Association between Family History of Cancer and Mutagen Sensitivity in Upper Aerodigestive Tract Cancer Patients[1]

Melissa L. Bondy,[2] Margaret R. Spitz, Susan Halabi, John J. Fueger, Stimson P. Schantz, Dory Sample, and T. C. Hsu

Departments of Epidemiology [M. L. B., M. R. S., S. H., J. J. F.], Head and Neck Surgery [D. S.], Cell Biology [T. C. H.], The University of Texas M. D. Anderson Cancer Center, Houston, Texas 77030, and Department of Head and Neck Surgery, Memorial Sloan Kettering Cancer Center, New York, New York 10021 [S. P. S.]

Abstract

This study evaluated the relationship between family history of cancer and bleomycin-induced mutagen sensitivity. The study included 108 patients who registered at The University of Texas M. D. Anderson Cancer Center from June 1987 to June 1991 with histologically confirmed and previously untreated squamous cell carcinoma of the upper aerodigestive tract. All patients underwent the mutagen sensitivity assay and completed a self-administered risk evaluation questionnaire, including a detailed family history. The patients reported having 650 first-degree relatives, including 54 cases with cancers. The patients were classified as mutagen sensitive (≥1 chromosome break/cell) or not mutagen sensitive (≤0.99 chromosome breaks/cell). Odds ratios (ORs) were calculated to test for significant associations between mutagen sensitivity and family history of cancer. We found a significant OR (OR = 2.63; 95% confidence interval = 1.06–6.53) for patients who were mutagen sensitive and had one first-degree relative affected with cancer. For mutagen-sensitive patients with two or more first-degree relatives affected with cancer, the OR increased to 6.59 (95% confidence interval = 1.69–25.72). Although 88% of the patients were ever smokers, cigarette smoking was not found to be related to mutagen sensitivity. The study findings suggest that patients who have defective DNA repair capability as evidenced by the mutagen sensitivity assay are significantly more likely to report a family history of cancer than patients who are not mutagen sensitive. Further studies are needed to confirm that mutagen-sensitive individuals have inherited an increased risk of cancer.

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[2] To whom requests for reprints should be addressed, at M. D. Anderson Cancer Center, University of Texas, 1515 Holcombe Boulevard, Box 189, Houston, TX 77030.

Introduction

Family history of cancer has been used to identify high-risk families that may be potentially important for determining candidate cancer genes (1). Familial aggregations of lung cancer have been documented (2–8), but there has been only one study of the possibility of such aggregation in relatives of patients with upper aerodigestive tract cancers (9). Because patients with upper aerodigestive tract cancers incur high risks of lung cancer as a second primary tumor, one might predict similar familial patterns of cancer. Such a study would be important because these cancers provide a paradigm for evaluating gene-environment interactions, since, as is the case for lung cancer, only a fraction of exposed individuals develop aerodigestive tract cancers.

Hsu has hypothesized a wide spectrum of DNA repair capability within the general population and developed an assay to provide an indirect measure of such repair capability. Case-control evaluation of upper aerodigestive tract cancer has shown that mutagen sensitivity (as quantified by ≥ 0.8 induced break/cell) is an independent risk factor, after controlling for smoking and alcohol use (10). Studies of patients with lung and colon cancer support these findings (11). If this mutagen sensitivity is genetically determined, one might predict that cancer would be more likely to aggregate in relatives of mutagen-sensitive patients than in those who are not mutagen sensitive (12).

This study investigated self-reported cancer histories of patients with upper aerodigestive tract cancer and used a mutagen sensitivity assay as an indirect measure of DNA repair capability (11). We predicted that mutagen-sensitive individuals who might have inherited deficiencies in DNA repair capability would be more likely to report a family history of cancer.

Materials and Methods

Study Population. This study was part of a case-control investigation of upper aerodigestive tract cancers designed to evaluate mutagen sensitivity as an independent risk factor (13) and included 108 patients with histologically confirmed and previously untreated squamous cell carcinoma of the upper aerodigestive tract who registered at the The University of Texas M. D. Anderson Cancer Center from June 1987 to June 1991. Upper aerodigestive tract sites included the oral cavity, pharynx, and larynx. The study patients completed a self-administered cancer risk evaluation questionnaire that elicited a detailed family history of the patient's first-degree relatives (parents, siblings, and offspring) and their medical histories; dates of birth, death, and cancer diagnosis; and age at diagnosis. Each patient was also asked to provide a blood sample before initiation of therapy.

