Risk Analysis, Vol. 14, No. 3, 1994

Significance of the Dermal Route of Exposure to Risk Assessment

David R. Mattie,[1,2] John H. Grabau,[1] and James N. McDougal[1]

Received December 9, *1993*

The skin is a route of exposure that needs to be considered when conducting a risk assessment. It is necessary to identify the potential for dermal penetration by a chemical as well as to determine the overall importance of the dermal route of exposure as compared with inhalation or oral routes of exposure. The physical state of the chemical, vapor or liquid, the concentration, neat or dilute, and the vehicle, lipid or aqueous, is also important. Dermal risk is related to the product of the amounts of penetration and toxicity. Toxicity involves local effects on the skin itself and the potential for systemic effects. Dermal penetration is described in large part by the permeability constant. When permeability constants are not known, partition coefficients can be used to estimate a chemical's potential to permeate the skin. With these concepts in mind, a tiered approach is proposed for dermal risk assessment. A key first step is the determination of a skin-to-air or skin-to-medium partition coefficient to estimate a potential for dermal absorption. Building a physiologically-based pharmacokinetic (PBPK) model is another step in the tiered approach and is useful prior to classical *in vivo* toxicity tests. A PBPK model can be used to determine a permeability constant for a chemical as well as to show the distribution of the chemical systemically. A detailed understanding of species differences in the structure and function of the skin and how they relate to differences in penetration rates is necessary in order to extrapolate animal data from PBPK models to the human. A study is in progress to examine anatomical differences for four species.

KEY **WORDS:** Skin; partition coefficients; permeability constants.

1. INTRODUCTION

1.1. Background

The skin, constituting about 10% of total human body weight, acts as the major interface between the homeostatic internal environment of the body and the comparatively unregulated and potentially hostile external environment. The skin primarily functions as a protective barrier that restrains entry of chemical substances into the body. The potential for occupational or accidental skin exposure to nonvolatile and volatile chemicals (both of which may penetrate the barriers of the skin) requires an experiment-based understanding of chemical absorption through the skin to adequately determine risks of such exposures.

Personnel working in an occupational environment are often exposed to a variety of chemicals. Maintenance, repair and fueling operations expose workers to engine oils, lubricants, fuels, hydraulic fluids, paints and solvents. All of these types of compounds present a potential for dermal exposure. Up to 40% of all occupational illness may involve the skin.[1] For some substances, cutaneous absorption is a major contributor to overall exposure.[2] The absorbed total uptake of xylene from hand skin contact with solvent mixtures for 15 min was greater than that from inhalation over a full 8-hr shift in auto body repair shops.[3] The dermal route was found

[1] Toxicology Division, Armstrong Laboratory, Wright-Patterson Air Force Base, Ohio 45433-7400.
[2] To whom all correspondence should be addressed.

0272-4332/94/0600-0277$07.00/1 © 1994 Society for Risk Analysis

to be the major contributor to total polychlorinated biphenyls body burden of transformer maintenance and repair personnel.[4] Gloves are of limited protection, as permeation of chemicals through glove material is known to occur.[5] Absorption of chemicals through the skin now appears to be of greater significance than previously suggested by industry or epidemiological experience.[6] The dermal route of exposure may not always be the most important route, but it may often contribute significantly to total exposure. For a highly fat soluble chemical such as dibromomethane, the body burden from dermal penetration compared with inhalation was approximately 6% in one rat study.[7] If respiratory protection were worn but the skin was exposed, absorption of this chemical vapor would still occur. A method to compare dermal vapor exposure to inhalation exposure at the same concentration has been described as a ratio of input functions for the contribution of each route of exposure, provided that the permeability constant, surface area of skin exposed, and alveolar ventilation rate are known or can be determined.[8]

Chemicals in the liquid state must also be considered because many exposure chemicals exist as a neat liquid or dissolved in a liquid medium such as water. Concentrations of pure liquid are much greater than in their vapor form. This results in greater total penetration through the skin. Even though the concentration on the skin is different between a vapor and the liquid form of a chemical, the solubility of a chemical in the skin should not be affected once the chemical enters the skin unless the liquid form of the chemical alters the skin barrier. Tsuruta[9] and others have reported on the percutaneous absorption of organic solvents. Morgan *et al.*[10] demonstrated that significant amounts of volatile organic chemicals (VOCs) can be absorbed through the skin during dermal exposure of rats to low levels of this class of chemical in aqueous solutions. Absorption to neat chemical did not appear to result in the same absorption rate as chemical in aqueous solution. Permeability constants were not determined by Morgan *et al.*,[10] but peak blood levels after neat chemical exposure were approximately an order of magnitude greater than after chemical in aqueous solution. Estimation of the significance of dermal absorption of VOCs from aqueous solutions based on data for pure liquids may not provide an accurate assessment of actual exposure levels.

