International Journal of Pharmacology

ISSN 1811-7775

science alert

ansinet
Asian Network for Scientific Information

International Journal of Pharmacology 9 (2): 164-169, 2013
ISSN 1811-7775 / DOI: 10.3923/ijp.2013.164.169

Study on the Pharmacokinetics Drug-drug Interaction of Danmo Capsules with Prednisone in Rats

[1]Bin Ren, [1]Binghua Wei, [1]Ruiming Li, [1]Liang Huang, [1]Xiaodan Hong,
[2]Xiaohua Fu and [1]Xiao Chen
[1]Department of Pharmacy, The First Affiliated Hospital, Sun Yat-sen University, Guangzhou 510080, China
[2]Department of Pharmacy, Guangzhou Xinhai Hospital, Guangzhou 510300, China

Abstract: The Danmo Capsule (DMC) is a widely used patent Chinese botanic drug, prepared with the extract of *Radix salviae miltiorrhizae* and *Ecliptae prostratae*. Drug-drug interaction of DMC and prednisone was investigated in rats via *in vivo* pharmacokinetic studies. After pretreatment with DMC at daily dosages of 0.432 g kg^{-1} for 14 consecutive days, there were significant decrease in the peak plasma concentration (C_{max}) of prednisone (from 174.6±32.9 ng mL^{-1} to 68.2±35.2 ng mL^{-1}, $p<0.01$) and in the area under the plasma concentration-time curve ($AUC_{0\text{-}2.5h}$) of prednisone (from 225.9±55.8 h.ng mL^{-1} to 128.8±37.5 h.ng mL^{-1}, $p<0.05$), respectively. In contrast to prednisone, the C_{max} of prednisolone was significantly increased from 155.9±40.4 ng mL^{-1} to 333.20±95.8 ng mL^{-1} ($p<0.05$) and the $AUC_{0\text{-}8h}$ of prednisolone was significantly increased from 467.5±35.3 h.ng mL^{-1} to 757.3±105.4 h.ng mL^{-1} ($p<0.01$), respectively. However, no statistically significant differences for the elimination half-life ($t_{1/2}$) and Mean Residence Time (MRT) of prednisone and prednisolone were detected. In conclusion, the increased C_{max} and AUC of prednisolone showed co-administration with DMC significantly increased exposure of prednisolone.

Key words: Danmo capsules, prednisone, prednisolone, interaction, pharmacokinetics

INTRODUCTION

Prednisone is one of the most commonly prescribed drugs in treatment of immunologic diseases such as rheumatoid arthritis, polymyositis, systemic lupus erythematosus and nephrotic syndrome. Prednisone is also an ester prodrug that is rapidly and extensively converted to the active 11β-hydroxylmetabolite, prednisolone, through the actions of 11β-hydroxysteroid dehydrogenase I (11β-HSD I) (Conti *et al*., 1994; Garg and Jusko, 1994; Jusko and Rose, 1980). Complex pharmacokinetics of prednisone and prednisolone includes hydroxylation by CYP3A4, hepatic conjugation, interconversion between prednisone and prednisolone, efflux by P-gp and renal excretion of unchanged drug (Frey and Frey, 1990; Karssen *et al*., 2002; Pichard *et al*., 1992; Yates *et al*., 2003). It has been reported that simultaneous use of drug metabolizing enzyme inducer, e.g., rifampicin or phenobarbital, with prednisolone can induce the metabolism and increase the clearence of prednisolone, worsen the symptoms of the disease being treated, e.g., nephrotic syndrome or rheumatoid arthritis (Bartoszek *et al*., 1987; Brooks *et al*., 1976; Hendrickse *et al*., 1979; McAllister *et al*., 1983).

