Development of a Mixed-Effect Pharmacokinetic Model for Vehicle Modulated *In Vitro* Transdermal Flux of Topically Applied Penetrants

JASON T. CHITTENDEN,[1] JAMES D. BROOKS,[1] JIM E. RIVIERE[2]

[1]Center for Chemical Toxicology Research and Pharmacokinetics, College of Veterinary Medicine, North Carolina State University, Raleigh, North Carolina 27607

[2]College of Veterinary Medicine, Coles 207, Department of Anatomy and Physiology, Kansas State University, Manhattan, Kansas 66506-5802

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ABSTRACT: Transient flux profiles from *in vitro* flow-through cell experiments exhibit different characteristics depending upon the properties of the penetrants and vehicle mixtures applied. To enable discrimination of the chemical properties contributing to these differences, a consistent mathematical model should first be developed. A mixed effects modeling framework was used so that models can be estimated with as few parameters as possible, while also quantifying variability and accounting for correlation in the data. The models account for diffusion and binding within the membrane as well as dynamics on the diffusion coefficient. The models explain key features of the data, such as: lag time, sharp peaks in flux, two terminal phases, and low flux profiles. The models with dynamic diffusivity fit the data better than those without—particularly the sharp peaks. The significance of changing diffusivity over time suggests that vehicle effects are transient and are more accurately estimated when dynamics are modeled. © 2014 Wiley Periodicals, Inc. and the American Pharmacists Association J Pharm Sci 103:1002–1012, 2014

Keywords: transdermal drug delivery; skin; passive diffusion/transport; mathematical model; *in vitro* models; formulation vehicle; diffusion

INTRODUCTION

For a topical exposure to a chemical or drug to result in a systemic exposure, the chemical (penetrant) must be absorbed across the dermal barrier. Whether in industrial or medical settings, the penetrant, in its pure form, will rarely come into contact with the skin. There is usually a vehicle that serves to solubilize the penetrant. In many cases, the permeability of the penetrant through the various layers of the skin is modulated by the constituents of the vehicle to the extent that it is the mixture that defines the ultimate level of exposure to the penetrant. Because of this interaction between the vehicle–penetrant mixture and the dermal barrier, there is marked interest in vehicle and penetrant interactions with respect to transdermal absorption.

The rate and extent of transdermal absorption depends on several characteristics of the penetrant: its partitioning into skin, diffusivity through the skin, and exposure at the skin surface.[1,2] Several studies have demonstrated the effects of vehicles on the absorption rates of topically applied compounds, by modulation of these characteristics. The permeability of halogenated methanes has been shown to be increased by up to 73-fold in corn oil compared with water.[3] In a variety of cases, the partition coefficient of the penetrant into the stratum corneum has been reported to be effected by vehicles ranging from oil, water emulsion, and petrolatum[4] to propylene glycol (PropGlyc), octonol, and ethyl decanoate[5] to isopropylmyristate.[6] Terpenes have been shown to enhance the diffusivity of 5-fluorouracil in excised epidermal membranes.[7]

Much progress has been made toward predicting transdermal absorption. Several models have been developed that take into account the chemical properties of the penetrant to describe its permeability coefficient. The original work of Potts and Guy,[8] based on a linear free-energy relationship (LFER) among molecular weight, lipophilicity, and permeability, has been further refined in more recent literature. Bunge and Cleek[9,10] have divided the Potts and Guy model into components representing resistances due to stratum corneum and epidermis. Hostynek and Magee[11] developed an LFER utilizing indicator variables to account for vehicle effects and also accounting for hydrogen bonding activity. In a comprehensive review of related literature, Abraham and Martins[12] rigorously developed a model across all of the data, utilizing descriptors for molecular size, hydrogen bonding potentials, polarity, and refractivity. An important limitation of this work, however, was that all of the compounds were exposed in an aqueous vehicle.

In the case of industrial and pharmaceutical exposure, one should also consider the effects of complex mixtures on absorption, where the multiple constituents of the vehicle may have cumulative or synergistic effects. Previous studies have demonstrated the effects of multicomponent mixtures on the rate and extent of transdermal absorption.[13–15] In particular, these metrics have been shown to be modulated by the specific composition of the vehicle. Riviere and Brooks[14,16] have used a mixture factor to account for the effect of composition of the mixture on permeability coefficient in an *in vitro* system, and total absorbed dose fraction in an *ex vivo* system. The mixture factor in these studies is a multivariate linear function that combines

Correspondence to: Jim E. Riviere (Telephone: +919-515-1912; Fax: +919-515-1909; Email: jriviere@ksu.edu)

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chemical descriptors of the penetrant and vehicle constituents in a quantitative structure to property relationship (QSPR). When applied as an additive factor to the existing LFERs, the mixture factor improves the predictability of the QSPR, demonstrating the effect of the vehicle composition.[17]

Previous analyses of these, and similar, data have modeled a summary statistic of the individual absorption experiments (like permeability coefficient or fraction absorbed). That approach may lose information about how the mixture affects the absorption of the penetrant. That is, one would like to isolate the effect to modifications in partition, diffusivity, or solubility of the penetrant. The difference would be seen in the ability to predict the time course of absorption.

