Reduced Food-Effect on Intestinal Absorption of Dronedarone by Self-microemulsifying Drug Delivery System (SMEDDS)

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The oral absorption of dronedarone (DRN), a benzofuran derivative with anti-arrhythmic activity, is significantly affected by food intake. The absolute bioavailability of the marketed product (Multaq®, Sanofi, U.S.) was about 4% without food, but increased to 15% when administered with a high fat meal. Therefore, to reduce the food-effect on the intestinal absorption of DRN, a novel self-microemulsifying drug delivery system (SMEDDS) was formulated and the comparative in vivo absorption studies with the marketed product were carried out using male beagle dogs either in the fasted or fed state. The SMEDDS consisted of the drug, Labrafail M 1944CS, and Kolliphor EL in a weight ratio of 1:1:2, rapidly formed a fine oil-in-water emulsion with a droplet size less than 50 nm. An in vivo absorption study revealed that the area-under-curve (AUC0–24h) and maximal plasma concentration (Cmax) were 10.4-fold (p<0.05) and 8.6-fold (p<0.05) higher, respectively, after the marketed product was orally administered to beagles in the fed state when compared to those in the fasted state. This food-effect was remarkably alleviated by SMEDDS formulation, with AUC0–24h and Cmax 2.9-fold (p<0.05) and 2.6-fold (p<0.05) higher in the fed state when compared to the fasted state, by facilitating intestinal absorption of DRN in the fasted state. The results of this study suggest that SMEDDS may decrease the differences in oral absorption of DRN between the prandial states, improving therapeutic efficacy as well as patient compliance.

Key words dronedarone; self-microemulsifying drug delivery system (SMEDDS); food-effect; oral absorption; solubilization

Dronedarone (N-[2-butyl-3-[4-(3-dibutylaminopropoxy)benzoyl]benzofuran-5-yl]methane sulfonamide; DRN) is a benzofuran derivative with anti-arrhythmic properties belonging to all four Vaughan–Williams classes.⁴,⁵ This drug, commercialized under the trade name Multaq® (Sanofi, U.S.), has been approved by the Food and Drug Administration (FDA) since 2009 in the tablet dosage form containing the hydrochloride salt of DRN (400 mg as base).⁶ In a clinical study, the anti-arrhythmic agent effectively reduced cardiovascular hospitalization or death from any cause by 24.2% compared to the placebo group.⁷ However, the oral administration of DRN has some problems such as extensive first-pass metabolism and food-effect on oral absorption.⁸ Although the original manufacturer (Sanofi, U.S.) formulated an oral dosage form (Multaq®) based on the solid dispersion (SD) system with a triblock copolymer of polypropylene glycol and polyethylene glycol to increase drug dissolution in the gastrointestinal (GI) tract,⁹ the oral absorption of this biopharmaceutics classification system (BCS) II compound is significantly affected by food intake. The absolute bioavailability (BA) of DRN is approximately 4% without food, which increased to approximately 15% when administered with a high fat meal.¹⁰ In this regard, Multaq® tablets should be taken shortly after a meal to increase its intestinal absorption.

Self-microemulsifying drug delivery systems (SMEDDS) are isotropic mixtures of oils and (co)surfactants, or alternatively, one or more hydrophilic solvents, which can be spontaneously dispersed to produce a fine oil-in-water (o/w) emulsion upon mild agitation in the gastrointestinal tract.¹¹ The lipophilic compounds can be maintained in a solubilized state by being incorporated in an internal phase of the o/w emulsion which provides a large interfacial surface area for drug absorption.¹² The advantages of a SMEDDS formulation include ease of production, outstanding solubilizing capacity, and increased intestinal permeation by fluidizing intestinal membranes, opening tight junctions, and inhibiting efflux pumps.¹³,¹⁴ In addition, SMEDDS was shown to significantly reduce the food-effect on intestinal absorption of poorly water-soluble molecules such as itraconazole and torcetrapib in human and/or animal models.¹⁵ While itraconazole capsules that are sold commercially exhibit a significant food-effect, the SMEDDS formula reduced the food-effects influencing on pharmacokinetic profiles of the drug.¹⁶ Perlman et al. also reported that SMEDDS consisting of medium chain triglyceride/Triacetin/Polysorbate 80/Capmul MCM increased the intestinal absorption of torcetrapib in the fasted state, reduced the food-effect from 5- to 3-fold in beagle dogs.¹⁷