The Danmo Capsule (DMC) is a widely used patent Chinese botanic drug, prepared with the extract of *Radix Salviae miltiorrhizae* and *Ecliptae prostratae*. In China, it is widely used with prednisone in clinics to treat renal diseases, such as glomerulonephritis, nephrotic syndrome and lupus nephritis, etc. DMC could produce a synergistic effect with prednisone and decrease glucocorticoids dosage. Complex *Radix Salviae miltiorrhizae* extract-drug interactions with warfarin (Lo *et al*., 1992; Wu and Yeung, 2010; Zhou *et al*., 2012), midazolam (Wang *et al*., 2010), diazepam (Jinping *et al*., 2003) via pharmacokinetic mechanisms were observed previously. Therefore, pharmacokinetic interaction between DMC and prednisone might occur and increase pharmacological effects of glucocorticoids. The present study was carried out to estimate the potential effects of repeated consumption of DMC on the pharmacokinetics of orally administered prednisone in rats.

Corresponding Author: Bin Ren, Department of Pharmacy, The First Affiliated Hospital, Sun Yat-sen University, Guangzhou, China, No. 58, Zhongshan No.2 Road, Guangzhou 510080, China Tel: +86 20 87755766-8442, Fax: +86 20 87334586

MATERIALS AND METHODS

Chemicals and reagents: Prednisone, prednisolone, dexamethasone and sodium carboxymethyl cellulose (CMC-Na) were purchased from Sigma-Aldrich Corporation (St. Louis, MO, USA). Danmo capsules (DMC, 0.4 g per capsule, registration No: Z20070864) were produced by Luoding Pharmaceutical Inc. (Guangdong, China). Plant extract from 20 Danmo capsules was suspended in 1% aqueous CMC-Na with the final concentration of 0.04 g mL^{-1}. Prednisone acetate tablets (5 mg per tablet) were obtained from Huanan Pharmaceutical Inc. (Guangdong, China). Sodium heparin injection was from Tianjing Biochem Pharmaceutical Inc. (Tianjing, China). Methanol and acetonitrile of HPLC grade were provided from TEDIA Company Inc. (Beijing, China). All other reagents were of analytical grade.

Animals: Male Sprague-Dawley rats (220-310 g) were obtained from the Laboratory Animal Service Center of Guangdong province (Guangzhou, China). The animals were maintained in a room with a light/dark cycle of 12/12 h and 55-60% relative humidity at 22-24°C. They were allowed to eat standard laboratory chow with water freely and fasted overnight before the experiments. All procedures were in accordance with the Regulations of Experimental Animal Administration issued by the Ministry of Science and Technology of the People's Republic of China.

Pharmacokinetic study in rats: Twelve rats were classified into two groups. In the experiment group, the rats received DMC orally twice a day (at a dosage of 0.432 g kg^{-1} per day of rat weight) for 14 consecutive days. The dosage was calculated from that of clinical use. The control group received 1% aqueous CMC-Na alone. On day 13, all the rats were anesthetized with ether and the right jugular vein was cannulated as described before (Qin *et al*., 2010). Later, the rats were kept individually in cage and not fed for 12 h prior to the pharmacokinetic study. On day 14, 15 min after the last dosing, a single-oral dose of 42 mg kg^{-1} prednisone acetate, suspended in 1% CMC-Na with the concentration of 5 mg mL^{-1}, was administered to all rats. Afterwards, all animals were deprived of water for 2 h and fasted for 4 h.

Blood samples (220 μL) were collected into heparinized tubes from the jugular vein, at 0, 0.083, 0.16, 0.25, 0.5, 0.75, 1.0, 1.5, 2.0, 2.5, 3.0, 4.0, 5.0, 6.0 and 8.0 h following administration of prednisone acetate. After that, the cannula was blushed with the same volume of heparinized saline solution (50 U mL^{-1}) for the purpose of preventing coagulation and to replace the blood loss. Blood was immediately processed for plasma by centrifugation at 6,000 g for 5 min. Plasma samples were frozen and maintained at −37°C until analysis.