Mutagen Sensitivity Assay. The assay has been described in detail elsewhere (10, 11). Briefly, on the third day of incubation, standard lymphocyte cultures were treated with bleomycin (0.03 units/ml), a radiomimetic drug, for 5 h, and in the fifth hour, the cells were treated with colcemid (0.04 μg/ml) to stop them in mitosis before they were harvested for preparation of conventional air-dried slides. The slides were coded and stained with Giemsa without banding, and the average number of chromosome breaks scored in 50 metaphases/sample was counted by one of us, who was unaware of the disease status of each individual. Only frank chromatid breaks or exchanges were recorded; chromatid gaps or attenuated regions were disregarded. Any individual who expressed 1 or more chromosome breaks/cell was considered mutagen sensitive, and persons with less than 1 break/cell were considered not mutagen sensitive. In the earlier paper (10) we used a less conservative estimate of 0.8 breaks/cell for classifying mutagen-sensitive individuals; however, we prefer a more conservative estimate of 1 break/cell for this study.

Statistical Analysis. The patients were classified as mutagen sensitive (≥1 break/cell) or not mutagen sensitive (≤0.99 chromosome breaks/cell) and ORs,[3] as an estimate of the relative risk, were calculated to test for significant associations between mutagen sensitivity and family history of cancer. Ninety-five % confidence limits were computed by the method of Woolf as detailed in Schlesselman (14).

We used standardized incidence ratios to compare overall observed and expected cancers in relatives to determine whether relatives of upper aerodigestive tract cancer patients had more cancers than would be expected in the general population. The expected number of cancers in the general population was determined using age-, sex-, and calendar year-specific rates from the Connecticut Tumor Registry incorporated in a computer program by Monson (15). Person-years at risk were determined from the date of birth to the date of death, cancer diagnosis, or interview, or to age 75, whichever came first. Individuals were considered at risk only until age 75 because of the poor reliability of cancer rates at older ages (16). The 95% CI for the standardized incidence ratio was determined by assuming a Poisson distribution for the observed number of cancers (17).

Results

The study included 108 subjects, 70 (65%) men and 38 (35%) women. Of these, one case had been adopted and thus dropped from the analysis. Using the mutagen assay, 49 (50%) patients were classified as not mutagen sensitive, 55 (47%) as sensitive, and 3 (3%) could not be classified. Therefore, the analysis was limited to 104 patients with mutagen sensitivity values. There were 41 subjects with cancer of the oral cavity, 31 subjects with cancers of the pharynx, and 32 were diagnosed with laryngeal cancer (Table 1). The mean number of breaks per cell for subjects with oral cavity cancer was 1.01, 1.05 for subjects with cancer of the pharynx, and 0.94 for subjects diagnosed with cancer of the larynx.

Table 1 Distribution of patient characteristics by mutagen sensitivity

Characteristic	Not mutagen sensitive (≤0.99 breaks/cell) (n = 49)	Mutagen sensitive (≥1.00 breaks/cell) (n = 55)
Sex		
Male	30 (61.2)[a]	38 (69.1)
Female	19 (38.8)	17 (30.9)
Age at diagnosis (years)		
<40	16 (32.7)	8 (14.5)
40–49	6 (12.2)	8 (14.5)
50–59	15 (30.6)	11 (20.0)
60–69	7 (14.3)	17 (31.0)
70+	5 (10.2)	11 (20.0)
Mean age at diagnosis (years)	50.1	56.7
Upper aerodigestive site		
Oral cavity	21 (42.8)	20 (36.4)
Pharynx	14 (28.6)	17 (30.9)
Larynx	14 (28.6)	18 (32.7)
Cigarette smoking		
Yes	43 (87.8)	49 (89.1)
Never	5 (10.2)	5 (9.1)
Unknown	1 (2.0)	1 (1.8)
No. of cigarettes per day		
1–14	6 (17.1)	3 (7.7)
14–24	15 (42.9)	18 (46.2)
25+	14 (40.0)	18 (46.2)
First-degree relatives with cancer		
None	35 (71.4)	23 (41.8)
1	11 (22.4)	19 (34.6)
2+	3 (6.2)	13 (23.6)

[a] Numbers in parentheses, percentage.

Table 2 Odds ratios for mutagen sensitivity

Variable	Odds ratio	95% CI
Family history of cancer		
1 first-degree relative	2.63	1.06–6.53
2+ first-degree relatives	6.59	1.69–25.72
Ever smoked cigarettes	1.14	0.31–4.20

Thirty patients (28%) had one first-degree relative affected with cancer, and 16 (15%) reported two or more affected relatives (Table 1). Overall, the patients identified 650 first-degree relatives (parents, siblings, and offspring) including 54 with reported cancers. There were no significant differences by gender or smoking status in the distribution of mutagen sensitivity (Table 1). Surprisingly, patients not mutagen sensitive were younger on average than were mutagen-sensitive patients. Almost one-third of those not sensitive were under 40 years of age compared with 14.5% of the patients who were mutagen sensitive (P = 0.05).

We compared family history and mutagen sensitivity status to determine whether mutagen-sensitive patients were more likely than patients who were not mutagen sensitive to report a family history of cancer. We found a significant OR (OR = 2.63; 95% CI = 1.06–6.53) for those patients who were mutagen sensitive and had one first-degree relative affected with cancer (Table 2). We

[3] The abbreviations used are: OR, odds ratio; CI, confidence interval.