Additional studies in this area have looked at chloroform, a VOC that contaminates chlorine-treated municipal tap water.[11] Individuals are, therefore, exposed to chloroform while showering with chlorine-treated tap water. In situations where water should not be consumed due to contamination with VOCs, individuals should also consider avoiding bathing with the water.

Dermal risk is a function of exposure penetration and toxicity. A toxic chemical that cannot penetrate the skin may be limited to local toxic effects on the skin. A chemical with a relatively low toxicity potential that readily penetrates the skin and enters circulation may have systemic effects or produce target organ toxicity. Therefore, it is necessary to know the capacity of a chemical for percutaneous absorption in order to assess its overall potential risk.

2. ESTIMATES OF DERMAL PENETRATION

Various methods have been used to measure the potential of a chemical to penetrate the skin. The permeability constant (K_p) of a chemical is a quantitative expression of the capacity of a chemical to enter and diffuse through the skin. Permeability constants are used to predict the absorption rate or flux, which is the mass of chemical absorbed per unit area of skin per unit time. The solubility of a chemical in skin (partition coefficient [PC]) is an important parameter for determining the permeability constant when the diffusion coefficient for skin is known.

Flux, or rate of penetration of a chemical across the skin, is determined by concentration at the skin surface, the surface area exposed, and solubility of chemical in the skin.[12,13] Skin-to-air PC values for a chemical are a measure of the solubility of the chemical in skin and should correlate with the permeability constant as shown in the following equation for flux:

$$\text{Flux} = \frac{Dk_mC}{l} = k_pC$$

where Flux is mg/cm²/hr, D is the diffusion constant of the chemical in the skin (cm²/hr), k_m is the solubility or PC of the chemical in skin (unitless), C is the concentration of chemical on the surface of the skin (mg/cm³), l is the skin thickness (cm), and k_p is the permeability constant (cm/hr).

Physical and chemical properties of chemicals such as solubility are important descriptors of skin penetration.[6,14,15] The PC for skin, a measure of the affinity of a chemical for skin tissue, is the ratio of concentrations at equilibrium between the tissue and an adjacent medium, such as air, water or other environmental vehicle. Various experimental methods have been reported in the literature for determining PC values for skin. One

method uses the octanol/water PC as a surrogate for partitioning between the skin (octanol phase) and the environment or vehicle (water phase).[16,17] Octanol/water PC values are typically determined by shaking the test compound in a mixture containing equal parts of water and octanol. After sufficient time for equilibration to occur, the ratio of the amount of test compound in each solvent is determined.[16] Hawkins and Reifenrath[18] compared octanol/water PC values to the percent of applied dose of pesticides and steroid hormones after exposure *in vitro* through pig and human skin. Kasting *et al.*[17] used octanol/water PC values in a mathematical model to estimate the flux of chemicals across the skin. Berner *et al.*[19] used octanol/water PC values to confirm skin permeation rates for a series of chemicals prior to examining the relationship between the pKa of these chemicals and acute skin irritation. Octanol/water PC values have been used to estimate dermal flux for setting a skin notation guideline for a threshold limit value-time weighted average.(") Although the octanolhater PC has been used extensively in estimating dermal penetration, it is an oversimplification of the process of chemical interaction with the skin. The octanol/water PC assumes that skin is homogenous with respect to octanol and that water is the environmental medium.

Surber *et al.*[15] measured stratum corneum (SC)/water and SC/isopropyl myristate PC values. In their study, PCs were determined as a function of equilibration time, initial concentration of drug in the vehicle, delipidization of stratum corneum, and source and preparation of stratum comeurn. The PCs were considered as predictors of percutaneous penetration for the purpose of conducting dermal risk assessments.[15]

3. TIERED APPROACH

A tiered approach is proposed for determining the potential hazard of a chemical for dermal risk assessment as shown in Fig. 1. This approach employs toxicity tests in an orderly sequence that can be stopped at various levels depending on the application and potential for exposure of the chemical, potential for full-scale development of the system of intended use, initial toxicity results, etc.

