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Effect of food components and dosing times on the oral pharmacokinetics of nifedipine in rats

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ABSTRACT

The present study was aimed to investigate the effect of food components and dosing time on the oral exposure of nifedipine in rats. Nifedipine was given orally to rats with and without food components at 8:00 a.m. (morning time) or 4:00 p.m. (evening time) during winter periods. Food components included milk, sodium chloride, oleic acid, and sodium taurocholate. Plasma concentration profiles of nifedipine showed double peak phenomena which were generally retained regardless of food components, vehicle types and the dosing time. Sodium chloride, milk and sodium taurocholate significantly increased the AUC while oleic acid did not, when drug was dosed in the morning time. After the dosing in the evening time, milk and sodium chloride significantly increased the plasma concentrations of nifedipine but oleic acid and sodium taurocholate decreased them. Overall, the systemic in vivo exposure of nifedipine was invariably lower with the evening dosing compared to the dosing in the morning, but this circadian rhythm dependency was not reversed by the multiple dosing of food components in rats. Food components and dosing time significantly altered the oral pharmacokinetics of nifedipine in rats, implying that the altered bioavailability and higher plasma concentrations in the morning time may influence dosing regimens of nifedipine for hypertension patients.

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1. Introduction

Nifedipine is a calcium channel-blocking agent that is widely used for the treatment of essential hypertension, coronary artery spasm, and angina pectoris (Sorkin et al., 1985). Although nifedipine has limited water-solubility (<10 mg/L), it is rapidly and completely absorbed from the gastrointestinal tract due to its lipophilicity (Abrahamsson et al., 1998), providing 50–70% oral bioavailability in man.

The bioavailability of nifedipine in immediate and sustained release forms is affected by the concurrent administration of food complexes such as high fat or high protein meals (Abrahamsson et al., 1998; Schug et al., 2002). Food intake can change the physicochemical properties of drugs, interact with formulation excipients, and change physiological and biopharmaceutical factors (Charman et al., 1997; Wagner et al., 2001; Deferme and Augustijns, 2003). Specific food components, including dietary fatty acids, milk, bile salts, sodium chloride, surfactants, grapefruit juice, and flavonoids, can change the solubility, intestinal permeability, P-glycoprotein activity, presystemic metabolism and bioavailability of drugs (Yamaguchi et al., 1987; Mithani et al., 1996; Charman et al., 1997; Grundy et al., 1998; Wagner et al., 2001; Vine et al., 2002; Deferme and Augustijns, 2003; Tran et al., 2009). For example, the intake of concentrated grapefruit juice increased the bioavailability of nifedipine by delaying gastric emptying and inhibiting metabolism (Grundy et al., 1998).

Circadian rhythms can also affect the pharmacokinetics and pharmacodynamics of certain drugs (Lemmer et al., 1991; Labrecque et al., 1997), which could become more complicated by the co-administration of food as some food additives can affect clock gene expression and daily rhythm (Horikawa et al., 2005; Angeles-Castellanos et al., 2007). The pharmacokinetics of nifedipine can be also influenced by circadian rhythm and the therapeutic window should be designed carefully to minimize the side effects associated with abnormal blood concentrations (Labrecque et al., 1997; Cao et al., 2005). It has been known that the environmental light/dark cycle and daily feeding are the most potent synchronizer of the circadian pacemaker in the suprachiasmatic nuclei. Peri-

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odic food feedings in birds and animals has been investigated in detail as a circadian zeitgeber (Horikawa et al., 2005; Mistlberger et al., 2003). The influence of meal time on salivary circadian cortisol rhythms and weight loss in obese women was also studied (Nonino-Borges et al., 2007). It is still not known how the circadian variation of nifedipine pharmacokinetics is related to the multiple feeding of food components.

The aim of the present study was to investigate the effect of food components and dosing time on the oral pharmacokinetics of nifedipine. In addition, potential changes in dosing time-dependent pharmacokinetics of nifedipine by the multiple dosing with food components were also examined. The food components such as oleic acid, sodium taurocholate, milk, and sodium chloride were tested in the present study since they have been widely used in the drug formulation. These components were fed for a week. Then, we investigated the oral pharmacokinetics of nifedipine in rats following the concurrent administration of nifedipine and food components in the morning time or in the evening time.

2. Materials and methods

2.1. Materials

Short-acting nifedipine powder was obtained from Jeil Pharmaceutical Co. (Seoul, Korea). Oleic acid and polyethylene glycol 400 (PEG 400) were purchased from Showa Chemical Co. (Tokyo, Japan). Sodium taurocholate, testosterone and sodium chloride were purchased from Sigma Chemical Co. (St. Louis, MO, USA). Pentane and dichloromethane were obtained from Junsei Chemical Co. (Tokyo, Japan). Acetonitrile (HPLC grade) was purchased from Fisher Scientific (Pittsburgh, PA, USA). All other chemicals were of reagent grade and used without further purification.

