Short Communication

A Modified Host Cell Reactivation Assay to Measure DNA Repair Capacity for Removing 4-Aminobiphenyl Adducts: A Pilot Study of Bladder Cancer

Jie Lin,1 Fred F. Kadlubar,2 Margaret R. Spitz,1 Hua Zhao,1 and Xifeng Wu1
1Department of Epidemiology, University of Texas M.D. Anderson Cancer Center, Houston, Texas and 2National Center for Toxicological Research, Jefferson, Arizona

Abstract

As DNA repair plays an important role in genetic susceptibility to bladder cancer, assessment of the DNA repair phenotype is critical for the molecular epidemiology of bladder cancer. In this study, we developed and applied an assay using the luciferase (luc) reporter gene in a host-cell reactivation assay to measure DNA repair capacity for DNA damage induced by 4-aminobiphenyl (4-ABP), a well-studied aromatic amine and a known bladder carcinogen. We observed a dose-response relationship for 4-ABP dosage and DNA repair capacity (luc activity). We then applied this assay to measure DNA repair capacity in a pilot study of 89 pairs of bladder cancer patients and healthy controls matched by age, gender, and ethnicity, and we found that DNA repair capacity was significantly lower in cases than in controls (13.0% versus 14.4%; \( P = 0.006 \)). Poor DNA repair capacity was associated with 3.42-fold increased bladder cancer risk. Further analysis revealed that intermediate and low levels of DNA repair capacity increased bladder cancer risk to 3.43-fold and 4.97-fold, respectively, compared with individuals with the most efficient DNA repair capacity. Moreover, ever smokers with suboptimal DNA repair capacity exhibited a 6.06-fold increased risk compared with never smokers with normal DNA repair capacity. In conclusion, our results support the hypothesis that deficient DNA repair capacity for 4-ABP induced DNA damage and increases bladder cancer risk. Our assay provides a new tool to specifically quantify DNA repair capacity in bladder cancer studies and, therefore, contributes to our goal of further elucidating bladder carcinogenesis. (Cancer Epidemiol Biomarkers Prev 2005;14(7):1832–6)

Introduction

Bladder cancer is a common cancer in the United States, especially in men (1). Cigarette smoking and occupational exposure have been reported as major risk factors for bladder cancer, but it is believed that genetic variation further modifies an individual’s susceptibility to the disease. Individuals with suboptimal DNA repair capacity, a genetically controlled phenotype, may be at increased risk of developing bladder cancer upon exposure to carcinogens that may have a lower carcinogenic potential in individuals with normal DNA repair capacity. Thus, developing functional assays that can quantify individual’s DNA repair capacity is crucial for identifying high-risk subgroups in the general population. To date, a variety of assays have been developed to assess DNA repair capacity (2-10). Among them, the host cell reactivation assay, an \textit{in vitro} assay, has the capability to assess the repair efficiency of specific excision repair pathways by using relevant challenge mutants. This assay has been implemented to quantify DNA repair capacity in studies of lung cancer (5, 6), skin cancer (7, 8), head and neck cancer (9), and prostate cancer (10). The aromatic amine, 4-aminobiphenyl (4-ABP), found in cigarette smoke, synthetic fossil fuels, and the by-products of the rubber, dye, textile, coal, and printing industries, has been unambiguously documented as a major human bladder carcinogen (11). 4-ABP can covalently interact with DNA to form adducts that induce mutation and initiate bladder carcinogenesis (12, 13). The concordance between the unique binding spectrum of metabolically activated 4-ABP along the human \( p53 \) gene and specifically at \( p53 \) mutational hotspots in bladder cancer provides molecular evidence linking 4-ABP and bladder cancer etiology (14). In this study, we developed a host cell reactivation assay to measure DNA repair capacity for removing 4-ABP–induced DNA damage by establishing the optimal 4-ABP dose and comparing repair capacity in normal and deficient cell lines. Because 4-ABP is a specific carcinogen for bladder cancer, we further applied the assay in a pilot bladder cancer case-control study to test the hypothesis that suboptimal DNA repair capacity of 4-ABP–induced DNA damage was associated with increased bladder cancer risk.

