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International Journal of Pharmaceutics 250 (2003) 147–156
international journal of pharmaceutics
www.elsevier.com/locate/ijpharm

Assessment of oral bioavailability enhancing approaches for SB-247083 using flow-through cell dissolution testing as one of the screens

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Received 20 February 2002; received in revised form 11 September 2002; accepted 11 September 2002

Abstract

SB-247083 is a potent, nonpeptidic, orally active, ETA-selective, endothelin receptor antagonist. The diacid form and three salts (monoarginine, diarginine and disodium) of SB-247083 were evaluated during the pre-clinical phase of development. The developability attributes (i.e. hygroscopicity, thermal behavior, aqueous solubility, and drug-excipient compatibility) of these compounds were evaluated. In addition to these attributes, the flow-through cell (FTC) dissolution testing (using USP Apparatus 4) was used as a screening technique to evaluate several SB-247083 formulations of the diacid and its salts. FTC dissolution testing offers two distinct advantages over the more traditional static-condition dissolution testing: (1) maintenance of sink conditions; and (2) the ability to change the dissolution medium during a dissolution run. The former advantage is especially important for poorly aqueous soluble drugs having associated dissolution-rate-limitations, and the latter advantage allows one to more closely simulate the pH gradient associated with transit through the GI tract. Based on the comparative dissolution data, three formulations were chosen for oral dosing in dogs. The reasonable correlation found between the FTC dissolution results and the oral bioavailability data demonstrate that FTC dissolution testing can be a valuable tool for aiding in salt (solid-state form) and formulation selection in the early stages of development of drug candidates.

Keywords: SB-247083; Salt selection; Flow-through cell dissolution; Bioavailability

1. Introduction

With the advent of combinatorial chemistry and high throughput screening techniques, an ever increasing number of potential drug candidates are being progressed into the early stages of

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0378-5173/02/$ - see front matter
PII: S0378-5173(02)00521-5

development. Hence, quick and meaningful techniques to aid in the selection of superior candidates are a must. When a compound has been selected from a given series, a critical aspect in its development if it is ionizable is the selection of an appropriate chemical form whether it be the free acid or base or a salt (Berge et al., 1977). For compounds having dissolution-rate limited oral bioavailabilities, improvement in their in-vivo performance may be achieved by selecting an appropriate salt form due to improvement in solubility and dissolution rate. Morris et al. (1994) used an integrated approach to select an optimal salt of BMS-180431. Their approach involved the comparative evaluation of the hygroscopicity, crystal form integrity, aqueous solubility and chemical stability of the different salts.

In the absence of direct in vivo comparisons, an additional comparative test which can add value to the selection process for ionizable compounds having dissolution-rate-limited oral bioavailabilities, is flow-through cell (FTC) dissolution testing (USP Apparatus 4). It offers two distinct advantages over the more traditional static-condition dissolution testing: (1) maintenance of sink conditions; and (2) the ability to change the dissolution medium during a dissolution run. The former advantage is especially important for poorly aqueous soluble drugs having associated dissolution-rate-limitations, and the latter advantage allows one to more closely simulate the pH gradient associated with transit through the GI tract (Thoma and Ziegler, 1998). As an added advantage, some investigators have demonstrated that good in vitro/in vivo correlations can be established using FTC dissolution testing (Derendorf et al., 1983; Aiache et al., 1987; Phillips et al., 1989).

