0022-202X/84/8301-0051$02.00/0
THE JOURNAL OF INVESTIGATIVE DERMATOLOGY, 83:51–56, 1984

Vol. 83, No. 1
Printed in U.S.A.

Aryl Hydrocarbon Hydroxylase, Epoxide Hydrolase, and Benzo[a]-pyrene Metabolism in Human Epidermis: Comparative Studies in Normal Subjects and Patients with Psoriasis*

DAVID R. BICKERS, M.D., HASAN MUKHTAR, PH.D., TAPU DUTTA-CHOUDHURY, M.S., CYNTHIA L. MARCELO, PH.D., AND JOHN J. VOORHEES, M.D.

Department of Dermatology, Case Western Reserve University and Veterans Administration Medical Center (DRB, HM, TD-C), Cleveland, Ohio, and University of Michigan Medical School (CLM, JJV), Ann Arbor, Michigan, U.S.A.

Prior studies have shown that human skin possesses a cytochrome P-450-dependent microsomal enzyme that is capable of metabolizing drugs and polycyclic aromatic hydrocarbon (PAH) carcinogens. This study characterized benzo[a]pyrene (BP) metabolism in human epidermis of normal and psoriatic individuals. The basal level of the cytochrome P-450-dependent microsomal enzyme aryl hydrocarbon hydroxylase (AHH) and epoxide hydrolase (EH) were measured in freshly keratomed epidermis from 12 normal individuals and from uninvolved skin sites of 12 patients with psoriasis. The induction response of AHH following the in vitro addition of the PAH benz[a]anthracene (BA) was also assessed. The basal activity (mean ± SE) of AHH in normal epidermis was 62.1 ± 5.6 units (fmol 3-hydroxybenzo[a]pyrene, 3-OH-BP/min/mg protein) whereas the activity in uninvolved skin of psoriatic individuals was 62.9 ± 5.1 units (NS), Epoxide hydrolase activity was 25.1 ± 1.1 (pmol BP 4,5-diol/min/mg protein) units in normal epidermis and 24.8 ± 2.1 units in epidermis from patients with psoriasis (NS). Following addition of BA (100 μM), in vitro, AHH activity in normal epidermis increased by a mean value of 165% whereas activity in nonlesional epidermis of psoriatic individuals increased 320%. Kinetic studies in normal epidermis revealed that the AHH reaction was linear up to 60 min and to 50 μg protein, had a pH optimum of 7.4, and the K_m for BP was 0.62 μM. High-performance liquid chromatography (HPLC) confirmed that the pattern of metabolism of BP was quite similar in epidermal microsomes prepared from normal and psoriatic individuals, insofar as the formation of diols, phenols, and quinones was concerned. These studies indicate that human epidermis is capable of metabolizing BP and that there is no significant difference between normal individuals and patients with psoriasis insofar as basal AHH activity or total BP metabolism is concerned. Furthermore, the epidermal enzyme system in patients with psoriasis has a greater responsiveness to environmental PAH than does that of normal individuals.

The skin is the major interface between the body and its environment and is exposed to a variety of potentially toxic chemical and physical agents. Some of these agents, such as ultraviolet radiation and selected PAH are known to be oncogenic for cutaneous tissue of humans and experimental animals [1,2]. In recent years it has become clear that the skin is not simply a passive structural barrier impeding the penetration of environmental agents, but rather this tissue possesses enzymes capable of metabolizing a variety of xenobiotics [3]. This metabolic capacity may be a major determinant of selected toxic responses such as chemical carcinogenesis. Major insights into the mechanism of chemical carcinogenesis have developed from the studies of the Millers who suggested that oncogenic chemicals first undergo metabolic alteration by membrane-bound cytochrome P-450-dependent monooxygenases [4]. These enzymes convert an inert procarcinogen into reactive electrophiles that can bind covalently to cellular macromolecules thereby initiating tumorigenesis [4,5]. In the case of PAH such as BP, 3 successive enzymatic steps are required for the generation of the ultimate oncogenic metabolite [6]. These enzymatic steps include AHH, which forms an epoxide, followed by EH which cleaves the epoxide into dihydrodiols. The dihydrodiol is then converted into diol-epoxides by AHH. These have been identified in the skin of tumor-susceptible as well as tumor-resistant mice following topical application of BP but are generally more inducible in the former [7–9]. AHH has been studied in organ cultures of human foreskin and its activity was shown to be inducible following in vitro incubation with BA [10]. Bickers and Kappas [11] subsequently showed that topical application to human skin of therapeutic coal tar solution, a mixture which contains numerous PAH, caused substantial induction of cutaneous AHH activity. Human skin microsomes have been shown to possess EH activity [12].

