IJBMS

Comparison of pharmacokinetic parameters of ranolazine between diabetic and non-diabetic rats

Habibeh Mashayekhi-sardoo [1], Hossein Kamali [2, 3], Soghra Mehri [1], Amirhossein Sahebkar [4, 5, 6], Mohsen Imenshahidi [1, 7]*, Amir Hooshang Mohammadpour [7, 8]*

[1] Department of Pharmacodynamics and Toxicology, School of Pharmacy, Mashhad University of Medical Sciences, Mashhad, Iran
[2] Targeted Drug Delivery Research Center, Pharmaceutical Technology Institute, Mashhad University of Medical Sciences, Mashhad, Iran
[3] Department of Pharmaceutics, School of Pharmacy, Mashhad University of Medical Sciences, Mashhad, Iran
[4] Biotechnology Research Center, Pharmaceutical Technology Institute, Mashhad University of Medical Sciences, Mashhad, Iran
[5] Applied Biomedical Research Center, Mashhad University of Medical Sciences, Mashhad, Iran
[6] Department of Biotechnology, School of Pharmacy, Mashhad University of Medical Sciences, Mashhad, Iran
[7] Pharmaceutical Research Center, Pharmaceutical Technology Institute, Mashhad University of Medical Sciences, Mashhad, Iran
[8] Department of Clinical Pharmacy, School of Pharmacy, Mashhad University of Medical Sciences, Mashhad, Iran

A R T I C L E   I N F O

A B S T R A C T

Objective(s): Diabetes mellitus (DM) affects the pharmacokinetics of drugs. Ranolazine is an antianginal drug that is prescribed in DM patients with angina. We decided to evaluate the effect of DM on the pharmacokinetics of ranolazine and its major metabolite CVT-2738 in rats.

Materials and Methods: Male rats were divided into two groups: DM (induced by 55 mg/kg Streptozotocin (STZ)) and non-DM. All animals were treated with 80 mg/kg of ranolazine for 7 continuous days. The blood samples were collected immediately at 0 (prior to dosing), 1, 2, 3, 4, 8, and 12 hr after administration of the 7th dose of ranolazine. Serum ranolazine and CVT-2738 concentrations were determined using the high-performance liquid chromatography (HPLC) method. Pharmacokinetic parameters were calculated using a non-compartmental model and compared between the two groups.

Results: The peak serum concentration (Cmax) and area under the curve (AUC) of ranolazine significantly decreased in DM compared with non-DM rats. DM rats showed significantly higher volumes of distribution (Vd) and clearance (CL) of ranolazine than non-DM rats. DM did not affect Ke, Tmax, and T1/2 of ranolazine. The concentration of metabolite was lower than the HPLC limit of detection (LOD).

Conclusion: It was found that streptozotocin-induced DM increased Vd and CL of ranolazine, thereby decreasing the AUC of the drug. Therefore, dosage adjustment may be necessary for DM patients, which requires further clinical studies.

Introduction

Diabetes mellitus (DM) as a growing healthcare challenge can affect the pharmacokinetics and pharmacodynamics of medications (1-3). Previous reports have shown that DM alters several physiological processes in humans, including micro- and macro vasculature, gastric mucosal blood flow, parasympathetic function, and intestinal hormone activity, which can affect the absorption of drugs (4-6). Moreover, poorly controlled blood glucose can cause gastropathy and changes in gastrointestinal pH. It ultimately leads to delays in gastric emptying, alterations in the lipophilicity of drugs, and decreased drug absorption (6, 7). Plasma protein binding of various drugs can change due to the modification in glycosylation of proteins after exceeding the produced plasma free fatty acids in the DM state (8). DM decreases vascular permeability through microvascular disorders (8). Previous clinical and experimental studies have revealed that DM impacts the CYP450 enzyme activity, but the findings have been inconsistent and

contradictory (1, 9). It has been reported that the expression and function of CYP3A4 are diminished in DM subjects (9). Nevertheless, experimental studies have reported the suppression of microsomal N-demethylase action and raised or even reduced levels of CYP450 enzymes. DM affects metabolism mainly by mechanisms such as increased reactive oxygen species (ROS) and oxidative stress (10). By affecting angiotensin-converting enzymes and micro- and macro vascular structure of the kidney, DM can alter the renal elimination of drugs. These changes may impair drug delivery to relevant tissues and contribute to nephropathy development (8, 11).

Ranolazine is an anti-ischemic and antianginal drug that is indicated for decreasing the symptoms of chronic stable angina pectoris, myocardial infarction, and mortality rate in DM patients with ischemic heart disease (12).