SB-247083, ((E)-α-[(1-butyl-5-[2-2(2-carboxyphenyl)methoxy]-4-chlorophenyl]-1H-pyrazol-4-yl]methylene]-2,3-dihydro-5-methoxy-6-benzofuranpropanoic acid; Fig. 1), is a potent class of nonpeptidic, orally active, ETA-selective endothelin receptor antagonist with a K_i of 0.41 nM (Willette et al., 1998). It has two carboxylic acid groups and is poorly soluble in water. Initial oral bioavailability studies in dogs were performed with a minimally formulated version of a crystalline form of the diacid of SB-247083 and yielded a bioavailability of 3.9%. Subsequent bioavailability studies supported the involvement of a dissolution-rate-limited component to the oral bioavailability. Hence, salt formation and formulation approaches were utilized in an attempt to improve the oral bioavailability of SB-247083. To aid in the selection of the best salt/formulation approach to progress, SB-247083 and its salts were evaluated by hygroscopicity, polymorphism, and excipient compatibility studies. Additionally, the FTC dissolution profiles of several formulations of SB-247083 was also compared to those of the SB-247083 salt forms. Lastly, based on the results of this comparative physicochemical screening, the most promising approaches were progressed into oral bioavailability studies performed in dogs.

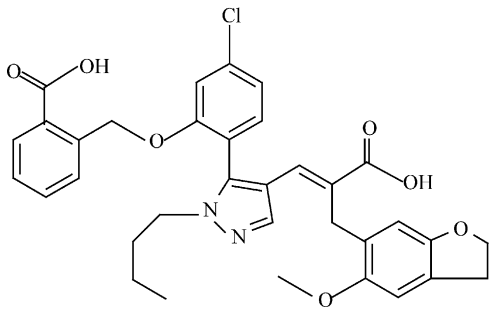

Fig. 1. The chemical structure of SB-247083.

2. Materials and methods

2.1. Materials

The diacid of SB-247083 (99.1% purity) was synthesized by the Chemical Development Department at GlaxoSmithKline Pharmaceuticals (King of Prussia, PA). Avicel PH-102 (FMC Corporation), Lactose Fast Flo and Lactose Impalpable (Foremost Farms), Starch 1500 (Colorcon), Starch 1551 (National Starch), PVP K30 (Kollidon 30, BASF), magnesium stearate (Mallinckrodt), L-arginine (Sigma), meglumine (Aldrich) and Tween 80 (Sigma) were used as received. The solvents used were HPLC grade. All other chemicals were of analytical grade, and the water used was

deionized then filtered through a Milli-Q Water Purification System prior to use.

2.2. Preparation of salts

The salts of SB-247083 were obtained from mixing the diacid and the corresponding molar ratios of bases in boiling methanol. Monoarginine salt was crystallized upon cooling while the disodium and diarginine salts were obtained by adding methylene chloride and n-hexane.

2.3. Differential scanning calorimetric (DSC) analysis

Using a Perkin Elmer DSC 7, the thermal behavior of the various forms of SB-247083 were examined over the temperature range of 30–250 °C and at a heating rate of 10 °C/minute. The materials (2–5 mg) were placed in crimped aluminum pans and evaluated under a nitrogen atmosphere.

2.4. Moisture sorption studies

The isothermal sorption of water by the various forms of SB-247083 were examined using a VTI vacuum microbalance (Model MB 300W). The materials (15–50 mg) were first dehydrated by drying under vacuum at 60 °C until there was less than a 5-μg weight change observed in three, consecutive, 5-min periods. After drying, the temperature was lowered to 25 °C, and the partial water vapor pressure over the sample was increased, in 5% relative humidity (RH) steps, from 0 to 95% RH. The sample was held at each RH step until no more than a 5-μg weight change was observed in three, consecutive, 5-min intervals. The weight gain or loss at each RH step was measured on the associated microbalance.

2.5. Aqueous solubility studies

The aqueous solubilities of the diacid form SB-247083 and its monoarginine salt were determined, in duplicate, at 25 °C. Excess solid (50 mg) was added to 10 ml glass centrifuge tubes with Teflon-lined screw caps. Five milliliters of water or aqueous buffered solutions were added to each tube then the tubes were placed on a VIBRO-Mixer (Chemap AG, Switzerland) which was set in a water bath maintained at 25 °C. The tubes were agitated for 24 h. (Preliminary studies showed that this was ample time to attain equilibrium solubility.) Then the solutions were filtered through 0.45-μm PTFE syringe filters. The pH of the subsequent filtrate was taken, and the filtrate was assayed by HPLC.

