, Gupta M, Kovar A, Meibohm B. The role of pharmacokinetics and pharmacodynamics in phosphodiesterase-5 inhibitor therapy. *Int J Impot Res*. **19**, 253—264 (2007).

6) Kohno H, Ichida F, Hirono K, Ozawa S, Yoshimura N, Nakamura T, Akita C, Ishida K, Taguchi M. Plasma concentrations of tadalafil in children with pulmonary arterial hypertension. *Ther Drug Monit*. **36**, 576—583 (2014).

7) Takahiro R, Nakamura S, Kohno H, Yoshimura N, Nakamura T, Ozawa S, Hirono K, Ichida K, Taguchi M. Contribution of CYP3A isoforms to dealkylation of PDE5 inhibitors: a comparison between sildenafil N-demethylation and tadalafil demethylation. *Biol Pharm Bull*. **38**, 58—65 (2015).
8) Lacroix D, Sonnier M, Moncion A, Cheron G, Cresteil T. Expression of CYP3A in the human liver—evidence that the shift between CYP3A7 and CYP3A4 occurs immediately after birth. *Eur J Biochem.* **247**, 625—634 (1997).

9) JCS 2018 Guideline on the Clinical Examinations for Decision Making of Diagnosis and Drug therapy in Pediatric Patients with Congenital Heart Disease and Cardiovascular Disorder.: <https://www.j-circ.or.jp/cms/wp-content/uploads/2020/02/JCS2018_Yasukochi.pdf>, accessed 28 August, 2020.

10) Yaroshenko DV, Grigoriev AV, Sidorova AA, Kartsova LA. Chromatographic Determination of Sildenafil in Blood Plasma Using Spectrophotometric and Mass-Spectrometric Detection. *J Anal Chem.* **68**, 801—808 (2013).

11) Nichols DJ, Muirhead GJ, Harness JA. Pharmacokinetics of sildenafil after single oral doses in healthy male subjects: absolute bioavailability, food effects and dose proportionality. *Br J Clin Pharmacol.* **53**(Suppl. 1), 5S—12S (2002).

12) Osman MA, El Maghraby GM, Hedaya MA. Intestinal absorption and presystemic disposition of sildenafil citrate in the rabbit: evidence for site-dependent absorptive clearance *Biopharm. Drug Dispos.* **27**, 93—102 (2006).

13) Muirhead GJ, Rance DJ, Walker DK, Wastall P. Comparative human pharmacokinetics and metabolism of single-dose oral and intravenous sildenafil. *Br J Clin Pharmacol.* **53**(Suppl. 1), 13S—20S (2002).

14) Okabe H, Miukami A, Taguchi M, Aiba T, Yasuhara M, Hashimoto Y. The increased intestinal absorption rate is responsible for the reduced hepatic first-pass extraction of propranolol in rats with cisplatin-induced renal dysfunction. *J Pharm. Pharmacol.* **55**, 479—486 (2003).

15) Fruhwald S, Holzer P, Metzler H. Intestinal motility disturbances in intensive care
patients pathogenesis and clinical impact. *Intensive Care Med*. **33**, 36—44 (2007).

16) Mutlu GM, Mutlu EA, Factor P. GI complications in patients receiving mechanical ventilation. *CHEST*. **119**, 1222—1241 (2001).

17) Fuder H and Muscholl E. Heteroreceptor-mediated modulation of noradrenaline and acetylcholine release from peripheral nerves. *Rev. Physiol. Biochem. Pharmacol.*, **126**, 265-412 (1995).

18) D’Ancona G, Baillot R, Poirier B, Dagenais F, Saez-de-Ibarra JI, Bauset R, Mathieu P, Doyle D. Determinants of gastrointestinal complications in cardiac surgery. *Tex Heart Inst J*. **30**, 280—285 (2003).

19) Morisawa T, Takahashi T, Sasanuma N, Mabuchi S, Takeda K, Hori N, Ohashi N, Ide T, Domen K, Nishi S. Passive exercise of the lower limbs and trunk alleviates decreased intestinal motility in patients in the intensive care unit after cardiovascular surgery. *J Phys Ther Sci*. **29**, 312—316 (2007).

20) Olguín HJ, Martínez HO, Pérez CF, Mendiola BR, Espinosa LR, Chávez -Pacheco JL, Pérez JF, Magaña IM. Pharmacokinetics of sildenafil in children with pulmonary arterial hypertension. *World J Pediatr*. **13**, 588—592 (2017).

