

# DNA repair and aging in basal cell carcinoma: A molecular epidemiology study

(skin cancer/case-control study/molecular epidemiology)

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**ABSTRACT** This molecular epidemiology study examines the DNA-repair capacities (DRCs) of basal cell carcinoma (BCC) skin cancer patients (88) and their controls (135) by using a plasmid/host-cell reactivation assay. In this assay UV-damaged expression vector plasmid is transfected into peripheral blood T lymphocytes from the subjects. The host-cellular repair enzymes repair the photochemical damage in the plasmid, and 40 hr later the plasmid-encoded reporter chloramphenicol acetyltransferase is measured. An age-related decline in this DRC, amounting to  $\approx 0.61\%$  per yr occurred in the controls from 20 to 60 yr of age. Reduced DRC was a particularly important risk factor for young individuals with BCC and for those individuals with a family history of skin cancer. Young individuals with BCC repaired DNA damage poorly when compared with controls. As the BCC patients aged, however, differences between cases and controls gradually disappeared. The normal decline in DNA repair with increased age may account for the increased risk of skin cancer that begins in middle age, suggesting that the occurrence of skin cancer in the young may represent precocious aging. Patients with reduced DRCs and overexposure to sunlight had an estimated risk of BCC  $>5$ -fold greater than the control group. Such a risk was even greater (10-fold) in female subjects.

The role of DNA repair in cancer is exemplified by the paradigm of the DNA-repair-defective autosomal-recessive disease xeroderma pigmentosum (XP) (1). XP cells are deficient in those gene products required for catalyzing the incision step in nucleotide-excision repair of damaged DNA (2, 3). Repair-deficient patients develop their first sunlight-induced skin cancers at early ages and have a  $>2000$ -fold increased risk of cancer compared with normal subjects (4).

The relationship of the persistence of UV-induced DNA damage to mutation fixation is epitomized by recent molecular studies with human skin tumor cells. Activation of the *Ha-ras* oncogene, for instance, was noted in skin tumors on sun-exposed body sites (5). Further, invasive skin cancers show strong structural correlations between p53 tumor-suppressor gene mutations and CC  $\rightarrow$  TT double-base changes and between pyrimidine dimer formation and C  $\rightarrow$  T mutation in DNA (6). The relationship of the gene mutation to changes specific for UV damage implicates sunlight as a cause of the cancers according to the investigators. On the other hand, the initial plateau on a repair dose-response curve (7) reflects an individual's DNA-repair capacity (DRC), and its limits represent the extremes of saturation for the repair machinery of the organism or cell. The saturation point of this plateau may, then, predict individual susceptibility to sunlight-induced DNA damage. We conducted a

molecular-epidemiological study to test the hypothesis that those individuals in the general population at risk for basal cell carcinoma (BCC) of the skin have impaired capacity to repair photochemically damaged DNA.

There are many different assays for measuring DNA repair, ranging from damage removal, damage or repair-dependent formation of single-strand breaks, and uptake of substrate for unscheduled DNA synthesis in response to DNA damage. Each assay may represent a unique rate-limiting step in the overall repair process. The test in this study used the subject's T lymphocytes as a surrogate cell to measure their overall DRC (7). The cellular DRC was determined by measuring the expression level of a nonreplicating recombinant plasmid DNA containing a UV-damaged chloramphenicol acetyltransferase (CAT) reporter gene in undamaged T lymphocytes. This CAT assay measures the entire progression of repair steps leading to the restoration of the biological properties of a reporter gene rather than a specific step within the process. Comparison of the overall process as well as identification of the rate-limiting step specific to a biological state should be of mechanistic importance.

## METHODS

**Subject.** The study population consisted of 88 patients with histopathologically confirmed primary BCC and 135 cancer-free controls. All subjects are Caucasians 20–60 yr of age who lived in Baltimore City or its suburban surroundings for most of their lives and worked indoors. Skin biopsies taken in 1987–1990 were diagnosed as BCCs at the dermatopathology laboratory of the Johns Hopkins Hospital, which serves multiple practicing dermatologists in Maryland. The subjects did not receive chemotherapy, radiation therapy, or blood transfusions; nor did they have diseases known to be related to DNA-repair deficiency, such as XP, ataxia telangiectasia, or Cockayne syndrome. In addition, eligible cases and controls did not have other forms of cancer. About 55% of the cases are males with an average age of 48.7 yr. The corresponding figures for controls are 50% and 46 yr. The purpose of selecting young subjects was to maximize the differences in risk factors between cases and controls. The control group had diagnoses of mild skin disorders such as intradermal nevus (53%), seborrheic keratosis (31%), subacute eczematous dermatitis (5%), or others (11%).