The oral bioavailability of ranolazine is 30–50% (13) and for Ranexa® tablets reaches 76% (14). Maximum (or peak) plasma concentrations of ranolazine are normally reached

within 1 hour. The steady-state concentrations will typically be achieved within 3 days after oral administration. Ranolazine binds to α-1-acid glycoprotein in the blood. Although the elimination half-life of pure ranolazine is 1.4 to 1.9 hr, for sustained-release tablet formulation (ranolazine SR) it is about 7 hr. This half-life prolongation is because of the flip-flop kinetics of the SR preparation (15).

Ranolazine is particularly metabolized by cytochrome P450 3A4 (CYP3A4; 70-75%) enzymes and to a lesser extent by CYP2D6 (less than 20%). The main metabolites of ranolazine are created through N-dealkylation of the piperidine ring (CVT-2738 and CVT-4786), O-demethylation (CVT-2514), methoxyphenoxy moiety O-dearylation (CVT-2512), and conjugation with glucuronide (CVT-5431). Consequently, 5% of ranolazine is excreted unaltered through the kidney (12, 16, 17) (Figure 1). Additionally, P-glycoprotein (P-gp) plays an important role in the reabsorption of ranolazine (18).

Since DM is known to affect the pharmacokinetics of drugs; the pharmacokinetics of ranolazine may also be altered in DM conditions, which will necessitate proper dose adjustment. For this reason, we decided to compare the pharmacokinetics of this drug and its metabolite CVT-2738 in DM and non-DM rats for the first time.

Materials and Methods

Ranolazine (Ranexa® 500 mg, $C_{24}H_{33}N_3O_4$) was obtained from A. Menarini Pharma (UK) Company. The standard working of ranolazine and CVT-2738 (1-[(2,6-dimethylphenyl) aminocarbonylmethyl] Piperazine; $C_{14}H_{21}N_3O$) was provided by Tinab Shimi Company (Mashhad, Iran). Acetonitrile gradient grade (CH₃CN, ≥ 99.9 % purity; CAT. No. 100030), Methanol gradient grade (CH₃OH, ≥ 99.9 %; CAT. No. 106007), Diethyl ether ($C_4H_{10}O$, ≥ 99.7 %; CAT. No. 100921), and Ortho-Phosphoric acid (H_3PO_4, 85%; CAT. No. 100573) were purchased from Merck Company (Rotexmeica, Germany). Potassium didrohygen phosphate (KH_2PO_4, >99.5%; CAT. No. CL00.1146) was obtained from Chem-Lab Company (Zedelgem, Belgium). Deionized water was used in the analytical experiment. Moreover, other solvents and chemicals used in the present study were of analytical grade.

Experimental animals

Male rats (Wistar strain; 260–300 g) were ordered from Mashhad University of Medical Sciences, School of Pharmacy, Iran, Mashhad. The animals were housed in husk-filled polycarbonate plastic cages (25 ×15× 40 cm),

Figure 1. Metabolism pathway of ranolazine

in a regular 24-hr interval and controlled light-dark cycle (12 hr-12 hr), temperature (21 ± 2 °C) circumstances. They freely had access to fresh water and a standard rodent chow diet (Behparvar Co, Karaj, Iran).

In vivo preclinical pharmacokinetic analyses

The male Wistar rats were divided into two groups: DM and non-DM. On day 0, the non-DM group was treated with an intraperitoneal (IP) injection of normal saline; and DM animals after 10 hr fasting received an IP injection of 55 mg/kg STZ in normal saline (19). Plenty of water was available for the animals to prevent dehydration. The blood glucose at 72 hr after STZ injection was checked by Easy Gluco (Germany) and AccuCheck glucometer (Per forma, Roche, USA). Any animal having FBS over 250 mg/dl was considered diabetic (20).

The DM and non-DM rats since day 10 after STZ or normal saline injection received 80 mg/kg ranolazine (oral; once a day) dissolved in normal saline (freshly prepared daily) for 7 days. Blood samples were collected immediately 0 (prior to dosing), 1, 2, 3, 4, 8, and 12 hr after administration of the 7th dose of ranolazine. Six rats were used for every time point. The animals were sedated and the blood via retro-orbital plexus was collected. The serum was separated and stored at −80 °C until the extraction phase of ranolazine.

