



SCHOOL of
GRADUATE STUDIES
EAST TENNESSEE STATE UNIVERSITY

East Tennessee State University
Digital Commons @ East
Tennessee State University

Electronic Theses and Dissertations

Student Works

8-2011

Biopharmaceutic and Pharmacokinetic Studies of Sucrose Acetate Isobutyrate as an Excipient for Oral Drug Delivery.

Martin Ray Tant

East Tennessee State University

Follow this and additional works at: <https://dc.etsu.edu/etd>

 Part of the [Medicinal and Pharmaceutical Chemistry Commons](#)

Recommended Citation

Tant, Martin Ray, "Biopharmaceutic and Pharmacokinetic Studies of Sucrose Acetate Isobutyrate as an Excipient for Oral Drug Delivery." (2011). *Electronic Theses and Dissertations*. Paper 1345. <https://dc.etsu.edu/etd/1345>

This Thesis - Open Access is brought to you for free and open access by the Student Works at Digital Commons @ East Tennessee State University. It has been accepted for inclusion in Electronic Theses and Dissertations by an authorized administrator of Digital Commons @ East Tennessee State University. For more information, please contact digilib@etsu.edu.

Biopharmaceutic and Pharmacokinetic Studies of Sucrose Acetate Isobutyrate
as an Excipient for Oral Drug Delivery

A thesis
presented to
the faculty of the Department of Physiology
East Tennessee State University

in partial fulfillment
of the requirements for the degree
Master of Science in Biomedical Sciences

by
Martin Ray Tant
August 2011

Tom W. Ecay, Ph.D., Chair
Kenneth E. Ferslew, Ph.D.
Mitchell E. Robinson, Ph.D.

Keywords: SAIB, Ibuprofen, Saquinavir, Clarithromycin

ABSTRACT

Biopharmaceutic and Pharmacokinetic Studies of Sucrose Acetate Isobutyrate as an Excipient for Oral Drug Delivery

by

Martin Ray Tant

Sucrose acetate isobutyrate (SAIB), a randomly substituted sucrose approximating sucrose diacetate hexaisobutyrate, is produced by Eastman Chemical Company for a variety of applications. SAIB is widely used in the food industry as a weighting agent to disperse flavoring oils in primarily citrus-based soft drink beverages. Additionally, SAIB is currently being marketed by another company as a parenteral drug delivery system. The studies reported here focused on investigating SAIB as an excipient, or delivery vehicle, for use in oral delivery of several drugs, including ibuprofen, saquinavir, and clarithromycin. Dissolution experiments were conducted using both ibuprofen and caffeine, and results suggest that SAIB can be used in dosage forms to control release rate. Pharmacokinetic studies in which laboratory rats were dosed with formulations containing drugs such as ibuprofen, saquinavir, and clarithromycin suggest that SAIB may act to reduce animal-to-animal variability in drug concentration profiles in some cases, and that it may also enhance gastroretention of the dosage forms. Finally, dosage form imaging studies suggest but do not reliably confirm that SAIB may aid in promoting gastric retention, which would make its use in dosage form formulation beneficial for administration of drugs whose action is intended to occur in the stomach.

DEDICATION

*To my late Father,
One of NASA's early explorers in the earth, atmospheric, and space sciences and
A pioneer in advancing international cooperation in space; and
To my Mother, who,
both figuratively and literally, kept the home fires burning during all the times he was away.
He kindled my desire to pursue a scientific and engineering career.
She made me believe that I could accomplish anything I should choose to do.*



NASA/DOD Scout Launch Vehicle (1960-1994)

ACKNOWLEDGEMENTS

Many people have contributed to this work both at Quillen College of Medicine and at Eastman Chemical Company. I wish to thank my major professor Dr. Tom Ecay for agreeing to take on a graduate student in his second 50 years of life and for all the help he has given me along the way. I also wish to thank Dr. Sador Black, director of my division at Eastman when I began this program and the person who made pursuing this degree possible while still working at Eastman. Mr. James Michalski of Eastman was also a prominent player in securing support for the research. Dr. Kevin Edgar of Eastman, now of Virginia Tech, was very involved in originating this particular research project. I also wish to thank my laboratory heads, Mr. Jeff Smith and Mr. Brad Snow, for their support and for allowing me to make up my lost work time by arriving early and working late. Finally, Dr. Stan Polichnowski of Eastman offered his support of this work by signing the research agreement and I wish to thank him.

People who played prominent roles in the research reported herein include my major professor Dr. Tom Ecay of the Department of Physiology, Dr. Peter Rice of the Department of Pharmacology and now at the College of Pharmacy at the University of Colorado, Dr. Michael Wempe of Eastman, now also of the College of Pharmacy at the University of Colorado, and Mr. James Little of Eastman. Dr. Jinghua Yuan and Ms. Nancy Clipse of Eastman were very helpful in the dissolution study and allowed me to work in their laboratory. As a member of my committee, Dr. Kenneth Ferslew of the Department of Pharmacology was involved in many of the discussions concerning the research and was prominent in my pharmacokinetics education. Ms. Janet Lightner was very helpful to me in Dr. Rice's laboratory. Finally, Dr. Gregory Hanley and his staff of the Department of Animal Resources provided much help to me, especially in performing the intravenous dosing. Dr. Mitchell Robinson has always been there to help things along and I thank him for serving on my committee as well. Ms. Beverly Sherwood, who knows everything about the program, has been very helpful in keeping me on track with all the requirements of the graduate school.

Having received my undergraduate and graduate training in chemistry and chemical engineering, respectively, learning about the biomedical sciences has been a challenging and very rewarding experience for me. When I began this program, I had not taken a biology course since freshman biology 30 years before. The quality teaching that exists among the faculty at Quillen College of Medicine has made learning a pleasant and fascinating experience.

CONTENTS

	Page
ABSTRACT	2
DEDICATION	3
ACKNOWLEDGEMENTS	4
LIST OF TABLES	7
LIST OF FIGURES	8
Chapter	
1. INTRODUCTION	11
2. METHODS AND MATERIALS	19
Dissolution Experiments	19
Ibuprofen	19
Caffeine	22
Pharmacokinetic Experiments	24
Ibuprofen	25
Saquinavir	25
Clarithromycin	26
Dosage Form Imaging Studies in Live Animals	26
Statistical Analysis	28
3. RESULTS	29
Dissolution Experiments	29
Ibuprofen	29
Caffeine	31
Pharmacokinetic Experiments	33
Ibuprofen	33
Saquinavir	40

Clarithromycin	49
Dosage Form Imaging Studies in Live Animals	57
4. DISCUSSION	64
Dissolution Experiments	64
Ibuprofen	64
Caffeine	65
Pharmacokinetic Experiments	66
Ibuprofen	66
Saquinavir	67
Clarithromycin	68
X-Ray Imaging Experiments	69
5. CONCLUSIONS	71
REFERENCES	73
VITA	79

LIST OF TABLES

Table		Page
1.	Pharmacokinetic Parameters for Rats (Normal Diet) Dosed Intravenously with Ibuprofen	38
2.	Kinetic Parameters for Rats (Normal Diet) Dosed Orally with Ibuprofen	38
3.	Kinetic Parameters for Rats (SAIB Diet) Dosed Orally with Ibuprofen	38

LIST OF FIGURES

Figure	Page
1. Dynamic relationship between the drug, dosage form, and pharmacologic effect	11
2. The USP-1 Dissolution Apparatus of the U.S. Pharmacopeia containing buffer solution and a rotating basket containing the dosage form.....	20
3. Absorption spectra for 6.8 pH buffer, standard solutions of ibuprofen in 6.8 pH buffer, and a capsule dissolved in the buffer.....	21
4. Calibration plot for analysis of ibuprofen during dissolution in 6.8 pH buffer.....	22
5. Calibration plot for analysis of caffeine during dissolution in 1.2 pH buffer.....	24
6. Standard curve of clarithromycin.....	27
7. Standard curve of decladinose clarithromycin.....	27
8. Absorbance at 266nm minus absorbance at 280nm during the dissolution in 6.8 pH buffer for several dosage forms	30
9. Concentration profiles vs. time for dissolution of ibuprofen in 6.8 pH buffer from several different dosage forms.....	30
10. Absorbance at 275 nm vs. time for dissolution of caffeine in 1.2 pH buffer from two different dosage forms – one containing SAIB and one without.....	31
11. Concentration-time dependence during dissolution of dosage forms in 1.2 pH buffer.....	32
12. Ibuprofen plasma concentration profiles for rats fed a normal diet and dosed intravenously.....	35
13. Ibuprofen plasma concentration profiles for rats fed a normal diet and dosed orally.....	36
14. Ibuprofen plasma concentration profiles for rats fed a diet consisting of and rat chow containing 4 wt% SAIB and dosed orally.....	37
15. Results of statistical analysis of C(0), A, and B for ibuprofen obtained using the open one-compartment model for (1) rats fed a normal diet and dosed intravenously and (2) rats fed a normal diet and dosed orally, and (3) rats fed a diet containing 4 wt% SAIB and dosed orally.....	39

16. Results of statistical analysis of elimination rate constant, K_{el} , for ibuprofen obtained using the open one-compartment model for (1) rats fed a normal diet and dosed intravenously and (2) rats fed a normal diet and dosed orally, and (3) rats fed a diet containing 4 wt% SAIB and dosed orally.....	41
17. Results of statistical analysis of absorption rate constant, K_a , for ibuprofen obtained using the open one-compartment model for rats dosed orally and (1) fed a normal diet or (2) fed a diet containing 4 wt% SAIB and dosed orally	42
18. Results of statistical analysis of areas under the ibuprofen plasma concentration-time curve obtained using the open one-compartment model for (1) rats fed a normal diet and dosed intravenously and (2) rats fed a normal diet and dosed orally, and (3) rats fed a diet containing 4 wt% SAIB and dosed orally.....	43
19. Saquinavir concentration profiles for the six rats dosed with saquinavir/CMC dosage forms.....	44
20. Saquinavir concentration profiles for the six rats dosed with saquinavir/CMC/SAIB dosage forms.....	45
21. Aggregate plots showing: (a) the concentration of saquinavir for rats dosed with saquinavir/CMC dosage forms and (b) the concentration of saquinavir for rats dosed with saquinavir/CMC/SAIB dosage forms.....	46
22. Saquinavir mean concentration profiles for the six rats dosed with either saquinavir/CMC or saquinavir/CMC/SAIB dosage forms.....	47
23. Results of statistical analysis of areas under the saquinavir plasma concentration-time curve for rats dosed either (1) saquinavir/CMC or (2) saquinavir/CMC/SAIB.....	48
24. Acid catalyzed hydrolysis of clarithromycin to decladinose clarithromycin.....	49
25. Clarithromycin concentration-time profiles for rats dosed orally with and without SAIB ($n = 3 \pm SEM$).....	50
26. Decladinose clarithromycin plasma concentration-time profiles for rats dosed orally with clarithromycin both with and without SAIB ($n = 3 \pm SEM$).....	50
27. Results of statistical analysis of areas under the clarithromycin plasma concentration-time curves for rats does with either (1) clarithromycin/CMC or (2) clarithromycin/CMC/SAIB.....	51

28.	Results of statistical analysis of areas under the decladinose clarithromycin plasma concentration-time curves for rats does with either (1) clarithromycin/CMC or (2) clarithromycin/CMC/SAIB.....	52
29.	<i>In vivo</i> metabolism from liver microsomal incubations.....	53
30.	Demethyl clarithromycin concentration-time profiles for rats dosed orally with clarithromycin both with and without SAIB (n = 3 ± SD).....	54
31.	Hydroxy-clarithromycin concentration-time profiles for rats dosed orally with clarithromycin both with and without SAIB (n = 3 ± SD).....	54
32.	Results of statistical analysis of areas under the demethyl clarithromycin plasma concentration-time curves for rats does with either (1) clarithromycin/CMC or (2) clarithromycin/CMC/SAIB.....	55
33.	Results of statistical analysis of areas under the hydroxy clarithromycin plasma concentration-time curves for rats does with either (1) clarithromycin/CMC or (2) clarithromycin/CMC/SAIB.....	56
34.	X-ray images showing the BaSO ₄ /CMC dosage form (top) and BaSO ₄ /CMC/SAIB (bottom) <i>in vivo</i> as a function of time following oral dosing.....	58

CHAPTER 1

INTRODUCTION

The general goal of oral drug administration is to simply, safely, and effectively deliver a drug to the target tissue, or biospace, within the body at the concentration and for the duration required to achieve the desired clinical or pharmacologic effect. The rate and extent to which the drug of interest reaches the biospace is a direct result of (1) the kinetics of the following processes: liberation from the dosage form; dissolution; absorption into systemic circulation; distribution to the target tissue; metabolism; and excretion; and (2) the dose size and frequency of dosing. The study of the rates of the processes given in (1) above encompasses the sciences of *biopharmaceutics* and *pharmacokinetics* (Van de Waterbeemd et al. 2003). Biopharmaceutics concerns the physicochemical properties of the drug and dosage form and involves such factors as (1) the stability of the drug within the dosage form, (2) the release of the drug from the dosage form, (3) the rate of dissolution of the drug, and (4) the systemic absorption of the drug (Shargel et al. 2005). Pharmacokinetics involves the study of the rates of absorption, distribution, metabolism, and excretion, as well as the interrelationships of these complex and dynamic processes (Ritschel et al. 2004). *Pharmacodynamics*, on the other hand, describes the rate at which the drug causes the pharmacologic effect once it has reached the biospace in sufficient concentration to induce the effect (Tozer and Rowland 2006). One way to explain the difference between pharmacokinetics and pharmacodynamics is that pharmacokinetics describes the effect of the body on the drug while pharmacodynamics describes the effect of the drug on the body.

The LADMER (liberation, absorption, distribution, metabolism, excretion, and response) acronym is often used to define the processes of interest in oral drug delivery (Shargel et al. 2005). The schematic diagram given in Figure 1 illustrates in a simplified way the dynamic

interrelationships between these complex processes and shows the general boundaries between the sciences of biopharmaceutics, pharmacokinetics, and pharmacodynamics. From a physical, chemical, and mathematical understanding of the biopharmaceutics and pharmacokinetics of a particular drug dosage form, as well as the pharmacodynamics of the specific drug, one can predict the dose and dosing frequency required to maintain the drug level needed in the biospace that will result in the desired clinical or pharmacologic effect.

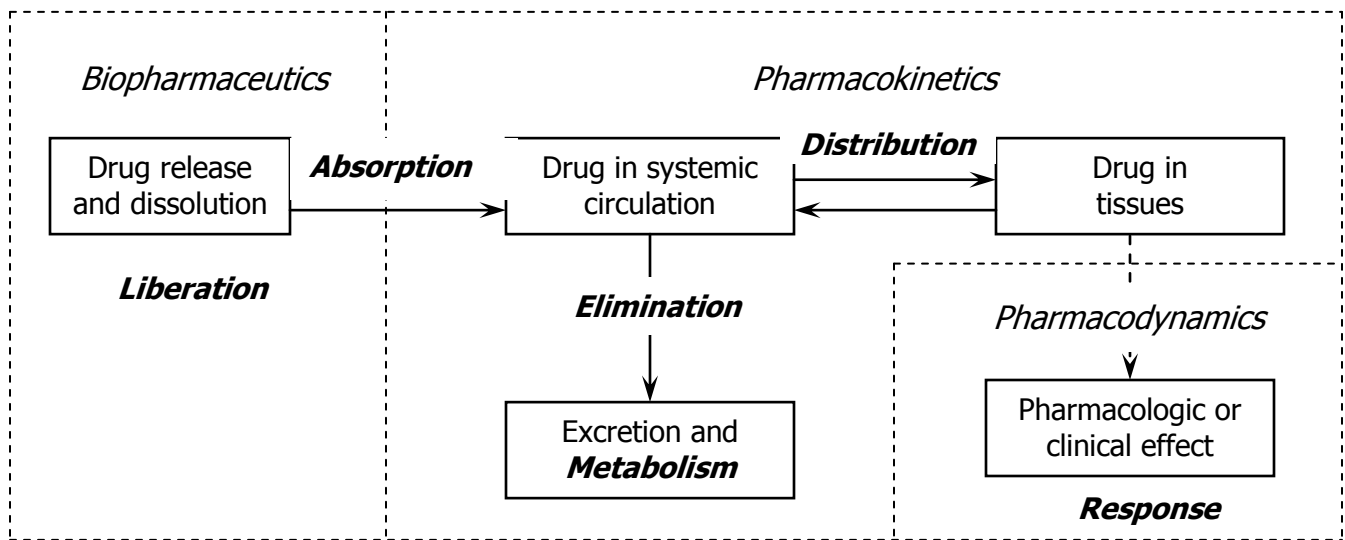


Figure 1. Dynamic relationship between the drug, dosage form, and pharmacologic effect (adapted from Shargel et al. 2005)

The general goal of pharmacokinetics is to describe these processes mathematically from the point of drug dissolution in the intestinal lumen to systemic circulation. Because elimination, which occurs by both metabolism and excretion, and distribution of the drug between the blood and tissue are continuous, these processes must be considered in addition to absorption. Basically, there are 3 different classes of approaches (Fournier 2007) for

mathematically describing the time-dependent plasma concentration of a drug administered to the body: (1) the compartmental approach, (2) the physiological approach, and (3) the model-independent approach. The compartmental approach is now very highly developed and is quite well described within the texts by Ritschel and Kearns (2004) and by Shargel et al. (2005). More recent approaches to quantitatively modeling pharmacokinetic processes have been physiologically based. In these approaches the processes of the different physiological systems are modeled mathematically, often requiring the use of higher mathematics than is required for more simplistic approaches to pharmacokinetics. The field of physiologically based pharmacokinetic modeling is currently evolving (Mancheras and Iliadis 2005, Reddy et al. 2006). The third approach for modeling pharmacokinetic data can be classified as phenomenological, as no attempt is made to describe the actual processes involved – only the resulting data. All of the pharmacokinetic modeling performed in the present work follows the compartmental approach.

Sucrose acetate isobutyrate (SAIB), a randomly substituted mixture of sucrose esters approximating sucrose diacetate hexaisobutyrate, is produced by Eastman Chemical Company (Kingsport, TN) under the trademark Sustain™. It is available in a variety of compositions for different applications. The most common application of SAIB is that of a weighting agent to disperse flavoring oils in primarily citrus-based soft drink beverages. SAIB acts as a density-adjusting agent to prevent separation and to maintain a cloudy dispersion throughout the beverage product, and is approved as a food additive in more than 40 countries. Due to the potential risk of acute and chronic exposure to this compound by humans, extensive toxicology studies using various animal subjects, including humans, were conducted beginning in the early 1960s. Many of these are summarized in 2 reviews published in 1998 in the journal *Food and Chemical Toxicology*. Here, Reynolds and Chappel (1998) reviewed toxicity studies published

prior to 1988, while Reynolds (1998) reviewed studies of the metabolism and pharmacokinetics of SAIB in rats, dogs, monkeys and humans. In the same issue appear articles concerning subchronic toxicity of SAIB in rats and dogs (Proctor and Chappel 1998), toxicity studies in the cynomolgus monkey (Blair and Chappel 1998), toxicity and carcinogenicity studies in the rat and the mouse (Mackenzie et al. 1998a), reproductive studies in the rat and teratology studies in the rat and rabbit (Mackenzie et al. 1998b), the effect of SAIB on the hepatobiliary function of humans (Chiang et al. 1998), and, finally, the genotoxic effects of SAIB (Myhr et al. 1998).

In the early 1970s, Reynolds and Chappel (1998) confirmed the finding of Morgareidge (1965) that ingestion of SAIB by dogs results in an increase in the weight of the liver and serum alkaline phosphatase levels resulting from interference of SAIB with biliary excretion. Further studies in dogs, which were reviewed by Proctor and Chappel (1998), suggest that effects of SAIB on the hepatobiliary system of the dog can appear after only one low dose. However, this effect is rapidly reversible following cessation of SAIB ingestion, suggesting that it is likely to be a pharmacological effect rather than a toxic one. Similar effects were not found in rats and monkeys.

The results of the toxicity studies of Blair and Chappel (1998) on the cynomolgous monkey showed that animals ingesting up to 2400 mg/kg body weight of SAIB per day showed no signs of specific toxicity, including a lack of the effects on the hepatobiliary function observed in dogs.

Hensley (1975) and Orr et al. (1976) conducted studies on humans. Hensley (1975) dosed human volunteers at a level of 10 mg/kg body weight per day for 14 days, while Orr et al. (1976) dosed a different sample of human volunteers at levels of 7 and 20 mg/kg body weight per day for the same time period. The 20 mg/kg per day dosage regimen is 4 times greater than the lowest dosage used in dogs which resulted in increases in BSP

(bromosulfophthalein) and ICG (indocyanine green). Chiang (1998) conducted further studies on the effect of SAIB on the hepatobiliary function of humans. These studies suggest that daily SAIB dosages of 20 mg/kg body weight in humans do not result in changes in biliary function as in the case of dogs.

Myhr et al. (1998) conducted a series of different *in vitro* genotoxicity assays that examined the interaction of SAIB with genetic material from metabolically competent liver cells at concentrations of 0.024 to 1000 µg/ml. These tests, which are capable of detecting point mutations, chromosomal damage, and the induction of DNA lesions, were negative, indicating that SAIB has no damaging effects on DNA. These workers concluded that, because of these negative results, and the fact that Tennant et al. (1987) had previously established that these *in vitro* genotoxicity assays are sensitive tests for potential carcinogenicity *in vivo*, SAIB is therefore not genotoxic and should not contribute to the dietary intake of mutagens.

Mackenzie et al. (1998) conducted a 3-generation reproductive and teratogenicity study of Fischer 344 rats receiving oral dosages of SAIB up to 2.0 g/kg for 10 weeks. A separate teratogenicity study was conducted on New Zealand white rabbits at oral dosages of up to 1.2 g/kg body weight. These workers concluded that, even at these high doses of SAIB, there were no observed effects in either case.