The first phase is conducted completely with *in vitro* tests and structure–activity comparisons. Dermal PCs are proposed as an important first step at this level. A procedure for determining skin:air PC values was developed in this laboratory and will be summarized in this paper. Exposure assessment is also an important early component of the tiered approach. Knowing the physical form of the chemical, the expected concentration, and possible environmental medium are essential in planning the appropriate tests to conduct initially as well as throughout the tiered approach.

The second phase involves acute *in vivo* toxicity studies such as a dermal limit test. The Phase I screen is used to eliminate as many chemicals as possible in order to decrease the number of animal studies.

Another relatively early step in the tiered approach is the development of a physiologically-based pharmacokinetic (PBPK) model with a skin compartment. The use of PBPK models will also be discussed, and an example of their use is presented in this paper. Physiologically-based pharmacokinetic model development spans *two* levels of the tiered approach because development of a model involves *in vivo* procedures. A PBPK model could still be developed without completing all of the endpoints for a Phase II screen. Completion of a PBPK model and short-term dermal exposure studies represents the Phase III screen.

Phase IV is the screening phase for genotoxicity and carcinogenicity. Completion of this phase would provide a comprehensive hazard assessment of potential dermal risk. It is possible that *in vitro* genotoxicity testing will need to be conducted prior to the completion of earlier phases.

4. SKIN:AIR PARTITION COEFFICIENTS

The headspace method for PC determination, developed by Sato and Nakajima[21] and modified by Gargas *et al.*,(=)has been used extensively in this laboratory for determining PCs of a variety of biological tissues. However, the methodology *was* not adequate for measuring the skin:air PC. A modification in the preparation of skin for the headspace method was developed in order to measure skin:air PC values.

Dibromomethane was used as a prototype for skin:air PCs. Male Fischer 344 (F-344) rats (Charles River Laboratories) were between 8 and 16 weeks old at the time the skin was collected for PC determination. Clipped dorsal skin was collected and cut into 1 x 0.5 cm strips. The pieces of skin were placed on the walls of scintillation vials (24.65 ml volume) without saline. Sample vials containing skin and the corresponding empty reference vials were injected with an equal concentration of chemical vapor. At equilibrium, vapor from the headspace of the sample and reference vials were measured on a gas chromatograph with a flame ionization detector.