2.2. Preparation of drug solution

Due to the low solubility in water, nifedipine was dissolved in a co-solvent of PEG400 and water (1:1, v/v) at 0.67 mg/ml. The food components were prepared and mixed with drug solution to final concentrations of whole milk (50%, v/v), sodium chloride (0.5%, w/v), oleic acid (2%, v/v) and sodium taurocholate (20 mM), based on literature values (Yamaguchi et al., 1987; Mithani et al., 1996; Charman et al., 1997). Aqueous drug suspension or drug in co-solvent without adding food component was used as a control. For comparison purposes, aqueous drug suspension (0.67 mg/ml) was also prepared and tested. The drug solution was then designated as follows: AS: aqueous suspension, Co: co-solvent of PEG400 and water (1:1 v/v), Co-Milk: milk in co-solvent, Co-NaCl: sodium chloride in co-solvent, Co-OA: oleic acid in co-solvent, Co-ST: sodium taurocholate in co-solvent.

2.3. Animal treatment

Male Sprague–Dawley rats weighing 280–350 g and aged 7–10 weeks were purchased from Dae Han Experimental Animal (Seoul, Korea). The rats were synchronized with a light–dark cycle (light period: 8:00 a.m. to 8:00 p.m.) to adjust circadian rhythm. Rats, four in a cage, were housed in a temperature-controlled room ($25 \pm 2\,^{\circ}C$) for more than 3 weeks prior to the study. Rats were fasted overnight prior to the experiment, but allowed free access to tap water.

2.4. Dosing scheme

A total of 72 rats were randomly divided into 12 different groups (six rats per group) to investigate the effect of food components and dosing time on the systemic exposure of nifedipine. One of four different food components in PEG 400-water co-solvent was orally given every morning or early evening (t.i.d.) for a week. Thereafter, drug was added to the co-solvent and then dosed to the rats for the evaluation of in vivo bioavailability. The drug co-solvent solution without adding any food components was dosed to the control group. The aqueous drug suspension was also given to rats without any pretreatment with food components for the comparison of the vehicle effect. All animal studies were conducted according to Guiding Principles in the Use of Animals in Toxicology adopted by the Society of Toxicology (http://www.toxicology.org/AI/FA/guidingprinciples.pdf).

2.5. Blood sampling

Under anesthesia by inhalation of ether, a polyethylene cannula (inner diameter, 0.58 mm; outer diameter 0.96 mm; dual plastics) was surgically introduced into the left femoral artery to obtain blood samples at various sampling times. After 2 h, 0.67 mg/kg nifedipine–food solution was orally administrated using an oral sonde. Approximately 0.4 ml of blood samples were collected from the indwelling cannula in heparinized tube at 5, 10, 20, 30, 45, 60, 90, 120, 240, and 360 min and then centrifuged at 3000 rpm for 10 min. Plasma samples were stored in a freezer at $-40\,^{\circ}C$ until HPLC analysis.

2.6. Treatment of plasma samples

A standard calibration curve for nifedipine (100–2000 ng/ml) in acetonitrile was constructed. A mixture of nifedipine standard and internal standard solution (2000 ng/ml testosterone in acetonitrile) was evaporated to dryness. The frozen plasma samples were melted at room temperature. Thereafter, 200 μl of rat plasma was added to each tube and vortexed for 5 s. Both 100 μl of 1 N sodium hydroxide solution and 1 ml of pentane-methylene chloride (7:3, v/v) were added and shaken for 5 min using a vortex mixer, and then centrifuged at 2500 rpm for 10 min. The organic phase was transferred into a tube by pipette and evaporated under reduced vacuum conditions to dryness at $45\,^{\circ}C$ for 30 min. The residue was reconstituted with 100 μl of the mobile phase and the resulting solution (20 μl) was injected into the HPLC system for analysis.

2.7. Pharmacokinetic and statistical analysis

Pharmacokinetic parameters were calculated using non-compartmental methods. The maximum peak plasma concentration (C_{max}) and the time to reach the maximum peak plasma concentration (T_{max}) were directly read from the plasma concentration-time profiles of nifedipine. The area under the plasma concentration-time curve ($AUC_{0-6\,h}$) was calculated by the classical trapezoidal rule method.

All data are presented as mean ± standard deviation. Statistical significance was assessed by two-way factorial analysis of variance test followed by Duncan's multiple comparison between groups (Steel and Torrie, 1980). A probability level of $p < 0.05$ was considered to be statistically significant.

3. Results

The mean plasma concentration–time profiles of nifedipine following a single oral administration of nifedipine with different food components in the morning and the early evening are shown in Fig. 1. The pharmacokinetic parameters were also summarized in Table 1.