Materials and Methods

Lymphoblastoid Cell Lines. We first constructed a dose-response curve to determine the optimal 4-ABP dosage using two repair-deficient and two repair-proficient EBV-immortalized human lymphoblastoid cell lines. The repair-deficient lines (GM02248, an XP-C line and GM02345, an XP-A line) were purchased from the Human Genetic Mutant Cell Repositories (Camden, NJ) of the National Institute for General Medical Sciences; and two repair-proficient lines were obtained from healthy blood donors. In addition, we also compared DNA repair capacity between normal and bladder cancer cell lines using 10 EBV-immortalized human lymphoblastoid cell lines: five from healthy blood donors and five from bladder cancer patients. The cells were cultured in RPMI 1640 supplemented with 15% FCS (Life Technologies, Gaithersburg, MD) at 37°C and 5% \( \text{CO}_2 \).

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Requests for reprints: Xifeng Wu, Department of Epidemiology, University of Texas M.D. Anderson Cancer Center, Unit 1340, 1155 Hermann Pressler Boulevard, Houston, TX 77303. Phone: 713/745-2485; Fax: 713/792-4657. E-mail: xwu@mdanderson.org

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Isolation of Lymphocytes and Cell Culture. The lymphocytes were isolated by Ficoll (Pharmacia Biotech, Inc., Piscataway, NJ) gradient centrifugation and suspended in freezing medium containing 50% fetal bovine serum, 40% RPMI 1640 (Life Technologies), and 10% DMSO (at 10^7 cells/mL). Aliquots of 2.0 mL were stored in an −80°C freezer and later thawed in batches for the host cell reactivation assays. The minimum amount of blood needed for the assay is 5 mL.

Plasmids and 4-ABP Treatment. The plasmid expression vector pGL-3 containing luciferase reporter gene was used for the LUC assays. The plasmid was treated with 4-ABP in Dr. Kadlubar’s laboratory (National Center for Toxicological Research, Jefferson, AR) and used for all assays in this study. In the 4-ABP treatment, 1 mL pGL-3 plasmid (500 µg/mL) was treated with 200, 400, and 800 µmol/L N-N-trifluoro-N-acetoxy-4-ABP (15), and incubated at 37°C for 3 hours. After incubation, the plasmid DNA was precipitated with 99% ethanol, washed with 75% ethanol, dissolved in 0.1 mol/L Tris-EDTA buffer at 0°C in a water bath and mixed (before the last trace of ice disappeared) with 5 mL of thawing medium (50% fetal bovine serum, 40% RPMI 1640, and 10% dextrose). Then, the cells were centrifuged at 900 rpm for 10 minutes and resuspended in RPMI 1640 (supplemented with 20% fetal bovine serum). Next, the cells were cultured in RPMI 1640 supplemented with 20% FCS and 56.25 µg/mL phytohemagglutinin (Murex Diagnostics, Norcross, GA) at 37°C and 5% CO2 and incubated for 72 hours. The lymphoblasts from each subject were divided into four aliquots, each containing 2.0 × 10^6 cells. The DEAE-dextran (Pharmacia Biotech) method was used to transfect two aliquots with undamaged pGL-3 and two with pGL-3 damaged by 4-ABP. The cultures were then incubated for 40 hours after transfection.

Transfection. The cells in each frozen vial were thawed quickly at 37°C in a water bath and mixed (before the last trace of ice disappeared) with 5 mL of thawing medium (50% fetal bovine serum, 40% RPMI 1640, and 10% dextrose). Then, the cells were centrifuged at 900 rpm for 10 minutes and resuspended in RPMI 1640 (supplemented with 20% fetal bovine serum). Next, the cells were cultured in RPMI 1640 supplemented with 20% FCS and 56.25 µg/mL phytohemagglutinin (Murex Diagnostics, Norcross, GA) at 37°C and 5% CO2 and incubated for 72 hours. The lymphoblasts from each subject were divided into four aliquots, each containing 2.0 × 10^6 cells. The DEAE-dextran (Pharmacia Biotech) method was used to transfect two aliquots with undamaged pGL-3 and two with pGL-3 damaged by 4-ABP. The cultures were then incubated for 40 hours after transfection.

LUC Assays. Briefly, the cell pellets were suspended in 50 µL of reporter lysis buffer (Promega Corp., Madison, WI), frozen, and thawed once in ethanol-dry ice and 37°C water baths and then centrifuged at 1,400 × g for 15 seconds. For each LUC assay, 20 µL of cell extract supernatant was mixed with 100 µL of Luciferase Assay Substrate (Promega) in a 12 × 50 mm tube at room temperature. LUC activity in arbitrary light-intensity units was measured with a luminometer (Lumat LB 9507; Berthold Technologies GmbH, Bad Wildbad, Germany). LUC light-intensity units were recorded for the cells with undamaged plasmids (control reading) and 4-ABP-damaged (repair reading) plasmids. DNA repair capacity (%) was calculated as the product of 100% and the ratio of the damaged plasmid values to the undamaged plasmid values.