Extensive studies in inbred mouse strains have shown that AHH activity and inducibility is a dominantly inherited trait and that inbred strains with inducible enzyme activity are more susceptible to the induction of skin tumors following topical application of PAH using standard initiation-promotion techniques [13]. These findings led to the hypothesis that AHH inducibility and susceptibility to skin cancer by the PAH may be related [14].

Recently, several studies have appeared in which epidermal AHH activity in patients with the dermatologic disease known as psoriasis was compared to that of normals and found to be decreased [15,16]. On the basis of these findings it was proposed that patients with psoriasis have defective AHH activity/inducibility and (a) that this defect could be somehow related to a fundamental biochemical abberration of the skin in this disease and (b) that patients with psoriasis are resistant to skin cancer from environmental chemicals because of the deficient

Manuscript received December 5, 1983; accepted for publication February 15, 1984.

Supported in part by NIH Grants ES-1900, AM 26009, and funds from the Veterans Administration.

* This was presented in part at The Society for Investigative Dermatology, April 1982, and appears in abstract form in Clin Res 30:260, 1982.

Reprint requests to: David R. Bickers, M.D., Department of Dermatology, Veterans Administration Medical Center, 10701 East Boulevard, Cleveland, Ohio 44106.

Abbreviations:
AHH: aryl hydrocarbon hydroxylase
BA: benz[a]anthracene
BP: benzo[a]pyrene
EH: epoxide hydrolase
HPLC: high-performance liquid chromatography
PAH: polycyclic aromatic hydrocarbons
THF: tetrahydrofuran

activity of AHH. Epidemiologic studies do not support this concept. Stern et al [17] have shown that patients with psoriasis who are intensively treated for many years with topically applied coal products and ultraviolet radiation are at substantially increased risk for the development of skin cancer than are individuals who are not so treated. Rawlins and Shuster [18] have reappraised their earlier observations and questioned the veracity of their earlier studies. In a recent publication, Finnen et al were unable to measure AHH activity in human epidermis obtained by the suction blister method [19].

In this study, we have measured AHH and EH activity in keratomed epidermis obtained from uninvolved skin of psoriatic individuals and compared it to epidermis obtained from age- and sex-matched normal nonpsoriatic controls. The inducibility of AHH was assessed by using a short-term organ culture system in which keratomed epidermis was incubated with a PAH. In addition, we also assessed the patterns of BP metabolism by skin microsomes of normal and psoriatic individuals. Our data indicate that there are no detectable differences between normal and psoriatic individuals in the basal and induced levels of these enzymes in human epidermis. Furthermore, enhanced inducibility of AHH by BA was observed in psoriatic epidermis as compared to normal epidermis.

MATERIALS AND METHODS

Epidermal Sampling

Twelve patients with plaque type psoriasis involving at least 20% of their body surface were studied at the University of Michigan Medical Center. Twelve normal individuals without any demonstrated dermatologic disease in whom no unusual environmental or occupational exposures could be identified served as controls. None of the controls or the patients with psoriasis had received any topical or systemic medication for at least 14 days prior to obtaining epidermis for study. Every effort was made to match the control population with the psoriatic population insofar as age, sex, and smoking history were concerned. A specially designed keratome was used to obtain epidermis as described previously [20]. Skin of the normal controls and visibly uninvolved skin of the patients with psoriasis were selected for study. The keratomed epidermal samples were obtained from the lower trunk or thigh. The skin was first cleansed thoroughly with a topical antiseptic and infiltrated with 1% lidocaine (Xylocaine) without epinephrine. Epidermal strips ~2.0 cm wide and ~4.5 cm long were obtained from each subject and immediately dropped into liquid nitrogen for later determination of enzyme activity. For the AHH and EH induction studies the freshly keratomed epidermis was immediately divided into 2 segments and added to an organ culture system as described previously [11]. Briefly, Eagle's minimum essential medium containing Earle's balanced salt solution (Grand Island Biological Co., Grand Island, New York), fetal bovine serum (Microbiological Associates, Walkersville, Maryland), glutamine, streptomycin, nystatin, and penicillin G were used. A solution of BA in acetone (0.05 ml) was dispersed in 10 ml of the fetal bovine serum which gave a final concentration of 100 nM. The resulting suspension was added to 190 ml of culture medium, shaken at 42°C for 16 h in the dark, and then passed through a 22-μm pore size Millipore filter (Millipore Corp., Bedford, Massachusetts). Control medium was prepared identically except that acetone alone was added.

One segment of the freshly keratomed epidermis obtained from the controls or from the uninvolved skin of psoriatic individuals was minced with surgical scissors and placed in control medium or in the BA-treated medium and incubated in the dark for 24 h at 37°C in a humidified atmosphere of 5% CO_2 in air. At the conclusion of the incubation, the medium containing the minced epidermal segments was centrifuged and the pellet washed 4 times with 0.1 M potassium phosphate buffer, pH 7.4. The epidermal pellet was overlaid with the same buffer and frozen immediately in liquid N_2. All samples were then shipped frozen in dry ice by air to Cleveland for the enzyme assays which were always performed within 3 days of obtaining the tissue.