Determination of prednisone and prednisolone in plasma samples: The analysis was processed on the reverse-phase HPLC system equipped with waters pump 600, an autosampler 717 and 2489 uv/vis is detector (Milford, MA, USA). To 100 μL of plasma, 20 μL internal standard solutions (IS, 200 μg mL^{-1} dexamethasone in acetonitrile) and 20 μL of 0.1 mol L^{-1} sodium hydroxide, 600 μL ethyl acetate were added, followed by vortex mixing for 1 min. After centrifugation (10,000 g, 4°C, 10 min), the organic phase was transferred to a clean centrifuge tube and evaporated to dryness in a thermostatic controller at 40°C under a slight of nitrogen. After that, the residue was dissolved in 20 μL of acetonitrile, vortexed for 1 min and centrifuged at 10,000 g for 10 min at 4°C and then 10 μL of supernatant was injected into the HPLC system.

The analyte was determined at 40°C on a reverse-phase Nucleodur™ 100-5 C18ec column (250 mm×4.6 mm; particle size, 5 μm). The mobile phases were consisted of methanol: 0.2% phosphoric acid in deionized water (55:45, v/v, A) and acetonitrile (B) and programmed by 100-100% (v/v) A at 0.0-15.0 min, 100-20% A at 15.0-18.0 min, 20-20% A at 18.0–23.0 min, 20-100% A at 23.0-28.0 min and 100-100% A at 28.0-35.0 min. The analysis was carried out at a flow rate of 1.5 mL min^{-1} with the detection wavelength set at 240 nm.

Pharmacokinetic analysis: Pharmacokinetic parameters were calculated by the WinNonlin program (Version 4.0.1, Pharsight Co., Mountain View, CA, USA) with the non-compartmental model. The peak plasma concentration (C_{max}) and time to reach C_{max} (T_{max}) were determined from the actual data obtained after oral administration. The elimination half-life ($t_{1/2}$) was calculated by 0.693/Ke, where Ke was estimated by plotting three terminal points of the plasma concentration profile with a log-linear regression equation using the least-squares method. The area under the plasma concentration-time curve to the last measurable concentration (AUC_{0-t}) was measured according to the linear trapezoidal rule. Mean Residence Time (MRT) was calculated from the equation MRT = $AUMC_{0-t}/AUC_{0-t}$. The Relative Bioavailability (RB%) was calculated by $(AUC_{with\ DMC}/AUC_{control})\times100$. The Metabolite-parent Ratio (MR) was estimated from $(AUC_{prednisolone}/AUC_{prednisone})$.

Statistical analysis: All data were expressed as Mean±SD. Comparison of data on the pharmacokinetic parameters between prednisone and prednisolone was performed by Student's t-test using SPSS version 12.0. The p-value<0.05 was considered statistically significant.

RESULTS AND DISCUSSION

HPLC method characteristics: Figure 1 showed the representative chromatographic profiles of blank plasma, a plasma sample spiked with prednisone, prednisolone and dexamethasone (internal standard, IS) and a plasma sample from a rat 0.5 h after oral administration of prednisone acetate with DMC. No endogenous interference was observed at the retention times of prednisone (9.2 min), prednisolone (11.4 min) and IS (16.7 min). The calibration curve for prednisone and prednisolone showed good linearity (r^2>0.99) over the concentration range from 10-5000 ng mL^{-1} with a weight of $1/x^2$ was applied to minimize the relative error for the curve fitting. The lowest limit of quantification of prednisone and prednisolone was 10 ng mL^{-1} with the accuracy ranged from 90.0-103.9%. The intra-and inter-day precisions (RSD%) for all quality control samples were estimated to be within 3.0-4.3% and 1.4-7.0% for prednisone, 1.4-5.1% and 1.3-7.4% for prednisolone, respectively. The extraction recovery from plasma was measured to be 68.6-74.3% for prednisone and 60.7-71.4% for prednisolone, respectively. Under routine laboratory conditions, stability samples were to be concluded stable. The data above showed that the assay was sensitive enough for pharmacokinetics study of prednisone and prednisolone *in vivo*.