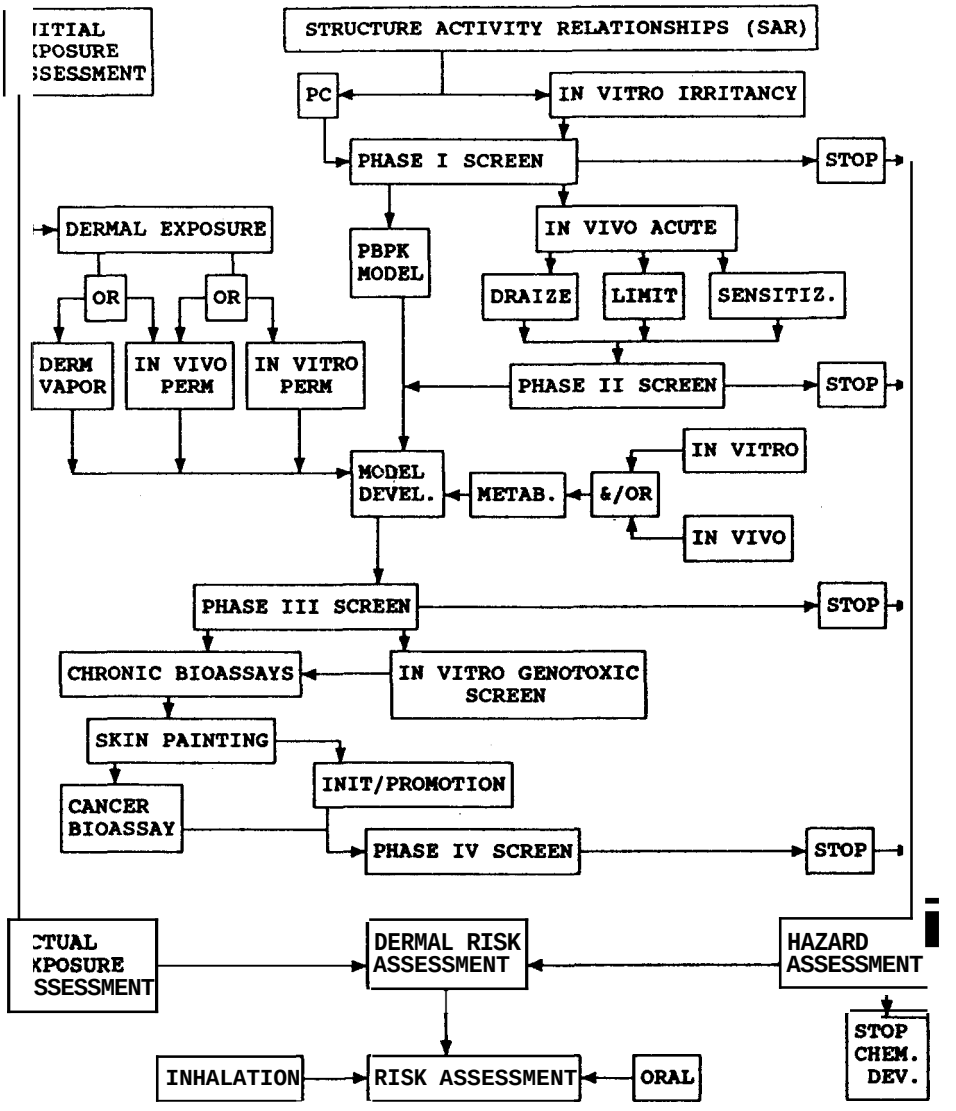

Fig. 1. Tiered approach to dermal risk assessment.

After measuring the amount of chemical as area counts from all of the reference and sample vials in a set, sample vials were compared with corresponding reference vials using the following equation modified from Gargas *et al.* [22]:

$$PC = \frac{(\text{reference vial area counts})(\text{vial volume}) - (\text{skin sample area counts})(\text{vial volume} - \text{sample volume})}{(\text{skin sample area counts})(\text{sample volume})}$$

After developing the technique for determining a skin:air PC value using dibromomethane, the procedure was used for other VOCs of interest (Table I).

The skin:air partition coefficient for dibromomethane and selected VOCs is shown in Table I. Approximately 19 samples were analyzed for each chemical. The most common equilibration time for this group of volatile chemicals was 4 hr. The skin:air PC values ranged from 1.9 for hexane to 91.9 for styrene. The octanol/water PC values[23] were compared with the corresponding skin:air PC values for 11 of the above chemicals. There was no correlation ($r^2 = 0.09$) between the octanol/water PC values and skin:air PC values (Fig. 2). Permeability constants were available for nine of the chemicals for which skin:air PC values were measured in this study.[7,13,24] There was good correlation (? = 0.93) between permeability constants and skin:air PC values (Fig.

Table I. Rat Skin:Air Partition Coefficients for Selected Volatile Organic Chemicals

Chemical	Skin:air PC (± SE)	N	Equilibration time (hr)
Dibromomethane	68.3 ± 3.1	10	4
Perchloroethylene	41.5 ± 1.2	16	4
Trichloroethylene	31.8 ± 1.5	19	4
Benzene	34.5 ± 1.9	19	4
Hexane	1.9 ± 0.1	18	4
Toluene	43.0 ± 1.8	16	4
Xylene	50.4 ± 1.7	24	2
Styrene	91.9 ± 6.8	20	3
Methylene chloride	13.6 ± 0.5	17	2
Carbon tetrachloride	12.4 ± 0.6	24	4
Methyl chloroform	10.8 ± 0.6	18	4
Halothane	10.6 ■ 0.7	17	3
Isoflurane	4.5 ■ 0.3	16	6

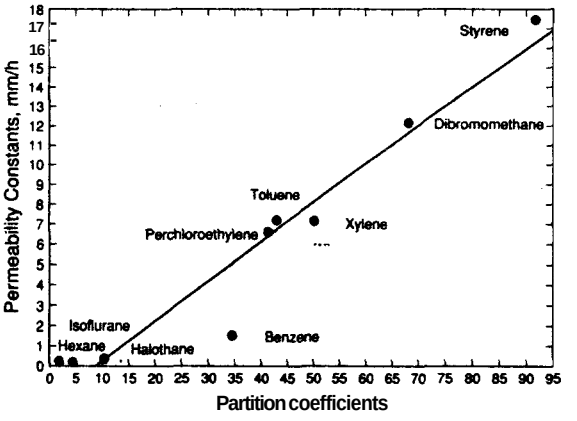

Fig. 3. Comparison of rat skin:air partition coefficients with rat permeability constants.