In a series of patents, Southern Biosystems made claims for inventions using SAIB as a parenteral depot delivery system under the trademark SABER™ (sucrose acetate isobutyrate – extended release), currently marketed by Durect (Tipton and Ewing 1996; Tipton and Holl 1996; Gibson et al. 1999). SABER™ can be classified as a biodegradable *in situ*-forming depot (ISFD) system (Tipton and Dunn 2000; Hatefi and Amsden 2002). Sullivan et al. (1998) tested a SABER system for release of progesterone and estradiol in seasonally anovulatory mares. Genentech, Inc. made claim for a specific formulation for parenteral dosing of growth

hormone, specifically human growth hormone (Okumu 2001). In 2002, Hatefi and Amsden and then Matschke et al. published reviews of injectable dosage forms that act as in situ forming drug delivery or depot delivery systems. Tipton (2003) published a review of the use of SAIB as an in situ forming drug delivery system.

Because SAIB is a mixed ester, it is not crystallizable and thus exists as an extremely highly viscous liquid. SAIB is very difficult to handle in its neat form due to both its high viscosity (>100,000 cP at 25°C) and high degree of tackiness. However, a 90%/10% SAIB/ethanol solution possesses such a dramatically lower viscosity (770 cP at 25°C) that it is injectable. In the SABER™ system, the drug of interest is dissolved in the low-viscosity SAIB-ethanol solution and injected into tissue. Upon injection, the ethanol rapidly diffuses away, leaving the high-viscosity, hydrophobic SAIB *in situ* depot containing the drug. The drug then diffuses out of the SAIB and into the surrounding tissue and the SAIB depot is slowly metabolized. As a result of that work, the question arose as to whether or not SAIB has potential as an excipient for oral drug delivery.

The absorption of most drugs delivered orally occurs primarily in the small intestine and the rate of absorption is dependent upon both the solubility and the permeability of the drug. Solubility might be enhanced by chemical means, e.g. by using a cosolvent that acts to solubilize the drug in the gastrointestinal environment (Li et al. 1999). The effective permeability of the drug through the gut wall may be enhanced by decreasing the degree of ionization of the drug (Palm et al. 1999; Kobate et al. 2008), by blocking metabolizing enzymes (Bai et al. 1996; Yusuf et al. 2000), or by blocking transporters that extrude the drug back into the intestinal lumen (Huisman et al. 2001,2003; Constantinides and Wasan 2006; Collnot et al. 2007). Thus the possibility exists that any enhancement of absorption of a drug in an SAIB delivery system could be due to effects on any of these processes. Effects on drug liberation

and solubility are easily tested, while effects on transport or metabolism require specific *in vitro* testing such as Caco-2 transport studies and microsomal metabolic studies (Tucker et al. 2001; Cummins et al. 2003). Identification of particular transporters and/or enzymes involved may be accomplished using recombinant transporters/enzymes or microsomal systems that differentially express the proteins of interest (Crespi et al. 2000).

An additional way that drug absorption might be enhanced is by using gastroretentive dosage forms (GRDFs) that prolong the lifetime and integrity of the delivery system within the stomach (Chawla et al. 2003; Hoffman et al. 2004; Streubel et al. 2006). This approach has been shown to be particularly effective for drugs having a narrow distribution window, such drugs being well absorbed in the proximal small intestine but to a lesser extent in the distal. Examples of such drugs are levodopa, metformin, furosemide, and ciprofloxacin. GRDFs have been shown to be effective for furosemide and ciprofloxacin, e.g. Depomed's RFD/ciprofloxacin recently marketed drug product, Ciprofloxacin GR™, for treatment of urinary infections. Gastroretentive dosage forms may also be the desirable delivery approach for certain drugs whose action is intended to occur, not in tissue, but within the gastric environment, e.g. the treatment of *Helicobacter pylori* in the stomach using clarithromycin (Conway et al. 2005; Bardonnet et al. 2006). In this latter case, gastric retention extends the duration of exposure of the drug to the bacteria in their gastric habitat and thus the exposure time of the bacteria to the minimum inhibitory concentration of antibiotic. Such an increase in the duration of exposure reduces frequency of dosing, improves compliance (fewer dosages), and greatly enhances the probability of eradication.

In this work, I hypothesized that sucrose acetate isobutyrate may enhance the absorption of some drugs and that this enhancement might occur by one or more of the following mechanisms: (1) modification of drug liberation from the dosage form, (2)

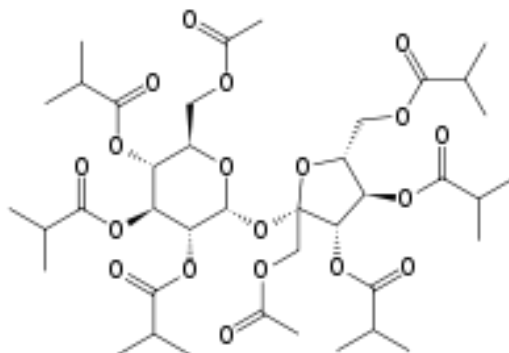
enhancement of drug solubility, (3) gastroretention of the dosage form, or (4) modification of the absorption mechanism. The question regarding mechanisms (1) and (2) above were addressed using dissolution studies and mechanisms (3) and (4) were addressed through pharmacokinetic studies and dosage form imaging studies in live animals. These studies reported here were directed at determining whether or not SAIB might be a useful excipient for oral drug delivery and, if so, which of the mechanisms does it affect.

CHAPTER 2
METHODS AND MATERIALS

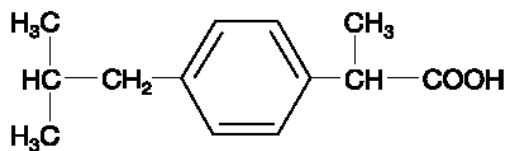
Dissolution Experiments

Ibuprofen

Sucrose acetate isobutyrate (SAIB), whose structure is



was supplied by Eastman Chemical Company in the form of Sustane™ SAIB ET-10 containing approximately 90 wt% SAIB and 10 wt% ethanol. As mentioned in the previous chapter, this particular form of SAIB was used because of its lower viscosity and ease of handling. In addition, ibuprofen, being soluble in ethanol, is easily dissolved in this system. Ibuprofen, having the chemical structure



was supplied in crystalline form by the Performance Chemicals and Intermediates Business Organization of Eastman Chemical Company.

Ibuprofen was initially dissolved in the Sustane™ SAIB ET-10 at 12.5 wt%. At this concentration the SAIB ET-10 was nearly saturated. Higher concentrations were made by adding additional ethanol to the mixture to solubilize the ibuprofen. Two additional compositions were then made: one containing 16.8% ibuprofen in a system of 82.6% SAIB/17.4% EtOH and 22.6% ibuprofen in a system of 79% SAIB/21% EtOH. Both solutions were essentially saturated with ibuprofen. Amounts of these 3 different Ibuprofen/SAIB/ EtOH solutions were then transferred to capsules for dissolution testing using a USP-1 dissolution apparatus (illustrated in Figure 1), specifically a Distek Dissolution System, in a 6.8 pH buffered solution. Exact amounts were measured to give a concentration of 50.0 ppm when fully dissolved in the dissolution apparatus. Three additional capsules were prepared: one empty capsule, one capsule containing only ibuprofen, and one capsule containing only SAIB. The capsules were placed in separate baskets and simultaneously immersed in different baths containing 6.8 pH buffer maintained at 37°C. The baskets were rotated at 50 rpm and 10 mL aliquots were removed at specific time intervals during the dissolution process.

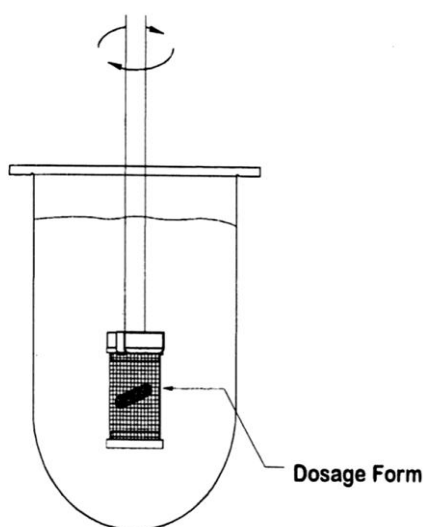


Figure 2. The USP-1 Dissolution Apparatus of the U.S. Pharmacopeia containing buffer solution and a rotating basket containing the dosage form

The samples were then analyzed immediately after collection using a Hewlett-Packard 8452A Diode Array Spectrophotometer. The analysis followed the procedure for ibuprofen outlined in the U.S. Pharmacopeia, which stipulates that the absorption at 280 nm be subtracted from the absorption at 266 nm. Clearly from Figure 3 below, one can see that the absorption at 266nm varies greatly with concentration while that at 280nm is much less a function of concentration. Plotting the difference in absorption at these two wavelengths vs. concentration gives a linear relationship as shown in Figure 4.

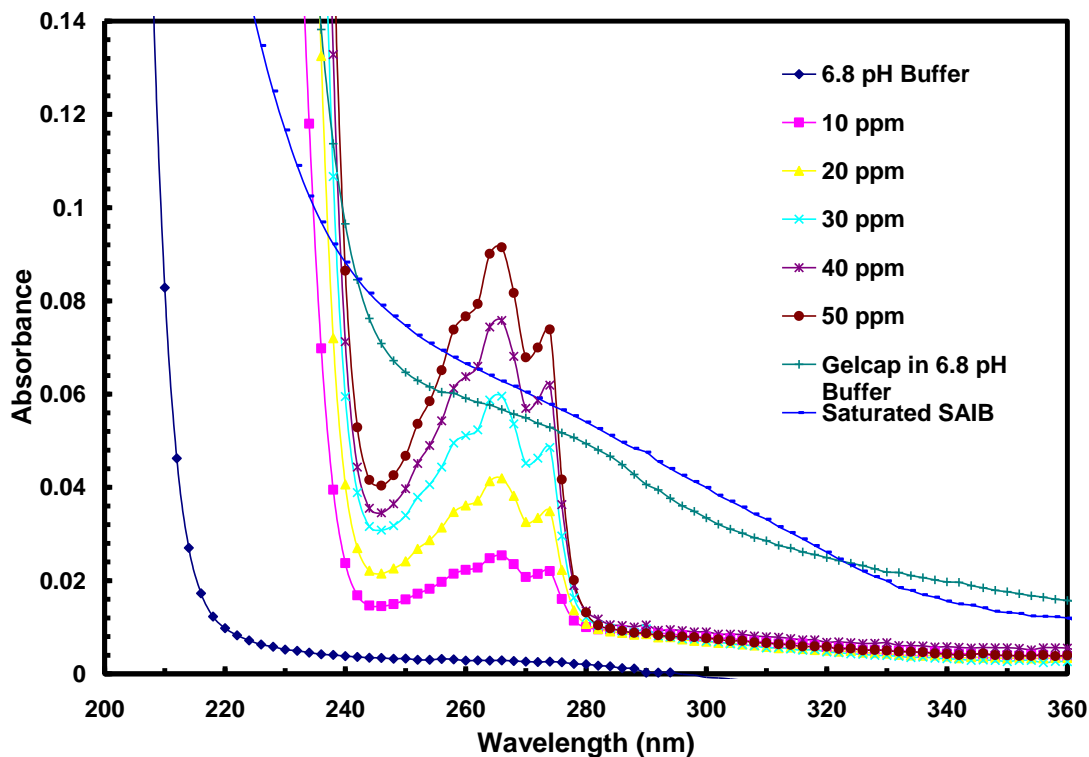


Figure 3. Absorption spectra for 6.8 pH buffer, standard solutions of ibuprofen in 6.8 pH buffer, and a capsule dissolved in the buffer

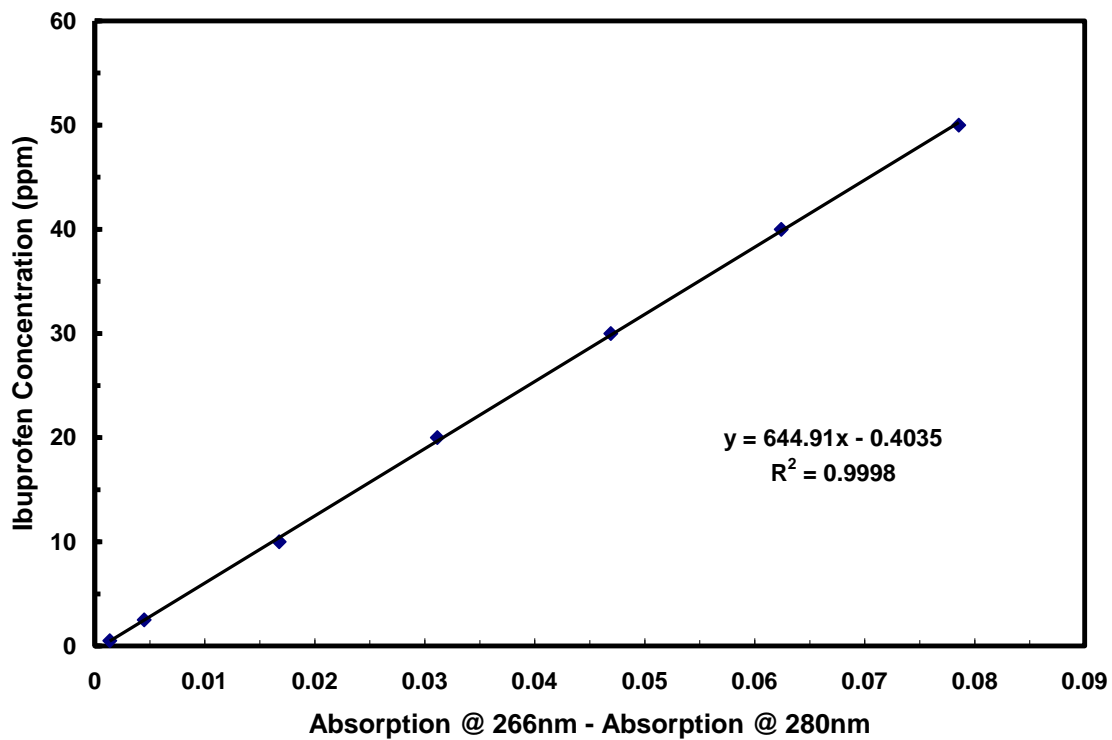
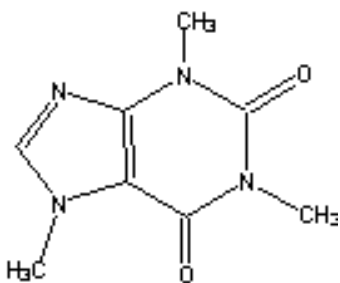


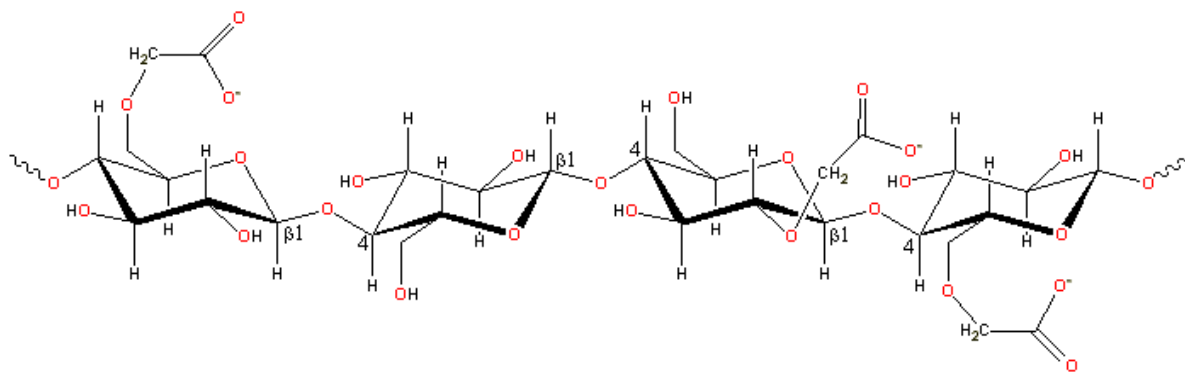
Figure 4. Calibration plot for analysis of ibuprofen during dissolution in 6.8 pH buffer

Caffeine

Caffeine, having the chemical structure



and carboxymethylcellulose (CMC), i.e.



were compounded together at a 25/75 weight ratio and caffeine/CMC/SAIB (not including ethanol) formulations were compounded at a 25/25/50 weight ratio. Ethanol was added to the formulation to facilitate mixing. Because SAIB ET-10 was used to prepare these formulations, calculations of weights to be used had to include allowance for the additional ethanol present. The formulations were then lyophilized overnight to remove ethanol and were reweighed to determine the exact composition and the amount of ethanol lost. They were then packed into size 9 porcine hard gelatin capsules, obtained from Torpac, for the dissolution study.

For the dissolution experiments, a 10 mL vial containing 9 mL of buffer solution was placed into a reciprocating water bath at 37°C for 5 minutes to reach temperature. A capsule was then placed into the vial and the bath was cycled at 60 cycles/min. Then 0.5 mL aliquots were removed at specific time increments, filtered, and stored. 0.5 mL of buffer was then replaced into the vial and it was returned to the reciprocating water bath. Following the dissolution and sample collection procedure, 200 μ L were removed from the solution taken at each time increment, transferred to a UV 96-well plate, and analyzed at 275 nm. A calibration curve, shown in Figure 5, was constructed and used to determine concentration as a function of the time following introduction of the capsule to the dissolution vial.

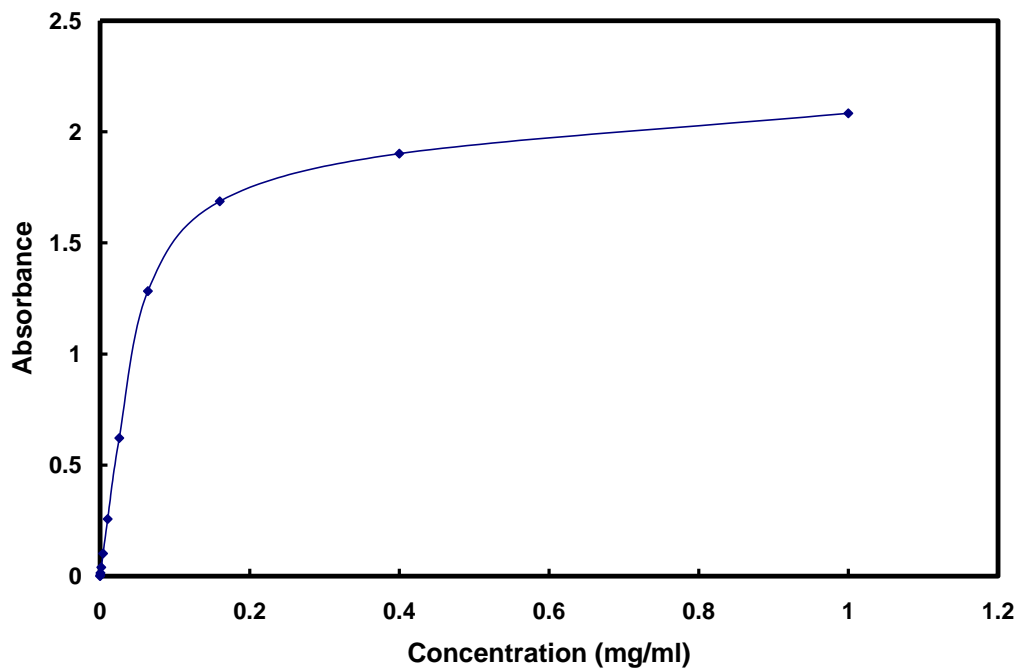


Figure 5. Calibration plot for analysis of caffeine during dissolution in 1.2 pH buffer (absorbance at 275nm)

Pharmacokinetic Experiments

Male Sprague-Dawley rats were used for all pharmacokinetic studies. The animals were housed in temperature-controlled rooms with, generally, three rats to a cage, having controlled 12-hr light and dark cycles in the ETSU Division of Laboratory Animal Resources on the College of Medicine campus. Water was provided during the entire period. Some studies included a 12-hr fasting period prior to dosing while some did not, the specifics of which are described below. All studies were reviewed and approved by the ETSU Committee on Animal Care.

Ibuprofen

For the pharmacokinetic (PK) studies with ibuprofen, SAIB was not formulated into the dosage form. For this initial PK study on rats, Sustain™ SAIB MCT containing 80 wt% SAIB and 20 wt% medium chain triglycerides was manually compounded into standard rat chow. Eighteen male Sprague-Dawley rats were included in the study and the rats were divided into 3 groups of 6 rats each. Group I and II rats were fed a normal diet of rat chow until and throughout dosing and blood collection. Group III rats were fed a diet of ground rat chow containing 4 wt% SAIB and 1 wt% MCT for 48 hrs before dosing and throughout the blood collection. Although animals are typically fasted prior to commencement of PK studies, these animals were not fasted as it was desired to determine the effect of diet (SAIB vs. no SAIB) on the pharmacokinetics of ibuprofen. Group I rats were dosed intravenously at the beginning of the pharmacokinetic experiment and Group II and Group III rats were dosed orally at about 10 mg/Kg as 0.45 mL. Approximately 125 µL of blood were collected into mini-capillary collection tubes containing EDTA dipotassium salt (SAFE-T FILL®; RAM Scientific Inc., Yonkers, NY USA), placed on dry ice, and kept frozen until analysis. Samples were analyzed by HPLC/MS by Mr. James L. Little at Eastman Chemical Company and data were analyzed using a GraphPad Prism, Version 5.0 software package.

Saquinavir

Two different saquinavir formulations were made for oral dosing: (1) a formulation containing 25 wt% saquinavir and 75 wt% CMC and (2) a formulation containing 25 wt% saquinavir, 25 wt% CMC, and 50 wt% SAIB. Animals were fasted beginning 12 hours prior to commencement of the pharmacokinetic study. Blood samples were collected in the same manner as for the ibuprofen study described earlier, and samples were analyzed for saquinavir

by Dr. Michael Wempe by HPLC/MS/MS at Eastman Chemical Company, Kingsport, Tennessee. Data were analyzed using the GraphPad Prism 4.0, Version 4 software package.