**Clinic Visit.** All participants were seen at the dermatology clinic, at which time the subjects completed a structured questionnaire, had blood drawn for T lymphocytes, and were examined by a dermatologist. The questionnaire included information on demographics, history of sunlight exposure, and medical history, including recent medication. The extent

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Abbreviations: BCC, basal cell carcinoma; DRC, DNA-repair capacity; XP, xeroderma pigmentosum; CAT, chloramphenicol acetyltransferase.

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of sunburns, including blistering, was listed to measure previous sunlight exposure. Family histories of all malignancies, as well as personal histories of cancers, were obtained from the questionnaire. The dermatologists examined the patients to verify current skin conditions, including skin lesions and signs of surgical removal of previous skin malignancies. Control individuals with a self-reported cancer history or clinical signs of skin cancer or other cancers were excluded.

**Laboratory Assays. CAT assay.** The peripheral blood lymphocytes isolated from each subject's peripheral blood were cryopreserved and assayed in batches, as described (7, 8). Briefly, the plasmid DNA containing the CAT gene was irradiated with UV of 254 nm at 0, 350, and 700 J/m<sup>2</sup> before transfection. The quick-thawed peripheral blood lymphocytes with a >95% viability were incubated for 72 hr with phytohemagglutinin and were then transfected with undamaged or damaged plasmid DNA. The standard assay for gene expression of CAT activity by measurement of [<sup>3</sup>H]acetylchloramphenicol with a Beckman scintillation counter (LS 3801) was done after another 40-hr incubation. The DRC was calculated based on scintillation counts as the percentage of residual CAT gene expression (percentage CAT activity) after repair of damaged DNA compared with undamaged plasmid DNA (100%) (7). Lymphoblast cell lines from group A XP patients (GM2345; XP-A, most severe), group D XP patients (GM2253; XP-D, severe), and group C XP patients (GM2246; XP-C, classic form) were used to generate the standard DNA repair-deficient curves because one pyrimidine dimer can inactivate the transfected CAT gene in XP cells (9). In addition, lymphoblasts from normal individuals (GM0131 and GM0892) were assayed in parallel with each batch of assays, serving as normal standards as well as quality control tests. The dose curves of 700 J/m<sup>2</sup>, 350 J/m<sup>2</sup>, and zero were done in triplicate. Because the dose curves based on these three doses were linear, the repair capacity at 700 J/m<sup>2</sup> [30 J/m<sup>2</sup> produces one pyrimidine dimer per plasmid (7)] was taken as the repair capacity for that individual. The repeatability of the assay (intraassay variability) was tested by using the same blood sample at different times. There is an ≈5% fluctuation in CAT activities in which the rank order of each subject's CAT activity is maintained (Table 1). Neither blastogenic rate nor level of CAT gene expression correlate with CAT activity (Table 2).

**CD4<sup>+</sup>/CD8<sup>+</sup> lymphocyte test.** An equal number of blood samples (*n* = 11) from both cases and controls spanning different ages were selected and assayed by S. Gore, Johns Hopkins School of Medicine, by using standard immunofluorescence methods with flow cytometry to count CD4- and CD8-positive lymphocytes. Although the control group had slightly higher blastogenic rates than the BCC group, the distribution of CD4<sup>+</sup> and CD8<sup>+</sup> cell types within T-lymphocyte fractions did not differ in BCC and control groups (Table

Table 1. Ranking of percentage CAT activity of repeated assays with the same blood sample from subjects

No.	Assay, % (rank)		
	First	Second	Third
51	5.4 (2)	6.1 (2)	5.8 (2)
121	8.2 (4)	9.8 (5)	
124	10.0 (6)	9.0 (4)	
152	8.9 (5)	11.2 (6)	
155	3.2 (1)	2.6 (1)	4.4 (1)
184	5.8 (3)	7.3 (3)	6.6 (3)
342	12.6 (7)	14.2 (7)	

CAT activity at UV dose of 700 J/m<sup>2</sup> measured by scintillation counts of radioactivity was standardized by CAT activity at zero UV dose. The assays were done in different weeks with one frozen 30-cc sample of peripheral blood from each individual labeled by number.