Based on these findings, we hypothesize that SMEDDS is effective in alleviating the food-effect on the intestinal absorption of DRN by facilitating intestinal absorption in the fasted state. In formulating SMEDDS, we used DRN free base, instead of the hydrochloride form, because the DRN free base exhibited better solubility in the oil/surfactant blends compared to the salt form. The lipophilic formulation was prepared by dissolving DRN in the preconcentrate, which consists of Labrafail M 1944CS (Oleoyl macrogol-6 glycerides) and Kolliphor EL (polyethoxylated castor oil). Dissolution profiles of the DRN-loaded SMEDDS were investigated under various conditions and compared with those of the marketed product (Multaq®). The pharmacokinetic profiles of DRN were
assessed after oral administration of DRN-loaded SMEDDS or the marketed product in beagle dogs in the fasted state and after a meal using a validated LC-MS/MS analysis.

MATERIALS AND METHODS

Materials DRN free base was provided from Dong-A Pharmaceutical Co. (Korea, purity over 97.0% w/w). Kolliphor EL and Labrafir M1944CS were kindly obtained from BASF Co., Ltd. (Ludwigshafen, Germany) and Gattefosse (Saint Priest, France), respectively. Verapamil (purity over 98.0% w/w) was used as an internal standard (IS) for LC/MS-MS analysis, and was purchased from Sigma Chemical Co. (St. Louis, MO, U.S.A.). Empty hard gelatin capsules (size #1) were obtained from Capsugel (Greenwood, SC, U.S.A.). Acetonitrile and methanol of HPLC grade were purchased from J.T. Baker (Phillipsburg, NJ, U.S.A.). All other chemicals used were of analytical grade.

Preparation of DRN-Loaded SMEDDS The DRN-loaded SMEDDS was prepared by dissolving 4000 mg of DRN free base in a mixture of oil (Labrafir M1944CS) and surfactant (Kolliphor EL, RH40, RH60, Labrasol, Tween 80, and d-locopheryl polyethylene glycol (PEG) 1000 succinate). The components were mixed using a magnetic stirrer at 1000 rpm until the drug was completely dissolved. The transparent and viscous solution containing 200 mg of DRN was placed in #1 hard gelatin capsules for an in vitro dissolution test and an in vivo pharmacokinetic assessment.

Physicochemical Characterization of DRN-Loaded SMEDDS Droplet size and surface charge: A DRN-loaded SMEDDS sample was diluted 1:100 with appropriate medium, and particle size distribution and zeta potential were assayed using a dynamic light-scattering particle size analyzer (Zetasizer Nano-ZS, Malvern Instrument, Worcestershire, U.K.) with a 50 mV laser at a scattering angle of 90°. All measurements were carried out in triplicate at 25°C.

Drug content: DRN-loaded SMEDDS samples containing 40 mg of the drug as a base was dissolved in 1000 mL of acetonitrile and sonicated for 10 min. The homogeneous solution was analyzed by HPLC analysis. The quantitative determination of DRN was performed by HPLC using acetonitrile–water–triethanolamine (900:100:1) as a mobile phase at a flow rate of 1.0 mL/min. The HPLC system consisted of a pump (L-2130), UV detector (L-2400), a data station (LaChrom Elite, Hitachi, Japan), and a 15 cm C18 column (Capcell Pak C18 MG column, Shiseido, Japan). The column eluent was monitored at 245 nm, and the peak at retention time of 4.5 min representing DRN.