Table 3 Cancers in relatives of upper aerodigestive tract cancer patients by mutagen sensitivity

Relationship to proband	≤0.99 breaks/cell				≥1.0 breaks/cell			
	No. of relatives at risk	Observed cancers	Expected[a] cancers	Obs./Exp.[b]	No. of relatives at risk	Observed cancers	Expected cancers	Obs./Exp.
Mother	49	5	8.37	0.60	55	8	10.27	0.78
Father	49	8	7.59	1.05	55	10	9.90	1.02
Sibling	110	2	6.87	0.29	130	18	14.92	1.21
Offspring	88	1	0.85	1.17	114	2	1.26	1.59
Total	296	16	23.67	0.68	354	38	36.35	1.05

[a] Person-years calculated through age 75 years.
[b] Observed/expected cancers.

observed a 6-fold increase in the OR when we compared patients with two or more affected first-degree relatives by sensitivity status (OR = 6.59; 95% CI = 1.69–25.72).

We also evaluated the relationship between cigarette smoking and mutagen sensitivity. Eighty-eight % of the patients had ever smoked cigarettes, of whom 49 were mutagen sensitive and 43 not mutagen sensitive. Only 10 patients had not smoked more than 100 cigarettes in their lifetime. Of these, five were mutagen sensitive and five were not sensitive (Table 1). Our study found that cigarette smoking was unrelated to mutagen sensitivity (OR = 1.14; 95% CI = 0.31–4.20) (Table 2).

Patients who were mutagen sensitive had twice as many relatives with cancer as patients who were not mutagen sensitive (38/354 = 11% compared with 16/296 = 5%). The mean age of the relatives at cancer diagnosis was 53.7 years for patients who were not mutagen sensitive and 57.3 for those who were mutagen sensitive. The difference was not statistically significant. The standardized incidence ratio analysis showed no overall excess of cancer (Table 3) or excess of specific kinds of cancer or cancer among relatives diagnosed before age 45, and there was no excess when the data were partitioned by proband's age at cancer diagnosis (data not shown). In addition, we partitioned the data by relatives with smoking-related cancers (respiratory tract, pancreas, esophagus, kidney, bladder, and cervix) and compared the groups by mutagen sensitivity. Again, there was no difference between the two groups.

Discussion

Our findings suggest that patients with upper aerodigestive tract cancers who are mutagen sensitive are six times more likely to have one or more first-degree relatives with cancer than are relatives of patients who are not mutagen sensitive. The mutagen sensitivity assay is an indirect measure of DNA repair capability, as indicated by the number of unrepaired breaks induced by the mutagen bleomycin, which causes direct DNA strand breakage in mammalian cells (11, 18). Liang *et al.* (19) reported one cancer-prone family in which all living relatives who had cancer were hypersensitive to chromosome breakage by bleomycin. They suggested that genetic defects that affect chromosomal breakage and repair may contribute to cancer development. Our results and the finding of Liang *et al.* (19) suggest that further studies are needed to test families at high risk for cancer to determine whether they exhibit mutagen sensitivity.

Our findings are consistent with Knudson's two-hit theory that two independent mutations are necessary for tumor development, therefore explaining variation in the occurrence of malignancy among genetically susceptible individuals (20). In our study population, the first hit may have had a genetic basis that could be identified by molecular studies of candidate genes. The second hit could be the result of multiple environmental exposures, together with a reduced ability to repair the resultant mutagenic damage.

Smoking has been documented as a major risk factor for upper aerodigestive tract cancer, and the population-attributable risk associated with cigarette smoking is 85% (21). However, only a fraction of tobacco-exposed individuals will develop smoking-related aerodigestive tract cancer. These interindividual differences in susceptibility seem to be heritable, and thus cancer may aggregate in families. Our study supported the finding that individuals with susceptible genotypes are more likely to report familial cancer aggregation.

This was a pilot study and has led to an effort to validate these findings in a larger study of lung cancer. Among the limitations to this hypothesis-generating study were small sample size and reliance on the proband's self-reports. Cancer information reported by the proband was not validated with medical documentation. Perhaps 10–15% of the cases may be inaccurately reported, since Bondy *et al.* (22) found 88% accuracy of reported cancer information in first-degree relatives compared with medical documentation. A more complete evaluation of the observed and expected cancers would also be important in a larger data set. The difficulty in determining the separate or combined roles of hereditary and environment also limits the study. Unmeasured environmental exposures in the relatives may confound the results, and other environmental exposures besides smoking might be responsible for the difference in mutagen sensitivity in the patients. A more conclusive study administering the mutagen sensitivity test to high-risk families would give a more precise description of gene-environment interactions. Because the potential preventive implications of validating biological markers of genetic susceptibility are immense, further determinations of the mutagen sensitivity of relatives of individuals with a family history of cancer are called for. High-risk individuals (and their relatives) can be targeted for intensive behavioral modification and screening surveillance. The utility of markers in recruiting such individuals into chemoprevention trials is also under investigation.

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