Clarithromycin

Two different clarithromycin formulations were made: (1) a formulation containing about 15 wt% clarithromycin and 85 wt% CMC and (2) a formulation containing about 20 wt% clarithromycin, 40 wt% CMC, and 40 wt% SAIB. Pharmacokinetic studies were conducted as described for saquinavir and samples were analyzed *via* LC/MS/MS by Dr. Michael Wempe for clarithromycin, decladinose clarithromycin (product of acid-catalyzed hydrolysis in the stomach), and the metabolites demethyl clarithromycin and hydroxyl clarithromycin. Standard curves for clarithromycin and decladinose clarithromycin are shown in Figures 6 and 7, respectively. Data were analyzed using the GraphPad Prism 4.0, Version 4 software package.

Dosage Form Imaging Studies in Live Animals

In order to determine if dosage forms containing SAIB persist in the gastrointestinal tract for longer periods of time thus leading to extended release of the drug *in vivo*, formulations containing (1) 25 wt% barium sulfate and 75 wt% CMC and (2) 25 wt% barium sulfate, 25 wt% CMC, and 50 wt% SAIB were made and packed into capsules in the same manner as for the pharmacokinetic studies. Animals were orally dosed, restrained, and then X-rayed in the animal facility at ETSU as a function of time following dosing. Images were collected using a Pinnacle Systems Dazzle imaging board and a personal computer and were visually compared to determine qualitative differences in rate of dispersion of the dosage form.

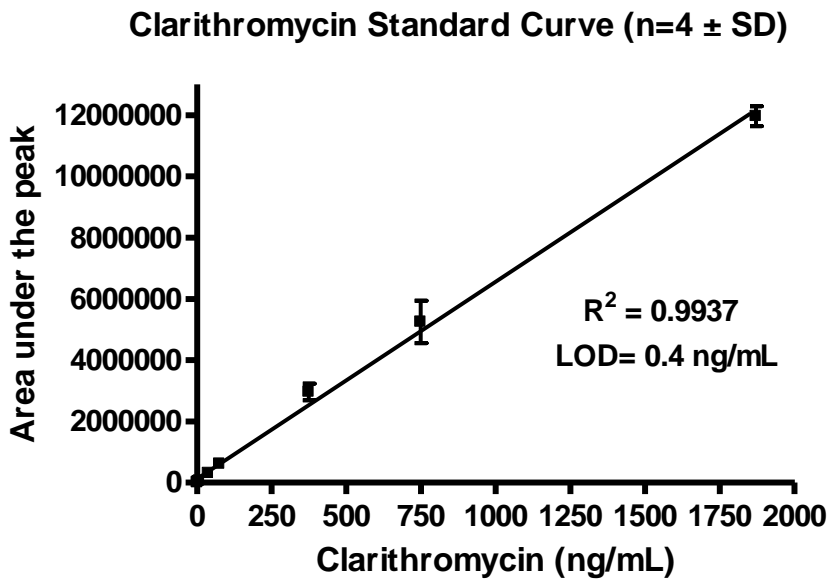


Figure 6. Standard curve of clarithromycin

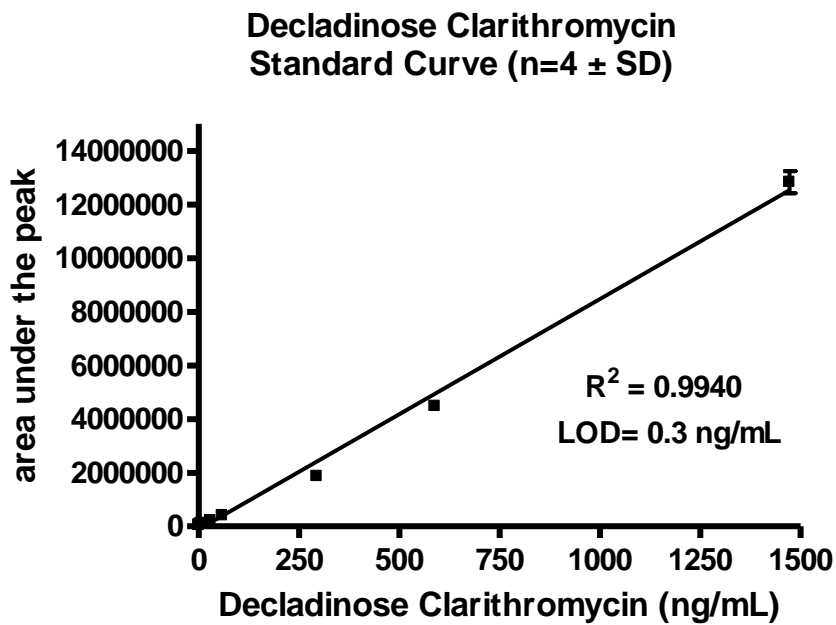


Figure 7. Standard curve of decladinose clarithromycin. LOD = limit of detection.

Statistical Analysis

Statistical analysis of the data for this work was accomplished using the JMP® statistical software package. A one-way analysis of variance (ANOVA) was used to compare groups that received different types of a dosage form containing a particular drug. Tukey-Kramer HSD (Honestly Significant Difference) comparisons were used to compare groups when the ANOVA was inconclusive. In cases where groups being compared had large differences in variability, we used a variance-stabilizing transformation to account for this variability and comparisons were made of the transformation to ensure that conclusions reached were not simply based on noise in the data.

CHAPTER 3

RESULTS

Dissolution Experiments

Ibuprofen

Ibuprofen was chosen as a model drug to begin investigating how sucrose acetate isobutyrate affects the release and dissolution kinetics of a drug when compounded into an oral dosage form. For these initial studies we used an SAIB/ethanol mixture without removing the ethanol from the dosage form. It is well known that when such a system is injected parenterally, the ethanol rapidly diffuses into surrounding tissue leaving the drug dissolved within the SAIB (Tipton 2003). The result is a depot delivery system from which the drug slowly diffuses out into the surrounding tissue. As described in the previous chapter, dissolution experiments for ibuprofen in 6.8 pH buffer involved measuring the absorption at 266 and 280nm and subtracting the latter from the former. These experiments resulted in the plots shown in Figure 8. Using the calibration curve from Figure 4 and accounting for withdrawal of buffer without replacement and for the presence of gelcap and/or SAIB, the concentration-time curves shown in Figure 9 were obtained.

Each of these dosage forms contained the same amount of ibuprofen – enough to give a 50.0 ppm solution of ibuprofen at full dissolution. Clearly the dissolution of the dosage form containing only ibuprofen in a gelcap reached 50.0 ppm at long times. The other 3, in which ibuprofen was dissolved in various compositions of SAIB and ethanol, did not, suggesting that not all of the ibuprofen was released from the dosage form even after about 3 days. These results will be discussed more thoroughly in Chapter 4.

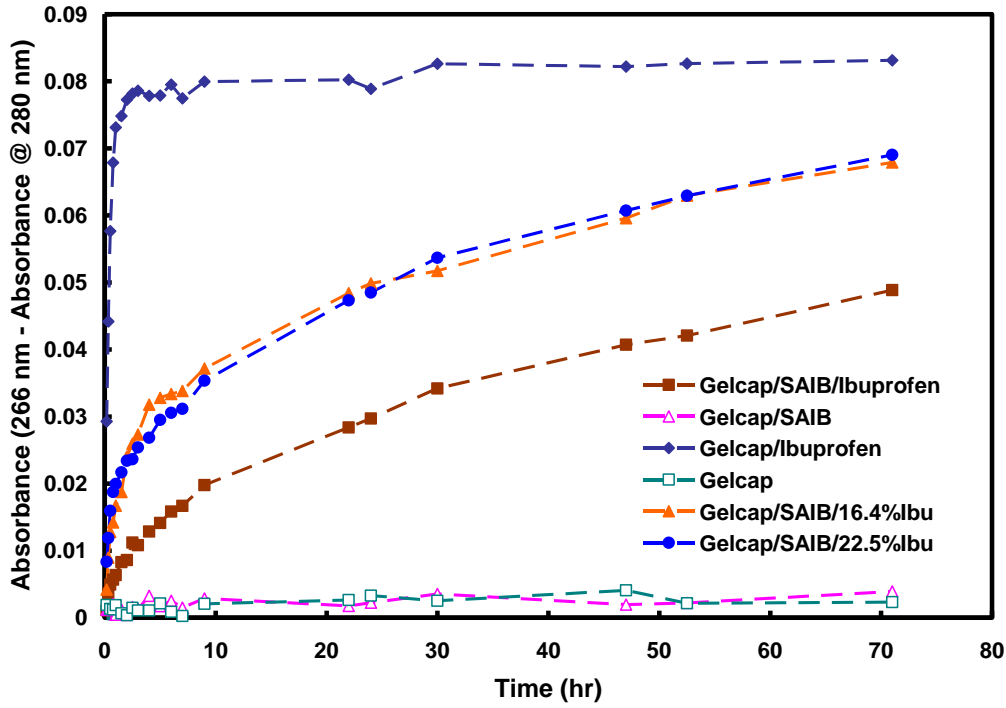


Figure 8. Absorbance at 266nm minus absorbance at 280nm during the dissolution in 6.8 pH buffer for several dosage forms

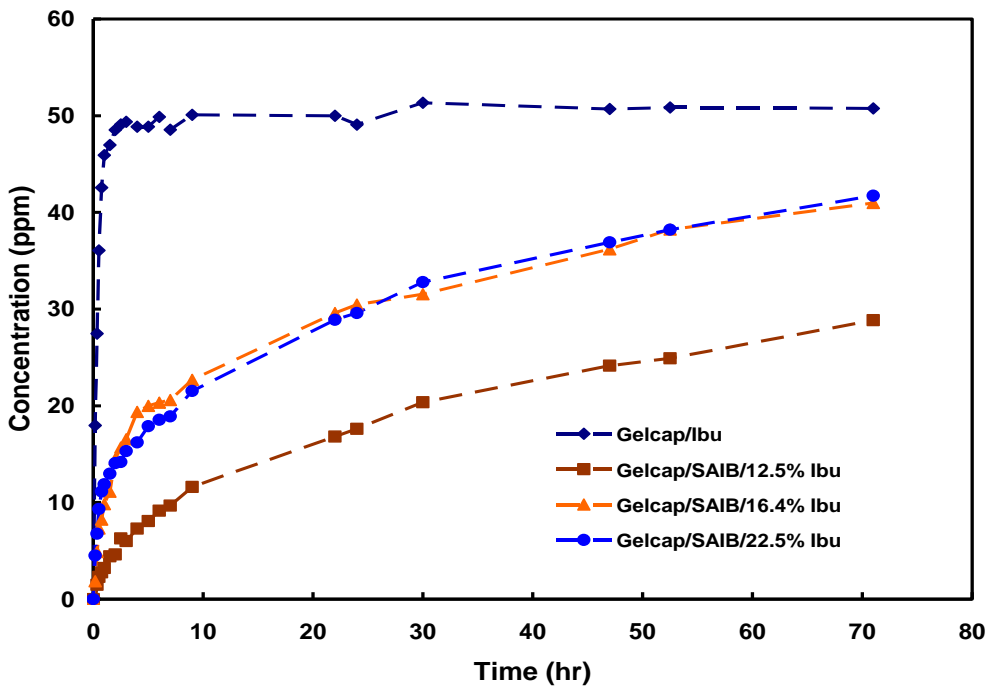


Figure 9. Concentration profiles vs. time for dissolution of ibuprofen in 6.8 pH buffer from several different dosage forms

Caffeine

While the dissolution work using solutions of ibuprofen in SAIB/ethanol provided useful information, further investigations of drug dissolution from solid drug forms containing SAIB were performed. Because carboxymethylcellulose (CMC) is often used as a binder in drug dosage forms, and we indeed did use it in pharmacokinetic work to be described later, we included it as well – primarily in the controls. Two dosage forms were made: (1) one containing 25% caffeine and 75% CMC as a control, and (2) one containing 25% caffeine, 50% SAIB, and 25% CMC. Experiments were performed as described previously, and the measurements of absorption vs. time are shown in Figure 10 below.

Converting absorbance to concentration using the calibration curve (Fig. 5) leads to the plot shown in Figure 11, where the results are shown with 2 different time scales. Clearly the concentration of caffeine reaches much higher values for the caffeine/SAIB/CMC dosage form than for the caffeine/CMC dosage form. This can be partially explained by the fact that the caffeine/SAIB/CMC dosage form contained more caffeine (7.45 mg) than the caffeine/CMC dosage form (4.525 mg). It should also be noted that the calibration curve is very flat in the high-concentration region and that concentration values are thus not as reliable in this region.

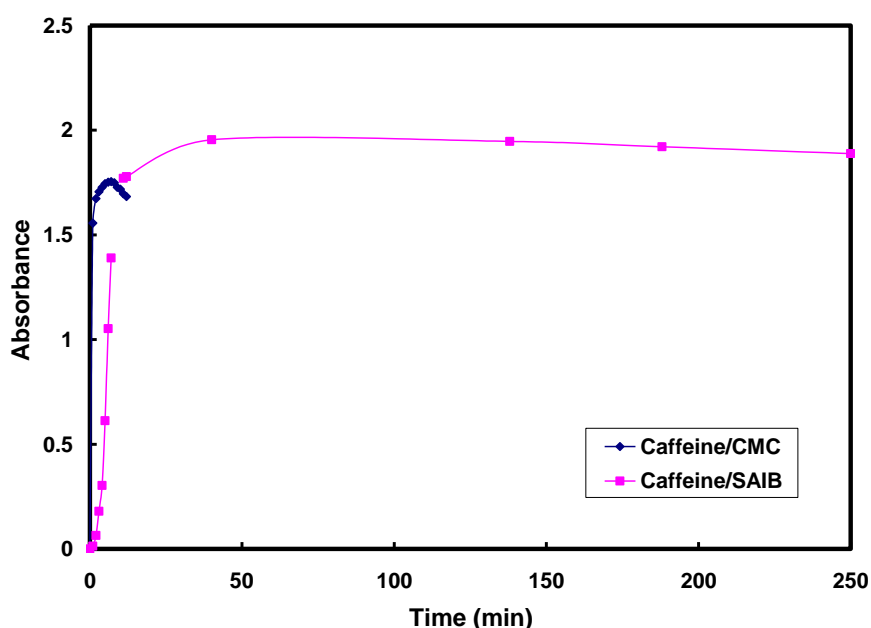


Figure 10. Absorbance at 275 nm vs. time for dissolution of caffeine in 1.2 pH buffer from 2 different dosage forms – one containing SAIB and one without

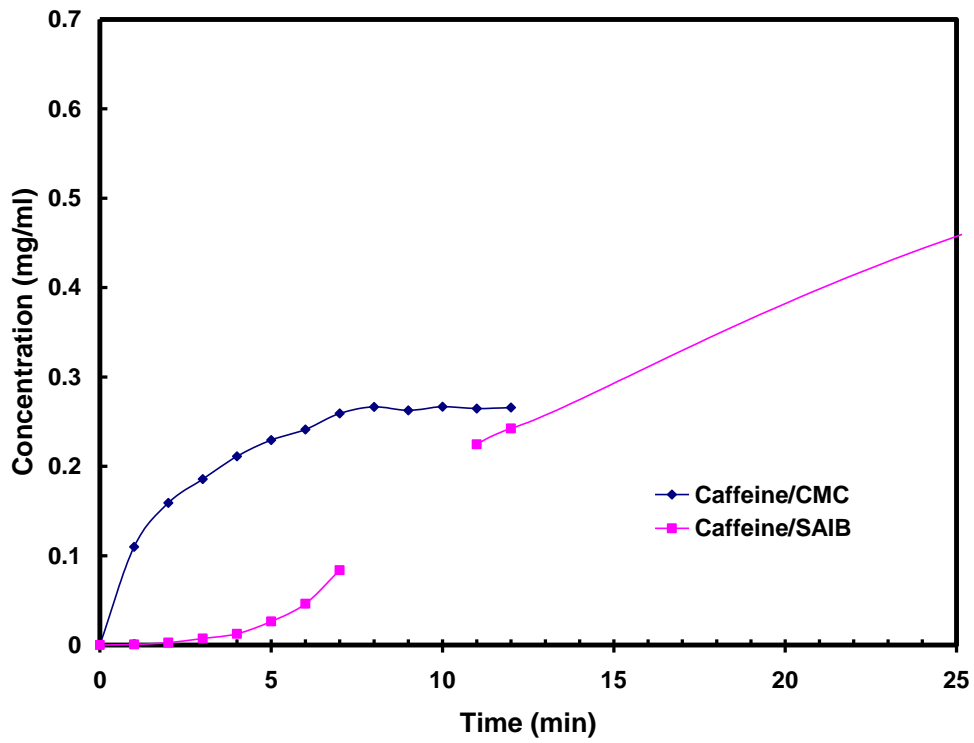
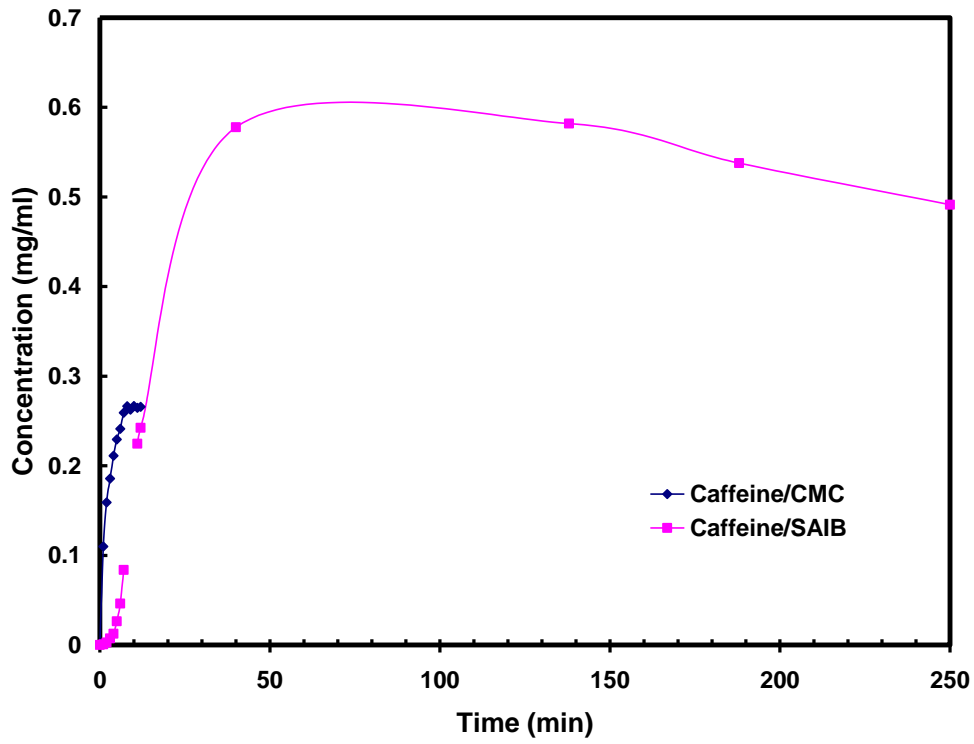


Figure 11. Concentration-time dependence during dissolution of dosage forms in 1.2 pH buffer

The dissolution results reported here on ibuprofen and caffeine dissolution experiments, both with and without SAIB as part of the dosage form, suggest that the inclusion of SAIB is observed to decrease the rate of release and/or dissolution of the drug. These results will be discussed more thoroughly in the Discussion section.

Pharmacokinetic Experiments

Ibuprofen

Figures 12-14 show plasma concentration-time profiles for rats in the three groups of six rats each dosed with ibuprofen in the following ways, respectively: Fig. 12 - rats fed a normal diet and dosed intravenously, Fig 13 – rats fed a normal diet and dosed orally, and Fig. 14 – rats fed a normal diet with 4 wt % sucrose acetate isobutyrate blended into the food. Data were fitted using an open one-compartment model, including elimination only for the intravenously dosed rats and including both absorption and elimination for the orally dosed rats. These equations are given as

$$C(t) = C(0) * e^{-k_{el}t} \quad (1)$$

for the rats dosed intravenously and

$$C(t) = B * e^{k_a t} - A * e^{-k_{el}t} \quad (2)$$

for the rats dosed orally, where

$C(t)$	=	drug concentration in blood at time t
k_a	=	absorption rate constant
k_{el}	=	elimination rate constant

and A and B are constants. GraphPad Prism 5.0 was used to model the data. The models converged for all experiments except rat #3 in Figure 14. Curve fits are shown in the figures and kinetic parameters are shown in Tables 1-3.

Although the open one-compartment model fit the data for many of the rats, the data for others appear to follow an open 2-compartment model. This behavior is observed when the kinetics of distribution between blood and other soft tissue, and distribution into deeper tissues, occur at different rates. In this case the equations for intravenous and oral administration become

$$C(t) = B * e^{-k_d t} + A * e^{-\alpha t} \quad (3)$$

and

$$C(t) = B * e^{-\beta t} + A * e^{-\alpha t} - C(0) * e^{-k_a t} \quad (4)$$

respectively, where

$C(0)$ = initial drug concentration

α = distribution slope

and β = overall elimination slope.

Such a model shows an initial rapid appearance of drug in the blood followed by a rapid decrease resulting from both distribution into the deeper compartment, i.e. tissue, and elimination through normal metabolic processes. As distribution into the tissue and redistribution back into the blood reaches a steady state, elimination becomes the controlling process, causing a decrease in drug concentration in the blood, and the concentration-time slope changes.