Table 2. Correlations between age, lymphocyte blastogenic rate, and CAT activity of undamaged and damaged plasmid (*n* = 223)

	Pearson correlation coefficient/ <i>P</i> value		
	CAT <sub>0</sub> (counts)	CAT <sub>700</sub> (%)	Blastogenic rate (%)
Age (in yr)	-0.091/0.174	-0.256/0.000	-0.058/0.387
CAT <sub>0</sub> * (counts)		0.014/0.836	0.096/0.153
CAT <sub>700</sub> † (%)		0.025/0.710	

Blastogenic rate represents the percentage of lymphocytes (blasts) responding to phytohemagglutinin within 72 hr.

\*Original scintillation counts of radioactivity of CAT assays from lymphocytes transfected with undamaged plasmid DNA containing CAT gene.

†CAT activity at UV dose of 700 J/m<sup>2</sup> measured by scintillation counts of radioactivity was standardized by CAT activity at zero UV dose.

3). In addition, the CD4<sup>+</sup>/CD8<sup>+</sup> ratios do not correlate with either CAT activity (*r* = 0.10; *P* > 0.05) or age (*r* = 0.35; *P* > 0.05). Therefore, we conclude that the CAT assay offers a DNA-repair measurement that is independent of immunological functions and culture conditions of the tested lymphocytes.

**Statistical Analyses.** Pearson correlation coefficients were calculated to evaluate the relationship between variables of interest. The group mean values of CAT activity were compared by using a two-tailed Student's *t* test. Simple and multiple linear-regression analyses were used to determine the best predictor for DRC. Logistic regression was used to calculate the adjusted odds ratio for estimating BCC risk. All computations were done with SAS Institute, Inc. statistical software on an International Business Machines mainframe computer.

## RESULTS

Distributions of age, sex, smoking, and medicine use are similar for BCC and control groups. However, the BCC patients are more likely than controls to have blue eyes and fair skin, factors associated with BCC risk (Table 4).

Multiple linear-regression analysis revealed that the DRC of control subjects declined with age from 20 to 60 yr (Fig. 1). Based on the estimates from the linear-regression model for the 135 controls, the decline is 0.61% per yr between 20 and 60 yr of age (Table 5, model 1: the estimated coefficient (-0.071) for current age relative to estimated intercept (baseline, 11.725) gives a rate of decline of -0.61%). The four-

Table 3. Comparison of blastogenic rate, baseline gene expression, and CD4<sup>+</sup>/CD8<sup>+</sup> ratio between BCC patients and cancer-free controls

Characteristic	BCC case, mean ± SD	Control, mean ± SD	<i>t</i> test, <i>P</i>
Mean of blastogenic rate, %*	61.7 ± 16.8	66.5 ± 16.5	0.037
Baseline CAT gene expression level, mean counts†	101,013 ± 40,266	102,019 ± 39,639	0.854
Mean of CD4, %	46.8 ± 6.4	45.2 ± 7.4	0.585
Mean of CD8, %	29.5 ± 7.7	27.3 ± 7.6	0.493
Mean of CD4/CD8 ratio	1.73 ± 0.7	1.79 ± 0.6	0.839

For the first two characteristics, *n* = 88 for BCC cases and 135 for controls; for the CD4 and CD8 studies, *n* = 11 BCC cases and 11 controls.

\*Percentage of lymphocytes responding to phytohemagglutinin within 72 hr.

†CAT<sub>0</sub> equals the scintillation counts of radioactivity of CAT assay from lymphocytes transfected with undamaged plasmid DNA containing CAT gene.