HPLC method
Chromatographic conditions

HPLC (A Shimadzu Prominence LC-20AD Liquid Chromatograph), an SPD-20A variable wavelength programmable UV/Vis detector, a CBM-20Alite system controller (Shimadzu Corporation, Gangnam-gu, Seoul, Korea). The analytical RP-C-18 column (Adamas® C18-Extreme; 5 µm, 250 mm x 4.6 mmID) was used in chromatography. The mobile phase consisted of phase A (phosphate buffer (pH = 2.2; 20 mM)) and phase B (acetonitrile); the pH was set up by ortho-phosphoric acid (1 mol/l). The gradient elution method was used as described in Table 1. The run time was adjusted to 16 min and the peak area of the samples was recorded by the Lab Solution Software. During the mobile phase preparation steps, the solvents were passed through a membrane filter and ultrasonically degassed before use. In all steps of the present experiment, the mobile phase was moved through the column at a flow rate of 1 ml/min and 40 °C. The UV detector at 214 nm was utilized. In each run, 20 µl of samples were injected.

Method validation

Linearity, precision, accuracy, the limit of detection (LOD), and limit of quantitation (LOQ) of the method were estimated according to the ICH Q2 (R1) guideline "validation and analytical procedures: text and methodology" (21).

Table 1. The Gradient elution method for the determination of ranolazine and CVT-2738

Time (mean)	Module	Command	Value
0.01	Pumps	Solvent B	0
3	Pumps	Solvent B	40
8	Pumps	Solvent B	40
11	Pumps	Solvent B	100
14	Pumps	Solvent B	0
16	Pumps	Solvent B	0
16	Controller	Stop	-

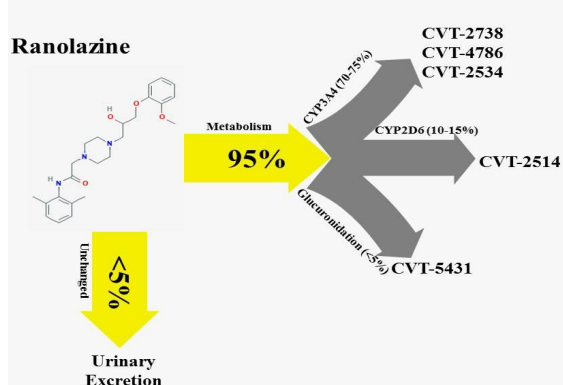The mobile phase consisted of phase A (phosphate buffer (pH = 2.2; 20 mM)) and phase B (acetonitrile)

Linearity

To evaluate the linearity and construction of the calibration curve, eight-point calibration curves in the range of 4.875–312 ng/ml of ranolazine and 1250–20000 ng/ml of CVT-2738 were determined with triplicate measurement of each peak area of ranolazine and CVT-2738 in different concentrations (22).

Limit of detection and limit of quantitation

LOD and LOQ of the present analytical method were estimated according to 3.3:1 and 10:1 signal-to-noise ratio, respectively, by the following equations (23):

$$LOD = \frac{std\ y\ intercept}{slope\ of\ calibration\ curve} \times 3.3$$

$$LOQ = \frac{std\ y\ intercept}{slope\ of\ calibration\ curve} \times 10$$

Accuracy and precision

Accuracy was analyzed by measuring the percentage of recovery of three concentration levels of ranolazine and CVT-2738 (50%, 100%, and 150%) at five different times. Precision was also calculated by measuring the standard deviation at intra-day and inter-day five times (24).

Sample preparation

Concerning the serum extraction, 0.5 ml of each rat serum was added to 0.5 ml of diethyl ether and vortexed for 10 min and then was sonicated for 15 min (two times). After that, the content was centrifuged at 10000 rpm for 15 min at 4 °C (three times). The upper separated layer was collected and evaporated by nitrogen gas (18). The rat serum residues were immediately reconstituted in methanol and then injected into the HPLC system.

Pharmacokinetic analysis

The non-compartmental pharmacokinetic model was carried out using the Microsoft Excel add-in program PKSolver (25). The pharmacokinetic parameters including the area under the curve of serum concentration (AUC) versus time, the maximum concentration recorded (C_{max}), the time taken to reach C_{max} (T_{max}), the half-life of elimination ($T\frac{1}{2}$), the volume of distribution (V_d), elimination rate (K_e), and total drug clearance (CL) were determined (26).