Study Population of the Pilot Study. For this pilot study, 89 incident urinary bladder cancer cases were selected from a pool of participants enrolled in an on-going case-control study of bladder cancer. Selection was based on the adequacy of stored lymphocytes. The case-control study started in 1999 and is on-going. The recruitment goal was 1,000 incident bladder cancer patients and 1,000 age-, gender-, and ethnicity-matched controls. To date, ~840 cases and 750 controls have been enrolled in the study. Bladder cancer patients were recruited from the Departments of Urology at University of Texas M.D. Anderson Cancer Center and Baylor College of Medicine (Houston, TX). To participate, patients had to have newly diagnosed and histologically confirmed urinary bladder cancer and no previous chemotherapy or radiotherapy. There were no recruitment restrictions on age, gender, or cancer stage. The controls were recruited in collaboration with the Kelsey-Seybold clinics, the largest private multispecialty physician group consisted of >23 clinics and over 300 physicians in the Houston metropolitan area. The potential controls were identified by reviewing short survey forms distributed to patients coming to the Kelsey-Seybold Clinics for annual health check-ups. The short questionnaire elicited willingness to participate in this study and provided basic demographic information for matching. Research interviewers reviewed the demographic information and identified potential controls that could possibly be matched (by age, gender, and ethnicity) with bladder cancer cases already recruited. Research interviewers then contacted eligible controls explaining the nature of our study, confirming willingness to participate and eligibility. If a potential eligible control agreed to participate, an in-person interview was scheduled at one of the Kelsey-Seybold clinics convenient to the participant. The participation rate was around 75%. After obtaining written informed consent, trained M.D. Anderson staff interviewers administered risk factor questionnaire to study participants. Other information gathered from the interview included demographics (age, gender, ethnicity) and smoking history. Following the interview, a 40 mL blood sample was drawn into coded heparinized tubes and immediately hand-delivered to the laboratory for lymphocyte isolation. From our long-term experience conducting this study, we obtained blood samples for over 99% eligible participants. Blood sample was collected before patients received chemotherapy or radiation therapy. Patients who had these treatments before were considered ineligible and were, therefore, not recruited. Human subject approval was obtained from the M. D. Anderson and the Kelsey-Seybold institutional review boards.

An individual who has never smoked or has smoked <100 cigarettes in his or her lifetime was defined as a never smoker. A former smoker was a person who had quit smoking at least 1 year before diagnosis (cases) or who had quit smoking at least 1 year before the interview (controls). A current smoker was someone who was currently smoking or who had stopped <1 year before being diagnosed with bladder cancer (cases) or interview (controls). Former smoker and current smoker combined were defined as ever smoker.

Statistical Analysis. The correlation of DNA repair capacity (%) between lymphoblastoid cell lines and lymphocytes was tested using the Spearman’s rank correlation analysis. Differences between cases and controls in the distributions of smoking status, gender, and ethnicity were tested using the χ² test. The Student’s t test was used to test for differences in continuous variables between the cases and controls. When the distribution of a variable deviated from normal, Wilcoxon rank sum test was used. For DNA repair capacity, we checked the normality of the distributions separately for cases and controls. The two distributions were not significantly different from normal in terms of kurtosis but slightly in skewness. There were no noticeable outliers that could distort the distribution or had substantial influence on the overall results. We first log-transformed the data, did the analyses based on both log-transformed data and untransformed data, and compared the results. Given that results obtained from log-transformed data were consistent with untransformed data, we only reported results based on untransformed data. Logistic regression was used to calculate odds ratios as an estimate of the relative risk associated with DNA repair capacity. Multivariate logistic regression was done to adjust for residual confounding effects of age, gender, ethnicity, and smoking status, where appropriate. All statistical analyses were done using the SAS 8.2 statistical software package (SAS Institute, Inc., Cary, NC). All tests were two-sided with a significance level of 0.05.