As shown in Fig. 1, the use of PEG400 as a co-solvent significantly increased the oral exposure of nifedipine compared to the aqueous dosing suspension. Furthermore, food components and dosing

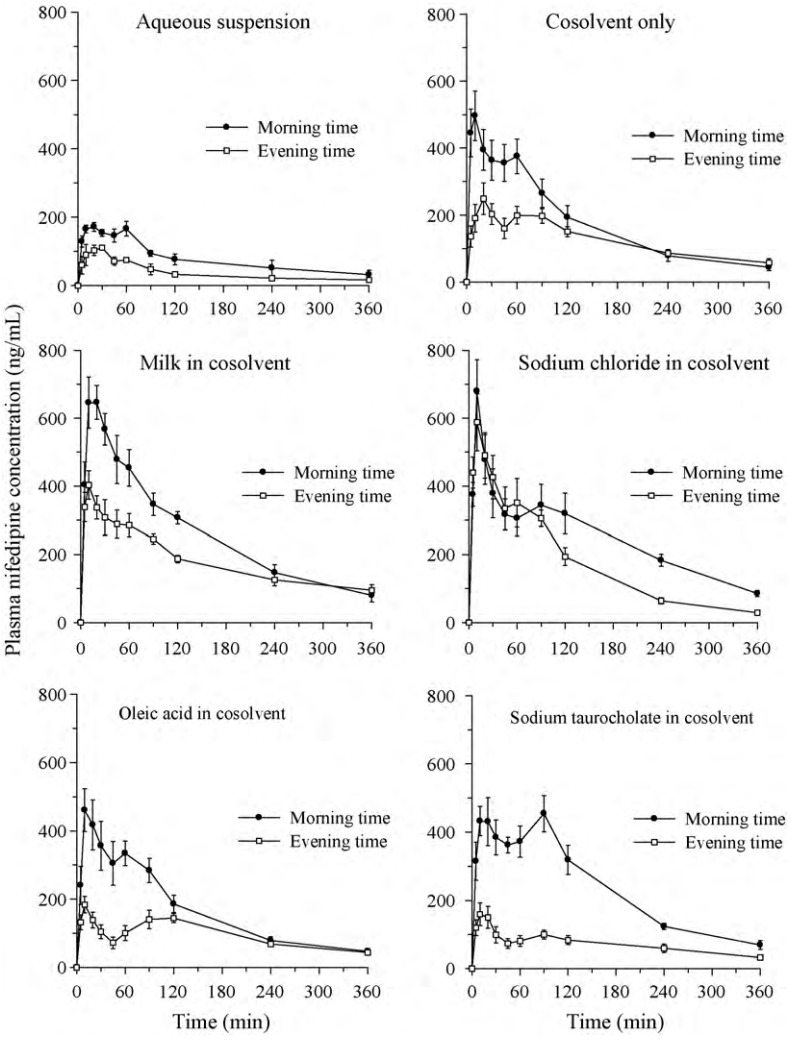

Fig. 1. Mean plasma concentration-time profiles of nifedipine following a single oral administration of drug solutions containing different food components to six rats in the morning and evening time (mean ± SD, $n = 6$).

time exhibited significant effect on the oral pharmacokinetics of nifedipine. Plasma concentration profiles of nifedipine showed a double peak phenomena (C_{max1} and C_{max2}), which were generally retained regardless of food components, vehicle types and the dosing time. As illustrated in Fig. 1, the oral exposures of nifedipine were invariably higher in the morning when dosed with food components. Nifedipine levels given with milk and sodium chloride in the morning were much higher than oleic acid and sodium taurocholate.

As shown in Table 1, food components such as sodium chloride, milk and sodium taurocholate significantly increased the AUC while oleic acid did not when drug was dosed in the morning. Although the intake of sodium chloride and milk increased C_{max1} significantly compared to the control group given drug only without food components, the oleic acid and sodium taurocholate tended to reduce C_{max1}. Food intake did not affect C_{max2} but both T_{max1} and T_{max2} appeared to be longer. In the case of evening dosing, milk and sodium chloride significantly increased the C_{max1}, C_{max2},

Table 1
Pharmacokinetic parameters of nifedipine following a single oral administration of drug solutions containing different food components to rats in the morning or evening (mean ± SD, $n = 6$).

	AS	Co	Co-Milk	Co-NaCl	Co-OA	Co-ST
Morning time						
C_{max1} (ng/ml)	171.6 ± 44.2	521.8 ± 152.0	670.7 ± 115.5	685.2 ± 187.6	480.6 ± 107.3	475.7 ± 94.6
C_{max2} (ng/ml)	160.3 ± 34.9	388.8 ± 110.5	386.0 ± 55.9	356.8 ± 121.7	338.8 ± 70.0	465.5 ± 106.0
T_{max1} (h)	0.31 ± 0.12	0.13 ± 0.05	0.28 ± 0.09	0.19 ± 0.07*	0.19 ± 0.07	0.25 ± 0.09
T_{max2} (h)	1.05 ± 0.24	0.97 ± 0.25	1.5 ± 0.45	1.75 ± 0.27	1.17 ± 0.26	1.42 ± 0.20
AUC $_{0\text{-}6h}$ (ng h/ml)	469.6 ± 112.1	1047.7 ± 310.1	1561.5 ± 153.1	1491.5 ± 324.4	1001.2 ± 117.4	1404.1 ± 250.3
Evening time						
C_{max1} (ng/ml)	110.9 ± 43.5	268.8 ± 85.5	413.9 ± 68.5	614.3 ± 153.4	188.4 ± 46.7	180.2 ± 73.2
C_{max2} (ng/ml)	74.9 ± 13.2	229.8 ± 38.8	331.1 ± 108.9	399.2 ± 148.8	167.1 ± 36.2	99.8 ± 27.3
T_{max1} (h)	0.55 ± 0.10	0.42 ± 0.09	0.15 ± 0.04	0.21 ± 0.08	0.15 ± 0.03	0.22 ± 0.09
T_{max2} (h)	1.09 ± 0.20	1.25 ± 0.27	0.97 ± 0.28	1.11 ± 0.28	1.75 ± 0.27	1.58 ± 0.20
$AUC_{0\text{-}6h}$ (ng h/ml)	227.1 ± 28.4	752.2 ± 112.1	1080.2 ± 119.0	1047.3 ± 156.4	565.9 ± 86.3	429.8 ± 86.5