2.6. Drug-excipient compatibility studies

To assess excipient compatibility, physical mixtures of the diacid of SB-247083 and common pharmaceutical excipients were prepared. The ratios of the diacid to a given excipient were as follows: 1:5 with Starch 1500, with Avicel PH-102, and with Fast Flo lactose, 1:3 with starch 1551 and PVP K-30, and 5:1 with magnesium stearate. The mixtures were stored at two conditions, 5 and 55 °C, for 3 weeks. (The 5 °C condition served as the 'control' condition.) HPLC and DSC testing were used to evaluate any chemical or physical changes occurring with SB-247083. Based on this work, the monoarginine salt and a prototype formulation of the diacid were selected for a further compatibility study. The wet granulated formulation contained drug (10%w/w), Avicel PH-102 (20%w/w), Starch 1551 (5%w/w), lactose impalpable (65%w/w) with water (35%v/w total weight of dry powder).

2.7. Preparation of formulations

Seven formulations (six using the diacid and one using the monoarginine salt) were prepared for evaluation (Table 1). For Formulations **1** and **7–9**, the drug was mixed with Avicel PH-102. For Formulations **2–6**, the ingredients were wet granulated using an aqueous solution containing Tween 80. After wet granulation, the formulations were tray dried at 50 °C for 2 h. All of the formulations were filled into Size 2, hard gelatin capsules for FTC dissolution testing. Lastly, an aqueous solution formulation in 0.1 M carbonate buffer (pH 8.0) was prepared for intravenous dosing in dogs

Table 1
Quantitative compositions of formulations of SB-247083 diacid and its salts

Formulation	Quantity (mg/capsule)								
	1	2	3	4	5	6	7	8	9
SB-247083[1]	10.0	10.0	10.0	10.0[2]	10.0	10.0	–	–	–
Monoarginine salt[3]	–	–	–	–	–	–	12.8	–	–
Disodium salt	–	–	–	–	–	–	–	10.7	–
Diarginine salt	–	–	–	–	–	–	–	–	15.6
Avicel PH-102	10.0	25.0	25.0	25.0	25.0	25.0	10.0	10.0	10.0
Lactose, impal.	–	58.0	59.5	59.5	56.3	53.1	–	–	–
Starch 1551	–	5.0	5.0	5.0	5.0	5.0	–	–	–
Tween 80	–	2.0	0.5	0.5	0.5	0.5	–	–	–
Meglumine	–	–	–	–	3.2	6.4	–	–	–

[1] Particle size: 41.1 μm (d_{90}), 19.5 μm (d_{50}), 5.6 μm (d_{10}).
[2] Material was micronized, particle size: 1.3 μm (d_{50}).
[3] Particle size: 56.7 μm (d_{90}), 14.6 μm (d_{50}), 5.30 μm (d_{10}).

to allow for the determination of the absolute oral bioavailabilities.

2.8. Flow-through cell (FTC) dissolution testing

An FTC dissolution apparatus with six cells, having an internal diameter of 22.6 mm, was used in all experiments (Sotax AG CH-4008 Basel). During testing, the dissolution medium was pumped through each cell. Hand-filled capsules of the various formulations ($n = 3$) were horizontally positioned in the sample holders in the cells and 6 g of glass beads (1 mm in diameter) were used to fill up the conical bottom part of each cell. Millipore filters (5 μm) were used in the filter-head in each experiment. The dissolution media were warmed up to 37 ± 1 °C, and a flow rate of 4 ml/min was used for the evaluation. During a dissolution run, the pH of dissolution medium was increased from 1.2 to 6.8 to mimic the pH of the GI tract. For each pH interval, samples were collected in 200 ml glass volumetric flasks. The sequence of dissolution media used for a given run was as follows: simulated gastric fluid (pH 1.2) was used for the 0–30-min interval, pH 3.0 phosphate buffer was used for the 30–60-min interval, pH 5.0 phosphate buffer was used for the 60–90-min interval, and pH 6.8 phosphate buffer was used for the 90–180-min interval. Post collection, the various fractions were adjusted to pH 6.8 using either 2 N NaOH or 2 N HCl solutions (20 ml of pH 6.8 phosphate buffer were added to pH 1.2 sample solutions before adjusted pH) then the solutions were diluted to 200 ml using pH 6.8 phosphate buffer. The samples were then assayed by UV spectrophotometry.

