Null Results in Brief

No Association between the XPD 312, 751, or XRCC1 399 Polymorphisms and K-ras Gene Mutation in Smoking Non-Small-Cell Lung Cancer

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Introduction

Lung cancer is strongly associated with exposure to tobacco smoke (1–3). Mutations in the K-ras gene have been found in 20–35% of lung adenocarcinomas of smokers (4–8), compared with about 5–7% in those of nonsmokers (7, 8), suggesting that their formation may be associated with exposure to tobacco smoke carcinogens.

DNA repair helps preserve the integrity of the cellular genome by repairing DNA damage induced by tobacco smoke carcinogens (9). Some polymorphic variants of DNA repair genes, such as the nucleotide excision repair gene xeroderma pigmentosum group D (XPD) polymorphisms at codons 312 and 751, and the base excision repair X-ray repair cross-complementing group 1 (XRCC1) polymorphism at codon 399, have been associated with an increased risk of lung cancer (10–13). In addition, our recent study showed that lung cancer cases who were smokers and carried the XPD 312 Asp/Asp genotype had a higher incidence of p53 mutations in their lung tumors (14).

In the present study, we analyzed K-ras mutations in lung tumors of these same lung cancer cases and investigated the relationship between the presence of these mutations and the status of DNA repair polymorphism, specifically the XPD Asp312Asn, XPD Lys751G1n, and XRCC1 Arg399Gln genotypes, of the cases.

Materials and Methods

Detailed description of the 204 smoking non-small-cell lung cancer Caucasian cases (153 adenocarcinomas, 40 squamous cell carcinomas, 7 adenosquamous carcinomas, and 4 large cell carcinomas) and the XPD 312, XPD 751, and XRCC1 399 genotypes have been published previously (14, 15). Briefly, these patients consisted of 130 males and 74 females. Patient age at diagnosis ranged between 38 and 92 years (median = 66). The genotypes were analyzed in genomic DNA from tumors using the ABI Prism 7700 sequence detector (TaqMan; Applied Biosystems, Foster City, CA). For quality control, genotype determinations were run as duplicates and we observed a 100% concordance rate. K-ras mutations occurring at codon 12 were analyzed by PCR-DGGE as described previously (8, 16). Statistical analysis of the data was carried out using STATA 6.0 software for Windows. Fisher’s exact and χ2 tests were employed to test the association between genotypes and K-ras mutation when appropriate. Odds ratios (ORs) and 95% confidence intervals (CIs) were calculated by stepwise unconditional multivariate logistic regression models controlling for age and sex. A trend test (a nonparametric test for trend across ordered groups) for the ordered groups generating from the genotype combination was also analyzed.

Results and Discussion

Mutations at codon 12 of the K-ras gene were detected in 64 of 204 (31%) lung cancer cases, including 39 (61%) G to T transversions, 22 (34%) G to A transitions, and 3 (5%) G to C transversions. The predominance of a G to T transversion is consistent with previous studies (4–7), suggesting that carcinogens in cigarette smoke may cause these mutations. Also, the mutation frequency observed in adenocarcinomas (37%, 57 of 153) was significantly higher than that in squamous cell carcinomas (15%, 6 of 40) (P = 0.008). This is consistent with results of several studies, which demonstrated that K-ras mutations were identified more frequently in lung adenocarcinomas than in squamous cell carcinoma (4, 6, 7).

The frequencies of the XPD 312 Asn (34%), XPD 751 Gln (37%), and XRCC1 399 Gln (36%) (14) were in Hardy-Weinberg equilibrium and were consistent with those reported in previous studies (10–12).

As shown in Table 1, there is no appreciable association between the presence of K-ras mutations in lung tumors and any of the XPD 312, XPD 751, or XRCC1 399 genotype. Individuals with the XPD 312 Asp/Asp or the XPD 312 Asn allele had a K-ras mutation...
Table 1. Comparison between XPD and XRCC1 polymorphisms and K-ras mutations

<table>
<thead>
<tr>
<th>K-ras mutation OR (95% CI)</th>
<th>XPD</th>
<th>Negative</th>
<th>Positive</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lys/Lys</td>
<td>55</td>
<td>27</td>
<td>1.00</td>
</tr>
<tr>
<td>Lys/Gln</td>
<td>67</td>
<td>27</td>
<td>0.81 (0.42–1.57)</td>
</tr>
<tr>
<td>Gln/Gln</td>
<td>18</td>
<td>10</td>
<td>1.12 (0.45–2.79)</td>
</tr>
<tr>
<td>Lys/Gln + Gln/Gln</td>
<td>85</td>
<td>37</td>
<td>0.88 (0.47–1.63)</td>
</tr>
<tr>
<td>XRCC1 Arg399Gln</td>
<td>Arg/Arg</td>
<td>57</td>
<td>28</td>
</tr>
<tr>
<td>Arg/Gln</td>
<td>67</td>
<td>23</td>
<td>0.71 (0.36–1.39)</td>
</tr>
<tr>
<td>Gln/Gln</td>
<td>16</td>
<td>13</td>
<td>1.80 (0.74–4.41)</td>
</tr>
<tr>
<td>Arg/Gln + Gln/Gln</td>
<td>83</td>
<td>36</td>
<td>0.91 (0.49–1.69)</td>
</tr>
</tbody>
</table>

*ORs and 95% CIs are adjusted for age and sex.

In summary, our previous study showed a relationship between an individual polymorphism or a combination of polymorphisms and the presence of p53 mutations (14). In this study, we observe no significant association (P > 0.05) between the three polymorphisms screened and the status of K-ras mutations in these lung cancer cases. The reason for this difference is not clear. We have previously identified p53 mutations among 20% of these same 204 lung cancer cases analyzed in this study (14). Among the 92 lung tumors positive for p53 and/or K-ras mutations, only 13% (12 of 92) had both mutations, suggesting that these two gene mutations primarily define separate events in lung cancer. One study demonstrated that glutathione S-transferase 1 (GSTM1) null genotype, which plays a role in the detoxification of B(a)P diol epoxide (BPDE) in tobacco smoke, was associated with K-ras gene mutations in lung adenocarcinoma of smokers (17). It is possible that deficiencies of the activities of susceptibility genes other than XPD and XRCC1 may contribute to the observed mutations at codon 12 of the K-ras gene.

References

Table 2. Comparison between the types of K-ras mutations and XPD and XRCC1 genotypes

<table>
<thead>
<tr>
<th>K-ras mutation</th>
<th>XPD</th>
<th>XPD 312</th>
<th>XPD 751</th>
<th>XRCC1 399</th>
</tr>
</thead>
<tbody>
<tr>
<td>Transversions</td>
<td>(n = 42)</td>
<td>Asp/Asp</td>
<td>Asp/Asn + Asn/Asn</td>
<td>Lys/Lys + Lys/Gln + Gln/Gln + Arg/Arg + Arg/Gln + Gln/Gln</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Transitions (n = 22)</td>
<td>Total</td>
<td>Total</td>
</tr>
<tr>
<td>Transversions</td>
<td>(n = 42)</td>
<td>Asp/Asp</td>
<td>Asp/Asn + Asn/Asn</td>
<td>Lys/Lys + Lys/Gln + Gln/Gln + Arg/Arg + Arg/Gln + Gln/Gln</td>
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