Table 4. Association of selected host characteristics with BCC in a clinic-based case-control study, Baltimore, Maryland, 1987-1990

Characteristic	Number, %		$\chi^2$ test, <i>P</i>
	BCC case ( <i>n</i> = 88)	Control ( <i>n</i> = 135)	
Age, yr			
20-40	19 (21.6)	36 (26.7)	
41-50	26 (30.0)	51 (37.8)	
51-60	43 (48.9)	48 (35.6)	0.141
Sex			
Male	48 (54.6)	68 (50.4)	
Female	40 (45.4)	67 (49.6)	0.542
Skin complexion*			
Type I/II	61 (69.3)	69 (51.1)	
Type III/IV	27 (30.7)	66 (48.9)	0.007
Smoking (100 cigarettes in a lifetime)			
Smoker	58 (65.9)	85 (63.0)	
Nonsmoker	30 (34.1)	50 (37.0)	0.654
Recent medication (at least 1 mo)			
Yes	29 (33.0)	45 (34.4)	
No	59 (67.1)	86 (65.7)	0.844

\*These data were recorded by a dermatologist according to Fitzpatrick skin typing: type I, severe burning (blistering) and no tanning; type II, moderate burning and mild tanning; type III, mild burning and moderate tanning; and type IV, no burning and intense tanning.

decade decline led to an accumulated 24.4% decrease in DRC. Although the age-related decline (apoptosis) in DNA-repair occurred in both BCC cases and controls, it only reached significance for the control group (Table 5, models 1 and 2). Model 2 is the only model that is not statistically different from the null model ( $P = 0.120$ ).

In keeping with the XP paradigm, those cases with first onset of BCC at young ages repair DNA photoproducts poorly when compared with controls. Cases developing their first cancers at later ages show little difference in DRC compared with their controls (see Fig. 1). This result suggests that poor DNA repair is associated with precocious skin aging, as manifested by an early age of first BCC. After adjusting for age at onset, the age-related decline in the repair of UV damage among the BCC cases was, at least, as sharp as that of controls ( $P = 0.003$ ). After controlling for age of subjects, the age at first onset of BCC correlated positively with DNA repair ( $P = 0.017$ ), indicating that the earlier the age of onset of BCC, the lower was their DNA repair (Table

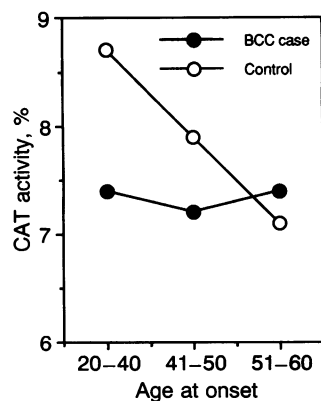


FIG. 1. Relationship between age at first BCC and DRC. The age-related decline in DRC among controls in comparison with that of age-matched cases is displayed. The linear-regression modeling and statistical tests of these data are presented in Table 5.

Table 5. Multiple linear-regression modeling of DNA repair related to risk factors among BCC and controls

Model*	Parameter	Estimate <sup>†</sup>	<i>t</i> value <sup>‡</sup>	<i>P</i> value <sup>§</sup>
1	Intercept	11.725	10.23	0.000
	Age in yr	-0.071	-3.47	0.000
	Sex	-0.409	-1.11	0.271
2	Intercept	10.445	6.96	0.000
	Age in yr	-0.047	-1.85	0.067
	Sex	-0.547	-1.26	0.211
3	Intercept	11.374	7.77	0.000
	Age in yr	-0.134	-3.09	0.003
	Sex	-0.360	0.84	0.404
	Age of onset	0.879	2.44	0.017
4	Intercept	11.569	12.81	0.000
	Age in yr	-0.067	-4.24	0.000
	Sex	-0.423	-1.51	0.133
	BCC FH	-0.645	-1.93	0.055

All models but model 2 significantly differ from a model with no risk factors according to the *F* test. The variables included in these linear-regression models are as follows: dependent variable is percentage of CAT activity at a UV dose of 700 J/m<sup>2</sup> (continuous variable); independent variables are age (at assay, in yr); age of onset (age at first BCC, in group): 1, <35; 2, 35-44; 3, 45-54; 4, 55-60; sex: 1, male; 2, female; BCC family history (FH): 1, without family history of BCC, 2, with BCC family history.