For the open one-compartment model, $C(0)=A=B$, and thus these constants can be easily compared to determine consistency between groups. A comparison of $C(0)$, A , and B obtained from the curve fits for each dosing method was conducted using a one-way analysis of variance (ANOVA) in the JMP® 9.0.0 statistics package. This comparison is shown in Fig. 15. The values of $C(0)$, obtained from curve fits for the rats dosed intravenously are much more tightly grouped than the values of A and B obtained from the curve fits for the rats dosed orally. Due to the amount of variability in A and B for the rats dosed orally, a variance-stabilizing transformation on $C(0)$, A , and B was used to determine if there are significant differences between the groups. Results of a Box-Cox transformation suggested that an inverse square transformation should be performed on $C(0)$, A , and B . The Tukey-Kramer HSD test on these transformed results revealed that the means of the inverse square roots are not significantly different. Thus, there is no difference between $C(0)$, A , and B . These constants should be the same for the open one-compartment model.

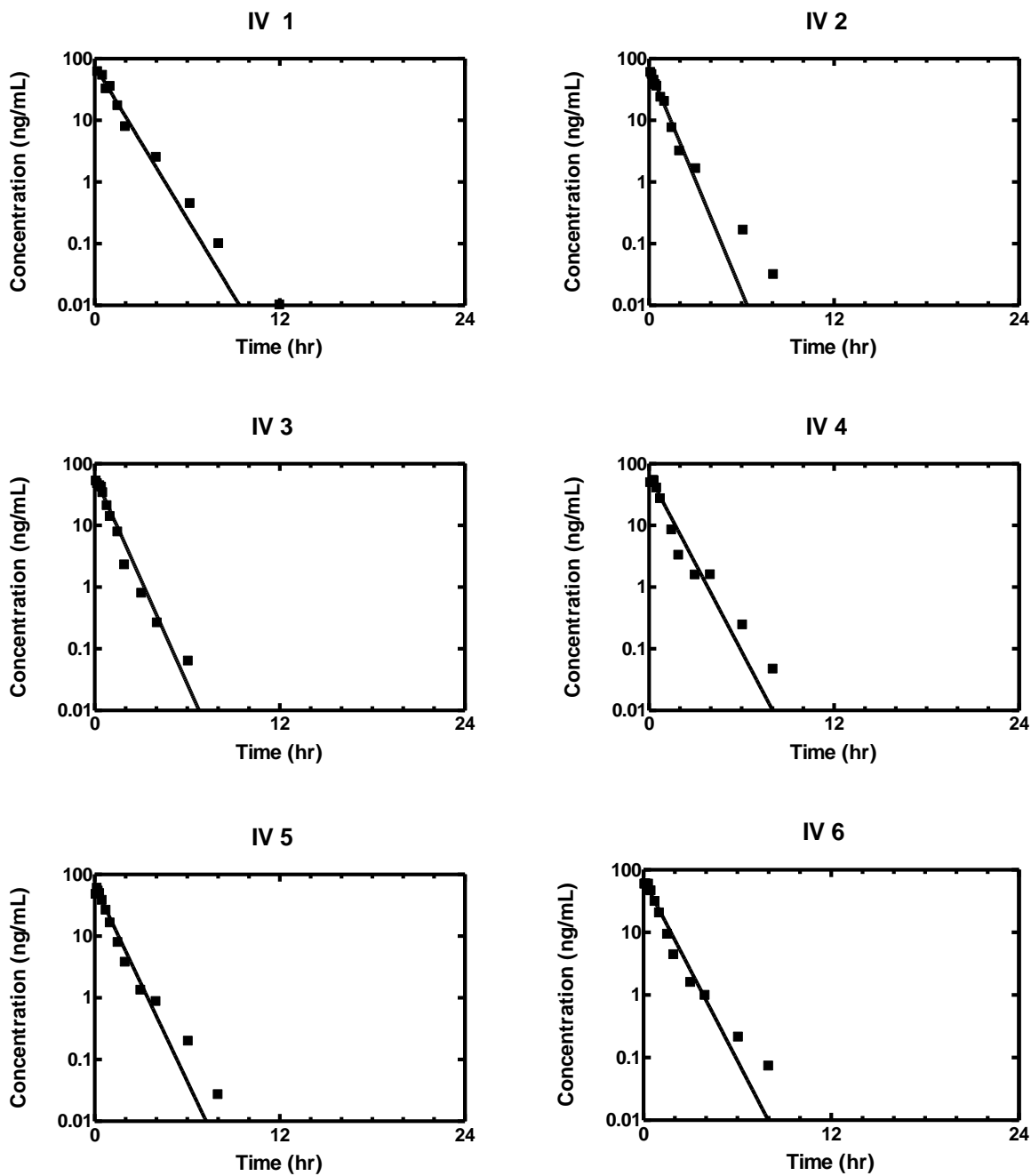


Figure 12. Ibuprofen plasma concentration profiles for rats fed a normal diet and dosed intravenously. Data were fitted using an open one-compartment intravenous model (elimination only)

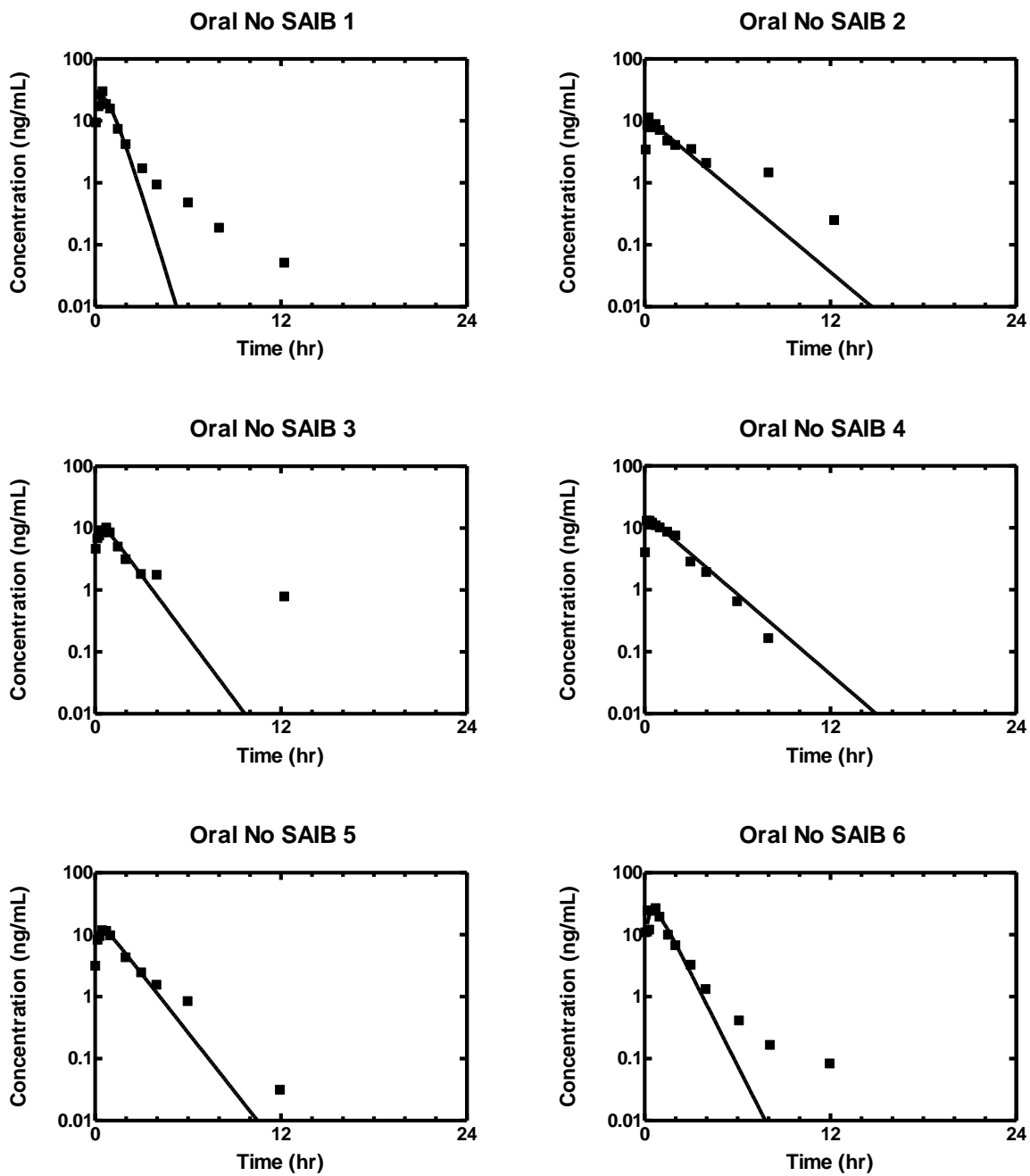


Figure 13. Ibuprofen plasma concentration profiles for rats fed a normal diet and dosed orally. Data were fitted using an open one-compartment oral model (absorption and elimination)

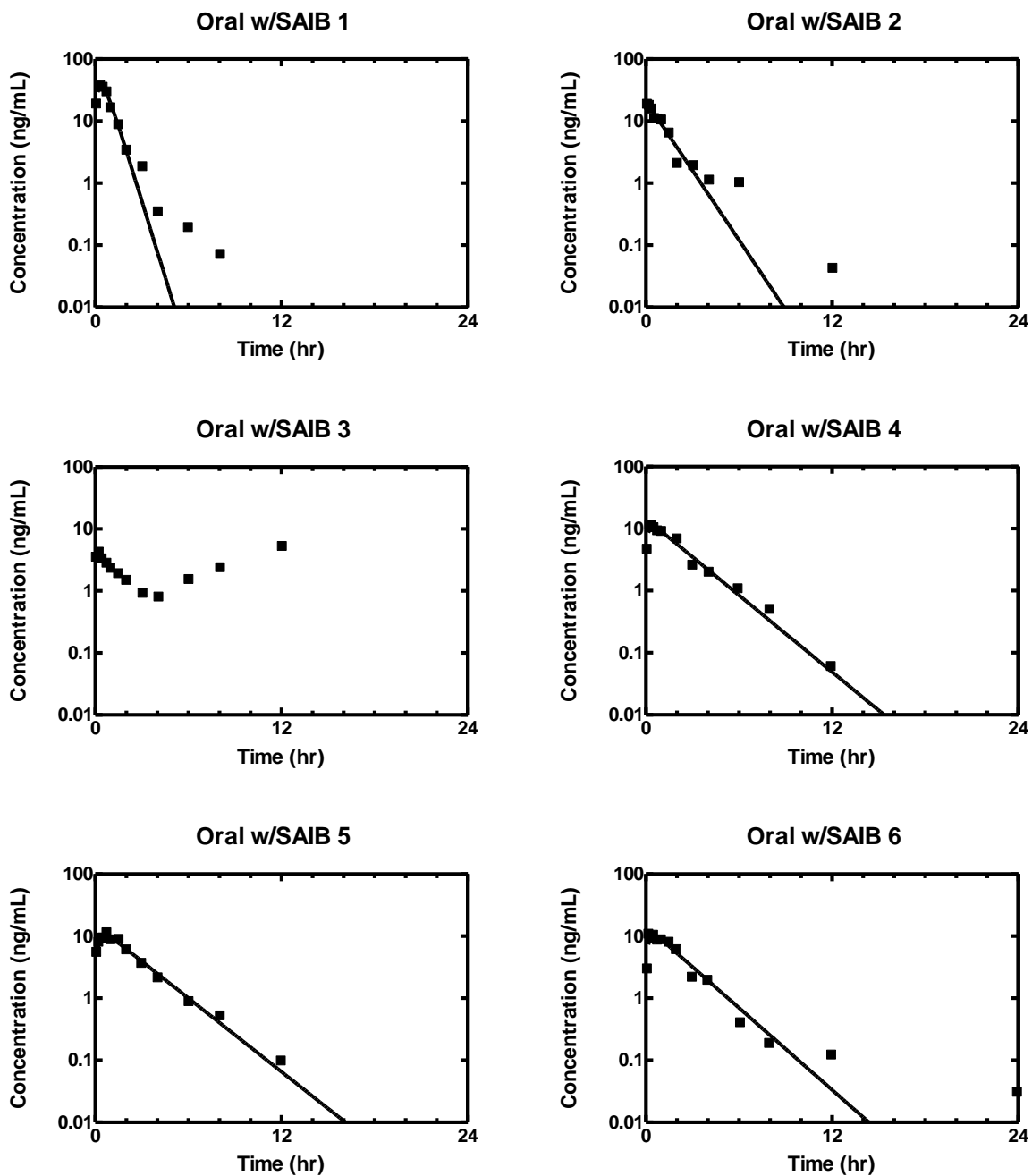


Figure 14. Ibuprofen plasma concentration profiles for rats fed a diet consisting of rat chow containing 4 wt% SAIB and dosed orally. Data were fitted using an open one-compartment oral model (absorption and elimination)

Table 1. Pharmacokinetic Parameters for Rats (Normal Diet) Dosed Intravenously with Ibuprofen

Parameter	IV1 Normal Diet	IV2 Normal Diet	IV3 Normal Diet	IV4 Normal Diet	IV5 Normal Diet	IV6 Normal Diet	Mean $\pm\sigma$
$C(0)$	77.75	71.18	63.51	65.06	66.30	70.35	69.0 \pm 4.7
k^{el}	0.954	1.391	1.292	1.095	1.217	1.111	1.18 \pm 0.14
R^2	0.976	0.996	0.983	0.965	0.969	0.974	

Table 2. Pharmacokinetic Parameters for Rats (Normal Diet) Dosed Orally with Ibuprofen

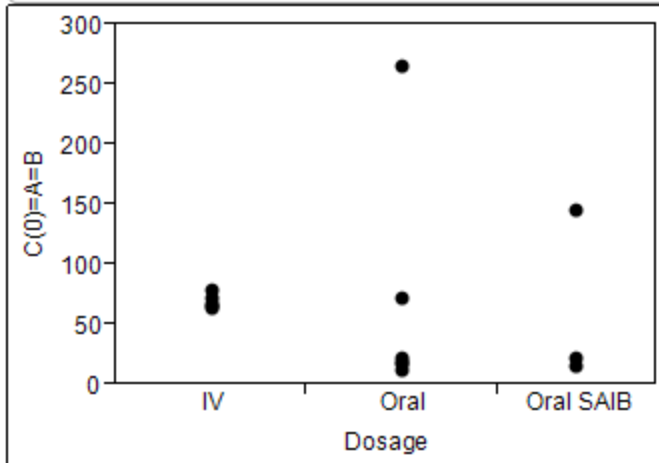
Parameter	Oral1 Normal Diet	Oral2 Normal Diet	Oral3 Normal Diet	Oral4 Normal Diet	Oral5 Normal Diet	Oral6 Normal Diet	Mean $\pm\sigma$
$A=B$	264.6	11.78	17.58	16.57	21.02	71.43	67.2 \pm 90.8
k^a	3.312	6.831	3.860	8.221	3.692	2.912	4.80 \pm 1.99
k^{el}	2.473	0.482	0.772	0.496	0.729	1.138	1.02 \pm 0.69
R^2	0.955	0.919	0.959	0.972	0.988	0.883	

Table 3. Pharmacokinetic Parameters for Rats (SAIB Diet) Dosed Orally with Ibuprofen

Parameter	Oral1 SAIB Diet	Oral2 SAIB Diet	Oral3 SAIB Diet	Oral4 SAIB Diet	Oral5 SAIB Diet	Oral6 SAIB Diet	Mean $\pm\sigma$
$A=B$	144.5	21.21	NC*	14.61	15.40	14.67	42.08 \pm 51.27
k^a	3.867	33.03	NC*	6.761	4.098	5.378	10.63 \pm 11.25
k^{el}	1.887	0.860	NC*	0.4746	0.455	0.507	0.84 \pm 0.55
R^2	0.994	0.982	NC*	0.978	0.976	0.943	

* NC = No Convergence

Oneway Analysis of C(0)=A=B By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
IV	6	69.0250	5.2170	2.130	63.55	74.50
Oral	6	67.1633	99.2110	40.503	-36.95	171.28
Oral SAIB	5	42.0780	57.3218	25.635	-29.10	113.25

Comparisons for all pairs using Tukey-Kramer HSD

Level	Mean
Oral SAIB A	0.21557135
Oral A	0.19557205
IV A	0.12057238

Levels not connected by same letter are significantly different.

Figure 15. Results of statistical analysis of C(0), A, and B for ibuprofen obtained using the open one-compartment model for (1) rats fed a normal diet and dosed intravenously and (2) rats fed a normal diet and dosed orally, and (3) rats fed a diet containing 4 wt% SAIB and dosed orally

The elimination rate constants, k_{el} , should be the same as well. Figure 16 shows the results of a similar statistical analysis on the elimination rate constants for all 3 rat groups. A one-way ANOVA of the data and a Tukey-Kramer HSD on the inverse square root were performed as before. There were no differences observed for the 3 groups.

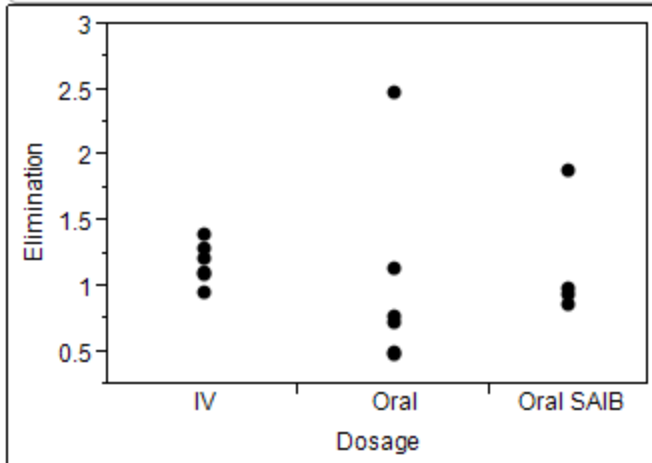
This statistical analysis was also performed on the integrated absorption rate constants, k_a , for each rat dosage group. A one-way ANOVA of the data and a Tukey-Kramer HSD on the inverse square root were once again performed and the results are shown in Figure 17. In this case, and inverse transformation of k_a was used and the Tukey-Kramer HSD indicated that there is no significant difference between the k_a 's for ibuprofen absorption for the rats fed different diets.

Finally, Figure 18 shows the results of the Tukey-Kramer HSD for the means of areas under the ibuprofen plasma concentration-time curves. The long horizontal line of the green diamond indicates the mean, the short horizontal lines indicate the standard error of the mean, and the upper- and lower-most points of the diamond indicate the standard deviations. The latter 2 are the pooled standard errors and standard deviations. The results suggest that the mean value of the area under the curve for the IV dosage form is significantly different from the mean values for the 2 groups dosed orally. However, the mean for the 2 groups dosed orally are not significantly different from each other.

Saquinavir

Figures 19 and 20 show the individual blood plasma concentration-time plots for rats dosed with saquinavir/CMC dosage forms and 6 rats dosed with saquinavir/CMC/SAIB dosage forms, respectively. We show these data individually for clarity. Figure 21 shows the open one-compartment model fits of these data. Because there is so much scatter, we decided to look at the data in a different way for this study. Figure 22 shows the mean values for the 6 rats in each of the 2 groups along with standard error of the means for each group at each time. This figure suggested that the area under the curve for the dosage form containing SAIB might be larger than that without SAIB. To investigate this we integrated the area under each of the curves and calculated a mean AUC for each of the 2 groups. Conducting a one-way analysis of variance and a Tukey-Kramer HSD, the results of which are shown in Figure 23, we determined that there is no significant difference in the mean areas under the curves for the 2 groups of rats.

Oneway Analysis of Elimination By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
IV	6	1.17667	0.155839	0.06362	1.0131	1.3402
Oral	6	1.01500	0.753019	0.30742	0.2248	1.8052
Oral SAIB	5	1.12880	0.426536	0.19075	0.5992	1.6584

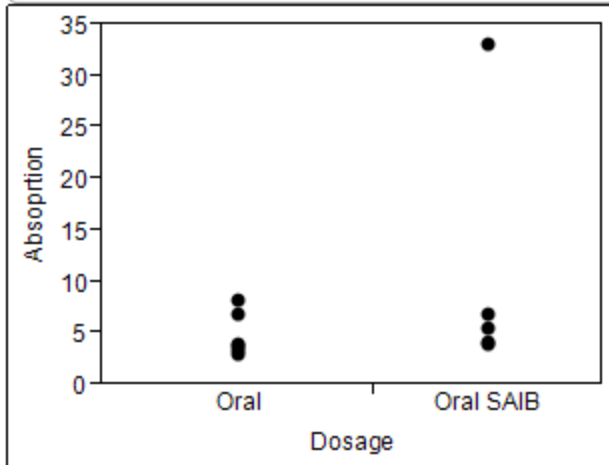
Comparisons for all pairs using Tukey-Kramer HSD

Level	Mean	
Kel Oral	A	1.1238221
Kel Oral SAIB	A	0.9718967
Kel IV	A	0.9270531

Levels not connected by same letter are significantly different.

Figure 16. Results of statistical analysis of elimination rate constant, K_{el} , for ibuprofen obtained using the open one-compartment model for (1) rats fed a normal diet and dosed intravenously and (2) rats fed a normal diet and dosed orally, and (3) rats fed a diet containing 4 wt% SAIB and dosed orally

Oneway Analysis of Absorption By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
Oral	6	4.8047	2.1780	0.8892	2.519	7.090
Oral SAIB	5	10.6268	12.5769	5.6246	-4.990	26.243

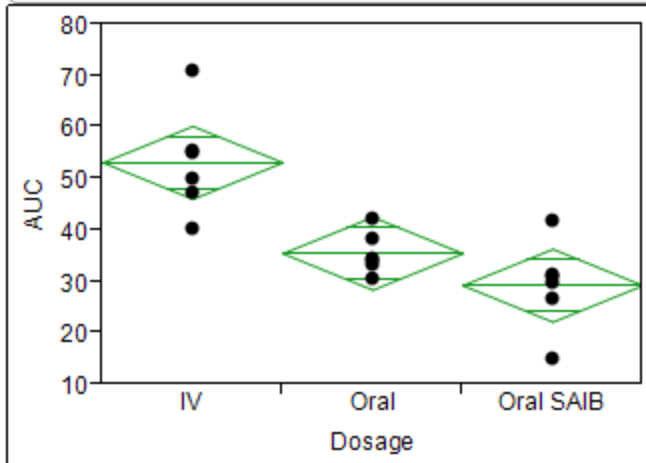
Comparisons for all pairs using Tukey-Kramer HSD

Level	Mean
Ka Oral	A 0.24054890
Ka Oral SAIB	A 0.17334904

Levels not connected by same letter are significantly different.