Tissue Preparation

The epidermis was thawed, washed once with 0.1 M phosphate buffer pH 7.40, weighed, and homogenized using a Polytron (Brinkmann) tissue homogenizer. The tissue was maintained in an ice-water bath to obviate warming during processing. This whole homogenate was poured into the tube of a ground-glass conical tissue homogenizer fitted with a ground-glass pestle and was homogenized for 6 passes with a rotary drill press [3]. The whole epidermal homogenate was then centrifuged at 800 *g* for 10 min; 800 *g* supernatants were centrifuged at 9000 *g*, and the 9000 *g* supernatant utilized for assay of AHH and EH. Epidermal segments used for the study of enzyme induction were similarly homogenized and 9000 *g* supernatant prepared. HPLC metabolism of BP was studied using human epidermal microsomes as the enzyme source. Epidermal microsomes were prepared essentially as described earlier [3] from pooled 9000 *g* supernatant from 6 normal subjects and 6 patients with psoriasis.

Enzyme Assays

AHH activity was determined by a slight modification of the fluorimetric method described by Nebert [21]. Briefly a typical incubation mixture consisted of 10 mM potassium phosphate buffer (pH 7.4), 200 nmol of NADPH, 300 nmol of $MgCl_2$, 12 ng of bovine serum albumin, and 100 μl of the enzyme source in a total volume of 0.2 ml. The reaction was initiated by adding 20 nmol BP in 10 μl of acetone and was incubated for 60 min at 37°C in a Dubnoff metabolic shaker. The reaction was terminated by the addition of 200 μl of cold acetone and 600 μl of hexane was then added. The quantitation of phenolic BP metabolites was compared to the fluorescence of 3-OH-BP Standard. EH activity with BP 4,5-oxide as substrate was measured as described previously [22]. Protein was determined by the method of Lowry et al [23]. Details of these procedures were described earlier [24].

All determinations were performed in triplicate and a zero time tissue blank was run for each assay. The values presented are the mean obtained from 3 determinations on each sample. The enzyme assays were carried out completely blind. All samples were labeled only with code numbers, which were assigned in Ann Arbor. At the completion of all studies the code was broken and the values assigned to the appropriate samples. These steps were taken to minimize possible bias. This was felt to be particularly important in these studies since the units of enzyme activity were quite low. The values obtained were then evaluated for statistical significance using Student's *t*-test. Comparison was made between normal and psoriatic individuals for basal and inducible activity of AHH and EH.

HPLC Analysis of BP Metabolism by Human Epidermal Microsomes

HPLC analysis of BP metabolism by human epidermal microsomes was studied by slight modifications of our procedure published on rodent skin [25]. The incubation mixture contained 2.0 mg microsomal protein, 0.10 mmol phosphate buffer (pH 7.4), 3 μmol $MgCl_2$, and 1 mg NADPH in a final volume of 0.96 ml. The reaction was initiated by the addition of 80 nmol [^{14}C]BP in 40 μl methanol. The samples were incubated for 60 min in the dark at 37°C in a Dubnoff metabolic shaker. The reaction was terminated by adding 1 ml of acetone followed by 2 ml of ethyl acetate. The mixture was vortexed for 1 min to extract any unreacted BP as well as the metabolites into the organic phase. The organic and aqueous layers were separated by centrifugation at 1500 rpm for 5 min. The radioactivity in the aqueous phase was less than 0.5% of the total radioactivity and was proportional to the total BP metabolized. The organic phase was then dehydrated over anhydrous $MgSO_4$, dried under a stream of N_2, and dissolved in 50 μl of methanol for HPLC analysis.

A Waters Associates model 440 liquid chromatograph, fitted with Waters Associate uBondapack C18 column (4.6 × 25 mm) was used for analysis of the radiolabeled BP metabolite mixtures. Identification of the metabolites was based on reference standard. The column was eluted at ambient temperature with a 30-min linear gradient of methanol:water:tetrahydrofuran (THF) (70:27:3, v/v) to methanol at a solvent flow rate of 0.5 ml/min. The eluates were monitored at 254 nm, fractions of approximately 0.2 ml were collected dropwise, and the radioactivity of each fraction determined on a Packard TriCarb 460 CD liquid scintillation spectrometer. The counting efficiencies of the early eluted fractions, containing a higher percentage of water, were about 2% lower than those of the fractions eluted with methanol.