\*Model: 1, control only (*n* = 135) with *F* value = 6.35 and  $R^2 = 0.088$  and  $P = 0.002$ ; 2, BCC case only (*n* = 88) with *F* value = 2.17 and  $R^2 = 0.049$  and  $P = 0.120$ ; 3, BCC case only (*n* = 88) with *F* = 3.51 and  $R^2 = 0.111$  and  $P = 0.019$ ; 4, all subjects (*n* = 223) with *F* = 7.44,  $R^2 = 0.092$  and  $P = 0.0001$ .

<sup>†</sup>Least-square estimate of regression coefficient from multiple linear-regression models.

<sup>‡</sup>Student's *t* test for the null hypothesis that the estimate is equal to zero.

<sup>§</sup>Two-sided Student's *t* test.

5, model 3). The group mean comparison revealed that the mean DRC of all BCC cases (*n* = 88) is 5% lower than that of all controls (*n* = 135), and the difference in means is of borderline significance ( $P = 0.103$ ). When the controls with a family history of BCC and with actinic keratosis are removed from comparison (Table 6), the mean of the cases is 8% lower than that of controls, and the difference between means is statistically significant ( $P = 0.047$ ). These findings suggest that heredity can influence DNA-repair levels in the general population. After further adjustment for age and sex, prior family history of BCC is a statistically significant indicator of the individuals' DRCs, regardless of whether they were BCC cases or control subjects (Table 5, model 4).

When the relationship between age at first occurrence of BCC and family history of the disease was examined, ≈45% of cases first diagnosed with BCC between the ages of 20 and 44 yr (*n* = 38) reported that their relatives had had BCC, whereas only 10% of those cases who had their first BCC at ages 55-60 yr (*n* = 21) had relatives with BCC. This trend was

Table 6. Comparison of DCC between BCC patients and cancer-free controls

	<i>n</i>	Mean ± SD*	% difference <sup>†</sup>	<i>t</i> test <sup>‡</sup>
Controls without FH or actinic keratosis	106	8.00 ± 2.2	0.0	Reference
Controls with FH or actinic keratosis	29	7.28 ± 2.2	-9.0	0.126
BCC case	88	7.35 ± 2.0	-8.1	0.047

FH, family history of BCC.

\*Mean percentage of CAT activity at UV dose to plasmid of 700 J/m<sup>2</sup> and its SD.

<sup>†</sup>Percentage reduction relative to controls (reference group), which were labeled as 0% difference.

<sup>‡</sup>Student's *t* test for comparison of means to reference group.

Table 7. Relationship of family history of BCC to the age of BCC onset

Subjects	<i>n</i>	BCC family history, no. (%)	$\chi^2$ test for distribution,* <i>P</i>
Controls	135	21 (15.6)	
BCC onset			
20–44	38	17 (44.7)	
45–54	29	10 (34.5)	
55–60	21	2 ( 9.5)	0.022

\*For BCC cases only.

statistically significant (Table 7). In contrast, only 16% of controls (*n* = 135) had a family history of BCC.

The proportion of those cases (36%) who experienced sunlight overexposure in their lifetime is double that of controls (17%). Those subjects who had six or more severe sunburns (blistering) in their lifetime had significantly lower DRCs in the BCC group than in the control group (a 15% difference, *P* = 0.044). Any measure of sunlight exposure affecting skin susceptibility—such as light skin type, poor tanning ability, or high burning tendency—showed a similar difference in DNA-repair between BCC cases and controls (8). These data suggest that if subjects are exposed to the genotoxic effects of environmental UV light at doses that traumatize skin, individuals with poor DRC are more likely to develop skin cancer than those with high DRC. These interactive effects between DNA-repair levels and UV exposure are shown graphically in Fig. 2. When the population is divided into high and low repair according to the median level of the controls, the estimated risk (odds ratio) of skin cancer after excessive sunlight exposure is almost five times higher in subjects with low repair but is only 1.9 times higher in those with high repair. Further stratification according to gender revealed that females with a history of six or more severe sunburns who had low DRC were at 10 times greater risk for BCC compared with those individuals with high DRC (Fig. 3). These data are consistent with the results in the previous small pilot study (7).