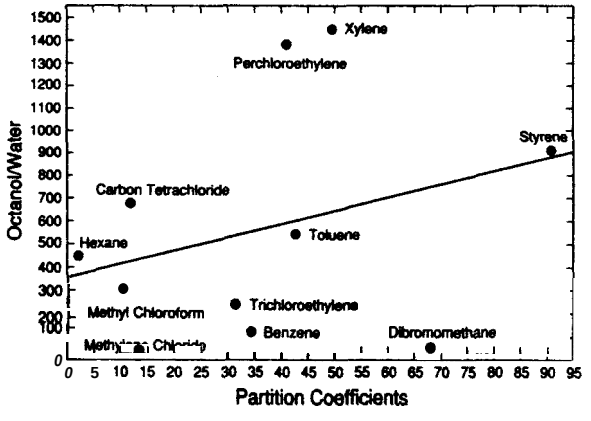

Fig. 2. Comparison of skin:air partition coefficients with octanol/water PC values.

3). When octanol/water PC values were compared directly to eight of the permeability constants (minus the octanol/water PC for isoflurane), the correlation was also poor ($r^2 = 0.04$). If a saline:air or water:air PC value is determined for a chemical, a skin:saline or skin:water PC value can be calculated for the chemical by dividing the skin:air PC value by the saline:air or water:air PC value. Comparison of octanol/water PC values with skin:saline PC values still resulted in a poor correlation ($r^2 = 0.20$).

The skin:air PC values were compared to both octanol/water PC values and permeability constants. Octanol/water PC values have been used as a qualitative measure of skin permeability.[16–18,25,26] Skin:air PC values for the chemicals tested showed a correlation with permeability constants but did not show as good a correlation with octanolhater PC values. Octanol/water PC values for the VOCs examined in this study appeared to be poor indicators of the solubility of these chemicals in skin. No single bulk solvent, such as octanol, precisely mimics the solvent properties of the stratum corneum transport barrier.[25] In addition, the skin:air PC values were determined for chemicals with poor water solubility. The predictive ability of octanol/water PC values is most likely lower for these volatile chemicals because octanol/water PC values are based on water representing the vehicle or environmental medium. The data in this study suggest that skin:air PC values are a better indicator of the relative skin permeability for the volatile chemicals examined in this study. Skin:saline PC values would be representative of permeability into skin from an aqueous environmental medium. Determining a skin:air PC or skin:saline PC is proposed as an initial screen to identify the potential for skin absorption of volatile chemicals with unknown permeability constants.

5. PBPK MODELING

In addition to indicating potential permeability, skin PCs are necessary €or developing the dermal compartment in a PBPK model. PBPK models mathematically describe the dynamics of chemicals in the body, including permeability of membranes and partitioning of chemicals into tissues. A PBPK model is developed by grouping various tissue types together based on similar blood flows and PCs. Each compartment has a measured physiological blood flow. Model parameters are determined from

laboratory studies or literature values and can be changed to extrapolate across species. Absorption, distribution, metabolism and elimination of a chemical are then mathematically described for each compartment which has such a process. The skin:air PC is essential for the rate equation in the dermal compartment describing the uptake of chemical from air into the skin. A skin:saline PC value is calculated, as described above, for the rate equation for uptake into skin from an aqueous medium. A PBPK model with a dermal compartment can then be utilized to determine the permeability constant for a chemical. The difference in concentration, surface area and exposure duration between the laboratory and an actual occupational situation can also be described using a PBPK model. In addition, metabolism of the chemical, which may be quantitatively or qualitatively different between experimental species and humans, can be estimated with existing methods and their impact on penetration described using a PBPK model. PBPK modeling provides the means to relate laboratory animal exposures to the human situation by extrapolating across exposure concentrations, routes of exposure and species.[8] Accurate extrapolation from animal exposures to personnel in the workplace will provide the means to more quickly and efficiently set safe, but not overly restrictive, dermal exposure standards.

For a PBPK model to accurately estimate a permeability constant for a chemical in skin, a number of conditions are important. A PBPK model with a dermal compartment must be validated based on exposure for a second route of exposure, such as the inhalation route. Skin PC data should be experimentally determined for the dermal compartment. Actual dermal exposures should be conducted in order to measure the uptake of chemical into the blood. The concentration of chemical in blood after dermal exposure is then used in model simulations to estimate the permeability constant for that chemical.