Statistical analysis

The analytical data were expressed as mean ± SD. Statistical analyses were conducted using GraphPad Prism 8.0 (GraphPad Prism Software Inc., San Diego, USA). The significance was determined by the independent samples *t*-test. The findings were significant at *P*<0.05 level.
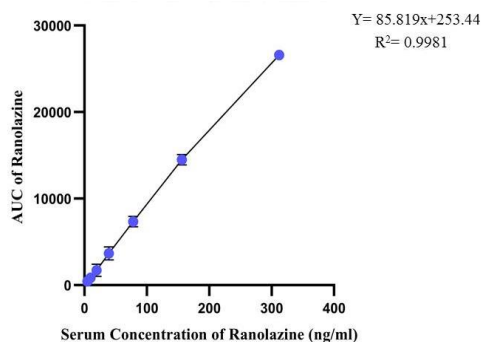

Figure 2. Calibration curve of ranolazine

Results

HPLC results

The findings of the chromatographic method validation of ranolazine and CVT-2738 were mean absolute recovery (91.8 and 92.4%; respectively); intra-and inter-day repeatability was less than 10% for serum concentrations. The standard curves represented excellent linearity for ranolazine and CVT-2738 with coefficients of correlation (r) more than 0.99 (Figures 2 and 3). The lower limits of detection (LOD) of ranolazine and CVT-2738 were 24.76 ng/ml and 940 ng/ml, respectively. The limits of quantitation (LOQ) of ranolazine and CVT-2738 were 51.59 ng/ml and 1200 ng/ml, respectively.

The pharmacokinetic profile of ranolazine and its metabolite CVT-2738 was determined after the 7th administration of ranolazine (80 mg/kg b.w., oral) in normal saline to DM and non-DM rats. The serum concentration of ranolazine versus the time profile appears in Figure 4.

Effects of DM on the absorption pharmacokinetic parameters of ranolazine (AUC, C_{max}, and T_{max})

The significantly lower AUC_{0-12} of ranolazine (3164.33 ± 501.87 ng/ml/min) was observed in DM compared with non-DM animals (8036.50 ± 2377.93 ng/ml/min) after oral administration of 80 mg/kg ranolazine (*P*<0.001).

The orally administered ranolazine showed significantly lower C_{max} (899.94 ± 387.85 ng/ml) in DM compared with non-DM rats (1911.25 ± 975.90 ng/ml; *P*<0.05).

The maximum serum concentration of ranolazine was achieved in about 1 ± 0.00 hr after oral administration in DM and 1 ± 0.55 hr in non-DM animals, and the difference was not statistically significant (*P*>0.05).

Effect of DM on the volume of distribution of ranolazine

A higher V_d of ranolazine was observed in DM animals (133.70 ± 2.81 L/Kg) compared with non-DM animals (60.77 ± 2.32 L/Kg; *P*<0.001).
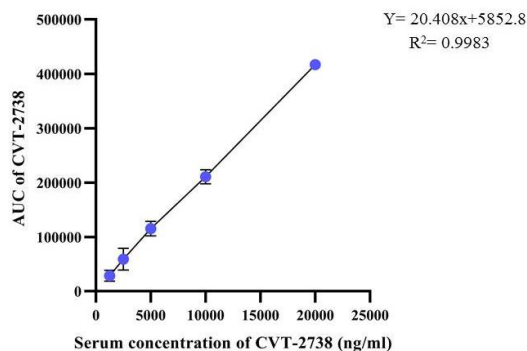

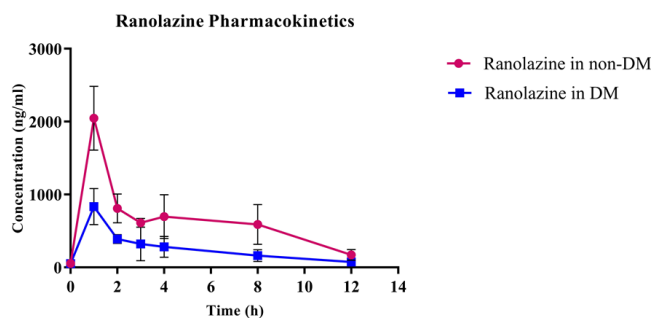**Figure 3.** Calibration curve of CVT-2738

Figure 4. The pharmacokinetic profiles of ranolazine after oral administration in DM and non-DM groups of rats

Table 2. Steady-state pharmacokinetic parameters after oral (80 mg/kg) administration of ranolazine to DM and non-DM rats