Results

The dose-response effect of DNA repair capacity (%) at the 40-hour time point for the 4-ABP DNA repair capacity assay was shown in Fig. 1. As the 4-ABP dosages increased from 0 to 600...
µmol/L, the LUC activity decreased proportionally in all cell lines. At the dosages of both 200 and 400 µmol/L, the curves were completely differentiated for repair-proficient and repair-deficient lines. At 400 µmol/L, the mean values reached 32.20% and 30.82% for two repair-proficient lines, but they reached only 11.71% and 12.65% for the deficient GM02345 and GM2248 lines, respectively. Each data point was based on an average of two independent experiments.

Five cell lines from bladder cancer patients and five age-, gender-, and ethnicity-matched cell lines from healthy blood donors were selected to compare DNA repair capacity (%) using the dosages of 200, 400, and 600 µmol/L (Fig. 2). The control cell lines exhibited significantly higher DNA repair capacity (%) compared with the case cell lines at the dosages of 200 and 400 µmol/L (both \( P < 0.05 \)). Although a similar pattern was observed at 600 µmol/L, the difference did not reach statistical significance \( (P = 0.18) \). Therefore, for all future experiments, we selected 400 µmol/L as the 4-ABP treatment dosage.

To test the correlation of DNA repair capacity (%) between lymphoblastoid cell lines and lymphocytes, we did the assay in 10 lymphoblastoid cell lines and 10 matched lymphocytes (from the same donors). A statistically significant correlation in DNA repair capacity (%) was observed between lymphoblastoid cell lines and lymphocytes \( (\rho = 0.79, P < 0.01) \), indicating the feasibility of applying the assay to lymphocytes. We further tested the reproducibility of the assay by repeating the assay twice using the 400 µmol/L dosage on different days with primary lymphocytes. The average coefficient of variation was 7.11%, indicating high reproducibility of the assay.

We next applied the assay in a pilot bladder cancer case-control study to test the hypothesis that deficient DNA repair capacity for removing 4-ABP–induced DNA damage is associated with increased risk of bladder cancer. A total of 89 patients and 89 controls were included and well matched by age, gender, and ethnicity. Males constituted 89.9% of the study population. Among the 89 case-control pairs, 80 (89.9%) were Caucasians, 6 (6.7%) were African Americans, and 3 (3.4%) were Mexican Americans (Table 1). The mean ages of the cases and controls were 60.1 years (range, 18-83 years) and 59.9 years (range, 22-82 years), respectively (Table 1). There were no significant differences in the distributions of ethnicity or gender between cases and controls. However, significant difference in the smoking status between the cases and controls was noted \( (P < 0.001) \). Specifically, 31.5% of cases compared with 5.6% of controls were current smokers; in contrast, 21.4% of cases and 42.7% of controls were never smokers. In addition, the self-reported median pack-years in smokers was significantly higher in cases than in controls (38.8 for cases and 15.9 for controls, \( P < 0.001; \) Table 1).

The 25th, median, and 75th percentiles of the DNA repair capacity were 10.37, 13.08, 17.22 for the controls and 9.10, 12.41, and 15.42 for the cases, respectively. The mean DNA repair capacity was significantly lower in cases (13.0%) than in controls \( (14.4, \ P = 0.006; \) Table 1). Using the 75% percentile DNA repair capacity in controls as the cutoff point (to form comparable groups of cases and controls to provide a stable estimate of risk), we observed a 3.42-fold \( [95\% \text{ confidence interval} (95\% \text{ CI}), 1.07-10.91] \) increased risk of bladder cancer associated with suboptimal DNA repair capacity after adjusting for age, gender, ethnicity, and smoking status in a multivariate logistic model (Table 2). We further divided the DNA repair capacity into low (<25th percentile), intermediate (≥25th and <75th percentile), and efficient (≥75th percentile) categories and assessed risks associated with each DNA repair capacity category. Using individuals with the most efficient DNA repair capacity (≥75th percentile in controls) as the reference group, intermediate and low levels of DNA repair capacity increased bladder cancer risk by 3.43-fold \( (95\% \text{ CI}, 1.07-10.97) \) and 4.97-fold \( (95\% \text{ CI}, 1.11-22.23) \), respectively, and significant risk was observed for low DNA repair capacity category compared with the intermediate DNA repair capacity category.

![Figure 1](image1.png)  
**Figure 1.** Dose-response curve of the LUC activity in lymphoblastoid cell lines. Each data point was based on an average of two independent experiments.