2.9. HPLC analysis

The HPLC analyses were performed on a Shimadzu system equipped with a 4.6×150 mm YMC proC18 column which was maintained at 40 °C in a column oven. The mobile phase consisted of a 50:50:0.1 (%v/v/v) mixture of water, acetonitrile and trifluoroacetic acid. The flow rate of the mobile phase was 1.5mL/min; the injection volume was 20 μl; the detection wavelength was 235 nm; and the run time was 25 min. Data acquisition and integration were performed with a Nelson System 6000 and Nelson Access*Chrom (version 1.8) software. The concentrations of SB-247083 were calculated using externally prepared standards.

2.10. UV spectrophotometric analysis

The UV spectrophotometric analyses were performed on an HP8452 system that was equipped with a 1-cm quartz cell. The concentrations of SB-247083 were calculated using externally prepared

standards and using the difference of the UV absorption at 286 nm (maximum) and 340 nm (baseline).

2.11. Oral bioavailability studies

Male beagle dogs (weighing 10–15 kg) were used for the oral bioavailability studies. The animals were fasted overnight prior to dosing, and the formulations were dosed either intraduodenally or orally. At pre-determined times, blood samples (~0.25 ml) were obtained from a catheter placed in the cephalic vein and transferred into heparinized 1.5-ml microcentrifuge tubes. Plasma (50 μl) was isolated from the blood samples by centrifugation, was transferred to new microcentrifuge tubes, and was quick frozen on dry ice. All samples were stored at −70 °C until analyzed.

SB-247083 was isolated from dog plasma by an on-line solid phase extraction method. Concentrations of SB-247083 in plasma samples were quantified using an LC/MS/MS method having a detection limit of ~10.0 ng/ml. The resulting data were analyzed using non-compartmental methods and pharmacokinetic analysis software (#Protocol Version 1.2). Plasma clearance was estimated by dividing the intravenous dose by the area under the plasma concentration versus time curve (AUC) from time zero extrapolated to infinity following intravenous dosing. Oral bioavailability was calculated from the ratio of time-matched dose-normalized AUCs following oral and intravenous dosing. The intravenous and oral half-lives were obtained by linear regression of log-transformed concentrations visually assessed to be on the linear portion of the terminal slope.

3. Results and discussion

Initial oral bioavailability studies were performed with a minimally formulated version of the highest melting, crystalline form of the diacid of SB-247083. (For these studies, the diacid was triturated in a mortar and pestle to yield a median particle size (d_{50}) of 19.5 μm). This material was mixed with microcrystalline cellulose (Avicel PH-102) in a 1:1 weight ratio. The resulting mixture was hand filled into hard gelatin capsules.) When this formulation was dosed orally in dogs, it yielded an oral bioavailability of 3.9%; in contrast, when a non-aqueous solution (99% PEG 400 and 1% DMSO) of the diacid was dosed intraduodenally in dogs ($n = 2$), it yielded an oral bioavailability of 14.5% which is close to the theoretical maximum value of 18% based on the associated enterohepatic extraction ratio of 0.82 and which suggests that the oral bioavailability of the solid dosage form was limited, at least in part, by the dissolution rate.

Based on these findings, a variety of salts and dissolution-enhancing formulation approaches were studied in an attempt to improve the oral bioavailability of SB-247083. To aid in the selection of the best approach to progress, the developability of SB-247083 and its salts (e.g. hygroscopicity, polymorphism, and drug:excipient compatibility) were evaluated. As an additional screen, the FTC dissolution testing of several formulations of the diacid of SB-247083 were compared to those of the SB-247083 salt forms. Based on the results of this physicochemical screening, the more promising approaches which progressed into oral bioavailability studies that were performed in dogs.