Dissolution Test Dissolution studies were performed according to the USP XXVIII paddle method using a VK 7000 dissolution testing station and VK 750d heater/circulator (Varian Industries, New Jersey, NJ, U.S.A.). SMEDDS and the marketed product containing 400 mg of DRN as a base were placed in the dissolution medium, e.g., distilled water, pH 1.2, pH 4.0, pH 6.8, fasted state simulated intestinal fluid (FaSSIF) and fed state simulated intestinal fluid (FeSSIF). The compositions and the preparation procedures of the biorelevant media such as FaSSIF and FeSSIF, have been described previously.22 FaSSIF was prepared with 3 mM of sodium taurocholate, 0.75 mM of lecithin, 3.9 g of KH2PO4, and 7.7 g of KCl in 1000 mL of distilled water (pH 6.5, osmolality 270 ± 10 mOsm·kg−1). FeSSIF was prepared with 15 mM of sodium taurocholate, 3.75 mM of lecithin, 8.65 g of acetic acid, and 15.2 g of KCl in 1000 mL of distilled water (pH 5.0, osmolality 670 ± 10 mOsm·kg−1). The stirring speed was set to 50 rpm, and the temperature was maintained at 37 ± 0.5°C. At predetermined times, 5 mL aliquots were withdrawn and filtered using a 0.45 µm glass membrane syringe filter (Whatman GD/X, GE Healthcare). Filtered samples were appropriately diluted with acetonitrile, and the drug concentration was assayed by HPLC as described earlier.

In Vivo Absorption Study in Beagle Dogs Drug administration: The animal study was performed in accordance with the NIH guideline “Principles of Laboratory Animal Care” (NIH publication No. 85-23, revised 1996) and approved by the Institutional Animal Care and Use Committee of Dong-A Pharmaceutical Co. in Seoul, Korea. The pharmacokinetic studies in beagle dogs were crossover studies to evaluate the oral BA after the oral administration of a single 400 mg DRN dose as the novel SMEDDS-loaded capsule and the marketed product (Multaq®) under fed and fasted conditions Male Beagle dogs (n = 20) weighing 6–12 kg were fasted overnight. The dogs were either fasted or fed solid chow (about 100 mg, fat 11%, protein 27%) just prior to receiving the SMEDDS-loaded capsule or the marketed tablet followed by 20 mL of water. Blood samples were collected in a heparinized syringe at 0, 0.5, 1, 2, 3, 4, 6, 8, 10, 12, and 24 h after oral administration. Those blood samples were ultracentrifuged at 12000 rpm for 10 time, and the plasma was collected and stored at −70°C. The DRN concentrations in the plasma were analyzed by the LC-MS/MS.

LC-MS/MS determination of DRN in beagle dog plasma: An LC-MS/MS assay was developed to determine the concentrations of DRN in beagle plasma. A 100 µL aliquot of plasma was transferred into a glass tube, followed by the addition of 100 µL of verapamil (200 ng/mL) as an internal standard and 200 µL of acetonitrile for protein precipitation. The mixture was vigorously vortexed for 2 min and then centrifuged at 13000 rpm for 5 min. Supernatant (20 µL) was injected into the LC-MS/MS system. Detection was performed with a Micromass Quattro Micro (Waters, U.S.A.) mass spectrometer with electrospray ionization (ESI) in positive ion mode for ion production. Chromatography was performed isocratically on an Xterra RP18 column (2.1 mm i.d.×100 mm, particle size: 5 µm). The mobile phase was 10 mM ammonium acetate–acetonitrile (2:8, v/v), pH 6.6, at a flow rate of 0.3 mL/min. Chromatography was performed at 40°C. The analytes were detected by monitoring the transitions m/z 557.4→100.1 and 455.0→165.0 for DRN and verapamil, respectively. The analytical time for each run was 4 min. The calibration equation was determined by least-squares linear regression (weighting 1/x) over the range 10–500 ng/mL in plasma. The precision and accuracy of the method were determined at five quality control sample levels.

