Gene polymorphisms of the DNA Repairing Genes APE1 and XRCC1 among Smoking Lung Cancer Egyptians


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Abstract

Lung cancer is the leading cause of cancer-related mortality worldwide and is thus a major public health problem. DNA base damage or losses caused by endogenous and exogenous agents occur constantly at a high frequency in human cells. The removal or repair of damaged bases is an important mechanism in protecting the integrity of the genome.

APE1 (Apurinic/Apyrimidinic Endonuclease 1) and XRCC1 (X-ray cross-complementing group1) are DNA repair proteins that play important roles in the base excision repair (BER) pathway. The focus of this work is limited to the association between polymorphisms in the DNA repair genes, (APE1 Asp148Glu (2197 T→G) and XRCC1 Arg399Gln (28152 G→A) genotypes), cigarette smoking and lung cancer. This study has included 131 cases affected with lung cancers include; 33 cases with small cell carcinoma (25.2%) and 98 cases with non-small cell carcinoma (74.8%).

They were recruited from oncology Center, Mansoura University, Egypt; in the period between April 2008 to March 2010. For comparison, a negative control group including 150 healthy individuals randomly selected from blood donors. Controls were selected by random sampling cancer-free individuals without a past history of cancer, who visited Mansoura University hospitals and provided peripheral blood between April 2008 and March 2010.

DNA was extracted from the whole peripheral blood using generation DNA purification capture column kit (Gentra system, USA) and genotyping for APE1 Glu148Asp and XRCC1 Arg399Gln polymorphisms was performed by a PCR–CTPP (PCR with confronting two-pair primers) method.

The collected data were organized and statistically analyzed using SPSS statistical computer package version 10 software. we observed that, There were no significant differences in the frequencies of the APE1 Asp148Glu (2197 T→G) polymorphism of all genotypes and alleles in all lung cancer cases compared to all healthy controls. Also, there were no significant differences in the frequencies of all genotypes and alleles in lung cancer cases compared to controls of all smoking status. While, in former smoking individuals; significant difference in the frequency of the mutant GG genotype in lung cancer cases compared to controls (p-value=0.003).

We observed that, There were no significant differences in the frequencies of the XRCC1 Arg399Gln (28152 G→A) polymorphism of all genotypes and alleles in all lung cancer cases compared to all healthy controls. Also, there were no significant differences in the frequencies of all genotypes and alleles in lung cancer cases compared to controls of all smoking status. While, in never smoking individuals; significant difference in the frequency of the mutant AA genotype in lung cancer cases compared to controls (p-value=0.0004).

Keywords: Gene, polymorphisms, DNA Repairing Genes, APE1, XRCC1, Lung Cancer.
Introduction
Lung cancer is the leading cause of cancer-related mortality worldwide (Edwards et al., 2005) and is thus a major public health problem. DNA base damage or losses caused by endogenous and exogenous agents occur constantly at a high frequency in human cells. The removal or repair of damaged bases is an important mechanism in protecting the integrity of the genome. Oxidized DNA bases and alkylated DNA bases can be removed and replaced with the correct ones in a localized burst of DNA synthesis by DNA base excision repair pathway, which mobilizes an array of proteins (Fortini et al., 2003). These proteins play an important role in repairing immediate DNA base damage caused by exposure to environmental agents and endogenous reactive oxygen species (ROS) as well as alkylating species (Christmann et al., 2003).

Defects in DNA repair can give rise to hypersensitivity to carcinogens and the accumulation of DNA lesions in the genome, and lead to the development of cancer. Cigarette smoke contains many carcinogens and reactive oxygen species that can induce various types of DNA damage (Hecht, 2002). Although cigarette smoking is a major risk factor in the development of lung cancer, only 10% to 15% of all smokers develop lung cancer, suggesting that variation in individual susceptibility to tumorigenesis of lung cancer (Mattson et al., 1987; Shields & Harris, 2000 and Shields, 2002). Susceptibility differences may be inherited in genes encoding for the metabolism of carcinogens or DNA repair molecules, which are essential in protecting the genomic integrity of the cells (Caporaso et al., 1991; Goode et al., 2002 and Spitz et al., 2003).