Chemicals

[^{14}C]BP (sp act 28.5 mCi/mmol) was purchased from New England Nuclear (Boston, Massachusetts) and was purified by a silica gel (Bio-Sil A, 100-200 mesh, BioRad Laboratories) column with hexane as the eluting solvent and subsequently by reverse phase HPLC using a DuPont Zorbax OBS column eluted with methanol:water (9:1, v/v). [^{14}C]BP was diluted with unlabeled BP (Sigma Chemical Co., St. Louis,

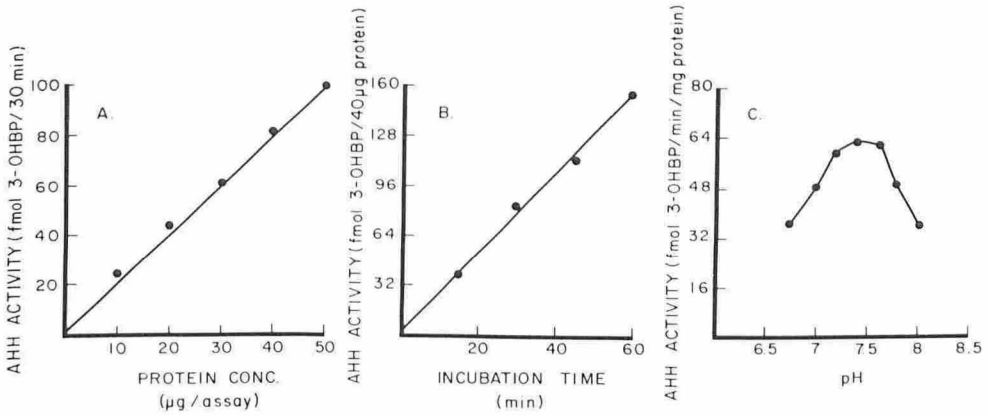

FIG 1. Optimal conditions for assaying AHH activity in human epidermis. The assay conditions of AHH activity are as described in the text with the exception of (a) protein concentration, (b) time of incubation, and (c) pH of the incubation mixtures which were varied as indicated.

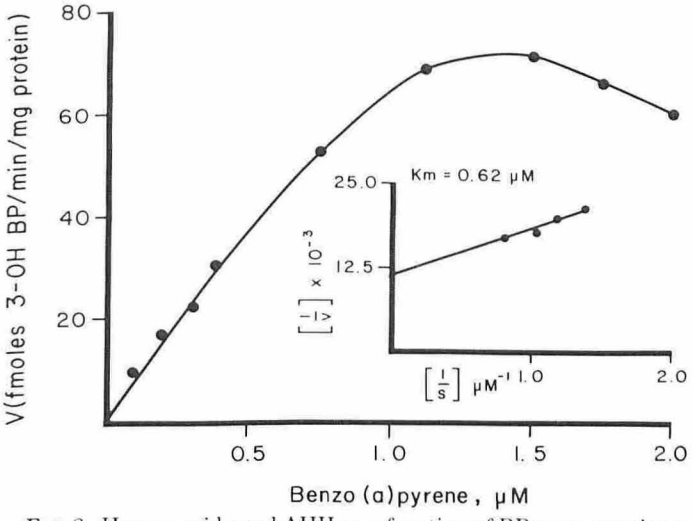

FIG 2. Human epidermal AHH as a function of BP concentrations. Enzyme activity was measured after 60 min as described in *Materials and Methods*. The concentration of BP was varied as indicated in the figure. A Lineweaver-Burk representation of the data for determination of an approximate K_m value is provided in the *inset*.

Missouri) to a sp act of 22.5 mCi/mmol. [^{3}H]BP 4,5 oxide (sp act 282.5 mCi/mmol; radiochemical purity greater than 99%) and chromatographically pure unlabeled BP 4,5-oxide, BP 4,5-dihydrodiol, and BP metabolite reference standards were obtained from the Chemical Repository Unit of National Cancer Institute, Bethesda, Maryland. All solvents and chemicals used were of the highest grade commercially available.

RESULTS

Characteristics of AHH in Human Epidermis

Since a modification of the standard AHH assay was developed for determination of enzyme activity in freshly keratomed human epidermis, initial experiments were performed to define the optimal conditions for the procedure. Fig 1 shows that the rate of the reaction in 9000 *g* supernatant of human epidermis was linear up to a protein concentration of 50 μg, up to an incubation time of 60 min, and that optimal catalytic activity occurred at a pH of 7.4. These enzyme characteristics are similar to those in the epidermis of neonatal rodents which were published previously [3]. The apparent K_m value for BP in human epidermis was 0.62×10^{-6} M. The V_{max} for human epidermal AHH was 70 fmol/min/mg protein (Fig 2).

Comparative AHH and EH Activity in Normal and Psoriatic Subjects

The data for AHH activity in keratomed epidermis obtained from individual normal subjects and from individual psoriatic patients are presented in Table I. There was a wide range of values in both normal and psoriatic individuals, though, as can be seen, they, for the most part, could be grouped into similar patterns. These differences in activity were consistent regardless of whether the results were calculated on the basis of units of enzyme activity per mg protein or wet weight. The mean AHH activity in epidermis obtained from the normal population was 62.1 ± 5.6, whereas the mean activity in noninvolved epidermis obtained from the psoriatic individuals was 62.9 ± 5.1 (Table I). There is no statistically significant difference between these two populations insofar as basal epidermal AHH activity is concerned.