Pharmacokinetic analysis: Pharmacokinetic analysis was performed using a BA Calc 2007 pharmacokinetic analysis computer program (Korea Food & Drug Administration). Area under the plasma concentration–time curve (AUC) was calculated using the linear trapezoidal rule by the program. The maximal plasma concentration of DRN (Cmax), the time needed to reach the maximum blood concentration (Tmax), and the half-life (t1/2) were also calculated by the program.
Table 1. Effect of Surfactant Added to Labrafil M1944CS on Mean Droplet Size in Fasted State Simulated Intestinal Fluid (FaSSIF) in a Weigh Ratio of 1:1

<table>
<thead>
<tr>
<th>Surfactants</th>
<th>Initial (nm)</th>
<th>After 2h (nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cremophor EL</td>
<td>93.3</td>
<td>151.4</td>
</tr>
<tr>
<td>Labrasol</td>
<td>571.7</td>
<td>—</td>
</tr>
<tr>
<td>Tween 80</td>
<td>1259</td>
<td>—</td>
</tr>
<tr>
<td>Cremophor RH 60</td>
<td>161.4</td>
<td>582</td>
</tr>
<tr>
<td>Cremophor RH 40</td>
<td>5264</td>
<td>—</td>
</tr>
<tr>
<td>Vit. E TPGS&lt;sup&gt;c&lt;/sup&gt;</td>
<td>341.2</td>
<td>309</td>
</tr>
</tbody>
</table>

<sup>a</sup>) Values represent mean (n=3, relative standard deviation <10%).
<sup>b</sup>) D-Tocopheryl polyethylene glycol 1000 succinate.
<sup>c</sup>) Not determined.

To demonstrate statistically significant differences, one-way ANOVA followed by least-squares difference test and the Student–Newman–Keul test was performed using SPSS software (v 12.0; SPSS Inc., Chicago, IL, U.S.A.).

RESULTS AND DISCUSSION

Formulation Development and Characteristics of DRN-Loaded SMEDDS

The novel self-emulsifying system of DRN was designed to improve its oral absorption, especially in fasted state, and to reduce the difference in drug absorption between the fasted and fed states. This formulation could be taken either in the fasted or the fed state with total exposures that are comparable to those of the marketed product dosed in the fed state. Ultimately, we expected that reducing the differences in oral BA between the prandial states would significantly improve patient compliance, as well as the therapeutic efficacy of the anti-arrhythmic agent. To formulate a SMEDDS for a poorly water-soluble compound like DRN, the selection of a suitable oil and surfactant is critical because the drug-loading capacity in the formulation predominantly depends on the solubility of the drug in the ingredients. To select a suitable oil for the DRN-loaded SMEDDS, oils in the generally recognized as safe (GRAS) category were screened for their ability to dissolve DRN at room temperature. DRN was most soluble in Labrafil M1944CS when compared to other oils such as Capryol 90, Maisin 35-1, Peceol, Labrafac PG and Laurolglycol 90 (data not shown). The droplet size and physical stability of various Labrafil M1944CS/surfactant 1:1 blends were evaluated after dilution in FaSSIF medium to grade the relative emulsification capability of each surfactant (Table 1). Kollidone EL and RH60 rapidly formed transparent microemulsions with the droplet size below 200 nm. Whereas, Labrasol, Tween 80, Kollidone RH40, and d-tocopheryl PEG 1000 succinate were not appropriate with Labrafil M1944CS as they produced coarse dispersion ranging from 350 to 1250 nm. The droplet size after emulsification in aqueous medium is a critical factor in the SMEDDS formulation because a smaller globular size yields a larger interfacial surface area for drug absorption with excellent colloidal stability. No change in the physical parameters such as droplet size and clarity was observed for test period in Labrafil M1944CS/ Kollidone EL formula (Table 1) and thus, Kollidone EL was selected as a surfactant. Previous literature revealed that DRN could be a potent P-glycoprotein (P-gp) substrate and thus, we also expected that the incorporation of Kollidone EL in SMEDDS, a well-known P-gp inhibitor, might facilitate the intestinal absorption of the anti-arrhythmic agent, especially in the fasted state. In healthy individuals in the fasted state, Kollidone EL dose-dependently (up to 5000 mg) increased the intestinal absorption of saquinavir, a P-gp substrate. The ratio of drug–oil–surfactant was determined based on preliminary studies, including and in vitro release test in FaSSIF medium and the measurement of droplet size. Figure 1 shows the dissolution profiles of DRN in the biorelevant medium from SMEDDS formulation at different ratios of drug–oil–surfactant. The drug release from the SMEDDS consisted of 1:0.5:0.5 in a weight ratio was only 20% after 2h. The SMEDDS composition in a weight ratio of 1:1:1 showed dissolution profile similar to that of 1:0.5:1; approximately 45% of the drug was released after 2h. On the other hand, the dramatic increase in the rate of release of DRN was obtained from SMEDDS consisted of 1:1:2, in which the cumulative release rate of DRN was approximately 90% after 2h. Therefore, the optimized SMEDDS formula was composed of DRN free base (400 mg), Labrafil M1944CS (400 mg), and Kollidone EL (800 mg) (Table 2). The formulation formed a transparent emulsion when it made contact with the aqueous medium, providing a small droplet size less than 50 nm with a neutral surface charge (Table 2). The low polydispersity index (PDI) of less than 0.3 indicates a narrow and homogeneous size distribution (Table 2). The prepared SMEDDS (800 mg)
contained 200 mg of DRN was placed in No. 1 hard gelatin capsules and two capsules (400 mg) of DRN was orally administered to beagles.