Table 2
Statistical significance of pharmacokinetic parameters between food components dosed in the morning time (M) or evening time (E) at a 5% significance level ($p < 0.05$).

Comparison of food components	C_{max1}		C_{max2}		T_{max1}		T_{max2}		AUC	
	M	E	M	E	M	E	M	E	M	E
AS vs Co	Yes	Yes	Yes	Yes	Yes	Yes	No	No	Yes	Yes
AS vs Co-NaCl	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes
AS vs Co-Milk	Yes	Yes	Yes	Yes	No	Yes	Yes	No	Yes	Yes
AS vs Co-OA	Yes	No	Yes	No	Yes	Yes	No	Yes	Yes	Yes
AS vs Co-ST	Yes	No	Yes	No	No	Yes	Yes	Yes	Yes	No
Co vs Co-Milk	Yes	Yes	No	Yes	Yes	Yes	Yes	No	Yes	Yes
Co vs Co-NaCl	Yes	Yes	No	Yes	No	Yes	Yes	No	Yes	Yes
Co vs Co-OA	No	No	No	No	No	Yes	No	Yes	No	No
Co vs Co-ST	No	No	No	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Co-Milk vs Co-NaCl	No	Yes	No	No	No	No	No	No	No	No
Co-Milk vs Co-OA	Yes	Yes	No	Yes	No	No	Yes	Yes	Yes	Yes
Co-Milk vs Co-ST	Yes	Yes	No	Yes	No	No	No	Yes	No	Yes
Co-NaCl vs Co-OA	Yes	Yes	No	Yes	No	No	Yes	Yes	Yes	Yes
Co-NaCl vs Co-ST	Yes	Yes	No	Yes	No	No	Yes	Yes	No	Yes
Co-OA vs Co-ST	No	No	Yes	No	No	No	No	No	Yes	No

Two-way analysis of variance was performed and subsequently all pairwise multiple comparison procedure was carried out by Duncan's method.

and AUC, but oleic acid and sodium taurocholate decreased them. T_{max1} became shorter while T_{max2} was variable and delayed significantly ($p < 0.05$) by oleic acid and sodium taurocholate. The detailed statistical analysis of the pharmacokinetic data between food components at each dosing time is compared in Table 2.

The oral exposure of nifedipine was also affected by the dosing time in addition to the food types. The AUC achieved from evening dosing were lower than those from morning dosing, regardless of food types (Fig. 2). C_{max1} was also higher with morning dosing except for sodium chloride which did not show dosing time dependency. C_{max2} showed similar behavior (Fig. 3). The T_{max} was variable depending on the dosing time and the type of food components. In the absence of food components, T_{max} was delayed with evening dosing but it was shorter by concurrent use of milk (Fig. 4). The detailed statistical analysis of the pharmacokinetic data between morning time and evening time according to the food components is warranted in Table 3.

Taken all together, the oral exposure of nifedipine was significantly affected by the food components in a dosing-time-dependent manner. Interestingly, multiple oral dosing of food components for 1 week did not change these time-dependent pharmacokinetic behaviors. Thus, circadian rhythm dependency was not reversed by the multiple dosing of food components in rats.

4. Discussion

Co-administration with food and pharmaceutical excipients that inhibit intestinal P-glycoprotein and presystemic metabolism can enhance absorption and bioavailability (Wagner et al., 2001; Deferme and Augustijns, 2003). The presence of food components within the gastrointestinal tract also impacts transit time, pH, and