Previous work with PBPK models in this laboratory has demonstrated their usefulness in extrapolation and the risk assessment process.[27-33] PBPK models were developed based on the work of McDougal *et al.*,[7,13,24] which described each of three different *in vivo* dermal exposures in rats: whole body dermal exposure to benzene vapor,[13] exposure to neat benzene from a closed cell on the dorsal skin (unpublished data and Ref. 10), and exposure to saturated solutions of benzene in water also from a closed cell.[10] The models were used to estimate the permeability constants of benzene from blood concentrations achieved during exposure to each form of the chemical. The estimated permeability constant for dermal vapor was 0.152 cm/hr, for neat benzene 0.0025 cm/hr, and for aqueous solutions 0.05 cm/hr. The physical form of the chemical and the presence of water resulted in different rates of absorption. The permeability constant for rat skin from aqueous solutions was one half the human permeability constant value used for dermal risk assessment, 0.111 cm/hr.[34] Rat skin has been reported to be more permeable than human skin by a factor of two to four,[8] so the rat permeability constant was expected to be at least twice as high as the human value for benzene.

6. SPECIES DIFFERENCES IN SKIN PENETRATION

In an attempt to better understand factors affecting dermal penetration and to be able to better extrapolate between animal species and humans, a study was initiated *to* quantitate selected anatomical differences in skin from a number of animal species. Anatomical differences that may affect permeability include density and size of hair follicles, density of sebaceous and apocrine glands, capillary density and distance from the surface, as well as thickness of the various layers of the epidermis and dermis. Anatomical differences in skin between species will be compared to permeability constants for three model chemicals to determine possible correlations between structure and permeability. Permeability constants will be determined using PBPK models for chloropentafluorobenzene, perfluoroheptane and dichlorobenzene.

Sections of dorsal skin were collected from IAF/HA hairless and Hartley guinea pigs, fuzzy and F-344 rats, B6C3F1 and Crl:SKH1 hairless mice, and farm pigs. Pieces of skin were processed at the same time and under identical conditions for standard histopathology sections in paraffin. One set of sections was stained with hematoxylin and eosin and another set with Massons trichrome. Image analysis was conducted on sections from each strain using an image analysis system. Parameters measured were thickness (stratum Corneum, stratum granulosum, viable epidermis and total epidermis); average depth and distribution of capillaries, venules and arterioles; surface area of each type of blood vessel relative to basement membrane of the epidermis; and depth and surface area of hair follicles and sebaceous glands relative to the basement membrane of the epidermis.

Exploratory data (Table 11) showed that the hairless guinea pig and farm pig have the thickest epidermal layers and the F-344 rat and the mouse the thinnest epidermal layers. There was a wide range for average depth of capillaries, venules and arterioles with the hairless guinea pig and mouse having capillaries and venules closer to the epidermis and the F-344 rat having all ves-

Table II. Anatomical Parameters in Skin from Four Animal Species (N=3)

	Mouse		Guinea Pig		Rat		Swine
	SKH1	B7C3F1	Hartley	Hairless	Fuzzy	F-344	farm
Total epidermis	26.4 ± 1.7	13.2 ± 1.5	26.8 ± 0.7	59.6 ± 3.9	47.0 ± 3.0	20.9 ± 2.2	52.2 ± 7.2
Capillary depth	299 ± 39	421 ± 55	446 ± 149	333 ± 48	519 ± 138	803 ± 83	511 ± 13
Veinule depth	389 ± 19	536 ± 20	903 ± 218	610 ± 59	723 ± 188	970 ± 57	623 ± 87
Arteriole depth	479 ± 20	410 ± 109	602 ± 275	743 ± 298	715 ± 89	1340 ± 63	792 ± 123
Follicle volume[a] ($\times 10^{-3}$)	1.7 ± 0.4	1.2 ± 0.6	1.5 ± 0.1	2.5 ± 0.1	2.0 ± 0.5	0.5 ± 0.1	6.4 ± 2.0
Gland volume[a] (x	1.4 ± 0.2	1.0 ± 0.3	0.3 ± 0.1	1.1 ± 0.2	2.4 ± 0.3	1.0 ± 0.2	5.1 ± 2.5

[a] Ratio of follicle or sebaceous gland area to total area.

sels farther away from the epidermis. The farm pig had the greatest volume of hair follicles and sebaceous glands; F-344 rat the least follicular volume and Hartley guinea pig the least gland volume.

Additional skin samples will be analyzed to confirm these preliminary data. A PBPK model will be built for each of the three strains showing the widest variation in anatomical parameters. Exploratory information suggests the use of F-344 rats and Hartley and hairless guinea pigs. A better understanding of the anatomical differences of skin in animal species may lead to a better extrapolation of animal skin data to human skin.

7. SUMMARY

The dermal route is an important potential route of exposure. There is still much research to be conducted to understand the skin and its significance in risk assessment.

ACKNOWLEDGMENTS

We would like to thank George Bates, Jill Solscheid, Peggy Parish, Jerry Nicholson and Gary Jepson for their technical assistance.