**Dissolution Test**

To increase the oral absorption of poorly water-soluble drugs with a reduced food-effect, it is very important to maintain high drug dissolution through the gastrointestinal tract, regardless of pH conditions and/or bile acid and salt concentrations. DRN was reported to have a considerable pH-dependent solubility profile, thus its release profiles were evaluated in media of different pH (distilled water, pH 1.2, 4.0, and 6.8) as well as biorelevant dissolution media (FaSSIF and FeSSIF). As shown in Fig. 2, the release profile of DRN from Multaq® was remarkably pH-dependent. Over 70% of the drug was released after 2 h in pH 4.0 and distilled water, whereas the cumulative dissolution rate of DRN in pH 1.2 and 6.8 was only 23% and 2%, respectively. These patterns were in good agreement with the solubility property reported. The solubility of the drug in a weak acidic environment (pH 3 to 5) is about 1 to 2 mg/mL, but it significantly decreases by one hundredth in gastric fluid (pH 1.2) and/or intestinal fluid (pH 6.8). Dissolution profiles of the marketed product in FaSSIF and FeSSIF were significantly different, displaying the extent of released of 3.0% and 28.8%, respectively (Fig. 2). FeSSIF increased the amount of DRN released, mainly due to the mixed-micelle effects. Differences in the dissolution rate between FaSSIF and FeSSIF indicate that food intake has the potential to exhibit a positive effect on the intestinal absorption of DRN by increasing drug solubility in the GI track. This difference was observed distinctively for dissolution profiles of BCS class ΙΙ molecules with positive food-effects, e.g., atovaquone, troglitazone.

On the other hand, dissolution profiles of DRN from SMEDDS formulation were quite different from that from the marketed product. Drug dissolution from SMEDDS formulation was significantly higher and faster compared to the Multaq® tablet, regardless of pH conditions (Fig. 2). In all media tested, more than 80% of DRN in the SMEDDS formula was released within 60 min. In particular, the amounts of DRN released from the SMEDDS formula in pH 1.2 and pH 6.8 were noticeably higher than those released from the marketed product, providing about 3.3-fold and 53.0-fold higher cumulative dissolution rate, respectively. In addition, there were no remarkable differences in the release rate and extent of dissolution between FaSSIF and FeSSIF, both of them releasing more than 80% drug within 60 min (Fig. 2). On the other hand, the globular size of SMEDDS in FaSSIF and FeSSIF medium were initially measured to approximately 50 nm and 120 nm, respectively. The droplet size was gradually increased in both biorelevant mediums, but it was maintained below 300 nm at least for 2 h (data not shown). These results indicate that SMEDDS could be spontaneously dispersed in the aqueous medium to produce a fine oil-in-water (o/w) emulsion upon mild agitation, maintaining the incorporated drug in a solubilized state. Based on the results of the dissolution test, we predicted that the DRN in a SMEDDS formula would reduce the food-effect, and increase BA in a fed and especially fasted state.