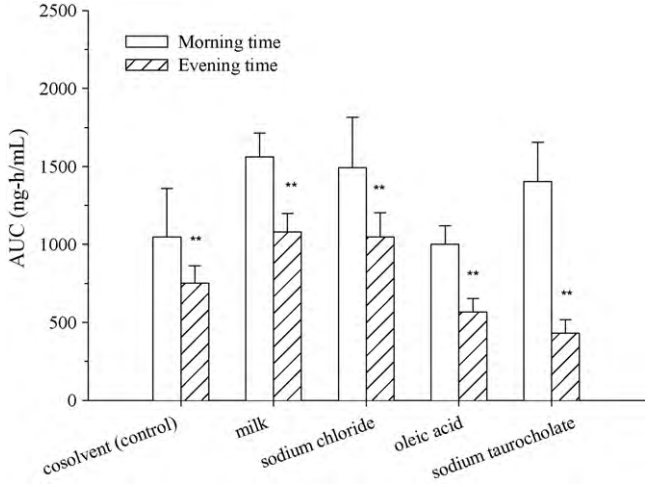

Fig. 2. The effect of food components on AUC of nifedipine in rats dosed at two different dosing times (mean ± SD, $n = 6$). $^{**}p < 0.05$.

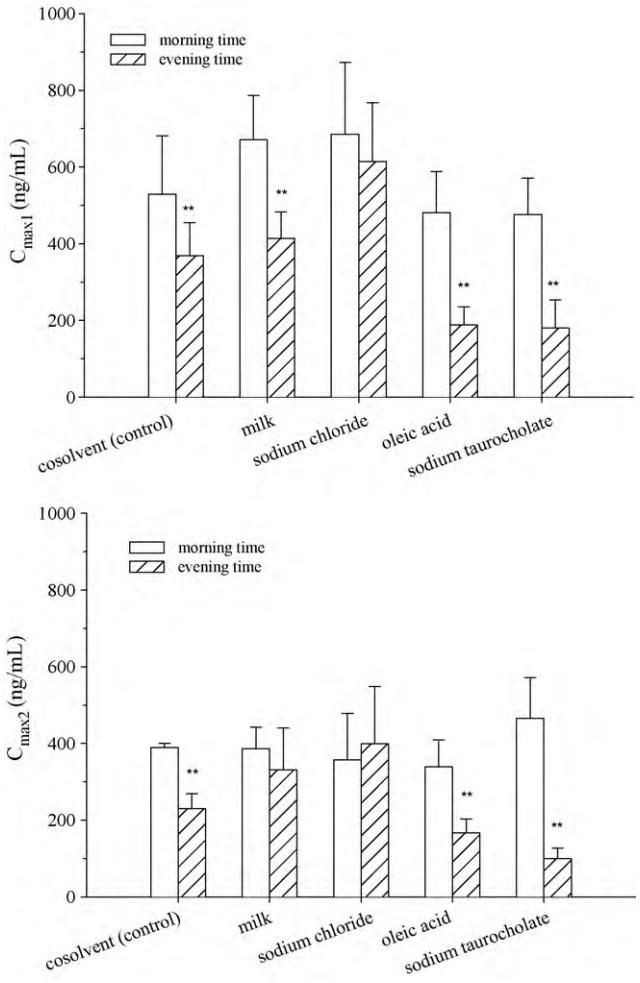

Fig. 3. The effect of food components on C_{max} of nifedipine in rats dosed at two different dosing times (mean ± SD, $n = 6$). $^{**}p < 0.05$.

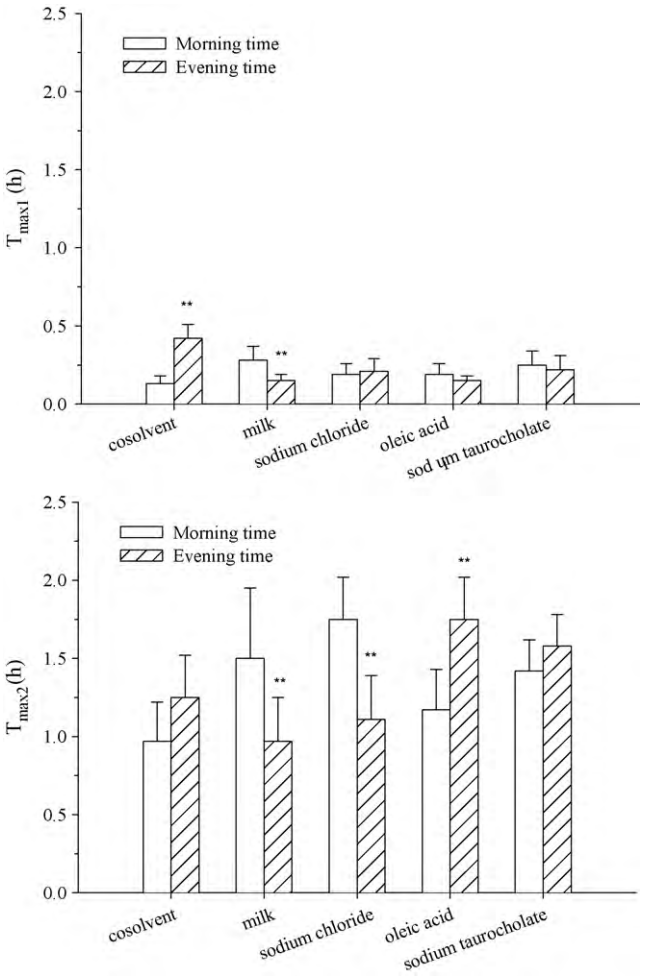

Fig. 4. The effect of food components on T_{max} of nifedipine in rats dosed at two different dosing times (mean ± SD, $n = 6$). $^{**}p < 0.05$.

solubilization capacity. The secretion of gastric acid, bile, and pancreatic fluid, as well as alterations of blood and lymphatic systems also significantly impact drug absorption (Charman et al., 1997). Intestinal membrane transporters also contribute to the absorption and distribution of drugs and nutrients. Thus, the mechanisms for how these food components change pharmacokinetics of drugs are relatively non-specific but should be investigated in detail.