Figure 17. Results of statistical analysis of absorption rate constant, K_a , for ibuprofen obtained using the open one-compartment model for rats dosed orally and (1) fed a normal diet or (2) fed a diet containing 4 wt% SAIB and dosed orally

Oneway Analysis of AUC By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
IV	6	53.2467	10.3071	4.2079	42.430	64.063
Oral	6	35.5750	4.1579	1.6974	31.212	39.938
Oral SAIB	6	29.3500	8.6687	3.5390	20.253	38.447

Comparisons for all pairs using Tukey-Kramer HSD

Level		Mean
IV	A	53.246667
Oral	B	35.575000
Oral SAIB	B	29.350000

Levels not connected by same letter are significantly different.

Figure 18. Results of statistical analysis of areas under the ibuprofen plasma concentration-time curve obtained using the open one-compartment model for (1) rats fed a normal diet and dosed intravenously and (2) rats fed a normal diet and dosed orally, and (3) rats fed a diet containing 4 wt% SAIB and dosed orally

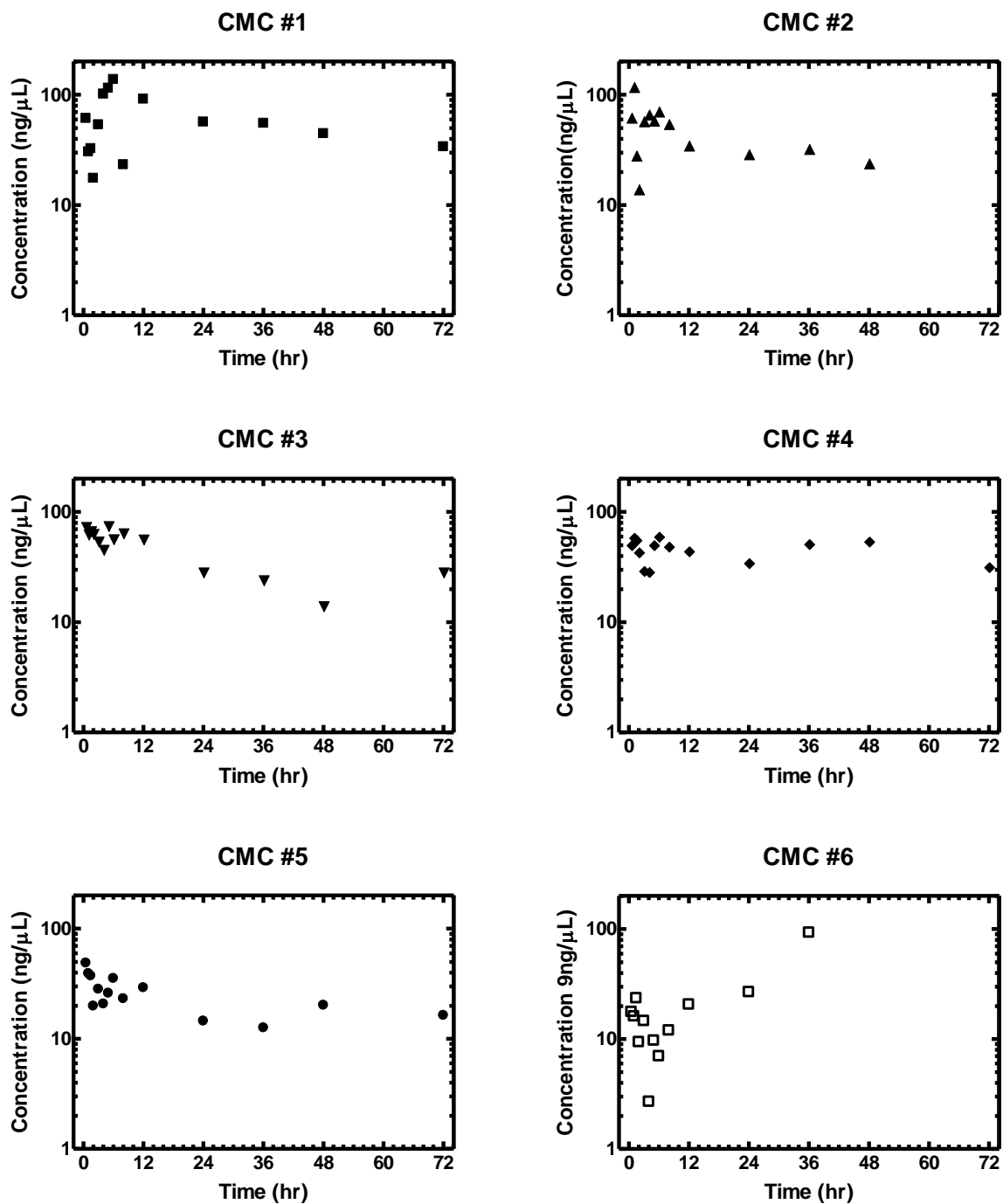


Figure 19. Saquinavir concentration profiles for the 6 rats dosed with saquinavir/CMC dosage forms

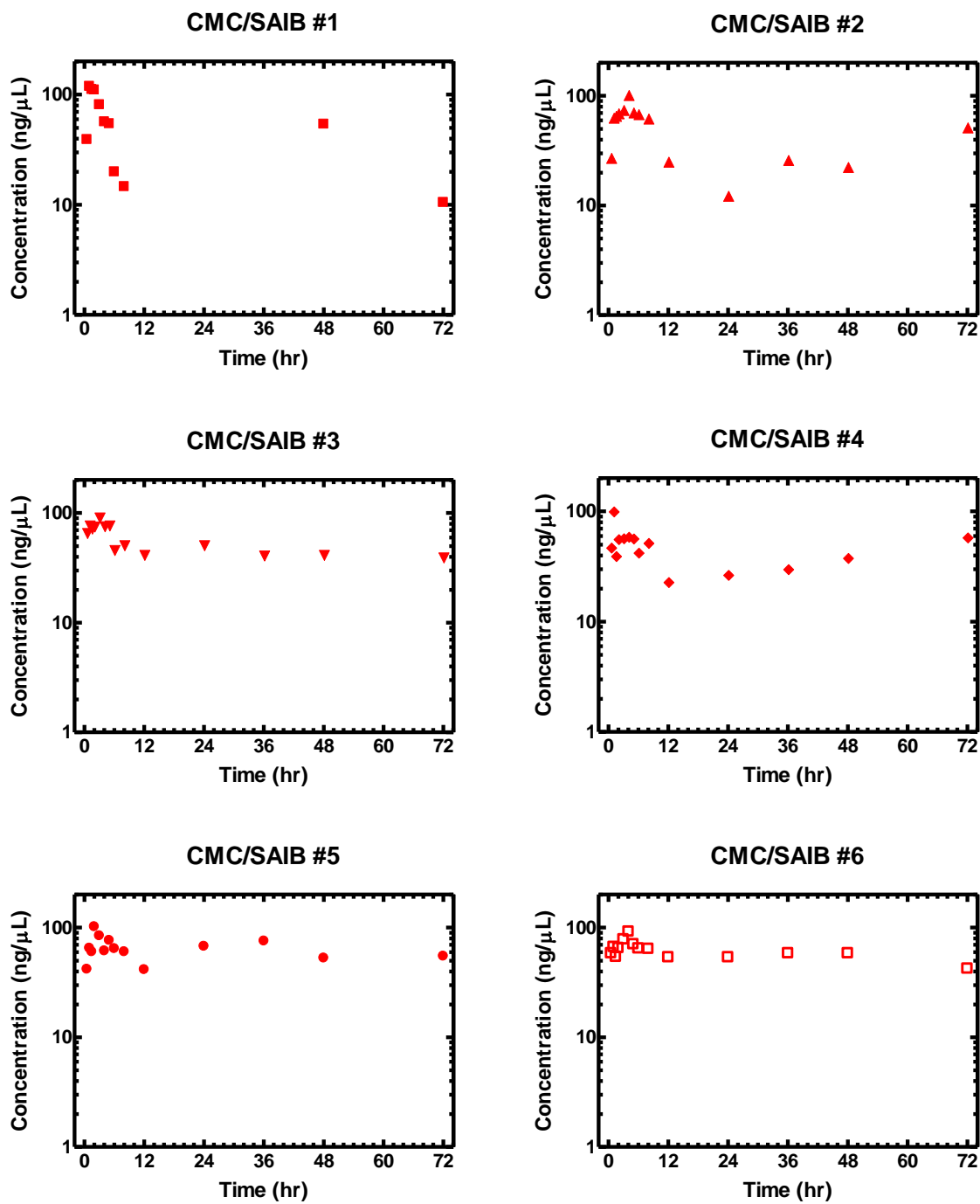


Figure 20. Saquinavir concentration profiles for the 6 rats dosed with saquinavir/CMC/SAIB dosage forms

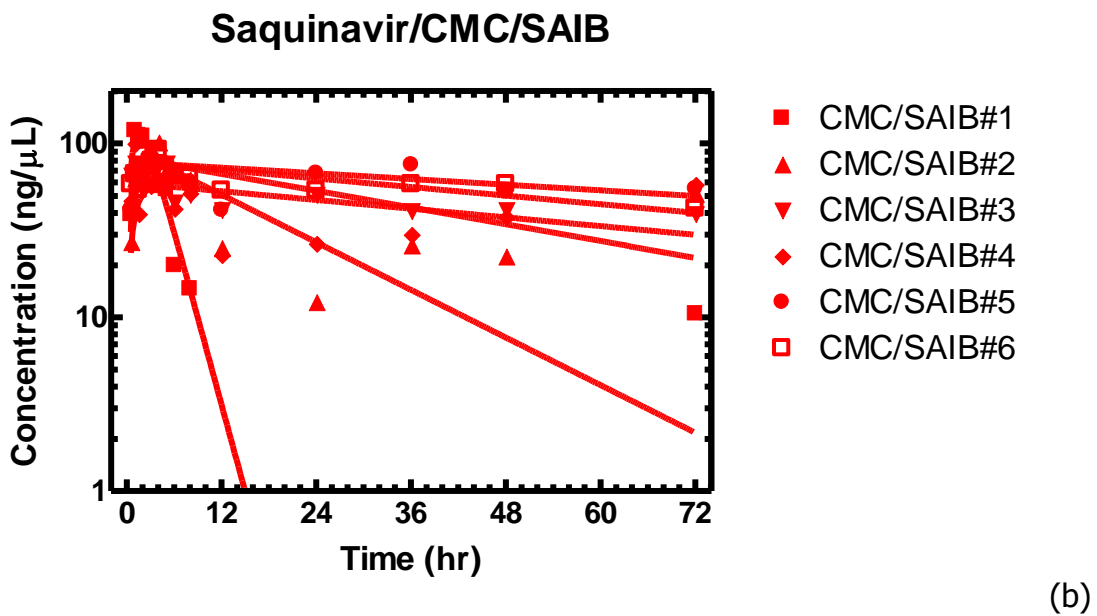
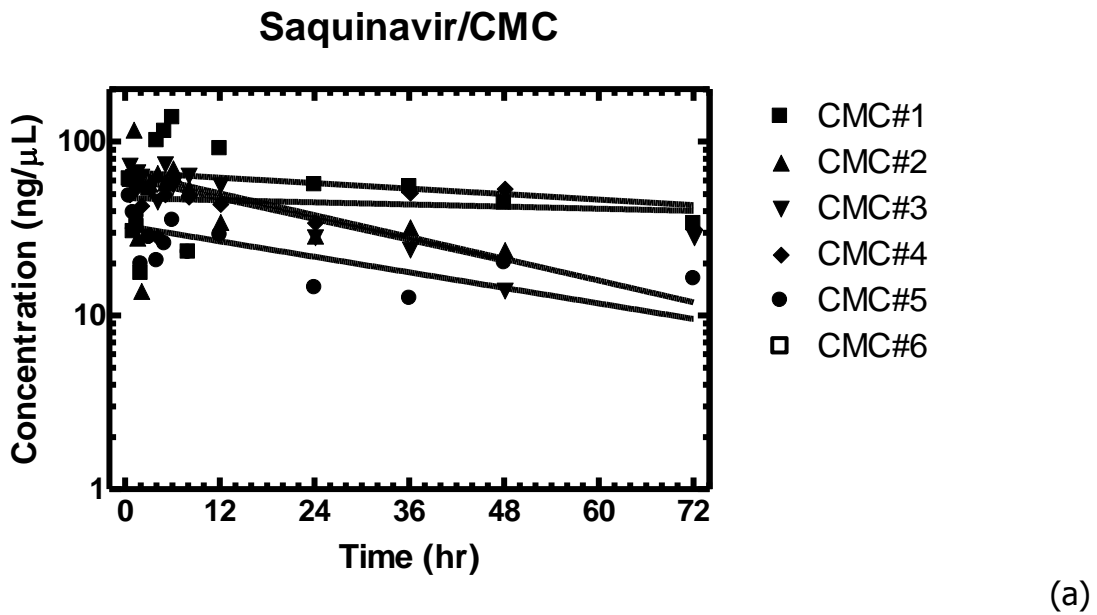


Figure 21. Aggregate plots showing: (a) the concentration of saquinavir for rats dosed with saquinavir/CMC dosage forms and (b) the concentration of saquinavir for rats dosed with saquinavir/CMC/SAIB dosage forms

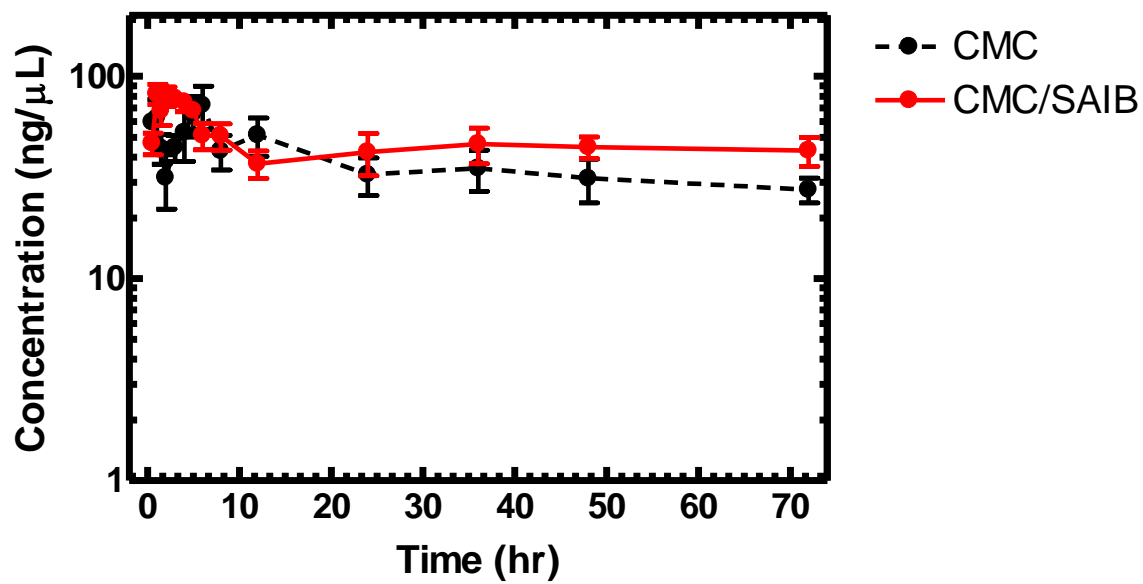
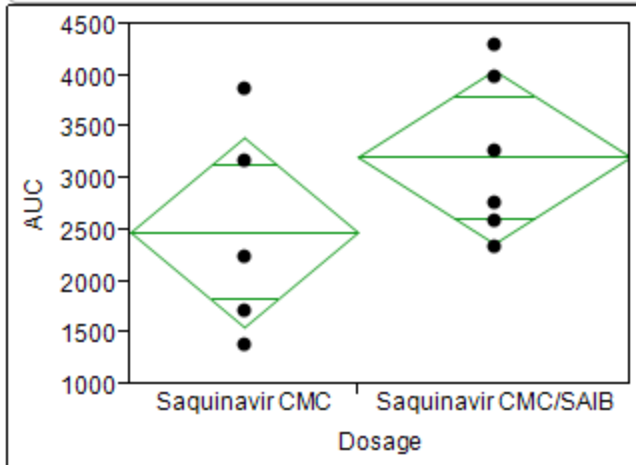


Figure 22. Saquinavir mean concentration profiles for the 6 rats dosed with either saquinavir/CMC or saquinavir/CMC/SAIB dosage forms. Shown are the means and standard error of the means for all rats of each population

Oneway Analysis of AUC By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
Saquinavir CMC	5	2481.40	1038.88	464.60	1191.5	3771.3
Saquinavir CMC/SAIB	6	3213.33	795.39	324.71	2378.6	4048.0

Comparisons for all pairs using Tukey-Kramer HSD

Level	Mean
Saquinavir CMC/SAIB A	3213.3333
Saquinavir CMC A	2481.4000

Levels not connected by same letter are significantly different.

Figure 23. Results of statistical analysis of areas under the saquinavir plasma concentration-time curve for rats dosed either (1) saquinavir/CMC or (2) saquinavir/CMC/SAIB

Clarithromycin

According to Wibawa et al. (2003), clarithromycin is hydrolyzed in an acidic environment (e.g. the stomach) to decladinose clarithromycin as shown in Figure 24. The pharmacokinetic studies that we conducted on clarithromycin included blood analysis for both clarithromycin and decladinose clarithromycin.

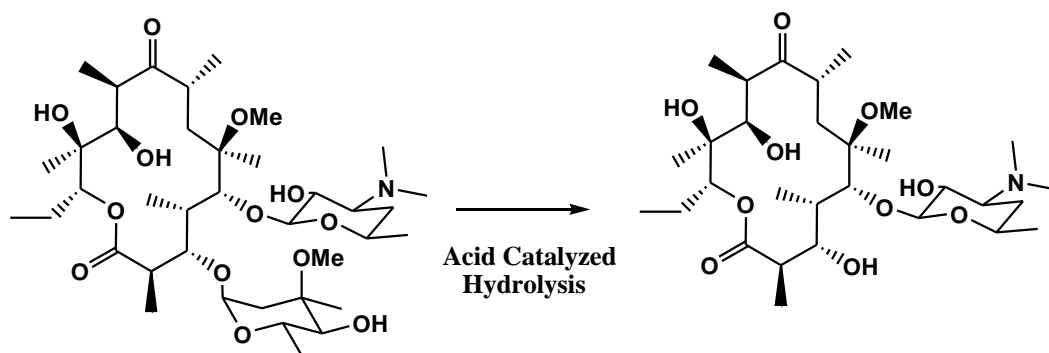


Figure 24. Acid catalyzed hydrolysis of clarithromycin to decladinose clarithromycin

Figure 25 shows the mean clarithromycin blood plasma concentration-time plots for three rats dosed with clarithromycin/CMC dosage forms and 3 rats dosed with clarithromycin/CMC/SAIB dosage forms. Figure 26 show the concentration-time profiles for decladinose clarithromycin. Clearly there is more variability in the results for the rats dosed with clarithromycin/CMC than the clarithromycin/CMC/SAIB dosage forms. Several statistical analyses of the areas under the curves for these 2 groups were performed, including a one-way analysis of variance and a Tukey-Kramer HSD. A t-test revealed a P-value of 0.747. These are shown in Figure 27 for clarithromycin. All the results confirm that there is no difference between the areas under the curves. The results for decladinose clarithromycin are shown in Figure 28. In this case, a natural log transformation was performed for the Tukey-Kramer HSD. The areas under the curves were found to be statistically significant for decladinose clarithromycin, the concentrations being much higher for the clarithromycin/CMC/SAIB dosage forms than for the clarithromycin/CMC forms.