Pharmacokinetic parameters	Non- rats DM	DM rats
AUC_{0-12} (ng/ml/h)	8036.50 ± 2377.93	3164.33 ± 501.87[***]
C_{max} (ng/ml)	1911.25 ± 975.90	899.94 ± 387.85[*]
T_{max} (h)	1 ± 0.55	1 ± 0.00
Half-life (h)	4.10 ± 1.93	4.83 ± 2.44
V_d (L/kg)	60.77 ± 2.32	133.70 ± 2.81[***]
CL (ng/ml)/h	9.91 ± 4.83	21.80 ± 5.00[**]
K_{el} (1/h)	0.16 ± 0.04	0.17 ± 0.05

Values are expressed as means ± SD (n=6). Statistical independent samples t-test was used to assess the level of statistical differences between the groups: ***= *P*<0.001, **=*P*<0.01, and *=*P*<0.05 for comparison with non-DM animals DM: diabetes mellitus, Kel: elimination constant; half-life: time half-life; AUC 0-12: area under plasma concentration/time plot until the last quantifiable value; Cl: clearance; Vd: volume of distribution; Ka: absorption constant; Cmax: maximum concentration; Tmax: time to reach Cmax

Effects of DM on clearance of ranolazine

The CL of ranolazine in DM rats (21.80 ± 5.00 ng/ml/h) was significantly higher than in non-DM rats (9.91 ± 4.83 ng/ml/hr; *P*<0.01). There were no significant changes in K_e of ranolazine in DM and non-DM rats (*P*>0.05).

The pharmacokinetic parameters of ranolazine were calculated and are presented in Table 2.

Discussion

Since ranolazine is prescribed in DM patients with angina (27), we decided to compare the pharmacokinetic profile of this drug and its metabolite CVT-2738 between DM and non-DM states. The findings indicated that DM significantly decreases AUC and C_{max} while it increases CL and V_d of this drug. Furthermore, DM did not affect K_e, $T_{1/2}$, and T_{max} of ranolazine.

In the present study, the AUC and C_{max} of ranolazine in DM animals were significantly lower than in non-DM animals. We suggest that DM as a P-gp expression inducer with no effect on metabolism (28) is able to reduce the AUC of ranolazine. Alfarisi *et al.* (29) reported a decline in plasma isoniazid and pyrazinamide concentrations in DM through several processes including elevation in intestinal motility, expression, and activity of P-gp. Adithan *et al.* (30) reported lower steady-state concentrations of phenytoin in patients with DM compared with controls. Nevertheless, the AUC of phlorizin was increased after down-regulation of P-gp and elevated intestinal tract permeability in DM rats (31).

Based on our previous study (32) showing inhibition of CYP3A2 in DM rats, we expected an increase in the serum concentration of ranolazine. However, in the current study, we observed that the increased V_d was an important factor influencing the pharmacokinetics of ranolazine, thereby decreasing its serum concentration. Different drugs have been shown with lower, equivalent, or higher distribution in DM versus non-DM conditions (33). DM can lead to an increase in V_d of drugs (34). Recently, Fediuk *et al.* (35) suggested that DM contributes to an enhancement in the apparent central volume of distribution (Vc/F) of ertugliflozin, thereby diminishing its C_{max} without any effect on the AUC value of the mentioned drug. Higher V_{dss} for torasemide and omeprazole in DM rats compared with control rats have also been reported (36). Patients with DM have increased V_d for paracetamol (acetaminophen) and theophylline (37, 38). In contrast, decreased V_1 and V_{dss} of glimepiride in type 2-DM rats in comparison with those of the control and type 1-DM groups were reported (39). DM by glycosylation of plasma proteins reduces the binding of the drug to the relevant protein and increases the concentration of the free form of the drug in the blood. Therefore, the free drug can be distributed from plasma to the tissues, thereby increasing V_d (40). The V_d of lipophilic drugs may be affected by DM, and the increase in the lipophilicity of drugs is proportional to their V_d (41). Since ranolazine is a drug with high lipophilicity (42), we propose that DM has led to an increase in the distribution of this drug in the current study.