21) Wang R, Burnett AL, Heller WH, Omori K, Kotera J, Kikkawa K, Yee S, Day WW, DiDonato K, Peterson CA. Selectivity of avanafil, a PDE5 inhibitor for the treatment of erectile dysfunction: implications for clinical safety and improved tolerability. *J Sex Med*. **9**, 2122—2129 (2012).
Table 1  Demographic data of children treated in the intensive care unit included in the study.

| Patient | Postnatal age (days) | Body weight (kg) | Body surface area (m$^2$) | Postoperative days (days) | Disease                                          |
|---------|----------------------|------------------|---------------------------|---------------------------|--------------------------------------------------|
| #1      | 440                  | 8.80             | 0.394                     | 3                         | Atrial septal defect                             |
| #2      | 744                  | 9.30             | 0.443                     | 2                         | Ebstein anomaly                                  |
| #3      | 48                   | 3.03             | 0.193                     | 5                         | Total anomalous pulmonary venous return          |
| #4      | 703                  | 8.74             | 0.419                     | 2                         | Ebstein anomaly                                  |
| #5      | 399                  | 6.37             | 0.345                     | 2                         | Pulmonary vein stenosis                          |
| #6      | 400                  | 7.97             | 0.388                     | 2                         | Single ventricle                                 |
| #7      | 623                  | 8.60             | 0.430                     | 1                         | Single ventricle                                 |
| #8      | 98                   | 4.66             | 0.261                     | 2                         | Cor triatriatum sinister                         |
| #9      | 35                   | 3.53             | 0.215                     | 3                         | Coarctation of the aorta                         |
| #10     | 12                   | 3.69             | 0.215                     | 5                         | Total anomalous pulmonary venous return          |
| #11     | 222                  | 6.37             | 0.347                     | 8                         | Hypoplastic left heart syndrome                  |
| #12     | 1150                 | 10.7             | 0.494                     | 5                         | Ebstein anomaly                                  |
| #13     | 131                  | 5.05             | 0.278                     | 3                         | Ventricular septal defect                        |

Mean ± S.D. 385 ± 345 6.67 ± 2.52 0.340 ± 0.094 3.3 ± 1.9
Table 2  The comparisons between patients with and without rapid absorption of SIL.

|                                      | Patients with rapid absorption of SIL (#3, #4, #8, #10) | Patients with reduced or delayed absorption of SIL (#1, #2, #5-7, #9, #11-13) |
|--------------------------------------|---------------------------------------------------------|---------------------------------------------------------------------------------|
| Postnatal age (days)                 | 215 ± 283                                               | 460 ± 323                                                                       |
| $C_{\text{max}}$ (ng/mL) SIL         | 67.9 ± 26.1                                             | 19.1 ± 6.5*                                                                     |
|                                      | 43.3 ± 26.4                                             | 27.2 ± 14.6                                                                      |
| $\text{AUC}_{0-6}$ (ng·h/mL) SIL     | 160 ± 47                                                | 53.5 ± 21.3†,*                                                                  |
|                                      | 112 ± 60                                                | 65.7 ± 32.3†                                                                    |
| SIL/DMS ratio‡                       | 4.08 ± 4.85                                             | 1.82 ± 1.94*                                                                    |

†The patient #1 was excluded due to deviation from the study protocol for blood sampling at 6 h (Fig. 1). ‡Corresponding to the observation depicted in Fig. 2. Values are expressed as the mean ± S.D. * p < 0.05 compared with values of patients with rapid absorption of SIL.
### Table 3  Pharmacokinetic parameters of sildenafil in rats after doses of 2.5 mg/body.

|                  | I.V. 1.5 mL/h for 30 min (n=6) | I.D. 4.5 mL/h for 10 min (n=6) | I.D. 0.75 mL/h for 60 min (n=6) |
|------------------|--------------------------------|--------------------------------|---------------------------------|
| **AUC<sub>0-∞</sub> (μg·min/mL)** | 514 ± 346                      | 119 ± 57                        | 51.0 ± 19.5*                    |
| **C<sub>max</sub> (μg/mL)**       | 6.99 ± 1.23                    | 3.46 ± 1.65                     | 0.990 ± 0.352**                 |
| **F (%)**             | —                              | 23.2 ± 11.1                     | 9.91 ± 3.79*                    |
| **k<sub>e</sub> (/min)**      | 0.00720 ± 0.00370              | 0.0120 ± 0.0050                 | 0.0110 ± 0.0110                 |
| **CL (mL/min/kg)**     | 25.8 ± 12.9                    | 25.5 ± 17.6                     | 22.3 ± 9.7                      |

I.V., intravenous administration; I.D., intraduodenal administration. Values are expressed as the mean ± S.D. * p < 0.05  and ** p < 0.01 compared with values of 4.5 mL/h for 10 min.
Fig. 1 Time course for the plasma concentration of SIL (A) and DMS (B) at a dose of 0.5 mg/kg SIL in 13 children treated in the intensive care unit.
Fig. 2 The ratio of parent-to-metabolite (SIL/DMS) in 13 Japanese pediatric patients. Closed circles represent patients who showed apparent peaks of SIL or DMS. Open circles represent patients who showed no apparent peaks. The parent drug-to-metabolite ratio was omitted when the concentration of SIL or DMS in plasma was below the quantitative limit of 1 ng/mL.
Fig. 3 Time course for the mean plasma concentration of SIL after intraduodenal administration (A) and intravenous administration (B) of SIL at doses of 2.5 mg/body in rats. (A) SIL was administered at two different rates (either 4.5 mL/h for 10 min (●) or 0.75 mL/h for 60 min(○)) in the duodenum of rats. (B) SIL was administered at a rate of 1.5 mL/h for 30 min in the jugular vein of rats. Bars represent mean ± S.D. of six rats.