3.1. Polymorphic screening

To investigate its 'polymorphic potential', the diacid of SB-247083 was recrystallized from several organic solvents. DSC assessment of the resulting materials revealed that the diacid can exist in at least four different polymorphic forms. As shown in Table 2, the different forms had melting onset temperatures of 173, 186, 206 and 214 °C. The form which melted at the highest temperature (214 °C) was selected for further development. Similar recrystallization studies were conducted on the monoarginine salt of SB-247083; however, only one form, as evidenced by a single melting onset temperature of 209 °C, was observed regardless of the recrystallization solvent used. The two other salt forms, the disodium and diarginine salts, were amorphous.

Table 2
Apparent aqueous solubilities of SB-247083 diacid and monoarginine salt at various pHs

pH	Medium	Apparent solubilities (mg/ml)	
		Monoarginine salt	Diacid
1.2	HCl	< DL[1]	–
4.0	Acetate buffer	< DL	–
6.8	–	0.37[2]	0.013[3]
7.0	Phosphate buffer	0.47	–
7.5	Carbonate buffer	2.1	0.026
7.9	Carbonate buffer	5.4	–
8.2	Carbonate buffer	7.1	–

[1] Less than detection limit of ~0.1 μg/ml.
[2] A saturated solution.
[3] Phosphate buffer.

3.2. Moisture sorption studies

Moisture adsorption can significantly impact the physical and chemical properties of a drug substance (e.g. Campen et al., 1980; Umprayn and Mendes, 1987); hence, an assessment of a compound's ability to adsorb moisture is an important developability criterion. Fig. 2 shows the moisture adsorption curves for the diacid of SB-247083 and three salts over a relative humidity range of 0–90%. As can be seen, the diacid and monoarginine salt were minimally hygroscopic, adsorbing less than 1 and 3.6% moisture, respectively. The DSC thermograms for both post-study materials were identical to those of the respective pre-study materials, consistent with no moisture-induced changes in the solid-state form. In contrast, the disodium and diarginine salts were both found to be hygroscopic, adsorbing 33.7 and 47.1% moisture, respectively. This finding was not unexpected based on the amorphous nature of these salts.

3.3. Aqueous solubilities studies

As shown in Table 2, the apparent solubilities for the diacid and monoarginine salt of SB-247083 under acidic conditions (i.e. pH < 5.0) are less than 0.1 μg/ml. Around neutral pH, it can be seen that the apparent solubility for the monoarginine salt is significantly greater than that of the diacid (approximately 28- and 80-fold greater at pH 6.8 and 7.5, respectively.)