**LC-MS/MS Determination of DRN in Beagle Dog Plasma**

The LC-MS/MS tool provided a robust determination of DRN in the beagle dog plasma. The retention times for DRN and IS, verapamil, were 2.6 and 1.9 min, respectively (data not shown). No endogenous blood components or pharmaceutical excipients were eluted at the retention times of the peaks of interest. The calibration curves were constructed by spiking blank plasma with known amounts of the benzofuran derivatives in the range of 10–500 ng/mL. The standard calibration curve (n=5) was linear, with a correlation coefficient of 0.997. In beagle dog plasma, intra-day precisions (CV %) ranged from 0.8 to 3.1%, and accuracies ranged from 86.5 to 108.3% (data not shown). Intra-day precision and accuracy were found...
to be within the acceptance criteria for assay validation guidelines.

**In Vivo Absorption Study in Beagle Dogs** The mean plasma DRN concentration–time profiles after oral administration of either the SMEDDS formula or the marketed product to the beagle dogs in the fed and fasted state groups are described in Fig. 3. For further statistical evaluation, relative differences in the pharmacokinetic parameters, such as \( \text{AUC}_{0-24\, \text{h}} \) and \( C_{\text{max}} \), after the administrations of SMEDDS or commercially available product in the fasted state or after a meal are summarized in Table 3. After oral administration of DRN as the marketed product in the fed state, \( \text{AUC}_{0-24\, \text{h}} \) and \( C_{\text{max}} \) were even 10.4-fold (\( p<0.05 \)) and 8.6-fold (\( p<0.05 \)) higher than those in the fasted state, respectively (Table 3). In the fed state, \( \text{AUC}_{0-24\, \text{h}} \) and \( C_{\text{max}} \) after Multaq® administration were 7956.5 ng·h/mL and 1297.9 ng/mL, respectively, compared to only 761.0 ng·h/mL and 150.8 ng/mL in the fasted state, respectively. These results are consistent with earlier studies that showed increased oral BA of DRN by food intake in healthy males. Food increased the drug BA of 800 mg of DRN by an average of approximately 2- to 5-fold.\(^{15}\) In the fed state, food increased the gastric residence period of the drug, while in the fasted state the drug passes through the empty stomach much faster with a lower solubility in the medium,\(^{10}\) as revealed in dissolution test. Furthermore, dietary components, especially fat, might increase bile secretion into the small intestine, which may solubilize lipophilic drugs within bile salt mixed micelles, and facilitate diffusion through the aqueous diffusion layer of the intestinal membrane in the fed state.\(^{19}\) The overall plasma DRN concentration-time profiles were quite similar regardless of food intake; the plasma concentration of DRN gradually rose and peaked between 2 and 3 h. The results are quite consistent with previous studies which demonstrated that food did not significantly prolong drug absorption; the \( T_{\text{max}} \) was the same at 5 h under fasted and fed conditions in healthy subjects.\(^{3}\)