![Figure 2](image2.png)  
**Figure 2.** Distribution of LUC activity in lymphoblastoid cell lines by case-control status at different dosages of 4-ABP treatments. Each data point was based on an average of two independent experiments.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Cases ((n = 89))</th>
<th>Controls ((n = 89))</th>
<th>(P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex, (n)%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>80 (89.9)</td>
<td>80 (89.9)</td>
<td>1.00*</td>
</tr>
<tr>
<td>Female</td>
<td>9 (10.1)</td>
<td>9 (10.1)</td>
<td></td>
</tr>
<tr>
<td>Ethnicity, (n)%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Caucasian</td>
<td>80 (89.9)</td>
<td>80 (89.9)</td>
<td></td>
</tr>
<tr>
<td>African American</td>
<td>6 (6.7)</td>
<td>6 (6.7)</td>
<td></td>
</tr>
<tr>
<td>Mexican American</td>
<td>3 (3.4)</td>
<td>3 (3.4)</td>
<td>&lt;1.00*</td>
</tr>
<tr>
<td>Smoking status, (n)%</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never smoker</td>
<td>19 (21.4)</td>
<td>38 (42.7)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Former smoker</td>
<td>42 (47.2)</td>
<td>46 (51.7)</td>
<td></td>
</tr>
<tr>
<td>Current smoker</td>
<td>28 (31.5)</td>
<td>5 (5.6)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Age, mean (SD)</td>
<td>60.1 (11.2)</td>
<td>59.9 (10.9)</td>
<td>0.94</td>
</tr>
<tr>
<td>Range</td>
<td>18.83</td>
<td>22.82</td>
<td></td>
</tr>
<tr>
<td>Pack-year, median (range)</td>
<td>38.8 (0-176)</td>
<td>15.8 (0-123)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>DNA repair capacity, (mean (SD))</td>
<td>13.0 (4.7)</td>
<td>14.4 (5.5)</td>
<td>0.006</td>
</tr>
</tbody>
</table>

*\(x^2\) test.  
†Student’s \(t\) test.  
‡Wilcoxon rank sum test.

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reach statistical significance ($P = 0.80$; Fig. 3). We then stratified the analysis by age, smoking status (never versus ever), and pack-years smoked. We observed significantly increased risk associated with poor DNA repair capacity in older subjects (age $\geq 61$ years) and ever smokers. The odds ratios were $2.83$ (95% CI, $1.02-7.83$) and $2.51$ (95% CI, $1.01-6.27$), respectively (Table 2). When stratified by pack-years, the risk was higher in heavy smokers (pack-years $\geq 15.9$) than in light smokers (pack-years $< 15.9$), although neither of the odds ratios was significant (Table 2).

We further assessed the joint effects of smoking and DNA repair capacity. Compared with the reference group (never smokers with efficient DNA repair capacity), never smokers with suboptimal DNA repair capacity showed a 1.93-fold increased risk (95% CI, 0.36-10.45). The risk was 2.46-fold (95% CI, 0.41-14.68) for ever smokers with high DNA repair capacity. However, the risk in ever smokers with suboptimal DNA repair capacity was substantially elevated (odds ratio, 6.06; 95% CI, 1.18-31.18) although the interaction term did not reach statistical significance ($P = 0.80$; Fig. 3).

### Discussion

It is well documented that cigarette smoking and exposure to industrially related aromatic amines are major risk factors for bladder cancer. These carcinogens cause DNA damage, which, if not repaired, can initiate tumorogenesis. Thus, a deficiency in DNA repair capacity can lead to genomic instability and carcinogenesis, and individuals with a suboptimal repair capacity may be at an increased risk of developing different kinds of cancer, including bladder cancer. In the case of 4-ABP, although the detailed molecular mechanisms involving the repair of 4-ABP induced DNA damage remain unclear, nucleotide excision repair is thought to be the major mechanism. Nucleotide excision repair mainly removes bulky DNA adducts, which are typically generated from exposure to environmental carcinogenic agents. In this study, we used a modified host cell reactivation assay to quantify the DNA repair capacity of 4-ABP–induced damage. Because 4-ABP is a bladder cancer–specific carcinogen, the application of our assay has great potential for identifying subgroups with high risk of bladder cancer.