The present study investigated how food (fatty acid, bile salt, milk, and sodium chloride) alters the pharmacokinetics of nifedipine. The PEG-water formulation showed higher plasma levels than water suspension because of enhanced drug solubility and a reduction in the gastrointestinal transit time (Basit et al., 2001). Fatty acids can change active and passive transport in the excised rat jejunum (Vine et al., 2002). In particular, long-chain fatty acids like oleic acid can enhance bioavailability by forming chylomicrons, which relates to the lymphatic transport of various poorly water-soluble drugs in the small intestine (Charman et al., 1997). These long-chain fatty acids in the formulation are re-esterified to triglycerides within the intestinal cell, incorporated into chylomicrons with lipoprotein, and then secreted into the lymph vessels.

Micellar complexes with endogenous or exogenous bile salts with drugs can change the bioavailability of drugs by increasing cellular permeability and solubilization within bile salt micelles (Mithani et al., 1996; Charman et al., 1997). The micellar complex of fatty acid with bile salts can decrease the intestinal absorption of nadolol (Yamaguchi et al., 1987), but increase the intestinal absorption of amphotericin B and halofantrine (Humberstone et al., 2000). Conversely, stabilized micellar complexes with bile salts may decrease absorption by decreasing the free fraction of drug and losing thermodynamic activity (Yamaguchi et al., 1987; Charman et al., 1997). We also found that the micellar complex of melatonin with bile salt decreased in vitro absorption through excised rat gut (Tran et al., 2009). However, the role of lymphatic delivery and micellar complexes seems insignificant here because oleic acid alone decreased nifedipine bioavailability (Table 1). Sodium taurocholate at the micellar concentration (20 mM) also decreased nifedipine C_{max} and AUC (Table 1), potentially by reducing the free fraction of the drug (Charman et al., 1997). The AUC of nifedipine was higher in the morning than the evening because of a larger secondary peak that resulted from delayed gastric transit time.

Milk does not affect the bioavailability of cyclosporine but increased the AUC of propranolol (Johnston et al., 1986; Ogiso et al., 1994). Here, whole milk increased C_{max1} by 28.4% and AUC by 49%, but not C_{max2}, and had a larger effect in the morning. Milk increased the C_{max1}, C_{max2}, and AUC by 53%, 44%, and 44%, respectively. Hypotonic sodium chloride also increased nifedipine bioavailability regardless of dosing times. Isotonic solution produces the best bioavailability for sulfafurazol (Marvola et al., 1980), and sodium chloride dose-dependently decreases the bioavailability of anti-arrhythmic drugs (Lee et al., 1997). In addition to osmotic pressure, the Na^+ concentration gradient may change the permeability of nifedipine by the Na^+/K^+ dependent ATPase in the intestinal membrane, as shown for glucose (Vine et al., 2002). Future work should examine how milk and sodium chloride increase nifedipine bioavailability by these mechanisms.

The double peak phenomena of the plasma concentration profiles were retained, regardless of food, vehicle, or dosing time. Sodium taurocholate increased the second peak height. Grapefruit juice or orange juice concentrate produced double peak profiles for nifedipine by delaying gastric emptying (Grundy et al., 1998), but the effects of other food components are unclear. Double peaks also occur after oral administration of other drugs. Enterohepatic recirculation or biliary excretion does not produce this double peak phenomenon because double peaks do not occur following intravenous administration of cimetidine (Takamatsu et al., 2002). Gastric emptying and gastrointestinal motility, as well as pH changes, are the major determinants of the secondary peak in both fasted and fed states in rats, dogs and humans (Takamatsu et al., 2002; Higaki et al., 2008; Tümer et al., 2008; Ozaki et al., 2009). PEG 400 enhances drug solubility and reduces gastrointestinal transit time, which could contribute to the double peak (Basit et al., 2001). This phenomenon is further complicated by the presence of food and discontinuous absorption sites in the gut.

It is also known that the pharmacokinetics of nifedipine display significant daily variations after oral dosing of both immediate release and controlled release preparations, with peak concentration and bioavailability higher with dosing in the morning (Lemmer 1991; Cao et al., 2005). Rats also show dose-timing variability in the nifedipine pharmacokinetics despite different rest-activity cycles from humans (Cao et al., 2005). In this work, the bioavail-

Table 3
Statistical significance of pharmacokinetic parameters between morning time and evening time according to the food components at a 5% significance level ($p < 0.05$).