In contrast to this food-effect observed with Multaq®, the low-fat meal has a less pronounced influence on the pharmacokinetics of DRN with the self-emulsifying formulation. DRN orally administered as the SMEDDS formulation in the fasted state significantly increased the pharmacokinetic parameters such as \( \text{AUC}_{0-24\, \text{h}} \) and \( C_{\text{max}} \) of DRN when compared to the marketed product, but there were no significant differences in the blood concentration at each observed time point between the SMEDDS and marketed product in the fed state (Fig. 3). After administering the SMEDDS formula in the fasted state, \( \text{AUC}_{0-24\, \text{h}} \) and \( C_{\text{max}} \) values were approximately 3.1-fold (\( p<0.05 \)) and 2.7-fold (\( p<0.05 \)) higher, respectively, than those of the marketed product. In case of BCS class II compounds such as DRN, the rate and extent of absorption are greatly governed by the solubility of compounds in GI tract. Thus, a remarkably higher dissolution rate of DRN by SMEDDS formulation in the pH range of 1.2 to 6.8 led to a greater absorption rate compared to the marketed product in the fasted state. As a result, when DRN-loaded SMEDDS was given to beagles in the fed state (Table 3), \( \text{AUC}_{0-24\, \text{h}} \) and \( C_{\text{max}} \) increased only by 2.9-fold (\( p<0.05 \)) and 2.6-fold (\( p<0.05 \)) when compared to the fasting state, respectively. Whereas the \( \text{AUC}_{0-24\, \text{h}} \) and \( C_{\text{max}} \) increased by 10.4-fold and 8.6-fold (\( p<0.05 \)) after administration of the marketed product in the fed state as compared to the fasting state. These data suggest that SMEDDS effectively alleviated the food-effect on DRN absorption. SMEDDS formulation immediately formed fine droplet of sufficiently small sizes for absorption in the stom-

![Fig. 3. Mean Plasma Concentration–Time Profiles of DRN Following a Single Oral Administration of (a) the Marketed Product (Multaq®) or (b) SMEDDS under Fasted Condition (○) or with a Low-Fat Meal (●). Each point represents mean±S.D. (n=10).](image)

| Table 3. Comparison of the Mean Pharmacokinetic Parameters of DRN after Single Oral Administration of SMEDDS or the Marketed Product (Multaq®) at a Dose of 400 mg to Beagle Dogs in the Fasted and Fed States |
|-----------------------------------------------|-----------------------------------------------|-----------------------------------------------|
| SMEDDS                                      | Multaq®                                    |
| Fed\(^{a}\)                                | Fed\(^{a}\)                                |
| Fed/Fasted\(^{b}\)                         | Fed/Fasted\(^{b}\)                        |
| \( \text{AUC}_{0-24\, \text{h}} \) (ng·h/mL) | \( \text{AUC}_{0-24\, \text{h}} \) (ng·h/mL) | \( \text{AUC}_{0-24\, \text{h}} \) (ng·h/mL) |
| 7091.4±2352.8                              | 7956.4±3013.2                              | 761.0±456.9                                  |
| 2396.6±1302.2                              | 456.9±10.4                                 | 10.4                                         |
| \( C_{\text{max}} \) (ng/mL)               | \( C_{\text{max}} \) (ng/mL)               |
| 1098.9±213.0                               | 1297.9±448.8                               | 150.7±88.7                                  |
| 412.1±167.3                                | 2.8±1.1                                    | 8.6                                          |
| \( T_{\text{max}} \) (h)                   | \( T_{\text{max}} \) (h)                   |
| 3.0±1.3                                    | 2.8±1.1                                    | 2.3±0.6                                      |
| \( t_{1/2} \) (h)                          | \( t_{1/2} \) (h)                          |
| 4.8±1.9                                    | 4.7±1.2                                    | 5.4±2.8                                      |
| \(^{a}\) Data are expressed as the mean±S.D. (n=10). \(^{b}\) The values were calculated by dividing mean values obtained in the fed state by average values obtained in the fasted state. \(^{c}\) Not determined.
ach. Since SMEDDS is sufficiently solubilized the compound by itself, the solubility and/or dissolution pattern of SMEDDS was not extensively dependent on the concentration of bile salts or pancreatic enzymes. This result is consistent with an earlier report which stated that a SMEDDS formulation of cyclosporin A (Neoral) improved oral BA in the fasted state, decrease the food effect, and increase dose linearity compared to the crude emulsion (Sandimmuno). It is also supported by an earlier report that itraconazole-loaded SMEDDS produced significantly higher plasma drug levels in rats, especially in the fasted state, than the marketed product, thus reducing the food-effect during intestinal absorption.