Considering that the K_e of ranolazine was similar in DM and non-DM groups, we assume that the elevated V_d of this lipophilic drug resulted in the increase of CL, thereby decreasing the AUC of ranolazine in DM animals. Apart from the fact that V_d can affect the CL of drugs, in the following, we will review studies regarding the effect of DM on the CL of drugs. DM predisposes to micro-and macrovascular disorders; hence it results in hyperfiltration and elevation in glomerular filtration rate. As shown in previous studies, a positive correlation exists between the doses of drugs and the glomerular filtration rate (28, 43). Through elevation of urine output, DM can increase the renal elimination of medications (44). Alteration of urine pH in DM conditions as a result of ketone production can affect urinary elimination of drugs (45). By speeding up the hepatic blood flow, DM increases the first-pass effect and then increases the hepatic CL, thereby increasing the total CL (46). A 3.4-fold increase in oral chlorzoxazone CL and then lower oral AUC were expressed in Type II-DM patients in comparison to healthy subjects (47). Likewise, after oral administration of clarithromycin to non-DM and DM rats, faster CL, thereby lower AUC of the mentioned drug in DM rats, was distinguished (48). Lee *et al.* in 2013 (49) reported similar urinary excretion of metoprolol in DM and non-DM rats. In contrast to our findings, DM rats exhibited lower CL but higher C_{max}/AUC of paclitaxel compared with non-DM rats (50).

In the current study, we expected an increase in the serum concentration of ranolazine, but the increased V_d was an important factor influencing the pharmacokinetics of the drug. This increased the CL of ranolazine and thereby decreased its serum concentration. The present findings imply that Streptozotocin-Induced DM can affect the pharmacokinetic profile of ranolazine. This finding highlights the need for dose adjustment of ranolazine in DM patients after clinical research.

Conclusion

The findings of our study indicated that increased V_d of ranolazine in DM rats resulted in the elevation of ranolazine CL and thereby a significant decrease in AUC and C_{max} of the drug. Further studies are required to explore the dosage adjustment of ranolazine in DM patients with chronic angina.

Acknowledgment

Authors' Contributions

HMS Performed acquisition, analysis, and drafting of the work. HK Helped with investigation, acquisition, and analysis. SM Helped write, review, and edit. AHS Proofread the paper. MI Reviewed and edited. AHM Designed the work and interpreted the data. HMS, HK, SM, AHS, MI, and AHM Approved the final version to be published.

Ethics Approval

Ethical number: IR.MUMS.PHARMACY.REC.1397.051

Conflicts of Interest

The authors state no conflicts of interest.

References

1. Mashayekhi-Sardoo H, Mohammadpour AH, Nomani H, Sahebkar A. The effect of diabetes mellitus on pharmacokinetics, pharmacodynamics and adverse drug reactions of anticancer drugs. J Cell Physiol 2019; 234:19339-19351.
2. Vahabzadeh M, Mohammadpour A-H. Effect of diabetes mellitus on the metabolism of drugs and toxins. J Clin Toxicol 2015; 5:2161-0495.1000233.
3. Mashayekhi-Sardoo H, Atkin SL, Montecucco F, Sahebkar A. Potential alteration of statin-related pharmacological features in diabetes mellitus. biomed res. Int 2021; 2021:6698743.
4. Hasler WL, Coleski R, Chey WD, Koch KL, McCallum RW, Wo JM, *et al.* Differences in intragastric pH in diabetic vs. idiopathic gastroparesis: relation to degree of gastric retention. Am J Physiol Gastrointest Liver Physiol 2008; 294:G1384-1391.
5. Dostalek M, Akhlaghi F, Puzanovova M. Effect of diabetes mellitus on pharmacokinetic and pharmacodynamic properties of drugs. Clin. Pharmacokinet 2012; 51:481-499.
6. Hussain S. Effect of acarbose on the bioavailability and pharmacokinetics of metronidazole in healthy and diabetic subjects. Br J Pharm Res 2012; 2:41-49.
7. Perea-Jacobo R, Muñiz-Salazar R, Laniado-Laborin R, Cabello-Pasini A, Zenteno-Cuevas R, Ochoa-Terán A. Rifampin pharmacokinetics in tuberculosis-diabetes mellitus patients: a pilot study from Baja California, Mexico. Int J Tuberc Lung Dis 2019; 23:1012-1016.
8. Mohd Sazlly Lim S, Sinnollareddy M, Sime FB. Challenges in Antifungal Therapy in Diabetes Mellitus. J Clin Med 2020; 1-9. Dostalek M, Court MH, Yan B, Akhlaghi F. Significantly reduced cytochrome P450 3A4 expression and activity in liver from humans with diabetes mellitus. Br. J. Pharmacol 2011; 163:937-947.
10. Cheguri S, Ajmera R, Ciddi V. Pharmacokinetic and pharmacodynamic interaction of quercetin and saxagliptin in normal and diabetic rats. Pharmacologia 2017; 8:90-94.
11. Caudle AS, Kim HJ, Tepper JE, O'Neil BH, Lange LA, Goldberg RM, *et al.* Diabetes mellitus affects response to neoadjuvant chemoradiotherapy in the management of rectal cancer. Ann Surg Oncol 2008; 15:1931-1936.
12. Nash DT, Nash SD. Ranolazine for chronic stable angina. The Lancet 2008; 372:1335-1341.
13. Zhao L, Li H, Jiang Y, Piao R, Li P, Gu J. Determination of ranolazine in human plasma by liquid chromatographic-tandem mass spectrometric assay. J Chromatogr Sci 2008; 46:697-700.
14. Acting Nitrates L. Abbreviated New Drug Evaluation: Ranolazine month/year of review: August 2012 End date of literature search: May 2012 Generic Name: Ranolazine Brand Name (Manufacturer): Ranexa®(CV Therapeutics). 2012.
15. Abdallah H, Jerling M. Effect of hepatic impairment on the multiple-dose pharmacokinetics of ranolazine sustained-release tablets. J Clin Pharmacol 2005; 45:802-809.
16. Jerling M. Clinical pharmacokinetics of ranolazine. Clin Pharmacokinet 2006; 45:469-491.