Pharmacokinetic studies: The plasma concentration-time curves of prednisone and prednisolone after a single oral dose of prednisone acetate (42 mg kg^{-1}) to rats, with and without DMC pretreatment, were given in Fig. 2 and 3, respectively. The pharmacokinetic parameters of prednisone and prednisolone were shown in Table 1 and Table 2. In rats pretreated with and without DMC, the prednisone concentration reached peak at 0.5 h and declined to an undetected level at 3.0 h. Prednisolone, the active metabolite of prednisone, was eliminated slower than prednisone and could still be detected till 8.0 h.

After pretreatment with DMC at daily dosages of 0.432 g kg^{-1} for 14 consecutive days, the C_{max} of prednisone was significantly decreased from 174.6±32.9 ng mL^{-1} to 68.2±35.2 ng mL^{-1} ($p<0.01$) and the $AUC_{0\text{-}2.5h}$ of prednisone was significantly decreased from 225.9±55.8 h.ng mL^{-1} to 128.8±37.5 h.ng mL^{-1} ($p<0.05$), respectively. The plasma concentrations of prednisone at 0.16-2.5 h in the DMC group were lower than those in the control group ($p<0.05$), whereas, the plasma

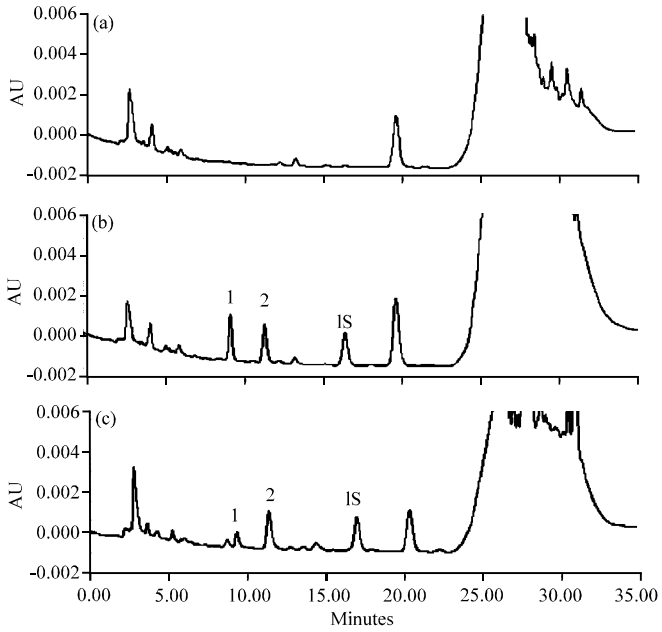

Fig. 1(a-c): HPLC chromatograms (a) Blank plasma, (b) Plasma spikes with prednisone, prednisolone and dexamethasone (internal standard, IS) and (c) Plasma sample obtained 0.5 h after oral administration of 42 mg kg^{-1} prednisone acetate, Peaks 1: Prednisone, 2: Prednisolone

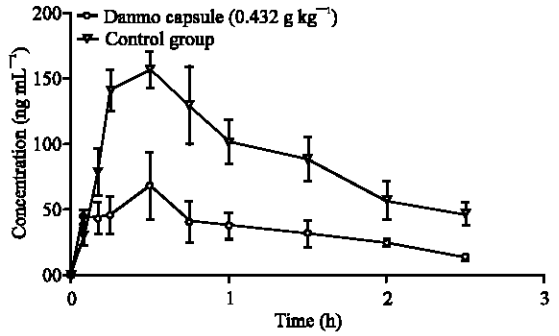

Fig. 2: Mean plasma concentration-time curves of prednisone after a single oral dose of prednisone acetate (42 mg kg^{-1}) to rats with or without orally administered Danmo capsule (0.432 g kg^{-1}) for 14 consecutive days. Each point represents the Mean±SD (n = 6)