The animals used in this study were handled in accordance with the principles in the *Guide for the Care and Use of Laboratory Animals*, prepared by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Animals Resources, National Research Council, DHHS, National Institute of Health Publication no.#86-23, 1985, and the Animal Welfare Act of 1966, as amended.

REFERENCES

1. K. F. Wheeler, "Barrier Lotions, Along With Gloves, Can Help Deter Occupational Dermatitis," *Occupational Health and Safety* Jan., 60–61 (1992).
2. G. Scansetti, G. Piolatto, and G. F. Rubino, "Skin Notation in the Context of Workplace Exposure Standards," *Amer. J. Indust. Med.* 14, 725–732 (1988).
3. W. Danieli, A. Stebbins, D. Kalman, J. F. O'Donnell, and S. W. Horstman, "The Contributions to Solvent Uptake by Skin and Inhalation Exposure," *Am. Ind. Hyg. Assoc. J.* 53,124129 (1992).
4. P. S. J. Lees, M. Corn, and P. Breysse, "Evidence for Dermal Absorption as the Major Route of Body Entry During Exposure of Transformer Maintenance and Repairmen to PCBs," *Am Ind. Hyg. Assoc. J.* 48,257–264 (1987).
5. J. L. Perkins and V. B. Knight, "Risk Assessment of Dermal Exposure to Polychlorinated Biphenyls Permeating a Polyvinyl Chloride Glove," *Am. Ind. Hyg. Assoc. J.* 50, A-171–172(1989).
6. P. Grandjean, A. Berlin, M. Gilbert, and W. Penning, "Preventing Percutaneous Absorption of Industrial Chemicals: The 'Skin' Denotation," *Amer. J. Indust. Med.* 14, 97–107 (1988).
7. J. N. McDougal, G. W. Jepson, H. J. Clewell III, and M. E. Andersen, "Dermal Absorption of Dihalomethane Vapors," *Taricol. AppL Pharmacol.* 19, 150–158 (1985).
8. J. N. McDougal and H. J. Clewell 111, "Dermal to Inhalation Extrapolation for Organic Chemicals," in T. R. Gerrity and C. J. Henry (eds.), *Principles of Route-to-Route Extrapolation for Risk Assessment* (Elsevier Science Publishing, New York, 1990), pp. 313–317.
9. H. Tsuruta, "Percutaneous Absorption of Organic Solvents. 1. Comparative Study of the *In Vivo* Percutaneous Absorption of Chlorinated Solvents in Mice," *Indust.* Health 13,227–236 (1975).
10. D. L. Morgan, S. W. Cooper, D. L. Carlock, J. J. Sykora, B. Sutton, D. R. Mattie, and J. N. McDougal, "Dermal Absorption of Neat and Aqueous Volatile Organic Chemicals in the Fischer 344 Rat," *Environ. Res. 55,* 51-63 (1991).
11. W. K. Jo, C. P. Weisel, and P. J. Lioy, "Chloroform Exposure and the Health Risk Associated with Multiple Uses of Chlorinated Tap Water," *Risk Analysis* 10, 581–585 (1990).
12. G. L. Flynn, S. H. Yalkowsky, and T. J. Roseman, "Mass Transport Phenomena and Models: Theoretical Concepts," *J. Pharm. Sci.* 63, 479–510 (1974).
13. J. N. McDougal. G. W. Jepson, H. J. Clewell III, M. L. Gargas, and M. E. Andersen, "Dermal Absorption of Organic Chemical Vapors in Rats and Humans," *Fundam.* Appl. *Taricol.* 14, 299–308 (1990).
14. L. K. Pershing, L. D. Lambert, and K. Knutson, "Partition Coefficient and Solubilities of Estradiol in a Variety of Vehicles Predict the *In Vivo* Flux Across the Human Skin Sandwich Flap," *Clin. Res.* 37, 727A (1989).