17. Dobesh PP, Trujillo TC. Ranolazine: a new option in the management of chronic stable angina. Pharmacotherapy 2007; 27:1659-1676.
18. Babu PR, Babu KN, Peter PL, Rajesh K, Babu PJ. Influence of quercetin on the pharmacokinetics of ranolazine in rats and in vitro models. Drug Dev Ind Pharm 2013; 39:873-879.
19. Seedevi P, Ramu Ganesan A, Moovendhan M, Mohan K, Sivasankar P, Loganathan S, *et al.* Anti-diabetic activity of crude polysaccharide and rhamnose-enriched polysaccharide from G. lithophila on streptozotocin (STZ)-induced in Wistar rats. Sci Rep 2020; 10:1-12.
20. King AJF. The use of animal models in diabetes research. Br J Pharmacol 2012; 166:877-894.
21. Guideline IHT. Validation of analytical procedures: text and methodology. Q2 (R1) 2005; 1:1-13.
22. Kurmi M, Kumar S, Singh B, Singh S. Implementation of design of experiments for optimization of forced degradation conditions and development of a stability-indicating method for furosemide. J Pharm Biomed 2014; 96:135-143.
23. Taleuzzaman M. Limit of blank (LOB), limit of detection (LOD), and limit of quantification (LOQ). Organic & Medicinal Chemistry International Journal, Juniper Publishers Inc., 7: 127-131.
24. Chen L, Chen J, Lu M, Stämpfli A. Simultaneous determination of elbasvir and grazoprevir in fixed-dose combination and mass spectral characterization of each degradation product by UHPLC-ESI-QTOF-MS/MS. J. Pharm. Biomed 2020; 178:112964.
25. Zhang Y, Huo M, Zhou J, Xie S. PKSolver: An add-in program for pharmacokinetic and pharmacodynamic data analysis in Microsoft Excel. Comput Methods Programs Biomed 2010; 99:306-314.
26. Ahmed T. Pharmacokinetics of Drugs Following IV Bolus, IV Infusion, and Oral Administration. 2015. p. 53-89.
27. Nguyen E, Coleman CI, Kohn CG, Weeda ER. Ranolazine in patients with type 2 diabetes and chronic angina: A cost-effectiveness analysis and assessment of health-related quality-of-life. Int J Cardiol 2018; 273:34-38.
28. Yao H, Gu J, Shan Y, Wang Y, Chen X, Sun D, *et al.* Type 2 diabetes mellitus decreases systemic exposure of clopidogrel active metabolite through upregulation of P-glycoprotein in rats. Biochem Pharmacol 2020; 180:114142.
29. Alfarisi O, Mave V, Gaikwad S, Sahasrabudhe T, Ramachandran G, Kumar H, *et al.* Effect of Diabetes Mellitus on the Pharmacokinetics and Pharmacodynamics of Tuberculosis Treatment. Antimicrob Agents Chemother 2018; 62: e01383-18.
30. Adithan C, Srinivas B, Indhiresan J, Shashindran CH, Bapna JS, Thakur LC, *et al.* Influence of type I and type II diabetes mellitus on phenytoin steady-state levels. Int J Clin Pharmacol Ther Toxicol 1991; 29:310-313.
31. Wang Z, Gao Z, Wang A, Jia L, Zhang X, Fang M, *et al.* Comparative oral and intravenous pharmacokinetics of phlorizin in rats having type 2 diabetes and in normal rats based on phase II metabolism. Food Funct 2019; 10:1582-1594.
32. Mashayekhi-Sardoo H, Mohammadpour AH, Mehri S, Kamali H, Sahebkar A, Imenshahidi M. Diabetes mellitus aggravates ranolazine-induced ECG changes in rats. J Interv Card Electrophysiol 2021; 63: 379-388.
33. Tran M, Elbarbry F. Influence of diabetes mellitus on pharmacokinetics of drugs. MOJ Bioequiv Availab 2016; 2:00016.
34. Lee JH, Yang SH, Oh JM, Lee MG. Pharmacokinetics of drugs in rats with diabetes mellitus induced by alloxan or streptozocin: Comparison with those in patients with type I diabetes mellitus. J Pharm Pharmacol 2010; 62:1-23.
35. Fediuk DJ, Zhou S, Dawra VK, Sahasrabudhe V, Sweeney K. Population pharmacokinetic model for ertugliflozin in healthy subjects and patients with type 2 diabetes mellitus. Clin Pharmacol Drug Dev 2020; 10: 696–706.
36.Lee DY, Lee MG, Shin HS, Lee I. Changes in omeprazole