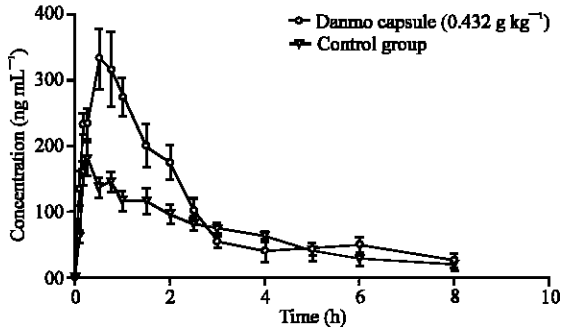

Fig. 3: Mean plasma concentration- time curves of prednisolone after a single oral dose of prednisone acetate (42 mg kg^{-1}) to rats with or without orally administered Danmo capsule (0.432 g kg^{-1}) for 14 consecutive days. Each point represents the Mean±SD (n = 6)

concentrations of prednisolone at 0.08-2.0 h were higher than those in the control group ($p<0.05$). In the elimination phase (2.5-8.0 h) of prednisolone, however, the concentration-time curves for the two groups were similar. In contrast to prednisone, after pretreatment with DMC at daily dosages of 0.432 g kg^{-1} for 14 consecutive days, the C_{max} of prednisolone was significantly increased from 155.9±40.4 ng mL^{-1} to 333.20±95.8 ng mL^{-1} ($p<0.05$) and the AUC_{0-8h} of prednisolone was significantly increased from 467.5±35.3 h.ng mL^{-1} to 757.3±105.4 h.ng mL^{-1} ($p<0.01$), respectively. These data showed co-administration with DMC significantly increased exposure of prednisolone. However, no statistically significant differences for $t_{1/2}$ and MRT of prednisone and prednisolone were detected. It suggested that the elimination of prednisone and prednisolone would not be affected by co-administration of DMC.

Table 1: Pharmacokinetic parameters of prednisone after a single oral dose of prednisone acetate (42 mg kg^{-1}) to rats with or without orally administered Danmo capsule (0.432 g kg^{-1}) 14 consecutive days

Parameters	Control group	With Danmo capsule
$t_{1/2}$ (h)	0.82±0.34	0.76±0.24
T_{max} (h)	0.49±0.22	0.51±0.08
C_{max} (ng mL^{-1})	174.6±32.9	68.2±35.2**
$AUC_{0-2.5h}$ (h.ng mL^{-1})	225.9±55.8	128.8±37.5*
$MRT_{0-2.5h}$ (h)	001.01±0.14	0.96±0.21
RB (%)	100	57.0

*p<0.05, **p<0.01 compared to control group, $AUC_{0-2.5h}$: Area under the plasma concentration-time curve from 0-2.5 h, C_{max}: Peak plasma concentration, T_{max}: Time to reach Cmax, $t_{1/2}$: Terminal half-life, MRT: Mean residence time, RB: Relative bioavailability, (Mean±SD, n = 6)

Table 2: Pharmacokinetic parameters of prednisolone after a single oral dose of prednisone acetate (42 mg kg^{-1}) to rats with or without orally administered Danmo capsule (0.432 g kg^{-1}) 14 consecutive days

Parameters	Control group	With Danmo capsule
$t_{1/2}$ (h)	2.35±1.02	1.91±0.61
T_{max} (h)	0.87±0.08	0.58±0.17
C_{max} (ng mL^{-1})	155.9±40.4	333.20±95.82*
AUC_{0-8h} (h.ng mL^{-1})	467.5±35.3	757.3±105.4**
MRT_{0-8h} (h)	2.64±0.38	2.02± 0.67
MR	2.15±1.23	5.82±1.80*
RB (%)	100	162.0