15. C. Surber, K.-P. Wilhelm, H. I. Maibach, L. L. Hall, and R. H. Guy, "Partitioning of Chemicals into Human Stratum Corneum: Implications for Risk Assessment Following Dermal Exposure," *Fund. Appl. Tox.* **15, 99–107 (1990).**
16. R. L. Bronaugh and E. R. Congdon, "Percutaneous Absorption of Hair Dyes: Correlation with Partition Coefficients," *J. Invest. Dermatol.* **83, 124–127 (1984).**
17. G. B. Kasting, R. L. Smith, and E. R. Cooper, "Effect of Lipid Solubility and Molecular Size on Percutaneous Absorption," *Phamacol. Skin* **1, 138–153 (1987).**
18. G. S. Hawkins and W. G. Reifenrath, "Influence of Skin Source, Penetration Cell Fluid, and Partition Coefficient on *In Vitro* Skin Penetration," *J. Pharmaceutical Sci.* **75, 378–381 (1986).**
19. B. Berner, D. R. Wilson, R. H. Guy, G. C. Mazzenga, F. H. Clark, and H. I. Maibach, "The Relationship of pK_a and Acute Skin Irritation in Man," *Pharmaceutical Res.* **5, 660–663 (1988).**
20. V. Fiserova-Bergerova, J. T. Pierce, and P. O. Droz, "Dermal Absorption Potential of Industrial Chemicals: Criteria for Skin Notation," *Amer. J. Indust. Med.* **17, 617635 (1990).**
21. A. Sato and T. Nakajima, "Partition Coefficients of Some Aromatic Hydrocarbons and Ketones in Water, Blood, and Oil," *Brit. J. Indust. Med. 36,* **231–234 (1979).**
22. M. L. Gargas, R. J. Burgess, D. E. Voisard, G. H. Cason, and M. E. Andersen, "Partition Coefficients of Low-Molecular-Weight Volatile Chemicals in Various Liquids and Tissues," *Toxicol. Appl. Phamacol.* **98, 87–99 (1989).**
23. A. Leo, C. Hansch, and D. Elkins, "Partition Coefficients and Their Uses," *Chem. Rev.* **71, 525–616 (1971).**
24. J. N. McDougal, G. W. Jepson, H. J. Clewell 111, M. G. MacNaughton, and M. E. Andersen. "A Physiological Pharmacokinetic Model for Dermal Absorption of Vapors in the Rat," *Toxicol. AppL Phamacol.* **85, 286–294 (1986).**
25. B. D. Anderson and P. V. Raykar, "Solute Structure-Permeability Relationships in Human Stratum Corneum," *J. Invest. Dermatol.* **93, 280–286 (1989).**
26. N. E. Tayar, R.-S. Tsai, B. Testa, P.-A. Carrupt, C. Hansch, and A. Leo, "Percutaneous Penetration of Drugs: A Quantitative Structure-Permeability Relationship Study," *J. Pharmaceutical Sci.* **80, 744–749 (1991).**
27. M. E. Andersen, H. J. Clewell III, M. L. Gargas, F. A. Smith, and R. H. Reitz, "Physiologically Based Pharmacokinetics and the Risk Assessment Process for Methylene Chloride," *Taricol. Appl. Phannacol.* **87, 185–205 (1987).**
28. H. J. Clewell III and M. E. Andersen, "Risk Assessment Extrapolations and Physiological Modeling," *Toxicol. Ind. Health.* **1, 111–131 (1985).**
29. H. J. Clewell III and M. E. Andersen, "Improving Toxicology Testing Protocols Using Computer Simulations," *Taricol. Lett.* **49, 139–158 (1989).**
30. R. A. Corky, A. L. Mendrala, F. A. Smith, D. A. Staats, M. L. Gargas, R. B. Conolly, M. E. Andersen, and R. H. Rietz, "Development of a Physiologically Based Pharmacokinetic Model for Chloroform," *Tox. Appl. Phan 103,* **512–527 (1990).**
31. J. W. Fisher, T. A. Whittaker, D. H. Taylor, H. J. Clewell 111, and M. E. Andersen, "Physiologically Based Pharmacokinetic Modeling of the Pregnant Rat: A Multiroute Exposure Model for Trichloroethylene and Its Metabolite, Trichloroacetic Acid," *Toxicol. AppL Pharmacol.* **99, 395–414 (1989).**
32. M. L. Gargas, M. E. Andersen, and H. J. Clewell III, "A Physiologically Based Simulation Approach for Determining Metabolic Constants from Gas Uptake Data," *Toxicol. AppL Pharmacol.* **86, 341–352 (1986).**
33. H. R. Reitz, J. N. McDougal, M. W. Himmelstein, R. J. Nolan, and A. M. Schumann, "Physiologically-Bad Pharmacokinetic Modeling with Methylchloroform: Implications for Interspecies, High Dose/Low Dose, and Dose Route Extrapolations," *Toxicol. Appl. Pharmacol.* **95, 185–199 (1988).**
34. I. H. Blank and D. J. McAuliffe, "Penetration of Benzene Through Human Skin," *J. Invest. Dermatol.* **85, 522–526 (1985).**