TABLE I. *Comparative activity of aryl hydrocarbon hydroxylase in epidermis from normal subjects and from patients with psoriasis*

Normal subjects	AHH[a]	Psoriatic patients	AHH[a]
A	80	A	56
B	85	B	67
C	83	C	107
D	66	D	52
E	44	E	52
F	50	F	37
G	83	G	67
H	53	H	80
I	24	I	63
J	68	J	49
K	64	K	58
L	45	L	67
Mean ± SE	62.1 ± 5.6		62.9 ± 5.1

[a] Aryl hydrocarbon hydroxlase, fmol 3-OH-BP/min/mg protein. Keratomed epidermis was obtained from normal volunteers and from visibly uninvolved skin of patients with psoriasis and assayed for AHH activity as described in *Materials and Methods*.

In further studies we attempted to determine the extent to which human epidermal AHH was inducible in vitro by BA. As shown in Table II, AHH activity in both normal and psoriatic epidermis was inducible by BA. AHH activity in the epidermis obtained from the normal controls increased by a mean value of 165 ± 26% whereas the epidermis obtained from noninvolved skin of psoriatic individuals increased by a mean of 320 ± 29%. Thus the responsiveness of epidermal AHH to BA added in vitro was approximately 2-fold greater in patients with psoriasis as compared to normal individuals.

EH activity was detectable in epidermis obtained from both the normal and the psoriatic populations. The mean ± SE activities in each were 25.1 ± 1.1 and 24.8 ± 2.1, respectively (Table III). There is no statistically significant difference between the two.

HPLC Profile of BP Metabolism in Human Epidermis

As shown in Table IV, HPLC analysis of human epidermal microsomal metabolism of BP revealed that diols, quinones,

TABLE II. *Comparative inducibility of epidermal microsomal AHH by BA in normal subjects and patients with psoriasis*

	AHH[a]		
	−BA	+BA	% Induction
	Normal subjects		
A	83	280	237
B	50	165	220
C	24	62	158
D	53	91	72
E	78	169	117
F	44	132	200
G	71	114	61
H	59	208	252
Mean ± SE	57.8 ± 6.9	152.6 ± 24.5	165 ± 26
	Psoriatic patients		
A	20	96	220
B	53	196	270
C	67	311	364
D	55	240	336
E	51	266	422
F	49	201	310
Mean ± SE	49.2 ± 6.0	218.3 ± 30.0	320 ± 29

[a] fmols 3-OH-BP/min/mg protein. BA = benz[a]anthracene; AHH = aryl hydrocarbon hydroxlase.

Freshly keratomed epidermis was immediately split into 2 segments and incubated for 24 h in tissue culture medium containing BA. Controls were incubated without the hydrocarbon. AHH activity was then determined as described in *Materials and Methods*.

TABLE III. *Microsomal epoxide hydrolase activity in epidermis of normal individuals and in patients with psoriasis*

Normal subjects	EH[a]	Psoriatic patients	EH
A	22.2	A	30.3
B	27.5	B	28.8
C	26.2	C	15.4
D	24.0	D	18.0
E	19.0	E	36.8
F	28.9	F	38.3
G	22.2	G	22.4
H	23.8	H	23.7
I	26.4	I	18.4
J	24.4	J	22.6
K	33.9	K	19.7
L	22.4	L	23.3
Mean ± SE	25.1 ± 1.1		24.8 ± 2.1

[a] Epoxide hydrolase, pmol BP 4,5-diol/min/mg protein.
For other details see Table I.

and phenolic metabolites of the carcinogen were formed by both normal and psoriatic individuals. Total metabolism and the overall pattern of metabolite formation were similar in each population. BP *trans*-7,8-diol is the precursor of the ultimate carcinogen of BP [6,8]. The formation of this particular metabolite by human epidermal microsomes was only slightly higher in psoriatic patients as compared to normal subjects. These data provide further confirmation of the similarity between normals and psoriatic individuals insofar as epidermal microsomal carcinogen metabolism is concerned.