In a new approach to the analysis of these data, a mixed effects modeling framework is applied to simultaneously estimate parameters using all of the data within a given experimental system, while modeling the full time course of absorption. This improves the accuracy of parameter estimates by accounting for random variations, and the commonality of parameters, between experimental units. In addition, data have been collected in two different membrane systems, which should allow for the identification of common dynamics in the system not related to the membrane such as binding within the apparatus, evaporation of the vehicle, and protein binding in the receptor well.

METHODS AND MATERIALS

Experimental Apparatus

Membrane and transdermal absorption data have been collected in two experimental systems: silastic membrane flow-through diffusion cell (FTDC); porcine skin FTDC. Both experimental systems share a common set of 12 chemical penetrants in (up to) 24 vehicle combinations that are topically applied. Each system should allow for the isolation and quantification of certain dynamics of the diffusional flux. This work examines several mathematical models for the flow-through systems to accurately describe the dynamics and extent of mass transfer with the goal of explaining them in terms of penetrant and vehicle effects.

The silastic or porcine skin is punched from prepared materials (1.6 and 1.9 cm diameter, respectively) and placed into a Bronough FTDC.[18] A combination of penetrant and vehicle totaling 20 μL is applied in the donor well on the surface of the membrane. Perfusate, containing bovine serum albumin, is pumped into the receptor well at a nominal rate of 4 mL/h. The receptor well is approximately 4 mm deep and the radius of the cell is 0.45 cm. Perfusate is sampled with a frequency of every 15 min through 120 min, then every 60–480 min. The penetrant mass in perfusate is assayed by scintillation counter and converted to an observed value of flux as percent of dose per hour. Further experimental details for the porcine skin diffusion experiment are described elsewhere,[19] and the complementary silastic data are presented here for the firsttime.

Penetrants

Table 1 lists the C^{14}-tagged chemicals used as penetrants in the studies. The abbreviations shown are used in this paper.

Table 1. Chemicals Used in Treatments

Abbreviation	Chemical Name
AZ	Atrazine
CP	Chlorpyrifos
EP	Ethylparathion
FN	Fenthion
MP	Methylparathion
NP	Nonylphenol
PC	Pentachlorophenol
PH	Phenol
PN	ρ-Nitrophenol
PZ	Propazine
SZ	Simazine
TZ	Triazine

Table 2. Vehicles Used in Treatments

Eth	PG
Eth+MNA	PG+MNA
Eth+MNA+SLS	PG+MNA+SLS
Eth+PG	PG+SLS
Eth+PG+MNA	W
Eth+PG+MNA+SLS	W+MNA
Eth+PG+SLS	W+MNA+SLS
Eth+SLS	W+PG
Eth+W	W+PG+MNA
Eth+W+MNA	W+PG+MNA+SLS
Eth+W+MNA+SLS	W+PG+SLS
Eth+W+SLS	W+SLS

Eth, ethanol; MNA, methyl nicotinate; W, water; PG, propolyne glycol; SLS, sodium laureth sulfate.

Vehicles

The dosing vehicles are combinations of five constituents: ethanol (EtOH), water, PropGlyc, methyl nicotinate (MNA), and sodium lauryl sulfate (SLS). Table 2 shows the combinations of these components as used in the experimental procedures.

Modeling Approach

The goal in modeling these data is to capture the dynamics of the flux profiles in parameters that can be related to physicochemical properties of the treatments. Such parameters are likely to be mixture properties of the chemical components of the treatments, and as such should relate to a physical, as opposed to empirical, model of the membrane system. Despite the differences in the physical systems between the two membrane systems studied, silastic and porcine skin, they are modeled simultaneously to allow for shared mechanics such as boundary effects in the donor and receptor wells. This approach should add some power in isolating and identifying boundary versus membrane mechanics.

A common model is developed that describes all of the data. Each replicate in each treatment has its own unique set of parameters. The similarity of replicates within a treatment should generate similar parameters, which will correlate to chemical properties of the treatment. Finding one model that describes all of the observed data while capturing treatment effects in the parameters is the goal of this endeavor.

The model must describe three primary shapes of flux profiles. Figure 1 shows an example of each. The atrazine in ethanol profile (AZ+EtOH) is typical of many of the profiles