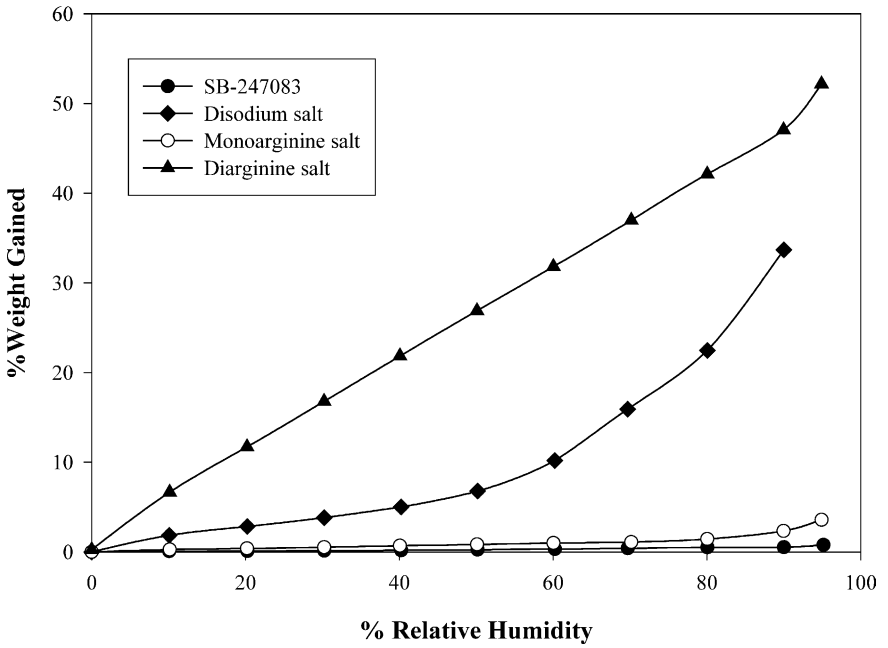

Fig. 2. Moisture adsorption curves for SB-247083 diacid and its salts.

Table 3
Extent of enhancement in FTC dissolution by various approaches and mean oral bioavailability for some formulations

Formulation	Dissolution enhancing approach	Amount of Tween 80 (%w/w)	Extent of enhancement in total % dissolved relative to formulation 1		F (%)
			pH 5	pH 6.8 (180 min)	
1	None	0	–	–	3.9 ± 0.5
2	Surfactant	2	1.3	2.8	–
3	Surfactant	0.5	1.2	3.2	2.3 ± 3.7[a]
4	Surfactant and micronized DS	0.5	1.4	3.7	–
5	Surfactant and alkalizing agent (1 mole eq.)	0.5	4.8	3.9	–
6	Surfactant and alkalizing agent (2 mole eq.)	0.5	6.9	4.5	6.3 ± 5.3[a]
7	Monoarginine salt	0	5.4	5.6	13.8 ± 7.6[b]
8	Disodium salt	0	8.7	3.3	–
9	Diarginine salt	0	12.3	5.1	–

[a] Not significant difference compared to F1 (Anova: single factor).
[b] Significant difference compared to F1 (Anova: single factor, P-value = 0.04).

3.4. Drug-excipient compatibility studies

In the drug-excipient compatibility studies, only magnesium stearate showed an incompatibility with the diacid of SB-247083. The combined mixture of drug and magnesium stearate showed an endothermic peak with an onset of 192 °C, but also showed three small endothermic with onsets of 93.6, 130.7 and 198.3 °C. These extra thermal events might be indicative of an incompatibility (Botha and Lötter, 1990); however, the HPLC testing showed no significant change in the levels of the diacid or the degradation products. These results are consistent with a physical, but not chemical, interaction occurring between magnesium stearate and SB-247083. Following exposure for a week at 55 °C, the loss in potencies, for the excipient mixtures containing the diacid and containing the monoarginine salt, was less than 0.5% (peak area response). This suggests that both forms of the compound may possess adequate chemical stability when formulated as a solid oral product. One noticeable difference between the excipient mixtures of the two forms was that those containing the monoarginine salt changed from an off-white to a yellow color. It is possible that this may be related to the interaction of lactose with an amino group of arginine via a Maillard-type reaction (Ellis, 1959).

3.5. Flow-through cell dissolution testing

For the FTC dissolution testing, the pH of the dissolution medium was increased from 1.2 to 6.8 during a given dissolution run in order to simulate the range of pH conditions that an orally dosed formulation would be exposed to while traversing through the gastrointestinal tract of a fasted human (e.g. Lui et al., 1986). The FTC dissolution of the seven formulations described in Table 1

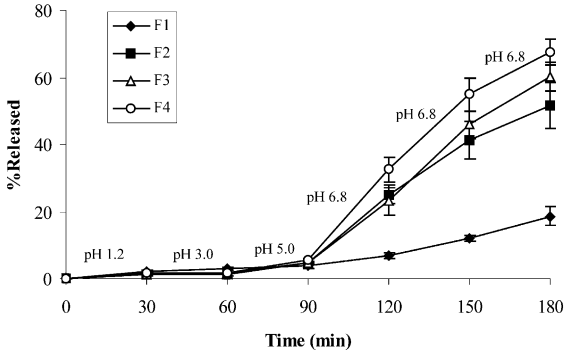

Fig. 3. Flow-through cell dissolution results for a series of formulations of SB-247083 diacid.