**DISCUSSION**

In this study, we found that DRC decreases as age increases at a rate of 0.61% per annum. In a 40-yr period, there is a cumulative 25% loss in repair ability among controls. In addition to this physiological decline, a family history of skin cancer is associated with reduced DNA repair. This reduction is associated with an early onset of BCC. The occurrence of skin cancer in the aged is due to the interactive effects of both the natural decline in DNA repair and excessive sunlight exposure. Hence, reduced DNA repair is a host susceptibility factor for BCC.

The age-related decline in DNA repair may result in an accumulation of persistent DNA damage. In turn, this should be manifested in an increase in mutation fixation. The mutation rate in the *HPRT* gene in human lymphocytes with increased age is reportedly between 1.3 and 1.6% per yr (10), which is compatible with the observed rate of decline in DNA repair. There are genetically linked repair-deficiency diseases—e.g., Cockayne syndrome and XP—in which probands also manifest premature aging (11–15). In XP patients, the defect in DRC is associated with age-related skin changes and development of skin cancer 20 yr earlier than the normal population (14). Other workers have reported age-related changes in unscheduled DNA synthesis in lymphocytes (16, 17) and epidermal cells (18) from elderly blood donors. The age-related decline in DNA-repair activity of normal lymphocytes has been seen in repair of UV (16, 19), x-ray (17, 20), and  $\gamma$  irradiation (21). A 30% reduction in DNA repair was seen previously in normal peripheral leukocytes from skin of 58 normal subjects aged 20–90 yr (16). These findings are consistent with the age-related decline

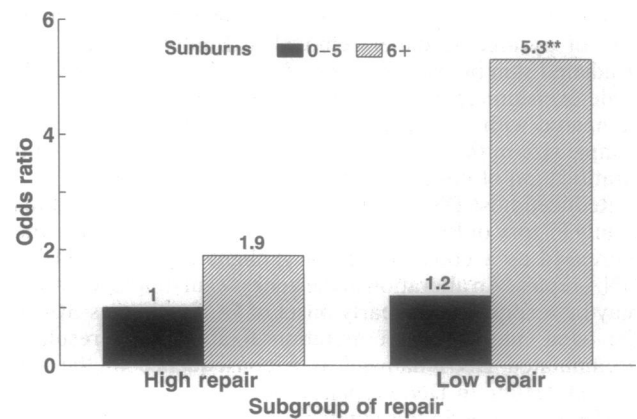


FIG. 2. Effect of DRC on risk of BCC: Relationship to number of severe sunburns in a lifetime. The reference group has an odds ratio of 1. The significantly increased age- and-sex-adjusted odds ratio [(5.32; 95% confidence interval (CI), 2.04–13.9)] is seen only in those who had both low DNA repair and six or more severe sunburns in their lifetime. \*\*, *P* < 0.01.

in DRC observed in this and other studies, although results have not been consistent (22–24).

A fundamental question is whether the genotoxicant–DNA-repair paradigm of XP is related to the carcinogenic response of individuals who possess marginal DRCs but who have oversaturated their repair system with excessive sunlight exposure. Having been exposed to sufficient sunlight to have caused six or more severe sunburns in a lifetime appears to be associated with an increased incidence of BCC in a large prospective study (25). In this study a high proportion of BCC patients had six or more severe sunburns, which probably reflects mutation fixation as a consequence of repair saturation and persistent photochemical DNA damage. After UV exposure, individuals with a family history of skin cancer and reduced DNA repair are likely to develop this disease sooner than others of the same age.

A few other studies have reported changes in the DNA repair of skin cancer patients. In an early BCC case–control study (26), a difference was seen in the UV-light induced unscheduled DNA synthesis of peripheral blood lymphocytes of cases (*n* = 29, aged 25–83 yr) and controls (*n* = 25, aged 25–83 yr). This difference was significant in patients with both BCC and squamous cell carcinoma (*n* = 10) but not in those