*p<0.05, **p<0.01 compared to control group, AUC_{0-8h}: Area under the plasma concentration-time curve from 0-8 h, C_{max}: Peak plasma concentration, T_{max}: Time to reach Cmax, $t_{1/2}$: Terminal half-life, MRT: Mean residence time, MR: The metabolite-parent ratio ($AUC_{prednisolone}/AUC_{prednisone}$), RB: Relative bioavailability, (Mean±SD, n = 6)

In the current study, systemic exposure of total prednisone (prednisone+prednisolone) was significantly increased when DMC existed. The AUC value of total prednisone was increased by 27.8% ($p<0.05$) after pretreatment with DMC at daily dosages of 0.432 g kg^{-1} for 14 consecutive days, compared to that of the control group. Passive diffusion and P-gp efflux pumps participate in the intestinal absorption of glucocorticoids. Moreover, glucocorticoids, such as methylprednisolone (Saitoh *et al*., 1998; Tomita *et al*., 2010), budesonide (Dilger *et al*., 2004), prednisone (Dilger *et al*., 2004; Karssen *et al*., 2002; Yates *et al*., 2003) are substrates of P-gp. Acting as the active transporter, P-gp plays an important role in the intestinal excretion of glucocorticoids, thus the bioavalability of hydrocortisone, prednisone and prednisolone can be elevated via the inhibition of intestinal P-gp by its inhibitors, such as verapamil (Farrell *et al*., 2002) and cyclosporin (Potter *et al*., 2004). It has been reported that tanshinone I and tanshinone IIA, two major active constituents of *Radix Salviae miltiorrhizae*, are substrates and inhibitors of P-gp (Li *et al*., 2008; Yu *et al*., 2007). Luteolin and apigenin, two major flavonoids isolated from *Eclipta prostrata*, also known as P-gp inhibitors, can invert multidrug resistance of cancer cells (Chan *et al*., 2006; Du *et al*., 2008; Molnar *et al*., 2004; Ugocsai *et al*., 2005). The bioavailability of total

prednisone increased by DMC might be due to increased intestinal absorption caused by the P-gp inhibition effect.

In the presence of DMC, the Metabolite-parent Ratio (MR) of prednisone increased from 2.15±1.23 (prednisone acetate alone) to 5.82±1.80 ($p<0.05$). It indicated that the presence of DMC facilitated metabolism of prednisone to prednisolone, significantly increased serum prednisolone generation and elevated the metabolite-parent ratio of prednisone. 11β-HSD I is the major metabolic enzyme which converts inactive prednisone to active prednisolone in liver and the plasma prednisolone concentrations are mediated by 11β-HSD I function. Thus, the conversion of oral prednisone to prednisolone in plasma (prednisolone generation study) was proposed as a biomarker to assess 11β-HSDI activity (Courtney *et al*., 2008) and serum prednisolone generation was reduced in patients with Cushing's syndrome after carbenoxolone (a 11β-HSDI inhibitor) administration (Tomlinson *et al*., 2007). The current study showed the increased prednisolone generation, which suggested elevated 11β-HSDI activity after DMC administration.

CONCLUSION

Co-administration of DMC significantly increased the systemic exposure of prednisolone via enhancing the metabolism of prednisone to prednisolone. As a result, obtaining same prednisolone concentration just need lower dosage of predinisone. The pharmacokinetic interaction might be taken into consideration when clincal use.

REFERENCES

Bartoszek, M., A.M. Brenner and S.J. Szefler, 1987. Prednisolone and methylprednisolone kinetics in children receiving anticonvulsant therapy. Clin. Pharmacol. Ther., 42: 424-432.

Brooks, P.M., W.W. Buchanan, M. Grove and W.W. Downie, 1976. Effects of enzyme induction on metabolism of prednisolone. Ann. Rheum. Dis., 35: 339-343.