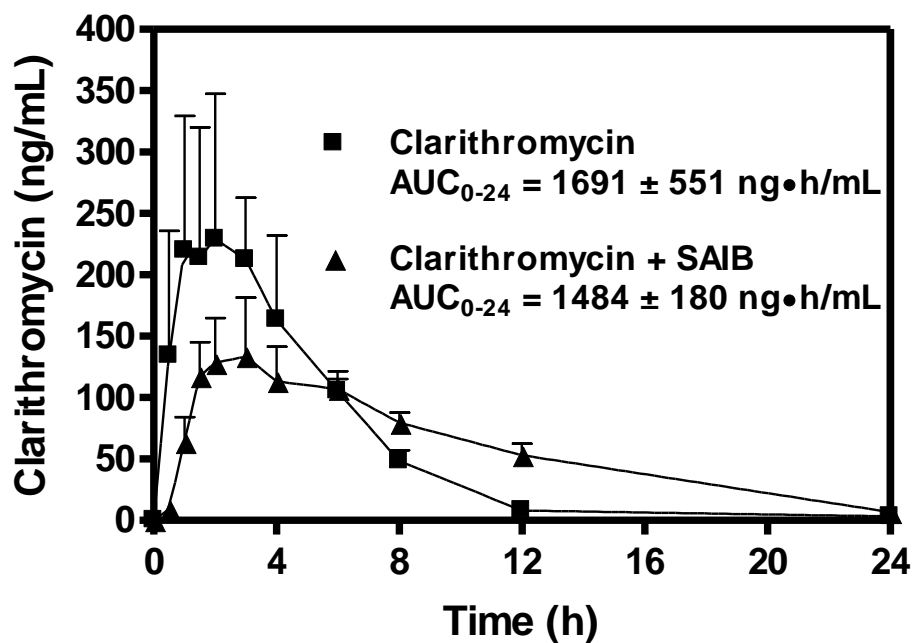


Figure 25. Clarithromycin concentration-time profiles for rats dosed orally with and without SAIB ($n = 3 \pm \text{SEM}$)

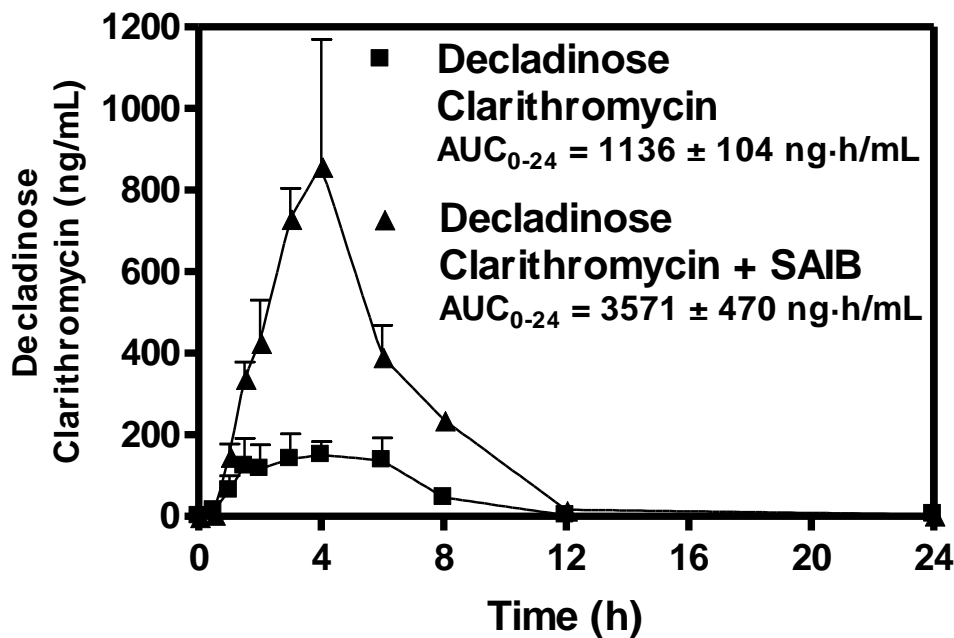
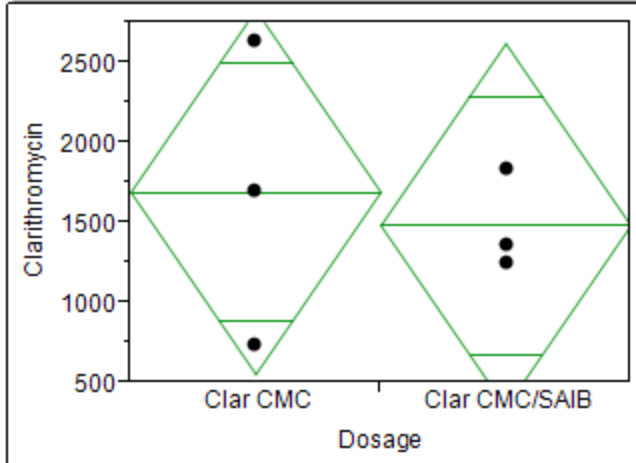


Figure 26. Decladinose clarithromycin plasma concentration-time profiles for rats dosed orally with clarithromycin both with and without SAIB ($n = 3 \pm \text{SEM}$)

Oneway Analysis of Clarithromycin By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
Clar CMC	3	1692.83	954.308	550.97	-677.8	4063.5
Clar CMC/SAIB	3	1483.67	311.632	179.92	709.5	2257.8

Comparisons for all pairs using Tukey-Kramer HSD

Level	Mean
Clar CMC A	1692.8333
Clar CMC/SAIB A	1483.6667

Levels not connected by same letter are significantly different.

t Test

Clar CMC/SAIB-Clar CMC

Assuming unequal variances

Difference	-209.2	t Ratio	-0.36088
Std Err Dif	579.6	DF	2.42175
Upper CL Dif	1911.4	Prob > t	0.7473
Lower CL Dif	-2329.7	Prob > t	0.6263
Confidence	0.95	Prob < t	0.3737

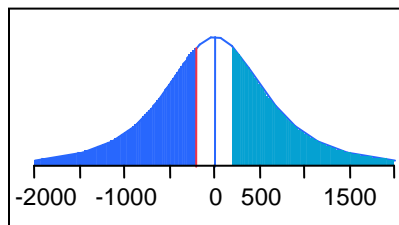
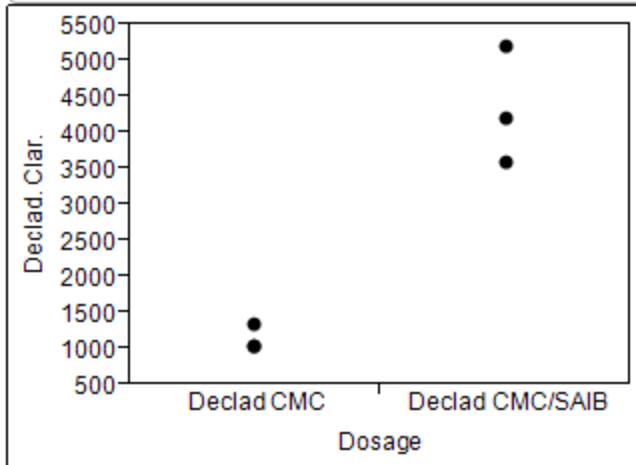


Figure 27. Results of statistical analysis of areas under the clarithromycin plasma concentration-time curves for rats does with either (1) clarithromycin/CMC or (2) clarithromycin/CMC/SAIB

Oneway Analysis of Declad. Clar. By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
Declad CMC	3	1135.67	180.672	104.31	686.9	1584.5
Declad CMC/SAIB	3	4315.33	814.267	470.12	2292.6	6338.1

Comparisons for all pairs using Tukey-Kramer HSD

Level	Mean
Declad CMC/SAIB A	8.3581940
Declad CMC B	7.0269531

Levels not connected by same letter are significantly different.

Figure 28. Results of statistical analysis of areas under the decladinose clarithromycin plasma concentration-time curves for rats does with either (1) clarithromycin/CMC or (2) clarithromycin/CMC/SAIB

Rat liver microsomal incubations performed at Eastman Chemical Company by Wempe (2006) demonstrated that 3 metabolites are formed from clarithromycin as shown in Figure 29. As a result of this work, both demethyl-clarithromycin and hydroxy-clarithromycin were also monitored and their blood plasma concentration-time plots are shown in Figures 30 and 31, respectively. For both of these metabolites, less variation again results from administration of clarithromycin with SAIB but there are no statistically significant differences, as demonstrated by the results shown in Figures 32 and 33.

The results of this study are consistent with the hypothesis that the formulation containing SAIB is retained in the stomach for a longer period of time and also suggest that in this particular study SAIB acts to decrease the animal-to-animal variability of drug concentration profiles.

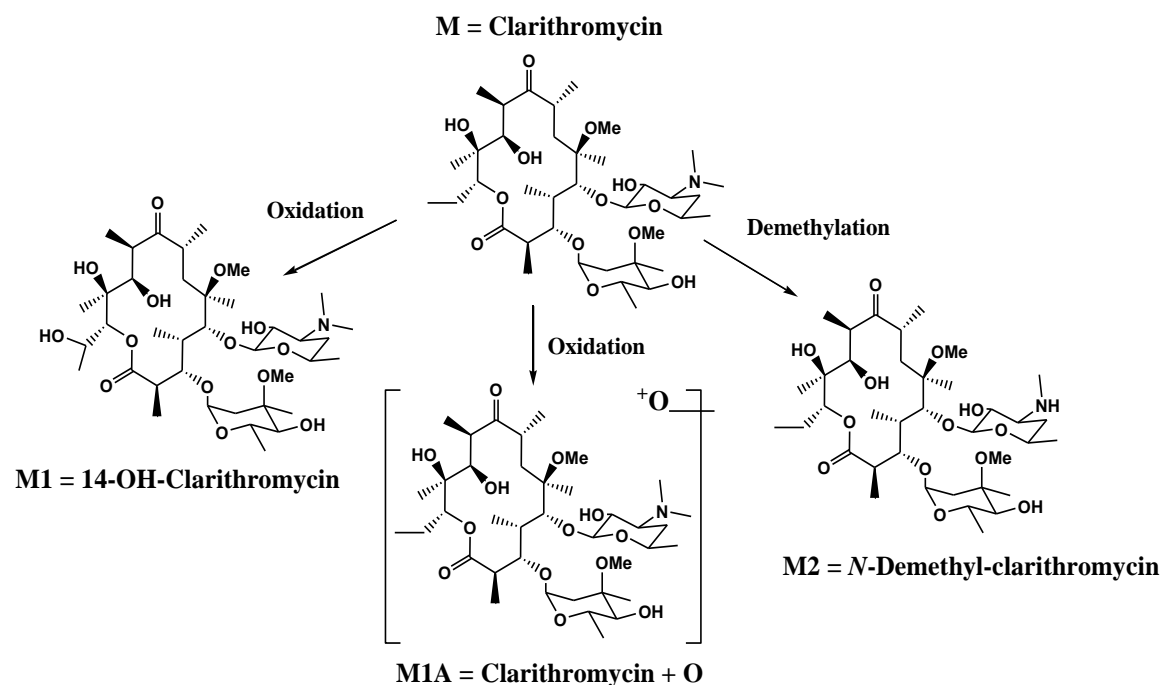


Figure 29. In vivo metabolism from liver microsomal incubations

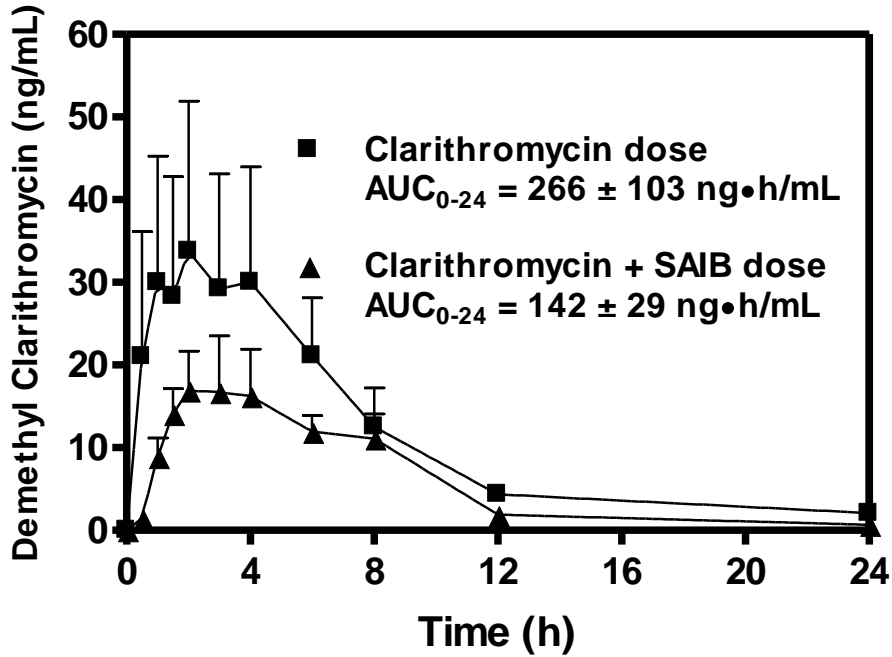


Figure 30. Demethyl clarithromycin concentration-time profiles for rats dosed orally with clarithromycin both with and without SAIB ($n = 3 \pm \text{SEM}$)

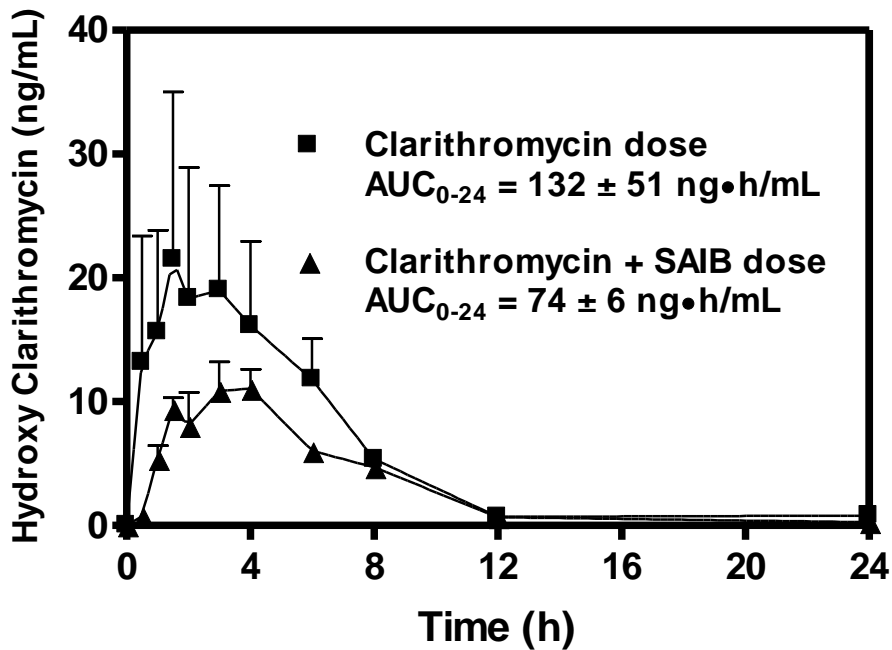
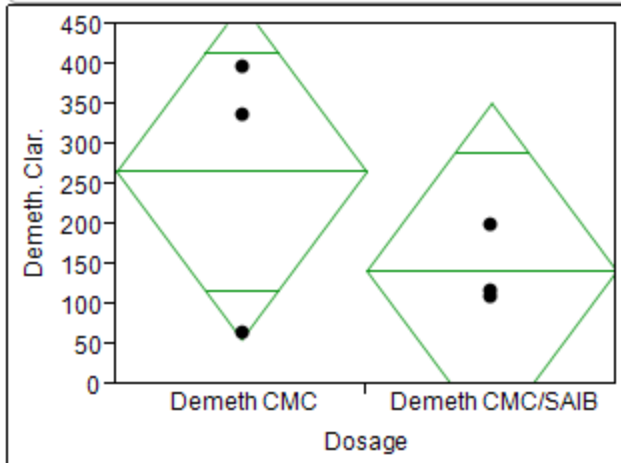


Figure 31. Hydroxy-clarithromycin concentration-time profiles for rats dosed orally with clarithromycin both with and without SAIB ($n = 3 \pm \text{SEM}$)

Oneway Analysis of Demeth. Clar. By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
Demeth CMC	3	266.457	178.137	102.85	-176.1	708.97
Demeth CMC/SAIB	3	142.367	49.874	28.79	18.5	266.26

Comparisons for all pairs using Tukey-Kramer HSD

Level	Mean
Demeth CMC A	266.45667
Demeth CMC/SAIB A	142.36667

Levels not connected by same letter are significantly different.

t Test

Demeth CMC/SAIB-Demeth CMC

Assuming unequal variances

Difference	-124.09	t Ratio	-1.16187
Std Err Dif	106.80	DF	2.311632
Upper CL Dif	280.90	Prob > t	0.3512
Lower CL Dif	-529.08	Prob > t	0.8244
Confidence	0.95	Prob < t	0.1756

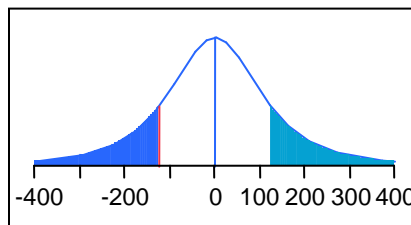
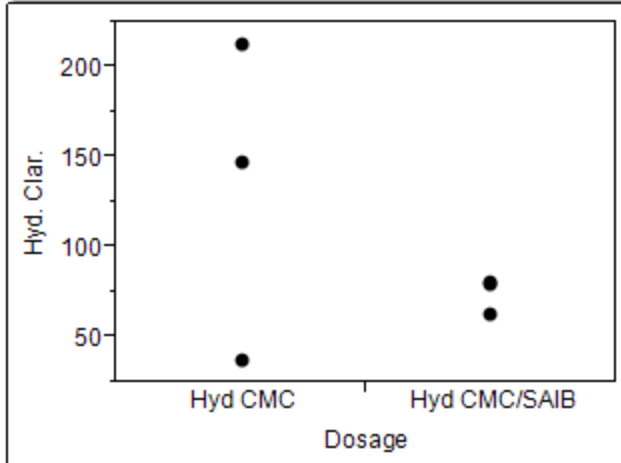


Figure 32. Results of statistical analysis of areas under the demethyl clarithromycin plasma concentration-time curves for rats does with either (1) clarithromycin/CMC or (2) clarithromycin/CMC/SAIB

Oneway Analysis of Hyd. Clar. By Dosage



Means and Std Deviations

Level	Number	Mean	Std Dev	Std Err		
				Mean	Lower 95%	Upper 95%
Hyd CMC	3	132.227	89.0835	51.432	-89.07	353.52
Hyd CMC/SAIB	3	74.370	9.8607	5.693	49.87	98.87

Comparisons for all pairs using Tukey-Kramer HSD

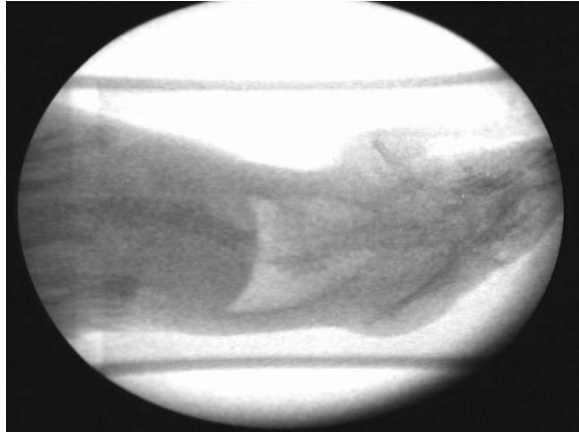
Level	Mean
Hyd CMC A	10.925092
Hyd CMC/SAIB A	8.610617

Levels not connected by same letter are significantly different.

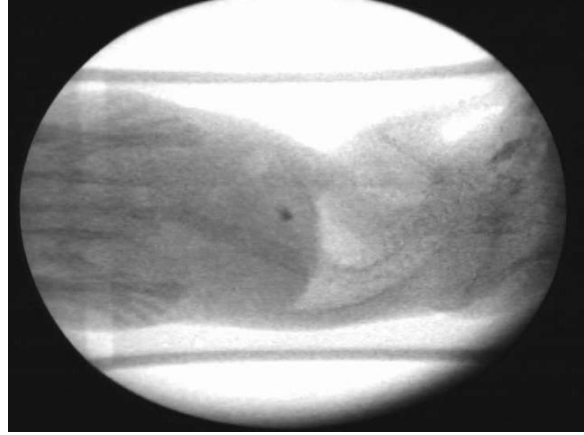
Figure 33. Results of statistical analysis of areas under the hydroxy clarithromycin plasma concentration-time curves for rats does with either (1) clarithromycin/CMC or (2) clarithromycin/CMC/SAIB.

X-Ray Imaging Experiments

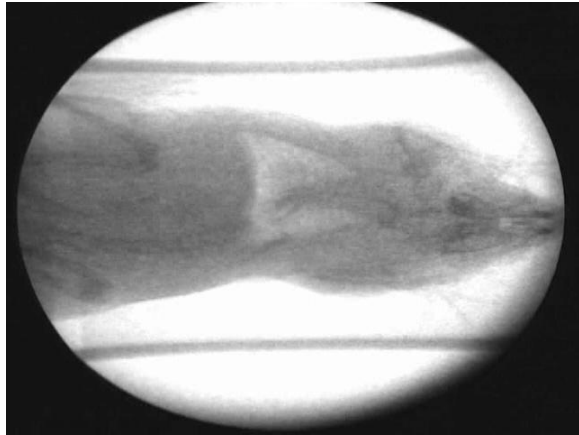
X-rays of rats dosed with (1) 25%BaSO₄/75%CMC and (2) 25%BaSO₄/25% CMC/50%SAIB are shown in Figure 34. Images are presented sequentially so that time moves from left to right and images from different groups can be compared directly at each time. These x-rays show that the dosage form containing SAIB persisted up to 100 minutes, while the dosage form that did not contain SAIB persisted only to 35 minutes. Had these been the only 2 dosing experiments performed, one would conclude that dosage forms containing SAIB retain their integrity for longer times. However, additional experiments were performed with mixed results using different dosage forms and different rats. Some dosage forms disappeared after relatively short times (~30 minutes), while others persisted to much longer times (>100 minutes). However, these results do suggest that perhaps through improvements in the design of SAIB dosage forms and in consistency of SAIB dosage form preparation, it may be possible to create SAIB dosage forms that retain their integrity for longer periods of time.