Food components	C_{max1}	C_{max2}	T_{max1}	T_{max2}	AUC
AS	No	No	Yes	No	Yes
Co	Yes	Yes	Yes	No	Yes
Co-Milk	Yes	No	Yes	Yes	Yes
Co-NaCl	No	No	No	Yes	Yes
Co-OA	Yes	Yes	No	Yes	Yes
Co-ST	Yes	Yes	No	No	Yes

Two-way analysis of variance was performed and subsequently all pairwise multiple comparison procedure was carried out by Duncan's method.

ability of nifedipine was much higher when dosed in the morning time. Nifedipine reached higher plasma levels in the morning than the evening. Both food effect and dosing time clearly changed the dosing-time-dependent pharmacokinetics of nifedipine.

Although the food effect on drug pharmacokinetics has been characterized, it is not still known whether this circadian variation is changed by the multiple dosing of food components. For example, periodic food feedings or intermittent fasting in animals has been also known as a circadian zeitgeber (Horikawa et al., 2005; Mistlberger et al., 2003). The meal time led to changes in weight, body composition, resting metabolic rate, and nitrogen balance but did not significantly alter salivary circadian cortisol rhythms (Nonino-Borges et al., 2007). Recently, intermittent fasting can affect circadian rhythms differently depending on the timing of food availability in rats (Froy et al., 2009). Thus, nighttime feeding yielded rhythms similar to those generated during ad libitum feeding unlike daytime feeding. However, time-dependent pharmacokinetic parameters of nifedipine in rats was not reversed by the multiple oral dosing of food components even though four food components and dosing time could modify nifedipine pharmacokinetics.

5. Conclusions

Plasma nifedipine levels were higher in the morning when dosed with food components, except sodium taurocholate, in rats. Food components also altered nifedipine bioavailability, which was much higher in the morning time as compared to the evening time. Interestingly, this circadian rhythm dependency was not reversed by the multiple dosing of food components in rats. Thus, meal composition and dosing times should be simultaneously considered in clinical trial when nifedipine is used in hypertensive patients.

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References

Abrahamsson, B., Alpsten, M., Bake, B., Jonsson, U.E., Eriksson-Lepkowska, M., Larsson, A., 1998. Drug absorption from nifedipine hydrophilic matrix extended release (ER) tablet-comparison with an osmotic pump tablet and effect of food. J. Controlled Release 52, 301–310.

Angeles-Castellanos, M., Mendoza, J., Escobar, C., 2007. Restricted feeding schedules phase shift daily rhythms of c-Fos and protein Per1 immunoreactivity in corticolimbic regions in rats. Neuroscience 144, 344–355.

Basit, A.W., Newton, J.M., Short, M.D., Waddington, W.A., Ell, P.J., Lacey, L.F., 2001. The effect of polyethylene glycol 400 on gastrointestinal transit: implications for the formulation of poorly-water soluble drugs. Pharm. Res. 18, 1146–1150.

Cao, Q.R., Kim, T.W., Choi, J.S., Lee, B.-J., 2005. Circadian variations in the pharmacokinetics, tissue distribution and urinary excretion of nifedipine after a single oral administration to rats. Biopharm. Drug Dispos. 26, 427–437.

Charman, W.N., Porter, C.J.H., Mithani, S., Dressman, J.B., 1997. Physicochemical and physiological mechanisms for the effects of food on drug absorption: the role of lipids and pH. J. Pharm. Sci. 86, 182–269.

Deferme, S., Augustijns, P., 2003. The effect of food components on the absorption of P-gp substrates: a review. J. Pharm. Pharmacol. 55, 153–162.

Grundy, J.S., Eliot, L.A., Kulmatycki, K.M., Foster, R.T., 1998. Grapefruit juice and orange juice effects on the bioavailability of nifedipine in the rat. Biopharm. Drug Dispos. 19, 175–183.

Froy, O., Chapnik, N., Miskin, R., 2009. Effect of intermittent fasting on circadian rhythms in mice depends on feeding time. Mech. Ageing Dev. 130, 154–160.

Higaki, K., Choe, S.Y., Löbenberg, R., Welage, L.S., Gordon, L., Amidon, G.L., 2008. Mechanistic understanding of time-dependent oral absorption based on gastric motor activity in humans. Eur. J. Pharm. Biopharm. 70, 313–325.

Horikawa, K., Minami, Y., Iijima, M., Akiyama, M., Shibata, S., 2005. Rapid damping of food-entrained circadian rhythm of clock gene expression in clock-defective peripheral tissues under fasting conditions. Neuroscience 134, 335–343.

Humberstone, A.J., Porter, C.J.H., Charman, W.N., 2000. A physicochemical basis for the effect of food on the absolute oral bioavailability of halofantrine. J. Pharm. Sci. 85, 525–529.