A single nucleotide polymorphism (SNP) is a common genetic variation. Some SNPs may have a functional impact on health outcome and contribute to the overall population risk of cancer. SNPs in DNA repair genes have been suggested to be risk factors for lung cancer but the current available results have been inconsistent (Kiyohara et al., 2002).

One of the DNA repair pathways is the DNA base excision repair pathway, which repairs the DNA damage caused by oxidation and alkylation and thus protects cells against the toxic effects of endogenous and exogenous agents (Hoeijmakers, 2001 and Fleck & Nielsen, 2004). DNA damage induced by reactive oxygen species (ROS) may be repaired by the base excision repair (BER) pathway (Mandal et al., 2012).

Specifically, the damaged bases of purine and pyrimidine are recognized and excised by specific DNA glycosylases, leaving abasic sites. Apurinic/ apyrimidinic endonuclease (APE) then incise the DNA 5'-to the abasic sites; further repair proceeds to short-patch (when the gap is only one nucleotide) or long-patch (when the gap is two or more nucleotides) sub-pathways of base excision repair (Krokan et al., 2000). The major human APE, APE1 (also known as APE, APEX, HAP1, and REF-1), plays a central role in the base excision repair pathway. APE1 is also known as a transcriptional co-activator for numerous transcription factors involved in cancer development (Bhakat et al., 2009) and is considered as a promising tool for anticancer
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therapy (Tell et al., 2010). As a member of APE, it initiates repair of apurinic/apyrimidinic sites in DNA produced either spontaneously hydrolyzing the 5'-phosphodiester bond of the apurinic/apyrimidinic site or after enzymatic removal of damaged bases. The repair activity of APE1 serves to protect the cell from the apurinic/apyrimidinic sites that can accumulate in DNA via endogenous and exogenous sources. In addition to APE activity, it can also act as a 3'-phosphodiesterase, initiating repair of DNA strands breaks with 3'-blocking damage, which are produced either directly by reactive oxygen species or indirectly through the enzymatic removal of damaged bases (Izumi et al., 2000 and Fortini et al., 2003).

**Material and methods**

**Studied patients:**

This study has included 131 cases affected with lung cancers include; 33 cases with small cell carcinoma (25.2%) and 98 cases with non-small cell carcinoma (74.8%). They were recruited from oncology Center, Mansoura University, Egypt; in the period between April 2008 to March 2010.

For comparison, a negative control group including 150 healthy individuals randomly selected from blood donors. Controls were selected by random sampling cancer-free individuals without a past history of cancer, who visited Mansoura University hospitals and provided peripheral blood between April 2008 and March 2010. They were confirmed not to have cancer by the hospital-based cancer registry system by the end of 2010.

**Genotyping procedure:**

DNA was extracted from the whole peripheral blood using generation DNA purification capture column kit (Gentra system, USA) and genotyping for APE1 Glu148Asp and XRCC1 Arg399Gln polymorphisms was performed by a PCR-CTPP (PCR with confronting two-pair primers) method (Hamajima et al., 2000).

For the APE1 Glu148Asp (2197 T to G) polymorphism, extracted DNA was amplified with the four primers by `Ampli Taq Gold' (Perkin-Elmer, Foster City, CA); F1, 50-CCT ACG GCA TAG GTG AGA CC-30; R1, 50-TCC TGA TCA TGC TCC TCC-30; F2, 50-TCT GTT TCA TTT CTA TAG GC G AT-30; and R2, 50-GTC AAT TTC TTC ATG TGC CA-30 . PCR conditions were 1-min denaturation at 95_C followed by 30 cycles of 95_C for 1 min, 60_C for 1 min, and 72_C for 1 min, with a 5-min extension at 72_C. Primer pairs F1 and R1 for the G allele (148Glu), F2