Chan, K.F., Y. Zhao, B.A. Burkett, I.L. Wong, L.M. Chow and T.H. Chan, 2006. Flavonoid dimers as bivalent modulators for P-glycoprotein-based multidrug resistance: Synthetic apigenin homodimers linked with defined-length poly (ethylene glycol) spacers increase drug retention and enhance chemosensitivity in resistant cancer cells. J. Med. Chem., 49: 6742-6759.

Conti, M., F.J. Frey, G. Escher, C. Marone and B.M. Frey, 1994. Renal handling of prednisolone/prednisone: Effect of steroid dose and 11 beta-hydroxysteroid dehydrogenase. Nephrol. Dial. Transplant, 9: 1622-1628.

Courtney, R., P.M. Stewart, M. Toh, M.N. Ndongo, R.A. Calle and B. Hirshberg, 2008. Modulation of 11beta-hydroxysteroid dehydrogenase (11betaHSD) activity biomarkers and pharmacokinetics of PF-00915275 a selective 11betaHSD1 inhibitor. J. Clin. Endocrinol. Metab., 93: 550-556.

Dilger, K., M. Schwab and M.F. Fromm, 2004. Identification of budesonide and prednisone as substrates of the intestinal drug efflux pump P-glycoprotein. Inflamm. Bowel. Dis., 10: 578-583.

Du, G.J., Z.H. Song, H.H. Lin, X.F. Han, S. Zhang and Y.M. Yang, 2008. Luteolin as a glycolysis inhibitor offers superior efficacy and lesser toxicity of doxorubicin in breast cancer cells. Biochem. Biophys. Res. Commun., 372: 497-502.

Farrell, R.J., M.J. Menconi, A.C. Keates and C.P. Kelly, 2002. P-glycoprotein-170 inhibition significantly reduces cortisol and ciclosporin efflux from human intestinal epithelial cells and T lymphocytes. Aliment. Pharmacol. Ther., 16: 1021-1031.

Frey, B.M. and F.J. Frey, 1990. Clinical pharmacokinetics of prednisone and prednisolone. Clin. Pharmacokinet., 19: 126-146.

Garg, V. and W.J. Jusko, 1994. Bioavailability and reversible metabolism of prednisone and prednisolone in man. Biopharm. Drug Dispos., 15: 163-172.

Hendrickse, W., J. McKiernan, M. Pickup and J. Lowe, 1979. Rifampicin-induced non-responsiveness to corticosteroid treatment in nephrotic syndrome. Br. Med. J., 1: 306-306.

Jinping, Q., H. Peiling, L. Yawei and Z. Abliz, 2003. Effects of the aqueous extract from *Salvia miltiorrhiza* Bge on the pharmacokinetics of diazepam and on liver microsomal cytochrome P450 enzyme activity in rats. J. Pharm. Pharmacol., 55: 1163-1167.

Jusko, W.J. and J.Q. Rose, 1980. Monitoring prednisone and prednisolone. Ther. Drug Monit., 2: 169-176.

Karssen, A.M., O.C. Meijer, I.C. van der Sandt, A.G. De Boer, E.C. De Lange and E.R. De Kloet, 2002. The role of the efflux transporter P-glycoprotein in brain penetration of prednisolone. J. Endocrinol., 175: 251-260.

Li, X.X., Z.W. Zhou and S.F. Zhou, 2008. Role of P-glycoprotein in the transport of tanshinone I one active triterpenoid from *Salvia miltiorrhiza*. Drug Metab. Lett., 2: 223-230.

Lo, A.C., K. Chan, J.H.K. Yeung and K.S. Woo, 1992. The effects of Danshen (*Salvia miltiorrhiza*) on pharmacokinetics and pharmacodynamics of warfarin in rats. Eur. J. Drug Metab. Pharmacokinet., 17: 257-262.