DISCUSSION

The etiology of psoriasis has been increasingly studied in recent years and in particular the possibility that a specific enzyme deficiency is responsible for the disease has intrigued some individuals. In prior studies Shuster and his associates suggested that patients with psoriasis had an intrinsic defect in the basal activity and the inducibility of the carcinogen-metabolizing enzyme, AHH, in the epidermis [15,16] and that

TABLE IV. *Patterns of metabolism of [^{14}C]benzo[a]pyrene by epidermal microsomes from normal individuals and from patients with psoriasis*

Metabolites	BP metabolism (pmol product/60 min/mg microsomal protein)	
	Normal subjects	Psoriatic patients
trans-9,10-diol	4.1	7.2
trans-4,5-diol	11.8	13.6
trans-7,8-diol	3.2	4.8
1,6-quinone	65.4	60.8
3,6-quinone	45.4	29.9
6,12-quinone	68.6	70.3
9-OH-BP	6.2	5.8
3-OH-BP	10.7	10.4
Total phenols	16.9	16.2
Total metabolites	215.4	202.8

Skin 9000 *g* supernatants from 6 subjects were pooled and microsomes prepared. Incubation mixture in 1 ml total volume contained 2.0 mg microsomal protein. Incubations were carried out at 37°C for 60 min and the metabolites extracted as described in *Materials and Methods*.

this defect was detectable in other tissues as well [26]. From these observations they proposed that defective AHH activity might be a marker for this disease [27]. Furthermore, on the basis of their clinical experience these workers suggested that the incidence of skin cancer in patients with psoriasis was low and that this was directly related to their observation that psoriatic epidermis had a reduced capacity to metabolize carcinogenic PAH [28].

The question of unique susceptibility of selected human populations to cancer as a result of exposure to environmental carcinogens was first raised in 1775 by the classical studies of a London surgeon, Percival Pott [29]. He pointed out his experience with scrotal cancer in chimney sweeps and made the seminal observation that this population of workers appeared to be uniquely susceptible to developing this specific type of neoplasm. Fron. his observations, Pott proposed that human skin cancer could be caused by exposure to environmental agents, a concept that remains valid until the present day. The major environmental oncogens for human skin that have been identified are solar ultraviolet radiation and selected chemicals, among them the PAH [30,31]. The PAH are generated whenever carbonaceous material such as fossil fuels are incompletely combusted. Both ultraviolet radiation and crude coal tar, a substance produced by destructive distillation of coal which is rich in PAH, are widely used in the management of psoriasis [32], a modality known as the Goeckerman regimen named for the individual who first described it [33].

In recent years much important new knowledge regarding the mechanism of chemical carcinogenesis has developed [5,6]. It is now well established that inducible membrane-bound enzymes such as AHH, present in many types of cells, including the skin [3], can convert selected biologically inert chemicals into highly reactive electrophilic metabolites which can bind to macromolecules thereby initiating tumor formation [4–6].

Studies in human populations have attempted to determine whether AHH inducibility or responsiveness can be correlated with susceptibility to cancer. Kellerman et al measured AHH inducibility in mitogen-stimulated lymphocytes obtained from patients with lung cancer and compared it to cells obtained from patients with other forms of cancer and from normal controls [34]. Their results indicated a trimodal distribution of AHH inducibility (low, intermediate, and high) in the normal population and showed that a disproportionate number of patients with lung cancer had intermediate or high inducibility of AHH in their lymphocytes. Subsequently, considerable controversy developed regarding the relationship of the inducibility of AHH in human lymphocytes to human cancer because it

was not possible to confirm these findings [35–37]. Furthermore, it became clear that the lack of reproducibility of the assay procedure in mitogen-stimulated lymphocytes was a major source of difficulty [38]. For example there is some evidence to suggest that there is seasonal variation in AHH activity in human lymphocytes with 10-fold higher activity in selected individuals during late summer and early fall [39]. Others have shown that the responsiveness of lymphocytes to mitogens is inhibited even in certain patients very early with (Stage I) lung cancer [40]. In such individuals diminished AHH inducibility could result from a change in the responsiveness of the cell being studied rather than as an inherited trait.

AHH activity and inducibility in psoriatic skin was studied by Chapman et al who obtained epidermis from involved and uninvolved skin of patients with psoriasis and from normal controls using a suction blister technique. The suction blisters were divided into two parts, one of which was incubated with BA for 18 h, and AHH activity measured [15]. From their data it was concluded that in the epidermis of patients with psoriasis there is defective AHH activity such that the basal level of the enzyme and its induction response to an inducer are both decreased, and it was suggested that deficient AHH activity could be a primary genetic abnormality in psoriasis.

The data presented here do not confirm the results of Chapman et al [15,26,27] and Shuster et al [16,28] regarding differences between normal and psoriatic individuals in the basal and inducible levels of epidermal AHH. During the course of our studies Rawlins and Shuster doubted the veracity of their earlier observation [18]. These investigators later suggested that AHH activity in suction blisters of human epidermis is not measurable [19]. Our data do indicate that keratomed human epidermis possesses measurable levels of AHH and EH activity and that there are no statistically significant differences between the basal levels of epidermal AHH and EH activities in normal and psoriatic individuals. However AHH in epidermis obtained from visibly uninvolved skin of psoriatic individuals is twice as responsive to BA as is the enzyme in normal epidermis. Our data also verify that the microsomal cytochrome P-450 is present and inducible in human epidermis. Furthermore we have shown that human epidermis is capable of metabolizing BP into a large number of derivatives. This suggests that topically applied drugs are likely to be substrates for microsomal monooxygenases in the epidermis. This type of catalytic activity could be an important determinant of drug efficacy in human skin.