Johnston, A., Marsden, J.T., Hla, K.K., Henry, J.A., Holt, D.W., 1986. The effect of vehicle on the oral absorption of cyclosporin. Br. J. Clin. Pharmacol. 21, 331–333.

Labrecque, G., Beauchamp, D., Vanier, M., Smolensky, M.H., 1997. Chronopharmacokinetics. Pharm. News 4, 17–21.

Lee, K.H., Xu, G.X., Schoenhard, G.L., Cook, C.S., 1997. Mechanism of food effects of structurally related antiarrhythmic drugs, disopyramide and bidisomide in the rat. Pharm. Res. 14, 1030–1038.

Lemmer, B., Scheidel, B., Behne, S., 1991. Chronopharmacokinetics and chronopharmacodynamics of cardiovascular active drugs: propranolol, organic nitrates, nifedipine. Ann. N.Y. Acad. Sci. 618, 166–181.

Marvola, M., Lehmussaari, L., Niinimaki, L., 1980. The effect of osmotic pressure on the intestinal absorption of sulfafurazol and ethanol in the rat. Arzneimitteforschung 30, 1631–1634.

Mistlberger, R.E., Antle, M.C., Kilduff, T.S., Jones, M., 2003. Food- and light-entrained circadian rhythms in rats with hypocretin-2-saporin ablations of the lateral hypothalamus. Brain Res. 980, 161–168.

Mithani, S.D., Bakatselou, V., TenHoor, C.N., Dressman, J.B., 1996. Estimation of the increase in solubility of drugs as a function of bile salt concentration. Pharm. Res. 13, 163–167.

Nonino-Borges, C.B., Borges, R.M., Bavaresco, M., Suen, V.M.M., Moreira, A.C., Marchini, J.S., 2007. Influence of meal time on salivary circadian cortisol rhythms and weight loss in obese women. Nutrition 23, 385–391.

Ogiso, T., Iwaki, M., Tanino, T., Kawafuchi, R., Hata, S., 1994. Effect of food on propranolol oral clearance and a possible mechanism of this food effect. Biol. Pharm. Bull. 17, 112–116.

Ozaki, K., Onoma, M., Muramatsu, H., Sudo, H., Yoshida, S., Shiokawa, R., Yogo, K., Kamei, K., Cynshi, O., Kuromaru, O., Peeters, T.L., Takanashi, H., 2009. An orally active motilin receptor antagonist, MA-2029, inhibits motilin-induced gastrointestinal motility, increase in fundic tone, and diarrhea in conscious dogs without affecting gastric emptying. Eur. J. Pharmacol. 615, 185–192.

Schug, B.S., Brendel, E., Chantraine, E., Wolf, D., Martin, W., Schall, R., Blume, H.H., 2002. The effect of food on the pharmacokinetics of nifedipine in two slow release formulations: pronounced lag-time after a high fat breakfast. Br. J. Clin. Pharmacol. 53, 582–588.

Sorkin, E.M., Clissold, S.P., Brogden, R.N., 1985. Nifedipine: a review of its pharmacodynamic and pharmacokinetic properties and therapeutic efficacy, in ischaemic heart disease, hypertension and related cardiovascular disorders. Drugs 30, 182–274.

Steel, R.G., Torrie, J.H., 1980. Principles and Procedures of Statistics, A Biometrical Approach, 2nd ed. McGraw-Hill Co., NY.

Takamatsu, N., Welage, L.S., Hayashi, Y., Yamamoto, R., Barnett, J.L., Shah, V.P., Lesko, L.J., Ramachandran, C., Amidon, G.L., 2002. Variability in cimetidine absorption and plasma double peaks following oral administration in the fasted state in humans: correlation with antral gastric motility. Eur. J. Pharm. Biopham. 53, 37–47.

Tran, H.T.T., Tran, P.H.L., Lee, B.J., 2009. New findings on melatonin absorption and alterations by pharmaceutical excipients using the Ussing chamber technique with mounted rat gastrointestinal segments. Int. J. Pharm. 378, 9–16.

Tümer, C., Oflazoğlu, H.D., Obay, B.D., Kelle, M., Taşdemir, E., 2008. Effect of ghrelin on gastric myoelectric activity and gastric emptying in rats. Regul. Pept. 146, 26–32.

Vine, D.F., Charman, S.A., Gibson, P.R., Sinclair, A.J., Porter, C.J.H., 2002. Effect of dietary fatty acids on the intestinal permeability of marker drug compounds in excised rat jejunum. J. Pharm. Pharmacol. 54, 809–819.

Wagner, D., Spahn-Langguth, H., Hanafy, A., Koggel, A., Langguth, P., 2001. Intestinal drug efflux: formulation and food effects. Adv. Drug Deliv. Rev. 50, 13–31.

Yamaguchi, T., Ikeda, C., Sekine, Y., 1987. Intestinal absorption of a β-adrenergic blocking agent nadolol. Enhancement of in situ and in vivo absorption of nadolol in rats. Int. J. Pharm. 37, 127–134.