McAllister, W.A., P.J. Thompson, S.M. Al-Habet and H.J. Rogers, 1983. Rifampicin reduces effectiveness and bioavailability of prednisolone. Br. Med. J. (Clin. Res. Ed), 286: 923-925.

Molnar, J., N. Gyemant, I. Mucsi, A. Molnar and M. Szabo *et al*., 2004. Modulation of multidrug resistance and apoptosis of cancer cells by selected carotenoids. *In vivo*, 18: 237-244.

Pichard, L., I. Fabre, M. Daujat, J. Domergue, H. Joyeux and P. Maurel, 1992. Effect of corticosteroids on the expression of cytochromes P450 and on cyclosporin A oxidase activity in primary cultures of human hepatocytes. Mol. Pharmacol., 41: 1047-1055.

Potter, J.M., B.C. McWhinney, L. Sampson and P.E. Hickman, 2004. Area-under-the-curve monitoring of prednisolone for dose optimization in a stable renal transplant population. Ther. Drug Monit., 26: 408-414.

Qin, X.L., H.C. Bi, C.X. Wang, J.L. Li and X.D. Wang *et al*., 2010. Study of the effect of Wuzhi tablet (*Schisandra sphenanthera* extract) on tacrolimus tissue distribution in rat by liquid chromatography tandem mass spectrometry method. Biomed. Chromatogr., 24: 399-405.

Saitoh, H., M. Hatakeyama, O. Eguchi, M. Oda and M. Takada, 1998. Involvement of intestinal P-glycoprotein in the restricted absorption of methylprednisolone from rat small intestine. J. Pharm. SSci., 87: 73-75.

Tomita, M., A. Watanabe, I. Fujinaga, T. Yamakawa and M. Hayashi, 2010. Nonlinear absorption of methylprednisolone by absorptive and secretory transporters. Int. J. Pharm., 387: 1-6.

Tomlinson, J.W., M. Sherlock, B. Hughes, S.V. Hughes and F. Kilvington *et al*., 2007. Inhibition of 11β-hydroxysteroid dehydrogenase type 1 activity in vivo limits glucocorticoid exposure to human adipose tissue and decreases lipolysis. J. Clin. Endocrinol. Metab., 92: 857-864.

Ugocsai, K., A. Varga, P. Molnar, S. Antus and J. Molnar, 2005. Effects of selected flavonoids and carotenoids on drug accumulation and apoptosis induction in multidrug-resistant colon cancer cells expressing MDR1/LRP. *In vivo*, 19: 433-438.

Wang, X., W.Y. Lee, X. Zhou, P.M. Or and J.H. Yeung, 2010. A pharmacodynamic-pharmacokinetic (PD-PK) study on the effects of Danshen (*Salvia miltiorrhiza*) on midazolam a model CYP3A probe substrate in the rat. Phytomedicine, 17: 876-883.

Wu, W.W.P. and J.H.K. Yeung, 2010. Inhibition of warfarin hydroxylation by major tanshinones of Danshen (*Salvia miltiorrhiza*) in the rat *In vitro* and *In vivo*. Phytomedicine, 17: 219-226.

Yates, C.R., C. Chang, J.D. Kearbey, K. Yasuda and E.G. Schuetz *et al*., 2003. Structural determinants of P-glycoprotein-mediated transport of glucocorticoids. Pharm. Res., 20: 1794-1803.

Yu, X.Y., S.G. Lin, Z.W. Zhou, X. Chen and J. Liang *et al*., 2007. Role of P-glycoprotein in the intestinal absorption of tanshinone IIA a major active ingredient in the root of *Salvia miltiorrhiza* Bunge. Curr. Drug Metab., 8: 325-340.

Zhou, X., K. Chan and J.H.K. Yeung, 2012. Herb-drug interactions with Danshen (*Salvia miltiorrhiza*): A review on the role of cytochrome P450 enzymes. Drug Metabol. Drug Interact., 27: 9-18.