It is of interest that recent studies in cultured lymphocytes of psoriatic patients found that there was no significant difference in the mean basal and induced AHH activities between these cells and cells obtained from normal individuals [41,42]. There is also epidemiologic evidence to suggest that psoriatic individuals are not resistant to the development of skin cancer [17].

In summary, there appear to be no differences in the patterns of epidermal carcinogen metabolism between normal individuals and patients with psoriasis. Further studies are needed to identify the specific enzymatic abnormalities that play a role in the pathogenesis of this disease.

We thank Ms. Betty Olson and Mrs. Sandra Evans for preparation of the manuscript.

REFERENCES

1. Blum HF: Ultraviolet radiation and skin cancer in mice and men. Photochem Photobiol 24:249–254, 1976
2. Hueper WC: Chemically induced skin cancers in man. Natl Cancer Inst Monogr 10:377–391, 1963
3. Bickers DR, Dutta-Choudhury T, Mukhtar H: Epidermis: a site of drug metabolism in neonatal rat skin. Studies on cytochrome P-450 content and mixed function oxidase and epoxide hydrolase activity. Mol Pharmacol 21:239–247, 1982
4. Miller EC, Miller JA: Biochemical mechanisms of chemical carcinogenesis, The Molecular Biology of Cancer. Edited by H Busch. New York, Academic Press, 1974 pp 377–402
5. Heidelberger C: Chemical carcinogenesis. Annu Rev Biochem 44:79–121, 1975
6. Gelboin HV: Benzo(a)pyrene metabolism, activation and carcinogenesis: role of regulation of mixed-function oxidase and related enzymes. Physiol Rev 60:1107–1166, 1980
7. Daudel PM, Duquesne M, Vigny P, Grover PL, Sims P: Fluorescence spectral evidence that benzo(a)pyrene-DNA products in mouse skin arise from diol-epoxides. FEBS Lett 57:250–253, 1975
8. Levin WA, Wood AW, Yagi H, Jerina DM, Conney AH: (±)-trans-7,8-dihydroxy 7,8-dihydrobenzo(a)pyrene: a potent skin carcinogen when applied topically to mice. Proc Nat Acad Sci USA 73:3867–3871, 1976
9. Koreeda M, Moore PD, Wislocki PG, Levin W, Conney AH, Yagi H, Jerina DM: Binding of benzo(a)pyrene 7,8-diol-9,10-epoxides to DNA, RNA and protein of mouse skin occurs with high stereoselectivity. Science 199:778–780, 1978
10. Alvares AP, Kappas A, Levin W, Conney AH: Inducibility of benzo(a)pyrene hydroxylase in human skin by polycyclic hydrocarbons. Clin Pharmacol Ther 14:30–40, 1973
11. Bickers DR, Kappas A: Human skin aryl hydrocarbon hydroxylase: induction by coal tar. J Clin Invest 62:1061–1068, 1978
12. Oesch F: Microsomal epoxide hydrolase, Enzymatic Basis of Detoxication, vol II. Edited by WB Jakoby. New York, Academic Press, 1980 pp 277–290
13. Nebert DW, Goujon FM, Gielen JE: Aryl hydrocarbon hydroxylase induction by polycyclic hydrocarbons: simple autosomal dominant trait in the mouse. Nature 236:107–110, 1972
14. Nebert DW, Benedict WG, Gielen JE, Oesch F, Daly JW: Aryl hydrocarbon hydroxylase, epoxide hydrase, and 7,12-dimethylbenz(a)anthracene-produced skin tumorigenesis in the mouse. Mol Pharmacol 8:374–379, 1972
15. Chapman PH, Rawlins M, Shuster S: Activity of aryl hydrocarbon hydroxylase in psoriatic skin. Lancet 1:297–298, 1979
16. Shuster S, Rawlins MD, Chapman PH, Rogers S: Decreased epidermal aryl hydrocarbon hydroxylase and localized pustular psoriasis. Br J Dermatol 103:23–26, 1980
17. Stern RS, Zierler S, Parrish JA: Skin carcinoma in patients with psoriasis treated with topical tar and artificial ultraviolet radiation. Lancet 1:732–735, 1980
18. Rawlins M, Shuster S: Aryl hydrocarbon hydroxylose in psoriasis. Lancet 2:271, 1982
19. Finnen MJ, Shuster S, Lawrence CM, Rawlins MD: Aryl hydrocarbon hydroxylose activity and psoriasis. Biochem Pharmacol 32:1707–1712, 1983
20. Voorhees JJ, Duell EA, Bass LJ, Powell JA, Harrell ER: Decreased cyclic AMP in the epidermis of lesions of psoriasis. Arch Dermatol 105:695–701, 1972
21. Nebert DW: Genetic differences in the induction of monooxygenase activities by polycyclic aromatic hydrocarbons. Pharmacol Ther 6:395–417, 1979
22. Mukhtar H, Elmamlouk TH, Bend JR: Epoxide hydrase and mixed-function oxidase activities of rat liver nuclear membranes. Arch Biochem Biophys 192:10–21, 1979
23. Lowry OH, Rosebrough NJ, Farr AL, Randall RJ: Protein measurement with Folin phenol reagent. J Biol Chem 193:265–275, 1951
24. Bickers DR, Marcelo CL, Dutta-Choudlury T, Mukhtar H: Studies on microsomal cytochrome P-450, monooxygenases and epoxide hydrolase in cultured keratinocytes and intact epidermis from balb/c mice. J Pharmacol Exp Ther 223:163–168, 1982
25. Bickers DR, Mukhtar H, Yang SK: Cutaneous metabolism of benzo(a)pyrene: comparative studies in C57BL/6N and DBA/2N mice and neonatal Sprague-Dawley rats. Chem Biol Interact 43:263–270, 1983
26. Chapman PH, Kersey PJ, Keys B, Shuster S, Rawlins M: Generalized tissue abnormality of aryl hydrocarbon hydroxylase in psoriasis. Br Med J 281:1315–1316, 1980
27. Chapman PH, Keys B, Rawlins M, Moss C, Shuster S: Antipyrine clearance in psoriasis. Br Med J 282:20–21, 1981
28. Shuster S, Chapman PH, Rawlins MD: Psoriasis and cancer. Br Med J 1:941–942, 1979
29. Pott P: Cancer scroti, Chiurgical Observations. The Chiurgical Works of Percival Pott. London, Hawes, Clark, and Collings, 1775
30. Blum HF: Carcinogenesis by Ultraviolet Light. Princeton, NJ, Princeton Univ Press, 1959
31. Henry SA: Occupational cutaneous cancer attributable to certain chemicals in industry. Br Med Bull 4:389–401, 1947
32. Harber LC, Bickers DR: Photochemotherapy: tar and ultraviolet A and B (TUVAB), Photosensitivity Diseases: Principles of Diagnosis and Treatment. Edited by DR Bickers, LC Harber. Philadelphia, WB Saunders, 1981, pp 56–64
33. Goeckerman WH: Treatment of psoriasis. Northwest Med 23:229–231, 1925
34. Kellermann G, Shaw CR, Luyten-Kellermann M: Aryl hydrocar-