In developing this assay, we established the best 4-ABP dosages that successfully differentiated repair-deficient cell lines from repair-efficient cell lines. Our data also showed a high correlation between DNA repair capacity obtained from cell lines and DNA repair capacity obtained from lymphocytes, suggesting the applicability of this assay in molecular epidemiologic studies. Unlike the cytogenetic assays or the comet assay that indirectly infer DNA repair capacity from cellular damage remaining after mutagenic exposure and recovery, the original host cell reactivation assay, upon which our new assay is based, quantifies the cellular ability to remove adducts from plasmids transfected into lymphocyte cultures by measuring the expression of damaged reporter genes. In the host cell reactivation assay, the lymphocytes are not treated and the cells remain undamaged, thus the DNA repair capacity derived from the assay objectively reflects intrinsic cellular repair capacity. Utilizing the host cell reactivation assay, Spitz et al. (6) reported that suboptimal DNA repair capacity was associated with increased lung cancer risk in a dose-response manner. Similar findings have been reported for other cancers (7-10). In these studies, DNA repair capacity was measured as the capability of the host cells to remove bulky DNA adducts derived from benzo(a)pyrene, a major constituent of tobacco smoke or to repair UV light–induced DNA damage. A unique feature of our study is our assessment of bladder cancer risk via the measurement of cellular DNA repair capacity specific to 4-ABP, a well-established bladder carcinogen. Instead of using the chloramphenicol (CAT) gene, a dose-response trend was apparent ($P_{\text{trend}} = 0.04$). We then stratified the analysis by age, smoking status (never versus ever), and pack-years smoked. We observed significantly increased risk associated with poor DNA repair capacity in older subjects (age $\geq 61$ years) and ever smokers. The odds ratios were $2.83$ (95% CI, 1.02-7.83) and $2.51$ (95% CI, 1.01-6.27), respectively (Table 2). When stratified by pack-years, the risk was higher in heavy smokers (pack-years $\geq 15.9$) than in light smokers (pack-years $< 15.9$), although neither of the odds ratios was significant (Table 2).

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### Figure 3.

Joint effects of DNA repair capacity and smoking in bladder cancer risk. DNA repair capacity (%) was dichotomized by the 75th percentiles in controls.

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**Table 2. DNA repair capacity and bladder cancer risk**

<table>
<thead>
<tr>
<th>Smoking status</th>
<th>Never smoker</th>
<th>Reference</th>
<th>Adjusted OR* (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4-ABP (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>17.22</td>
<td>12 (13.5)</td>
<td>22 (24.7)</td>
<td>3.42 (1.07-10.91)</td>
</tr>
<tr>
<td>22.22</td>
<td>11 (12.8)</td>
<td>27 (29.5)</td>
<td>2.83 (1.02-7.83)</td>
</tr>
</tbody>
</table>

*Multivariate logistic regression adjusted for age, gender, smoking status, and ethnicity where appropriate.

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**NOTE:** In stratified analysis, age was dichotomized by median and pack-year was dichotomized by median in controls.

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**Abbreviations:** OR, odds ratio; DRC, DNA repair capacity.
we used the luciferase (LUC) gene, which has several advantages over CAT as the reporter gene: The LUC assay is not radioactive and less labor intensive, it results in less interassay variation, and it requires fewer cells (16). These features make the LUC assay more suitable for molecular epidemiologic studies than the CAT assay.

The potential of our assay was tested in a pilot case-control study. We found that bladder cancer patients exhibited significantly lower DNA repair capacity compared with healthy controls. More importantly, our results showed that suboptimal DNA repair capacity of 4-ABP-induced DNA damage was associated with significantly increased risk of bladder cancer in a dose-dependent manner. Further stratified analysis showed that the risk was only significant in ever smokers, suggesting a possible joint effect of DNA repair capacity and smoking in bladder cancer risk. In addition, our data showed that ever smokers with suboptimal DNA repair capacity were at a >6-fold increased risk of bladder cancer compared with never smokers with efficient DNA repair capacity, supporting that poor DNA repair capacity may act in concert with smoking to increase bladder cancer susceptibility. However, due to the relatively small sample size, our findings should be interpreted cautiously and need to be validated in a larger molecular epidemiologic study.

Our assay is the first assay to link intrinsic cellular repair capacity and bladder cancer risk. It is, therefore, a step toward our objective of identifying subgroups with low DNA repair capacity and elevated cancer risk. Our finding that suboptimal DNA repair capacity is associated with increased bladder cancer risk adds to the existing literature of different cancer sites, strengthening the hypothesis that deficient DNA repair capacity acts as a risk factor for a variety of human epithelial cancers, and that the assay can be tailored to specific cancer sites with well-established carcinogen etiologic links.

References
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