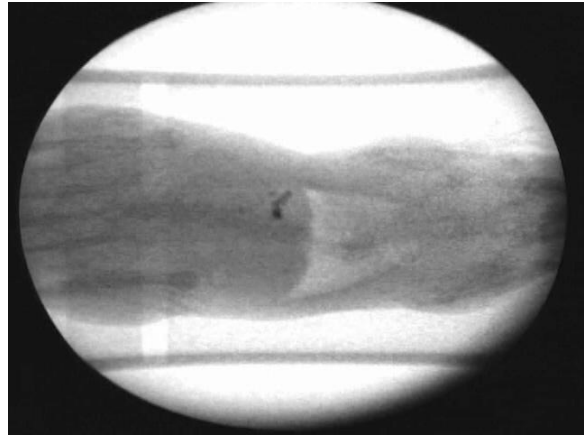
Pre-dosing (BaSO₄/CMC)



2 min (BaSO₄/CMC)

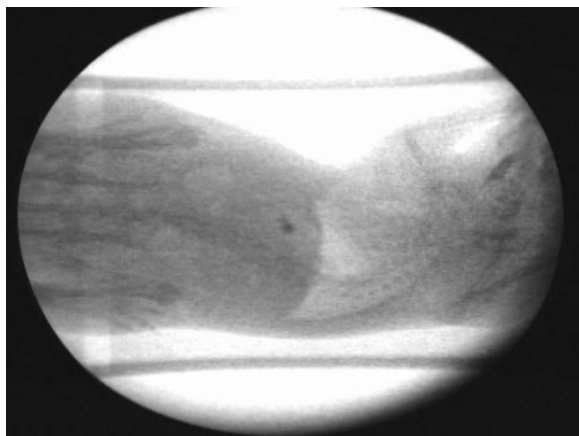


Pre-dosing (BaSO₄/CMC/SAIB)

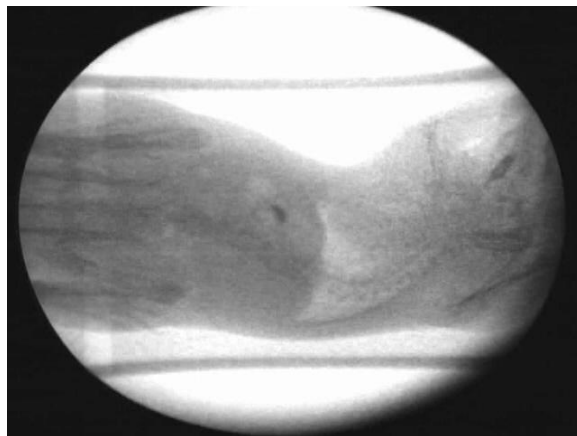


2 min BaSO₄/CMC/SAIB

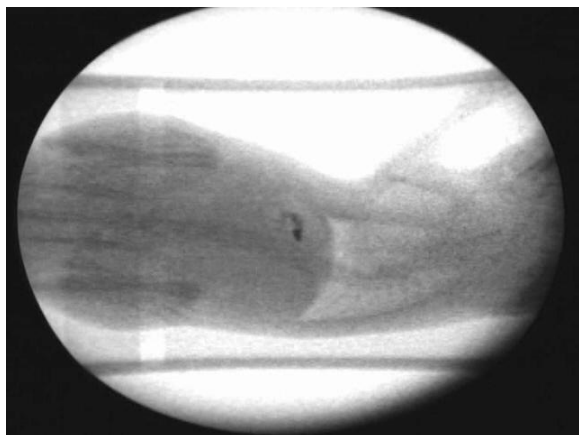
Figure 34. X-ray images showing the BaSO₄/CMC dosage form (top) and BaSO₄/CMC/SAIB (bottom) *in vivo* as a function of time following oral dosing



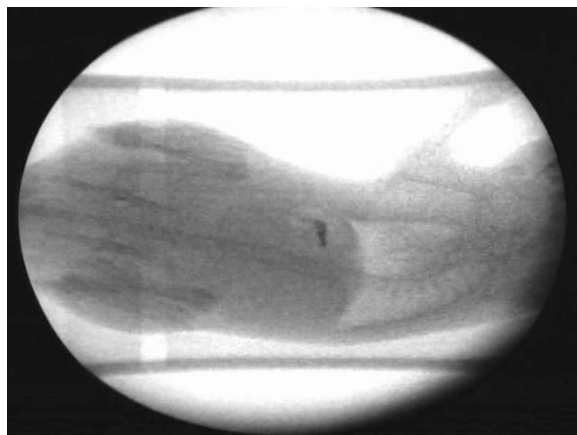
5 min (BaSO_4/CMC)



10 min (BaSO_4/CMC)

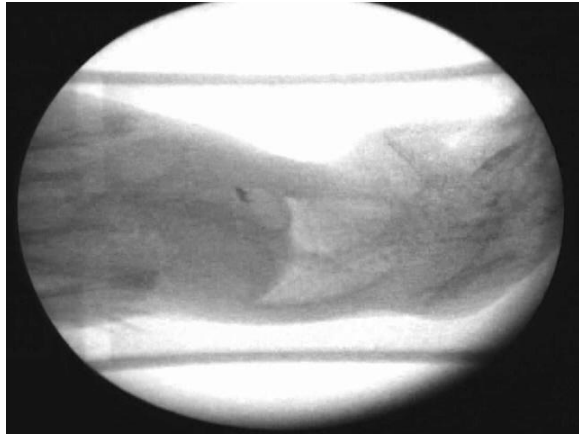


5 min ($\text{BaSO}_4/\text{CMC}/\text{SAIB}$)

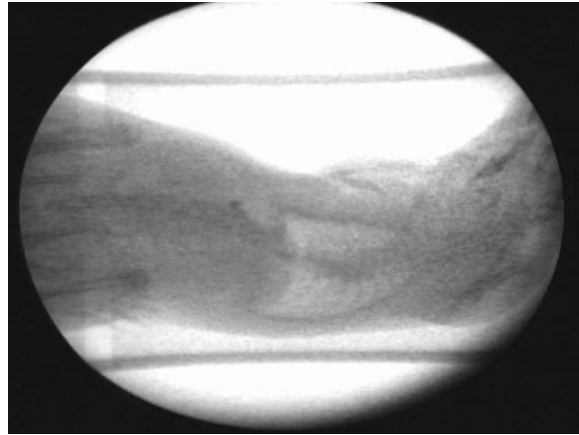


10 min $\text{BaSO}_4/\text{CMC}/\text{SAIB}$

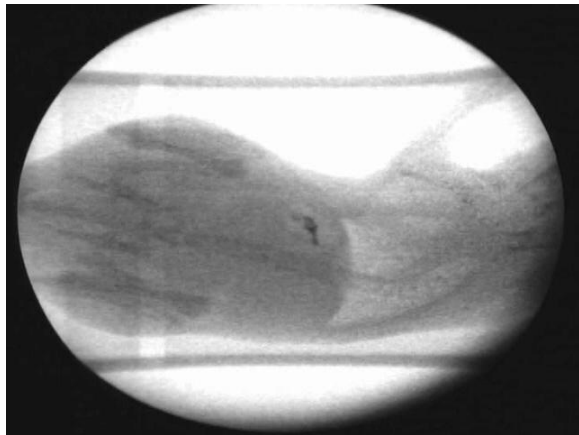
Figure 34 (continued). X-ray images showing the BaSO_4/CMC dosage form (top) and $\text{BaSO}_4/\text{CMC}/\text{SAIB}$ (bottom) *in vivo* as a function of time following oral dosing



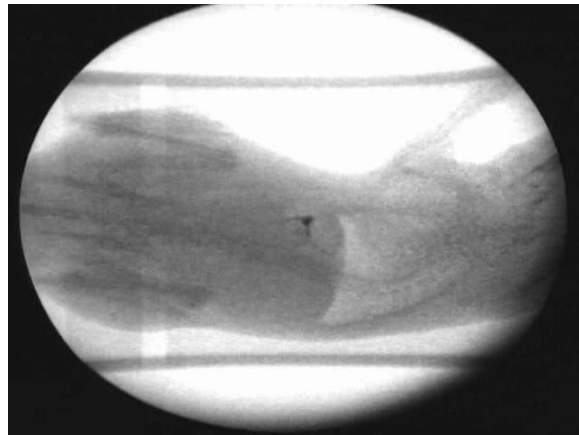
15 min (BaSO_4/CMC)



20 min (BaSO_4/CMC)

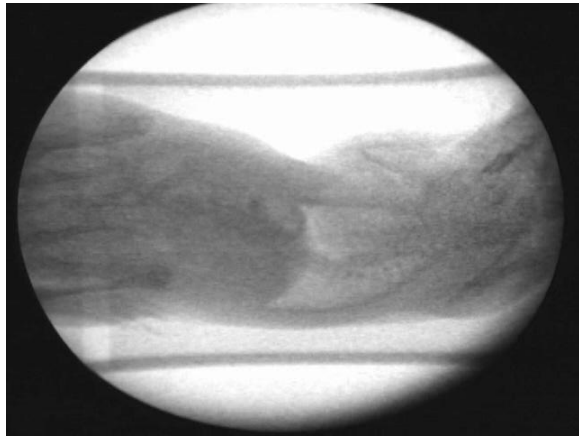


15 min ($\text{BaSO}_4/\text{CMC}/\text{SAIB}$)

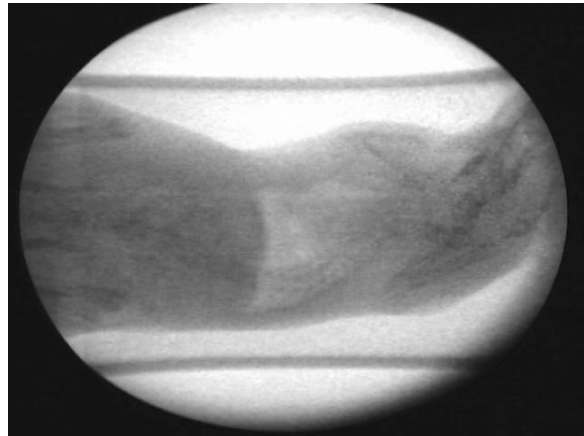


20 min $\text{BaSO}_4/\text{CMC}/\text{SAIB}$

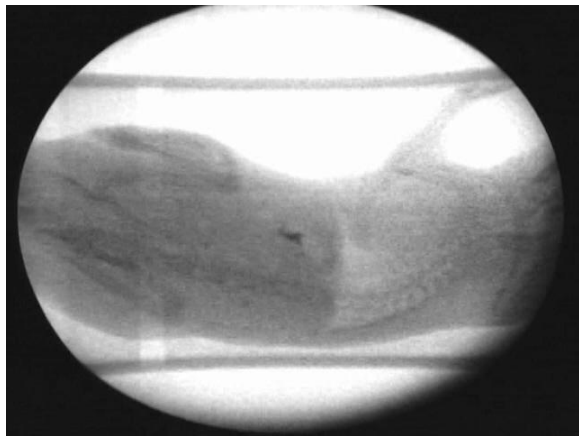
Figure 34 (continued). X-ray images showing the BaSO_4/CMC dosage form (top) and $\text{BaSO}_4/\text{CMC}/\text{SAIB}$ (bottom) *in vivo* as a function of time following oral dosing



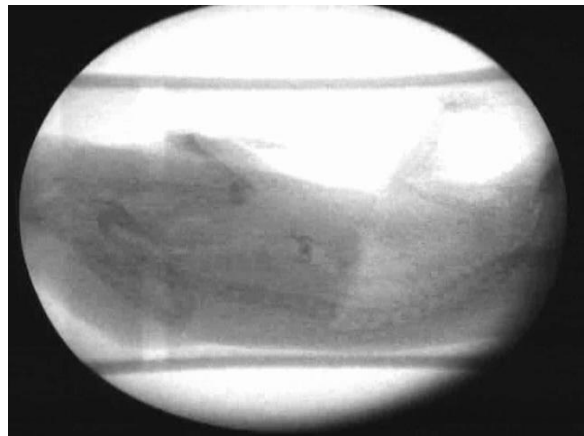
30 min (BaSO_4/CMC)



40 min (BaSO_4/CMC)

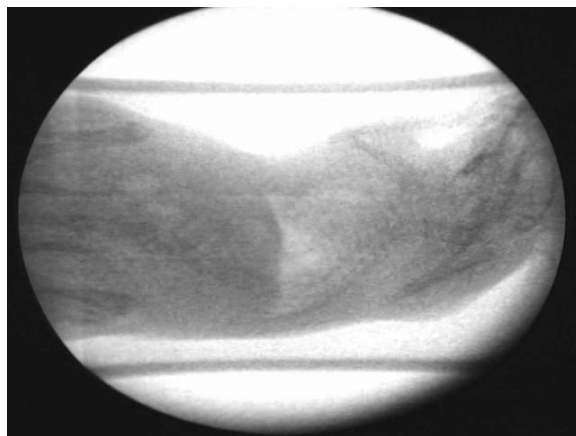


30 min ($\text{BaSO}_4/\text{CMC}/\text{SAIB}$)

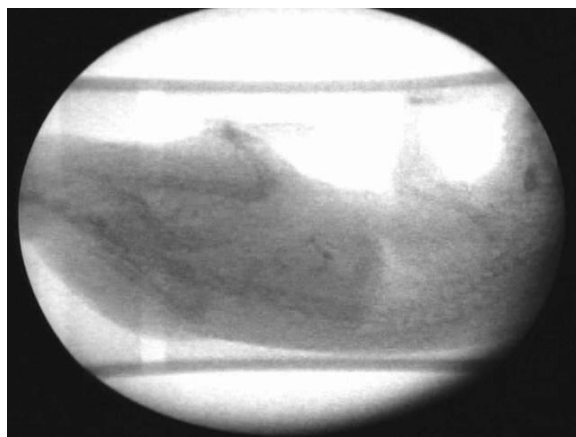


40 min $\text{BaSO}_4/\text{CMC}/\text{SAIB}$

Figure 34 (continued). X-ray images showing the BaSO_4/CMC dosage form (top) and $\text{BaSO}_4/\text{CMC}/\text{SAIB}$ (bottom) *in vivo* as a function of time following oral dosing

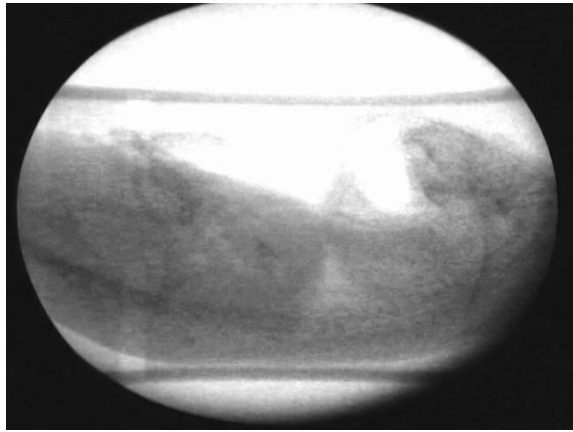


50 min (BaSO₄/CMC)

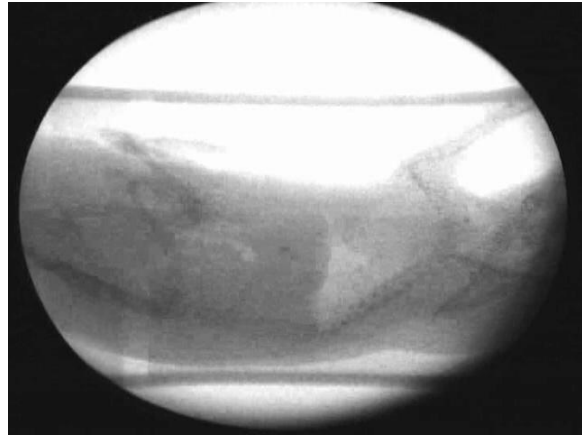


50 min (BaSO₄/CMC/SAIB)

Figure 34 (continued). X-ray images showing the BaSO₄/CMC dosage form (top) and BaSO₄/CMC/SAIB (bottom) *in vivo* as a function of time following oral dosing



75 min (BaSO₄/CMC/SAIB)



100 min BaSO₄/CMC/SAIB

Figure 34 (continued). X-ray images showing the BaSO₄/CMC/SAIB *in vivo* as a function of time following oral dosing

CHAPTER 4

DISCUSSION

As discussed in Chapter 1, the processes involved in oral drug delivery are complex and interdependent. Absorption of a chemical substance into the bloodstream for distribution to various tissues of the body that contain the receptors that stimulate the pharmacologic response cannot occur until the drug has been dissolved in the fluids of the intestinal lumen. The amounts of drug administered and the rates of the processes of drug dissolution, absorption, and distribution must therefore be quantified in order for drug delivery to be understandable and predictable. As the goal of this work was to begin to gain an understanding of how sucrose acetate isobutyrate might be used to control processes involved in oral drug delivery, we focused on understanding various aspects of these processes for several different drug and dosage systems – primarily dissolution and pharmacokinetics.

Dissolution Experiments

Ibuprofen

As described in Chapters 2 and 3, initial dissolution studies were conducted using capsules containing ibuprofen dissolved in a solution of sucrose acetate isobutyrate and ethanol. Figures 8 and 9 show the results of the dissolution experiments conducted on several ibuprofen dosage forms. Figure 8 shows the absorption results of aliquots take from the dissolution apparatus at different times following introduction into the apparatus and Figure 9 shows the calculated ibuprofen concentration. The upper curve in each figure, shown by blue diamonds and dashed line, is that of the control sample – a capsule containing only ibuprofen powder. In this case, the concentration of ibuprofen dissolved in the buffer is dependent upon the kinetics of 2 consecutive processes: (1) the dissolution/disintegration of the capsule and (2) the dissolution of the ibuprofen powder as it is exposed to the buffer solution. As mentioned previously, all dosage forms contained precisely enough ibuprofen to give a 50.0 ppm solution at full dissolution. Clearly, for this dosage form, the ibuprofen powder had been exposed to the buffer within the first 10 minutes after being introduced to the buffer as 35% of the ibuprofen was in solution by that time. Therefore the capsule had dissolved/disintegrated

enough to allow escape of the powder into the buffer solution within the first 10 minutes after introduction to the dissolution bath. At 45 minutes into the dissolution experiment about 85% of the ibuprofen was dissolved, and all of it was in solution within 6 hours. This sample containing only ibuprofen powder thus provided an excellent control to compare with those containing both ibuprofen and sucrose acetate isobutyrate. The dissolution of the dosage form containing 12.5 wt% ibuprofen dissolved in 90%SAIB/10%EtOH is shown by red squares and red dashed line. It would be assumed that release of the ibuprofen/SAIB/EtOH following dissolution/disintegration of the capsule would occur at about the same time as for the control sample. However, 6 hours into the dissolution experiment only about 18.3% of the ibuprofen had been released into solution. Visual inspection of the dosage form revealed that the SAIB remained together and attached to the rotating basket during the experiment. Clearly the diffusion of the ibuprofen out of the SAIB is a relatively slow process. The other 2 curves represent dissolution of dosage forms containing higher concentrations of ibuprofen (16.4 and 22.5 wt%). The amount of ethanol in these 2 dosage forms was higher in order to dissolve more ethanol. The release rates for these 2 dosage forms were obviously virtually the same. The conclusion from these experiments is that, while dissolution of ibuprofen at 6.8 pH is very rapid, its release/dissolution rate can be controlled through design of the dosage form using SAIB/ethanol as an excipient.

Caffeine

The initial dissolution experiments on ibuprofen used ethanol as a solvent for the SAIB and the ibuprofen. Our next experiments, which were conducted on caffeine due to its stability and solubility, did not use ethanol as a cosolvent. Instead, caffeine was compounded with a mixture of SAIB and carboxymethylcellulose (CMC) as described in the experimental section. The results of these experiments are shown in Figures 10 and 11. The absorbance at 275 nm is shown as a function of time in Figure 10. The calibration curve shown in Figure 5 was then used to convert absorbance to concentration, which is plotted in Figure 11. Similar to the observation from the previous study on ibuprofen, caffeine from the dosage form compounded with SAIB and CMC goes into solution more slowly than from the one in which caffeine was compounded with CMC alone. As mentioned earlier, final concentration levels are much higher for the caffeine/CMC/SAIB dosage form than for the caffeine/CMC dosage form because it contained more caffeine (7.45 mg vs. 4.525). Also, concentrations above about 0.2 mg must

be viewed with caution as the calibration curve becomes very flat in this concentration range and small deviations in absorbance can have a very large effect on the calculated concentration. The more interesting results of this experiment are, however, at very early times (<5 min) and low concentrations (<0.2 mg/mL). Clearly, the dosage form containing SAIB releases caffeine much more slowly than the dosage form that does not. In fact, the dissolution curve for the dosage form containing SAIB is concave upward while the dissolution curve for the dosage form that does not contain SAIB is concave downward. One can easily calculate an initial dissolution rate for both dosage forms from the first data point obtained for each. At 1 min following introduction of the dosage forms, the concentration of caffeine for the caffeine/CMC dosage form is 0.11 mg/ml while that of the caffeine/SAIB/CMC dosage form is 0.0006 mg/ml. Even accounting for the difference in the amount of caffeine present in the 2 dosage forms, the dissolution rate of caffeine from the caffeine/CMC dosage form was initially many times faster than the dissolution of caffeine from the caffeine/CMC/SAIB dosage form. Visual observation of the dissolution experiments confirmed that the caffeine/CMC/SAIB dosage forms maintained their integrity longer, as was observed in the ibuprofen dissolution experiments, thus allowing the drug to be released more slowly into the buffer solution. Clearly, by manipulating the relative amounts of drug, CMC, and SAIB one should be able to vary the release rate of the drug from the dosage form and thus control how and to what extent the drug is released in the body.

Pharmacokinetic Experiments

Ibuprofen

Initial pharmacokinetic experiments were performed using ibuprofen as the drug. Instead of using a dosage form containing SAIB compounded with the drug, it was initially decided to feed the rats a diet either containing or not containing SAIB as a component. In this way the rats that were fed a diet containing 4 wt% SAIB would always have SAIB throughout their digestive system while the rats fed a normal diet would not. This would then serve as a test of whether or not SAIB might improve absorption of ibuprofen. As stated earlier, blood samples were analyzed by Mr. James L. Little of Eastman Chemical Company. Figures 12-14

show the results of the pharmacokinetic study. Data were fitted using an open one-compartment model with elimination only for the intravenously dosed rats and using an open one-compartment model with both absorption and elimination for the orally dosed rats.

The calculated pharmacokinetic parameters obtained using GraphPad 5.0 are tabulated in Tables I-III along with R^2 values for each curve fit and mean values and sample standard deviations for each set of rats. It should be noted that the values of the parameters for the rats dosed intravenously are much more reproducible than for the animals dosed orally. Figure 15 shows the results of a one-way analysis of variance and a Tukey-Kramer HSD for the pre-exponential constants for all of the curve fits. The results confirm that there is no difference between $C(0)$, A, or B for the different rat groups, as should be the case. The elimination rate constant, k_{el} , for all 3 rat groups should be the same as well. The same statistical analyses were performed on k_{el} and, as shown in Figure 16, k_{el} is not statistically different for the 3 groups. The same 2 statistical comparisons were performed on the k_a values for the 2 rat groups that were dosed orally, one being fed an SAIB diet and the other fed a normal diet. These results are shown in Figure 17 and indicate that there is no difference in the absorption rate constant for these 2 groups, as should be the case.