It is worth noting that, in spite of significant differences in the in vitro dissolution rate of DRN between SMEDDS and Multaq® tablet in FeSSIF medium, the plasma concentration–time profile of two formulas was almost equivalent in terms of AUC<sub>0–24h</sub> and C<sub>max</sub> under fed state. Although, drug dissolution from SMEDDS formulation was significantly higher and faster compared to that of Multaq® tablet in FeSSIF medium, there were no significant differences in the plasma concentration–time profile between the SMEDDS and the marketed product in the fed state. This discrepancy between the in vivo oral BA and in vitro dissolution test could be associated by physiological changes in the GI tract by food intake, which might have not been fully considered in designing in vitro dissolution tests. It was reported that food induces the alternations in the GI tract such as the secretion levels of gastric acid and/or bile and pancreatic fluids, gastric and intestinal motility patterns, and visceral blood and lymph flow. Especially, the delayed gastric emptying by food was reported to have the potential to increase absorption of poorly water-soluble drugs including DRN by increasing the time available for drug dissolution.

This high dissolution rate of DRN in weak acid in both formulations also might be contributed to diminish the formulation effect on intestinal absorption under post-prandial state. It was reported that DRN is well absorbed (≈70% to 94%) after oral administration of the marketed product, although the absolute BA of DRN is only 15% when given with food in healthy subjects. From these findings, we concluded that DRN was well absorbed through GI tract, regardless of formulation variable under fed state. However, the employment of self-emulsifying system could provide a benefit in oral therapy of DRN because it reduces the differences in oral absorption between the prandial states, by improving BA of DRN in the fasted state. Furthermore, the SMEDDS formula is expected to reduce inter- and intra-individual variability in drug absorption, by keeping the solubilized state of the compound under GI circumstances with lesser sensitivity to food intake as previously reported.

Actually, DRN-loaded SMEDDS provided lesser inter-individual variability in terms of AUC<sub>0–24h</sub> and C<sub>max</sub> compared to the marketed product under fed state (Table 3).

This study emphatically found that self-emulsifying formulation could be a promising alternative for DRN therapy, by improving oral BA and reducing the food-effect on drug absorption. Nevertheless, further studies are needed to recommend the optimal SMEDDS formulation for use in clinical applications. Reformulation will include reducing the amount of Kolliphor EL in the SMEDDS formula and replacing it with safer pharmaceutical excipients. The recommended dosage of DRN is 400 mg twice daily in adults, therefore, the amount of Kolliphor EL intake is 1600 mg per day in case of the SMEDDS formula. However, the dose of the surfactant in oral formulations ranges from 2 to 600 mg per day in currently approved products, because of adverse effects of the surfactant such as anaphylactoid hypersensitivity reactions, axonal demyelination and acute toxicity of the heart and thymocytes.

In addition, pharmacokinetic evaluation studies are supplementary needed to determine DRN absorption after a high fat-meal. In a previous clinical study, a high fat meal had a slightly greater effect on DRN absorption (ca. 40% increase) than a low fat meal. In vitro dissolution behavior cannot completely explain potential food-effect with DRN-loaded SMEDDS formulations, thus studies including the lipolysis of SMEDDS formula are needed to thoroughly explain potential food interactions with the formulations.

In conclusion, the SMEDDS formula consisting of Labrafac M 1944CS and Kolliphor EL was easily dispersed in aqueous medium to spontaneously produce fine o/w emulsion with the formulations. 26,27) The high dissolution rate of DRN in weak acid by increasing the time available for drug dissolution including DRN by increasing the time available for drug dissolution.

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**Conflict of Interest** The authors declare no conflict of interest.

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