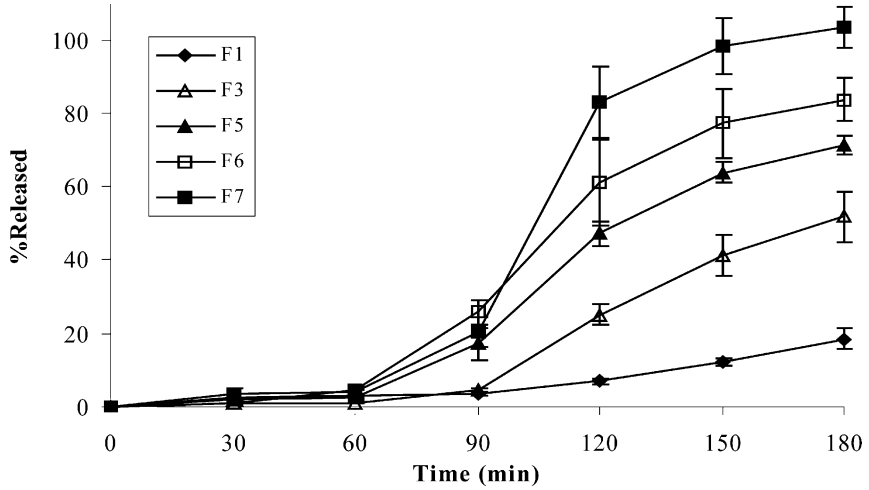

Fig. 4. Flow-through cell dissolution results for a series of formulations of SB-247083 diacid, and monoarginine salt.

were evaluated, and the results are shown in Figs. 3 and 4.

Fig. 3 shows the FTC dissolution profiles obtained for the formulation approaches that used either the micronized or unmicronized diacid of SB-247083 and that incorporated a nonionic surfactant (Tween 80) as the solubilization/dissolution aid. As can be seen, the total percentage of SB-247083 dissolved for the control formulation (i.e. Formulation 1, a 1:1 mixture of the unmicronized diacid and Avicel) under the varying pH conditions was about 19%, whereas all of the test formulations (i.e. Formulations 2, 3, and 4 that contained surfactant and in one case micronized drug) displayed significantly higher total percentages of the diacid dissolved. The differentiation in the dissolution profiles of the control and test formulations occurred when the pH of the dissolution medium was increased to 6.8. The increase in the extent of dissolution seen by incorporating a small amount of nonionic surfactant is generally due to a decrease in the interfacial tension between the drug substance and dissolution medium (Finholt and Solvang, 1968; Bakatselou et al., 1991).

A comparison of the profiles for Formulations 2 and 3 reveals that increasing the surfactant level from 0.5 to 2% w/w had a modestly negative effect on the extent of dissolution, whereas a comparison of the profiles for Formulations 3 and 4 reveals that micronization of the drug substance resulted in a further increase in the extent of dissolution. (Formulation 4 was identical to Formulation 3 except that the drug was micronized prior to formulating. The d_{50} values for the particle sizes of the unmicronized and micronized materials were 19.5 and 1.3 μm, respectively.)

An additional formulation approach that was studied, in an attempt to improve the dissolution rate of the diacid, was the incorporation of an alkalinizing agent (i.e. N-methylglucamine) into the formulation. To achieve this, Formulation 3 was modified by incorporating either 3.2% (one molar equivalent) or 6.4% (two molar equivalents) of meglumine (with a compensatory decrease in the amount of lactose) in the formulation to produce Formulations 5 and 6, respectively. As shown in Fig. 4, the formulations containing meglumine displayed higher total percentages of SB-247083 dissolved than the comparable formulation that did not contain meglumine. As expected, the increase in the extent of dissolution was 4.5-fold (Table 3) higher for the formulation containing two molar equivalents of meglumine relative to the formulation containing one molar equivalent (3.8-fold). The differentiation in the dissolution profiles of the control and test formulations occurred when the pH of the dissolution medium was increased to 5.0 (i.e. at a lower pH than seen with the surfactant containing formula-

tions). Mechanistically, an alkalinizing agent can raise the microenviromental pH surrounding the dissolving formulation leading to an increased rate of dissolution for acidic compounds (Lesson and Carstensen, 1974).