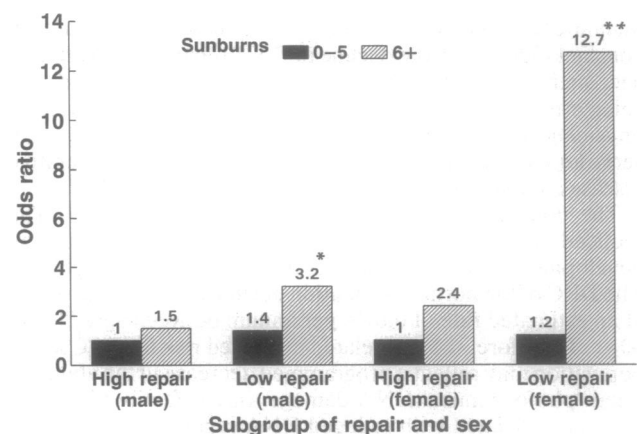


FIG. 3. Effect of DRC on risk of BCC by gender: Relationship to number of severe sunburns in a lifetime. The data in this graph are processed as for Fig. 2. The significantly increased age-adjusted odds ratio is seen in both males (3.22; 95% confidence interval, 0.97–10.7) and females (12.7; 95% confidence interval, 2.28–71.1) in those who had both low DNA repair and six or more severe sunburns in their lifetime. \*\*, *P* < 0.01; \*, *P* < 0.05.

with BCC lesions only ( $n = 19$ ). A recent study, in which the rate of pyrimidine dimer removal in fibroblasts from UV-irradiated skin biopsies was tested, found that reduced nucleotide-excision repair occurred in patients ( $n = 22$ ) with BCC compared with cancer-free subjects ( $n = 19$ ) (27). The small sample size in these studies did not permit age adjustment or stratification of known risk factors for skin cancer.

Reduced host DRC as a susceptibility factor is linked to a family history of BCC in this study. The risk of BCC is further increased as a consequence of sunlight exposure and low DNA repair. An alteration in the genes regulating DNA repair may be responsible for early onset of BCC, whereas BCC in the aged may be from mutation fixation as a result of accumulated DNA damage as a consequence of the age-related decline in DNA repair.

The assay used in this study has a number of technological advantages. One benefit of using the CAT assay to measure DNA repair is that it is the transfecting DNA that is damaged and not the host cell. This advantage minimizes any cytotoxic effects of these damaging agents that may indirectly compromise the endogenous repair mechanisms of the cell. The study focuses on skin cancer, a condition which is nonlethal and treated surgically without any confounding factors associated with chemotherapy or radiation treatment. In addition, this study emphasizes cancers in young individuals in whom the disease is rare and the effects of any risk factors can be maximized.

The findings from this study contribute to the understanding of the role of DNA repair in carcinogenesis in the general population. The probability of the early onset of mutation fixation because of reduced DRC suggests that the paradigm of XP applies to skin cancer in the general population. The age-related decline in DRC eventually will place all individuals at risk when their repair mechanisms can no longer accommodate the excess cumulative genotoxic damage. The maximum rate of pyrimidine dimer repair in a normal skin cell is reported to be  $5 \times 10^4 \text{ cell}^{-1}\text{hr}^{-1}$  (28), which is barely sufficient to cope with the rate at which damage is imposed on skin in full sunlight (29). Hence, a 25% decline in DNA repair in the 40-yr period between 20 and 60 yr places DNA repair at a sensitive focal point in the accumulation of persistent damage. This sensitivity may be responsible for tumor-suppressor gene mutations seen in skin cancer (6).

The consequences of accumulated damage may be particularly important in those postmitotic tissues where the dilution of damage by replicative mechanisms is severely diminished. There is a postnatal loss of DNA polymerases under developmental conditions in which repair enzymes remain relatively constant (30). Given the decline in DNA repair as a function of age, such tissues should be particularly vulnerable during the aging process because of their lack of regeneration. Therefore, physiological differences, such as those reported here, may serve to explain the elevated incidence of neurodegenerative diseases in many DNA-repair deficiency diseases (31).

The conclusions from our study are as follows. (i) The reduced host DRC of T lymphocytes correlates with the development of BCC in subjects overexposed to sunlight. (ii) The DRC of the normal population declines with increased age at an estimated rate of 0.61% per annum between ages 20 and 60 yr. Therefore, the age-related increased risk for BCC in the population may reflect a programmed decrease in the ability to repair photochemical DNA damage. (iii) A family history of BCC is associated with reduced DNA repair and early onset of this disease. (iv) The results of the study suggest that the persistence of DNA damage that leads to skin cancer may be directly attributed to reduced DRC, either from hereditary predisposition or from cumulative effects due to aging.

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