XRCC1, one of more than twenty genes that participate in the base excision repair pathway, has multiple roles in repairing DNA base damage and single-strand DNA breaks (Thompson & West, 2000). Inconsistent results have been reported regarding the associations between the Arg399Gln (exon 10) polymorphism of XRCC1 and either functional significance or the risk of tobacco associated cancers (Ratnasinghe et al., 2001; Butkiewicz et al., 2001; David-Beabes & London, 2001 and Park et al., 2002). We studied the association between genetic variation of DNA base excision repair pathway genes (APE1 and XRCC1), smoking and lung cancer.
and R2 for the T allele (148Asp) produced respectively, as well as a 360-bp common band. For XRCC1 Arg399Gln (28152 G to A), extracted DNA was amplified with the four primers by ‘Ampli Taq Gold’ (Perkin-Elmer, Foster City, CA); F1, 50-TCC CTG CGC CGC TGC AGT TTC T-30; R1, 50-TGG CGT GTG AGG CCT TAC CTC C-30; F2, 50-TCG GCG GCT GCC CTC CCA-30; and R2, 50-AGC CCT CTG TGA CCT CCC AGG C-30. PCR conditions were 1-min denaturation at 94_C followed by 30 cycles of 94_C for 1 min, 59_C for 1 min, and 72_C for 1 min, with a 10-min extension at 72_C. Primer pairs F1 and R1 for the G allele (399Arg) and F2 and R2 for the A allele (399Gln) produced allele-specific bands of 447- and 222-bp, respectively, as well as a 630-bp common band.

Statistical analysis:
The collected data were organized and statistically analyzed using SPSS statistical computer package version 10 software.

Statistical measurements included:
1- Mean ± SD: Descriptive data included mean and standard deviation of variables of cases.
2- Genotype frequency and percentage frequency by counting subjects with certain genotype polymorphisms and dividing by the number of total studied subjects either cases or controls.
3- Allele frequency and percentage frequency by counting subjects with certain allele polymorphisms (2 for each subject) and dividing by the number of studied chromosomes.

Results
The Descriptive data of the studied lung cancer cases and healthy controls are shown in table (1). The average age of the lung cancer patients was 55.2 ± 10.5 while that of the control group was 51.5 ± 11.5

| Table (1): Descriptive data of the studied lung cancer cases and healthy controls. |
|-------------------------------------|-------------|-----------------|
| Total:                              | Cases n(%)  | Controls n (%)  |
|                                    | 131         | 150             |
| Sex:                                |             |                 |
| Male                                | 92 (70.2)   | 140 (93)        |
| Female                              | 39 (29.8)   | 10 (7)          |
| Age at diagnosis(years):            |             |                 |
| <=49 years                          | 34 (26)     | 60 (40)         |
| 50-59 years                         | 47 (35.9)   | 48 (32)         |
| >=60 years                          | 50 (38.1)   | 42 (28)         |
| Mean age ± SD                      | 55.2 ± 10.5 | 51.5 ± 11.5     |
| Diagnosis:                          |             |                 |
| Small cell carcinoma                | 33 (25.2)   | -               |
| Non-small cell carcinoma            | 98 (74.8)   | -               |
| Smoking status:                     |             |                 |
| Never smokers                       | 37 (28.2)   | 8 (5.3)         |
| Former smokers                      | 11 (8.4)    | 15 (10)         |
| Current smokers                     | 83 (63.4)   | 127 (84.7)      |

n= number of studied cases, (%) = percentage of studied cases.

The genotype and allele distributions of the APE1 Asp148Glu (2197 T→G) polymorphism in lung cancer cases and controls were shown in table (2). Between cases and control groups displayed no significant differences in the frequencies of all genotypes and alleles (p-value>0.05).
Table (2): Frequencies of APE1 Asp148Glu (2197 T→G) genotypes and allelic polymorphisms in lung cancer cases compared to healthy controls.