In addition to formulation approaches, a salt forming approach was also investigated as a means of increasing the extent of dissolution. Although the amorphous disodium and diarginine salts of SB-247083 were ruled out for further development because of their lack of crystallinity and hygroscopic natures, there was an interest in seeing how they performed, relative to the other approaches, on FTC dissolution testing. Among the three salts, the monoarginine salt (Formulation 7) displayed the highest total percentage of SB-247083 dissolved under the varying pH conditions. Only the dissolution profile of monoarginine salt is shown in Fig. 4. Table 3 summarizes the extent of dissolution enhancement by various approaches at pH 5.0 and pH 6.8 (180 min) with respect to Formulation 1.

3.6. Oral bioavailability studies

Based on the in vitro FTC dissolution results (Table 3), three formulations were progressed into the oral bioavailability studies: (1) the formulation containing the diacid and 0.5% Tween 80; (2) the formulation containing the diacid, 0.5% Tween 80, and two molar equivalents of meglumine; and (3) the formulation containing the monoarginine salt. Table 3 summarizes the results of this testing. (As a point of comparison, the maximum oral bioavailability of SB-247083 in dogs is estimated to be 18% based on an experimentally determined enterohepatic extraction ratio of ~ 0.82.)

If the FTC dissolution results (Fig. 4) are indicative of the what occurs in vivo, all three test formulations would be expected to have a higher oral bioavailability than the simple capsule formulation of the diacid. However, the mean oral bioavailability of the formulation containing the diaicd and 0.5% Tween 80 was slightly lower than that of the diacid alone (2.3 vs. 3.9%). It is possible that the presence of physiological surfactants in the GI tract may have a leveling effect on the wetting ability of the 0.5% Tween 80 in the formulation (Finholt and Solvang, 1968; Weintraub and Gibaldi, 1969) especially in dog where the total bile salt concentrations and secretion rates are high (Kararli, 1995). In contrast, the other two formulations followed the expected trend even though one is not statistically significant. The mean oral bioavailabilities for the formulation containing the diacid, 0.5% Tween 80, and two molar equivalents of meglumine and for the formulation containing the monoarginine salt were 1.6- and 3.5-fold higher, respectively, than the formulation containing the diacid alone. Additionally, the mean oral bioavailability of the monoarginine salt was consistent with the value found for intraduodenal dosing of a non-aqueous solution of the diacid.

These findings with the monoarginine salt were confirmed with additional testing performed in rats (Smith et al., 1998). An aqueous suspension of the diacid yielded an oral bioavailability of $20.7 \pm 10.0\%$ when dosed orally in rats; a non-aqueous solution (99% PEG 400 and 1% DMSO) of the diacid yielded an oral bioavailability of $59.8 \pm 6.3\%$ when dosed intraduodenally in rats; and an aqueous suspension of the monoarginine salt yielded an oral bioavailability of $45.6 \pm 13.1\%$. The mean oral bioavailability of the monoarginine salt suspension was lower than that of a non-aqueous solution of the diacid dosed id but was 2.2-fold higher than that of a diacid suspension.

4. Conclusions

Based on the physicochemical and biological data, the monoarginine salt of SB-247083 was selected for further development. Because of a potentially destabilizing chemical interaction, lactose will be excluded from future formulations. Lastly, for SB-247083, which is ionizable and where there is a significant dissolution-rate limiting component to the oral bioavailability, the flow-through cell dissolution testing was a useful predictor of the oral bioavailability of various formulations.

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