pharmacokinetics in rats with diabetes induced by alloxan or streptozotocin: faster clearance of omeprazole due to induction of hepatic CYP1A2 and 3A1. J Pharm Pharm Sci 2007; 10:420-433.

37. Adithan C, Danda D, Swaminathan RP, Indhiresan J, Shashindran CH, Bapna JS, *et al.* Effect of diabetes mellitus on salivary paracetamol elimination. Clin Exp Pharmacol Physiol 1988; 15:465-471.

38. Adithan C, Danda D, Shashindran CH, Bapna JS, Swaminathan RP, Chandrasekar S. Differential effect of type I and type II diabetes mellitus on antipyrine elimination. Methods Find Exp Clin Pharmacol 1989; 11:755-758.

39. Kiriyama A, Kimura S, Banba C, Yamakawa M, Yajima R, Honbo A, *et al.* Pharmacokinetic-pharmacodynamic analyses of anti-diabetes, glimepiride: comparison of the streptozotocin-induced diabetic, GK, and Wistar rats. J Drug Metab Toxicol 2017; 8: 1-6.

40. Gour A, Dogra A, Sharma S, Wazir P, Nandi U. Effect of disease state on the pharmacokinetics of bedaquiline in renal-impaired and diabetic rats. ACS Omega 2021; 6: 6934-6941.

41. Robert G. Smith DPM M, RPh, CPed. Understanding How Diabetes Affects Patient Response To Medications. Podiatry Today 2012; 25:14-20.

42. Abdelsayed M, Ruprai M, Ruben PC. The efficacy of ranolazine on E1784K is altered by temperature and calcium. Sci Rep 2018; 8:1-20.

43. Meeme A, Kasozi H. Effect of glycaemic control on glomerular filtration rate in diabetes mellitus patients. Afr Health Sci 2009; 9 Suppl 1:S23-26.

44. Chiou WL. A new simple approach to study the effect of changes in urine flow and/or urine pH on renal clearance and its applications. Int J Clin Pharmacol Ther Toxicol 1986; 24:519-527.

45. Ryan JP, Irwin JA. Practical studies on urine demonstrating principles of clinical and veterinary significance. Biochem Mol Biol Educ 2002; 30:98-100.

46. Gwak EH, Yoo HY, Kim SH. Effects of Diabetes Mellitus on the Disposition of Tofacitinib, a Janus Kinase Inhibitor, in Rats. Biomol Ther (Seoul) 2020; 28:361-369.

47. Wang Z, Hall SD, Maya JF, Li L, Asghar A, Gorski JC. Diabetes mellitus increases the in vivo activity of cytochrome P450 2E1 in humans. Br J Clin Pharmacol 2003; 55:77-85.

48. Kim YC, Lee JH, Kim SH, Lee MG. Effect of CYP3A1(23) induction on clarithromycin pharmacokinetics in rats with diabetes mellitus. Antimicrob Agents Chemother 2005; 49:2528-2532.

49. Lee U, Lee I, Lee BK, Kang HE. Faster non-renal clearance of metoprolol in streptozotocin-induced diabetes mellitus rats. Eur J Pharm Sci 2013; 50:447-453.

50. Lee JH, Lee A, Oh JH, Lee YJ. Comparative pharmacokinetic study of paclitaxel and docetaxel in streptozotocin-induced diabetic rats. Biopharm Drug Dispos 2012; 33:474-486.