bon hydroxylase inducibility and bronchogenic carcinoma. N Engl J Med 289:934–937, 1973
35. Paigen B, Gurtoo HL, Minowada J, Houten L, Vincent R, Paigen K, Parker NB, Ward E, Hayner NT: Questionable relation of aryl hydrocarbon hydroxylase to lung-cancer risk. N Engl J Med 297:346–350, 1977
36. McLemore TL, Martin RR, Pickard LR, Springer RR, Wray NP, Toppell KL, Mattox KL, Guinn GA, Cantrell ET, Busbee DL: Analysis of aryl hydrocarbon hydroxylase activity in human lung tissue, pulmonary macrophages and blood lymphocytes. Cancer 41:2292–2300, 1978
37. Prasad N, Prasad R, Harrell JE, Thornby J, Lumb JH, Hudgins PT, Guinn GA: Aryl hydrocarbon hydroxylase inducibility in lung cancer patients undergoing radiotherapy. Int J Cancer 24:415–420, 1979
38. Fletcher KA, Price Evans DA, Canning MV: Inducibility of aryl hydrocarbon hydroxylase in cultured human lymphocytes. A study of repeatability. J Med Genet 15:182–188, 1978
39. Paigen B, Ward E, Reilly A, Houten L, Gurtoo HL, Minowada J, Steenland K, Havens MB, Sartori P: Seasonal variation of aryl hydrocarbon hydroxylase activity in human lymphocytes. Cancer Res 41:2757–2761, 1981
40. Jett JR, Moses LL, Branum EL, Taylor WF, Fontana RS: Benzo(a)pyrene metabolism and blast transformation in peripheral blood mononuclear cells from smoking and nonsmoking populations and lung cancer patients. Cancer 41:192–200, 1978
41. Ruzicka T, Vizethum WW, Jacobs A, Goerz G: Aryl hydrocarbon hydroxylase activity in lymphocytes of patients with psoriasis. Lancet 1:1142–1143, 1980
42. Karki N, Karvonen J, Pelkonen O: Aryl hydrocarbon hydroxylase activity in cultured lymphocytes of psoriatic patients. Arch Dermatol Res 273:103–109, 1982