The total amount of drug absorbed from an oral dosage is typically calculated as the area under the concentration-time curve and then compared with area under the curve calculated for the intravenously dosed rats. This was done using the trapezoidal rule approach in GraphPad Prism 5.0 and the results are shown in Figure 18 along with the results of the Tukey-Kramer statistical analysis. It was found that there is a statistical difference between the rat group dosed intravenously and the 2 rat groups dosed orally, with the IV-dosed rats absorbing more ibuprofen since all of the drug was injected into the circulatory system. However, there was no statistically significant difference in the total amount of drug absorbed between the 2 rat groups dosed orally and thus the SAIB diet had no discernible effect on drug absorption.

Saquinavir

Because the inclusion of SAIB in the diet had no detectable effect upon absorption of ibuprofen in rats, it was decided to investigate SAIB as an actual component of the dosage form itself and to compare the pharmacokinetics of a drug in rats dosed with such a dosage form to the pharmacokinetics of the drug in rats dosed with a dosage form not containing SAIB.

Saquinavir was chosen as the model drug for this study and the measured blood concentration levels following oral dosing are shown in Figures 19 and 20. Clearly there is a great deal of scatter in the results. It is interesting, however, that high saquinavir blood plasma concentration levels are maintained for both dosage groups for up to 3 days. Figure 21 shows the compiled results with curve fits assuming an open one-compartment model. Again, the scatter is great and no real conclusion can be drawn from the data. If the data within each of the 2 groups are compiled together we get more data points at each time and can analyze it statistically. Calculating standard errors of the mean for each group at each time increment separately leads to Figure 22. Lines are drawn to connect the means and standard errors are shown. The only difference between the 2 curves occurs at longer times where the rats dosed with dosage forms containing SAIB appear to maintain higher blood concentration levels of saquinavir. This result indicates the potential of SAIB as a possible sustained release agent. Further manipulation of the dosage form composition should be carried out to investigate this potential. These data were analyzed further by calculating the areas under the concentration-time curves for each rat and applying a Tukey-Kramer HSD comparison. These results are shown in Figure 23 and show that there is no difference in the areas under the curves for the 2 rat groups. Therefore, we were not able to determine an effect of having SAIB as a component of the dosage form on the total absorption of saquinavir.

Clarithromycin

The most interesting pharmacokinetic study carried out as part of this work was conducted on clarithromycin. As stated previously, an acidic environment such as that found in the stomach is expected to catalyze clarithromycin to dechloroquine clarithromycin. Figures 25 and 26 show the blood concentration levels of clarithromycin and dechloroquine clarithromycin, respectively, following dosing for 2 groups of rats dosed with clarithromycin dosage forms containing SAIB or not. Figure 25 shows that, while there is not statistical difference in blood level concentrations at times up to 6 hours, beyond that time there is a statistically higher blood concentration level of clarithromycin for the rats dosed with dosage forms containing SAIB. This supports the previously discussed saquinavir results that suggested a possible sustained or prolonged release of drug from the dosage forms containing SAIB. Areas under the concentration-time curves were again calculated and compared. The results shown in Figure 27

reveal that, although the pharmacokinetics of clarithromycin is apparently different, the total amount of clarithromycin absorbed by the 2 rat groups cannot be differentiated. The results presented in Figure 26 show that the blood concentration of dechloranose clarithromycin, which would likely be formed in the acidic environment of the stomach, is always higher for the rats dosed with dosage forms containing SAIB. The Tukey-Kramer HSD comparison, as shown in Figure 28, reveals that these rats produced much more of the dechloranose form, suggesting that the SAIB dosage forms may be retained in the stomach for longer periods of time than dosage forms that do not contain SAIB. This could obviously be useful for a drug such as clarithromycin, as it is widely used for treatment of gastric *Helicobacter pylori*, and gastric retention of the drug could maximize exposure to the bacteria and thus be beneficial.

Blood concentrations of the metabolites demethyl clarithromycin and hydroxyl clarithromycin are shown in Figures 30 and 31, respectively. These amounts indicate a portion of the clarithromycin that has been absorbed and metabolized by the body. The Tukey-Kramer HSD comparison for the areas under the curves are shown in Figure 32 and 33 for the demethyl and hydroxy forms, respectively. It was found that the differences between the blood levels resulting from oral dosage via an SAIB-containing dosage form and one not containing SAIB are not statistically significant. However, there is clearly less variation for the SAIB dosage forms. This result was also observed for clarithromycin itself. Apparently, SAIB imparts an improved level of control to the release of clarithromycin in the body therefore rendering concentration levels of the drug itself, as well as the metabolites demethyl clarithromycin and hydroxyl clarithromycin, potentially more predictable.

X-Ray Imaging Experiments

As described earlier, rats were dosed with capsules containing (1) 25%BaSO₄/75%CMC and (2) 25%BaSO₄/25% CMC/50%SAIB. X-ray images were obtained as a function of time after dosing to follow break-up and dispersion of the dosage forms in the gastrointestinal system. The dosage forms are clearly observable following dosing and persist for a variety of durations. However, the length of time that they are observable by x-ray is highly variable and does not seem to be related to whether or not SAIB is a component of the dosage form.

Hence we have not been able to determine a difference in gastrointestinal integrity and longevity for dosage forms prepared with and without SAIB from this work. However, it is important to point out the fact that the dosage form not being observable by this method is not proof that it does not retain some integrity. We do believe that by improvements in the design of dosage forms and consistency in preparation, the longevity of dosage forms containing SAIB may be enhanced.

CHAPTER 5

CONCLUSIONS

These biopharmaceutic and pharmacokinetic studies of sucrose acetate isobutyrate in experimental dosage forms were aimed at determining whether or not SAIB offers advantages over other excipients in the oral delivery of drugs. Several different types of studies have been conducted in an effort to address this question: (1) drug dissolution studies using ibuprofen and caffeine as model drugs directed at determining if SAIB influences the release (liberation) of dissolution of the drug; (2) pharmacokinetic studies in live laboratory rats using ibuprofen, saquinavir, and clarithromycin as model drugs to determine if SAIB affects blood concentration profiles of the drug following administration, e.g. by altering the rate or extent of drug release or absorption; and (3) dosage form imaging studies in live laboratory rats directed at determining whether or not SAIB improves dosage for integrity or promotes retention of the dosage form in the stomach. The results of these studies led to several conclusions regarding the effects of SAIB in oral drug delivery and, in other cases, suggest further potential work to verify the results and conclusions.

The results of dissolution studies of ibuprofen/SAIB/ethanol dosage forms suggest that this system may behave in a way similar to the reservoir system developed by Durect; i.e. once the capsule is dissolved away, the SAIB/ethanol matrix acts as a depot delivery system, slowly releasing ibuprofen over time. By varying the concentrations of drug, SAIB, and ethanol the release rate of the drug can be controlled. The dissolution studies of caffeine/SAIB/CMC (carboxymethylcellulose) and caffeine/CMC dosage forms suggest that the incorporation of SAIB into the dosage form leads to a reduction in the initial release rate of the drug. Further studies in this area should include different ratios of SAIB and CMC to learn more about the effects of this ratio on release rate.

The pharmacokinetic studies of ibuprofen were directed at determining if the presence of SAIB in the gastrointestinal tract of the rats would affect absorption of the drug. It was found that the amount of ibuprofen absorbed was less for the rats fed an SAIB diet than for those fed a normal diet. This suggests that SAIB simply being present in the digestive system at the levels used does not result in enhancement of drug absorption in the case of ibuprofen. Further studies could be directed at other types of drugs. Pharmacokinetic studies of

saquinavir/SAIB/CMC and saquinavir/CMC dosage forms did not reveal a difference in the overall amount of drug absorbed, but data results suggest that higher blood concentration levels of saquinavir may be maintained at long times when SAIB is included as an excipient. Further studies should be conducted to confirm this. Finally, pharmacokinetic studies of clarithromycin/SAIB/CMC and clarithromycin/CMC dosage forms show that dechloranose clarithromycin levels in the blood are much higher in the rats dosed with dosage forms containing SAIB than without. Dechloranose clarithromycin is much more likely to be formed in the acidic environment of the stomach, suggesting that the dosage forms containing SAIB are retained in the stomach for longer periods of time. Further studies should be conducted to confirm this finding. In addition, all of the results for the clarithromycin study show that the clarithromycin blood levels are more controlled, i.e. more reproducible, when SAIB is included in the dosage form.

Finally, the dosage form imaging studies were not conclusive in confirming results from other studies that dosage forms containing SAIB persist for longer periods of time. Though some dosage forms containing SAIB were longer-lived than some without SAIB, this observation was not consistent. It is possible that inconsistencies in dosage form preparation led to this result. Future work should be directed at improving dosage form preparation and making the preparation process more consistent. It could not be determined whether or not SAIB promotes gastric retention of the dosage form.

REFERENCES

- Bai, J. P. F. and Chang, L. L. 1996. Effects of enzyme inhibitors and insulin concentration on transepithelial transport of insulin in rats, *J. Pharm & Pharmac.* 48: 1078-1082.
- Bardonnet, P. L., Faivre, V., Pugh, W. J., Piffaretti, J. C. and Falson, F. 2006. Gastroretentive dosage forms: Overview and special case of *Helicobacter pylori*, *J. Controlled Release* 111: 1-18.
- Blair, M. and Chappel, C. I. 1998. 4-Week range-finding and 1-year oral toxicity studies of sucrose acetate isobutyrate (SAIB) in the cynomolgus monkey, *Food Chem. Tox.* 36: 121-126.
- Chawla, G., Gupta, P., Koradia, V. and Bansal, A. K. 2003 (July). Gastroretention: A means to address regional variability in intestinal drug absorption, *Pharm. Tech.*, 50-68.
- Chiang, M., Gray, K. and Chappel, C. I. 1998. Effect of sucrose acetate isbutyrate (SAIB) ingestion on the hepatobiliary function of normal human male and female volunteers, *Food Chem. Tox.* 36:141-144.
- Collnot, E-M., Baldes, C., Wempe, M., Kapl, R., Httermann, J., Hyatt, J. A., Edgar, K. J., Schaefer, W. F. and Lehr, C-M. 2007. Mechanism of inhibition of P-glycoprotein mediated efflux by vitamin E TPGS: Influence on ATPase activity and membrane fluidity, *Mol. Pharmaceutics* 4: 465-474.
- Constantinides, P. P. and Wasan, K. M. 2006. Lipid formulation strategies for enhancing intestinal transport and absorption of P-glycoprotein (P-gp) substrate drugs: *In vitro/In vivo* case studies, *J. Pharm. Sci.* 96: 235-248.
- Conway, B. R. 2005. Drug delivery strategies for the treatment of *Helicobacter pylori* infections, *Curr. Pharmaceut. Des.* 11: 775-790.

- Crespi, C. L., Fox, L., Stocker, P., Hu, M. and Steimel, D. 2000. Analysis of drug transport and metabolism in cell monolayer systems that have been modified by cytochrome P450A4 cDNA expression, *Eur. J. Pharm. Sci.* 12: 6368.
- Cummins, C. L., Jacobsen, W., Christians, U. and Benet, L. Z. 2004. CYP3A4-transfected caco-2 cells as a tool for understanding biochemical absorption barriers: Studies with sirolimus and midazolam, *J. Pharm. Exp. Ther.* 308: 143-155.
- Fournier, R. L. (2007) *Basic Transport Phenomena in Biomedical Engineering*, Taylor & Francis, New York, NY.
- Gibson, J. W., Sullivan, S. A., Middleton, J.C. and Tipton, A. J. Sustained release formulations, U.S. Patent 6,413,536, July 2, 1999.
- Hatefi, A. and Amsden, B Biodegradable injectable in situ forming drug delivery systems, *J. Control. Release* 2002; 80: 9-28.
- Hensley, W. J. 1975. A brief report on the use of sucrose acetate isobutyrate in human volunteers, Unpublished report, Royal Prince Alfred Hospital, Australia.
- Hoffman, A., Stepensky, D., Lavy, E., Eyal, S., Klausner, E. and Friendman, M. 2004. Pharmacokinetic and pharmacodynamic aspects of gastroretentive dosage forms, *Inter. J. Pharmaceut.* 277: 141-153.
- Huisman, M. T, Smit, J. W. Wiltshire, H. R., Hoetelmans, R. M. W., Beijnen, J. H. and Schinkel, A. H. 2001. P-glycoprotein limits oral availability, brain, and fetal penetration of saquinavir even with high doses of ritonavir, *Mol. Pharmacol.* 59: 806-813.
- Huisman, M. T, Smit, J. W. Wiltshire, H. R., Hoetelmans, R. M. W., Beijnen, J. H. and Schinkel, A.H. 2003. Assessing safety and efficacy of directed p-glycoprotein inhibition to improve the pharmacokinetic properties of saquinavir coadministered with ritonavir, *J. Pharmacol. Exp. Thera.* 304: 596-602.

- Kokate, A., Li, X. and Jasti, B. 2008. Effect of drug lipophilicity and ionization on permeability across the buccal mucosa: a technical note, *AAPS PharmSciTech.* 9: 501-504.
- Li, L., Zhao, L. and Yalkowsky, S. H. 1999. Combined effect of cosolvent and cyclodextrin on solubilization of nonpolar drugs, *J. Pharm. Sci.* 88: 1107-1111.
- Lu, Y., Tang, Y.L., Cui, Y., Zhang, Y., Qin, F. and Lu, X. 2008. In vivo evaluation of respiridone-SAIB *in situ* system as a sustained release delivery system in rats, *Eur. J. Pharm. Biopharm.* 68: 422-429.
- Mackenzie, K. M., Henwood, S. M., Tisdell, P. J., Boysen, B. G., Palmer, T. E. Scardein, J. L., West, A. J. and Chappel, C. I. 1998. Sucrose acetate isobutyrate (SAIB): Three-generation reproduction study in the rat and teratology studies in the rat and rabbit, *Food Chem. Tox.* 36: 135-140.
- Mackenzie, K. M., Tisdell, P. J., Hall, R. L., Boysen, B. G., Field, W. E. and Chappel, C. I. 1998 Oral toxicity and carcinogenicity studies of sucrose acetate isobutyrate (SAIB) in the Fischer 344 rat and B6C3F₁ mouse, *Food Chem. Tox.* 36: 111-120.
- Matschke, C., Isele, U., van Hoogevest, P. and Fahr, A. 2002. Sustained-release injectables formed *in situ* and their potential use for veterinary products, *J. Controlled Release* 85: 1-15.
- Mancheras, P. and Illiadis, A. (2005) *Modeling in Biopharmaceutics, Pharmacokinetics, and Pharmacokinetics: Homogeneous and Heterogeneous Approaches*, Springer-Verlag, New York, NY.
- Morgareidge, K. 1965. Subacute (90 day) feeding studies with SAIB in dogs. Unpublished report No. 86501, Food and Drug Research Laboratories, Inc. Maspeth, NY.

- Myhr, B. C., Cifone, M. A., Ivett, J. L., Lawlor, T. E. and Young R. R. 1998. Lack of genotoxic effects of sucrose acetate isobutyrate, *Food Chem. Tox.* 36: 127-134.
- Okumu, F. (Genenech, Inc.), Sustained release formulations, U.S. Patent 6992065, January 31,2006. (PCT International Applic. No. WO 01/78683, April 29, 2001.)
- Okumu, F.W., Dao, Le N., Fielder, P.J., Dybdal, N., Brooks, D., Sane, S. and Cleland, J.L. 2002. Sustained delivery of human growth hormone from a novel gel system: SABER™ *Biomaterials* 23: 4353-4358.
- Orr, J. M., Marier, G. and Chappel, C. I. 1976. Fourteen day feeding and tolerance study of sucrose acetate isobutyrate (SAIB) in human volunteers, Unpublished report no. 4479, Bio-Research Laboratories Ltd., Pointe Claire, Quebec.
- Palm, K., Luthman, K., Ros, J., Grasjo, J. and Artursson, P. 1999. Effect of molecular charge on intestinal epithelial drug transport: pH-dependent transport of cationic drugs, *J. Pharma. & Exper. Ther.* 291: 4345-443.
- Proctor, B. G. and Chappel, C. I. 1998. Subchronic toxicity studies of sucrose acetate isobutyrate (SAIB) in the rat and dog, *Food Chem. Tox.* 36: 101-110.
- Reddy, M. B., Yang, R. S. H., Clewell, H. J. III and Anderson, M. E. (2006) *Physiologically Based Pharmacokinetic Modeling: Science & Applications*, Wiley, Hoboken, NJ.
- Reynolds, R. C. 1998. Metabolism and pharmacokinetics of sucrose acetate of sucrose acetate isobutyrate (SAIB) and sucrose octaisobutyrate (SOIB) in rats, dogs, monkeys or humans: a review, *Food Chem. Tox.* 36: 95-99.
- Reynolds, R. C. and Chappel, C. I. 1998 Sucrose acetate isobutyrate (SAIB): Historical aspects of its use in beverages and a review of toxicity studies before 1988, *Food Chem. Tox.*36: 81-93.

- Ritschel, W. A. and Kearns, G. L. (2004) *Handbook of Basic Pharmacokinetics...including Clinical Applications*, 6th Ed., American Pharmacists Association, Washington, DC.
- Shargel, L., Wu-Pong, S. and Yu, A. B. C. (2005) *Applied Biopharmaceutics & Pharmacokinetics*, 5th Ed., McGraw-Hill, New York.
- Sullivan, S.A., Gibson, J.W., Burns, P.J., Franz, L., Squires, E.L., Thompson, D.L. and Tipton, A.J. 1998. Sustained release of progesterone and estradiol from the *SABER*[™] delivery system: *In vitro* and *in vivo* release rates, paper #572, The 25th International Symposium on Controlled Release of Bioactive Materials, Park, K. and Potts, K.O. Chairs, Controlled Release Society, Inc., Las Vegas, NE.
- Streubel, A., Siepmann, J. and Bodmeier, R. 2006. Drug delivery to the upper small intestine using gastroretentive technologies, *Curr. Opin. Pharm.* 6: 501-508.
- Streubel, A., Siepmann, J. and Bodmeier, R. 2006. Gastroretentive drug delivery systems, *Expert Opin. Drug Deliv.* 3: 217-233.
- Tennant, R.W., Margolin, B.H., Shelby, M. D., Zeiger, E., Haseman, J.K., Spalding, J., Caspary, W., Resnick, M., Stasiewicz, S., Anderson, B. and Minor, R. 1987. Prediction of chemical carcinogenicity in rodents from *in vitro* genetic toxicity assays, *Science* 236, 933-941.
- Tipton, A. J. and Holl, R.J. (1996) High viscosity liquid controlled delivery system, U.S. Patent 5,747,059.
- Tipton, A.J. and Dunn, R.L. 2000. In situ gelling formulations, *Sustained-Release Injectable Products*, Senior, J. & Radomsky, M., Eds., Taylor & Francis, Philadelphia.
- Tipton, A. J. and Ewing, J. L. High viscosity liquid controlled delivery system, PCT International Applic. No. WO 96/39995, December 19, 1996.

- Tipton, A. J. (2003) Sucrose acetate isobutyrate (SAIB) for parenteral delivery, *Modified-Release Drug Delivery Technology*, Marcel Dekker, New York.
- Tozer, T. N and Rowland, M. (2006) *Introduction to Pharmacokinetics and Pharmacodynamics: The Quantitative Basis of Drug Therapy*, Lippincott, Williams & Wilkins, Baltimore, MD.
- Tucker, G. T. Houston, J. B. and Huang, S-M. 2001. Optimizing drug development: Strategies to assess drug metabolism/transporter interaction potential – towards a consensus, *Pharm. Res.* 18: 1071-1080.
- Van de Waterbeemd, H., Lennernäs, H. and Artursson, P. (2003) *Drug Bioavailability: Estimation of Solubility, Permeability, Absorption & Bioavailability*, Wiley-VCH, Weinheim, Germany.
- Wempe, M.F. 2006. Eastman Chemical Company restricted internal report.
- Wibawa, J. I. D., Shaw, P. N. and Barrett, D. A. 2003. Quantification of clarithromycin, its 14-hydroxy and decladinose metabolites in rat plasma, gastric juice and gastric tissue using high-performance liquid chromatography with electrochemical detection, *J. Chromat. B* 783: 359-366.
- Yusuf, S., Sleight, P., Pogue, J., Bosch, J., Davies, R. and Dagenais, G. 2000. Effects of angiotensin converting enzyme inhibitor, ramipril, on cardiovascular events in high-risk patients, *N. Engl. J. Med.* 342: 145-153.

VITA

MARTIN RAY TANT

Personal Data	Date of Birth: April 10, 1953 Birthplace: Seneca, South Carolina
Education	Old Dominion University, Norfolk, Virginia B.S. Chemistry 1975 Virginia Tech, Blacksburg, Virginia M. S. Chemical Engineering 1979 Virginia Tech, Blacksburg, Virginia Ph.D. Chemical Engineering 1986 East Tennessee State University, Johnson City, Tennessee M. S. Biomedical Sciences 2011
Professional Experience	Chemist, Naval Mine Engineering Facility Yorktown, Virginia 1975-1976 Graduate Teaching Assistant, Virginia Tech Blacksburg, Virginia 1976-1978 Graduate Research Assistant, Virginia Tech Blacksburg, Virginia 1978-1979 Chemical Engineer, Naval Surface Warfare Center Dahlgren, Virginia 1979-1982 Pratt Presidential Engineering Fellow, Virginia Tech Blacksburg, Virginia 1982-1985 Cunningham Dissertation Year Fellow, Virginia Tech Blacksburg, Virginia 1985-1986 Senior Research Engineer, Dow Chemical Company Freeport, Texas 1986-1988 Senior Research Chemical Engineer, Eastman Kodak Company Kingsport, Tennessee 1988-1993 Principal Research Chemical Engineer & Research Associate Eastman Chemical Company Kingsport, Tennessee 1993-Present