<table>
<thead>
<tr>
<th>Total</th>
<th>APE genotypes</th>
<th>Individual Alleles</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Asp/Glu (TG)</td>
<td>Asp/Asp (TT)</td>
</tr>
<tr>
<td></td>
<td>N (%)</td>
<td>N (%)</td>
</tr>
<tr>
<td>Controls</td>
<td>37 (24.7)</td>
<td>92 (61.3)</td>
</tr>
<tr>
<td>Cases</td>
<td>48 (36.6)</td>
<td>68 (51.9)</td>
</tr>
<tr>
<td>p -value</td>
<td>0.129</td>
<td>0.377</td>
</tr>
</tbody>
</table>

N= number; %= percentage; *p=< 0.05 significant; **p=<0.001 highly significant.

There were no significant differences in the frequencies of all genotypes and alleles in lung cancer cases compared to controls of all smoking status. While, in former smoking individuals; significant difference in the frequency of the mutant GG genotype in lung cancer cases compared to controls (p-value=0.003) as shown in table (3).

Table (3): Frequencies of APE1 Asp148Glu (2197 T→G) genotypes and allelic polymorphisms in lung cancer cases compared to healthy controls in the smoking status.

<table>
<thead>
<tr>
<th>Smoking status</th>
<th>APE genotypes</th>
<th>Individual Alleles</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Asp/Glu (TG)</td>
<td>Asp/Asp (TT)</td>
</tr>
<tr>
<td></td>
<td>N (%)</td>
<td>N (%)</td>
</tr>
<tr>
<td>Never Controls</td>
<td>2 (25)</td>
<td>5 (62.5)</td>
</tr>
<tr>
<td>Cases</td>
<td>14 (37.8)</td>
<td>20 (54.1)</td>
</tr>
<tr>
<td>p -value</td>
<td>0.106</td>
<td>0.437</td>
</tr>
<tr>
<td>Former Controls</td>
<td>5 (33.3)</td>
<td>6 (40)</td>
</tr>
<tr>
<td>Cases</td>
<td>5 (45.5)</td>
<td>5 (45.5)</td>
</tr>
<tr>
<td>p -value</td>
<td>0.169</td>
<td>0.552</td>
</tr>
<tr>
<td>Current Controls</td>
<td>30 (23.6)</td>
<td>81 (63.8)</td>
</tr>
<tr>
<td>Cases</td>
<td>29 (34.9)</td>
<td>43 (51.8)</td>
</tr>
<tr>
<td>p -value</td>
<td>0.139</td>
<td>0.264</td>
</tr>
</tbody>
</table>

N= number; %= percentage; *p=< 0.05 significant; **p=<0.001 highly significant.
Figure (1): PCR amplification for APE1 Asp148Glu (2197 T→G) polymorphisms using PCR-CTPP method; lane M: DNA size marker, lanes 1, 3, 5: Asp/Asp (TT) genotype; lanes 4, 7: Asp/Glu (TG) genotype; and lanes 2, 6: Glu/Glu (GG) genotype.

Figure (2): PCR amplification for XRCC1 Arg399Gln (28152 G→A) polymorphisms using PCR-CTPP method; lane M: DNA size marker, lanes 2, 6, 7: Arg/Arg (GG) genotype; lanes 1, 5: Arg/Gln (GA) genotype; and lanes 3, 4: Gln/Gln (AA) genotype.

Table (4): Frequencies of XRCC1 Arg399Gln (28152 G→A) genotypes and allelic polymorphisms in lung cancer cases compared to healthy controls.

<table>
<thead>
<tr>
<th>XRCC1 polymorphism</th>
<th>Arg/Gln (GA) N (%)</th>
<th>Arg/Arg (GG) N (%)</th>
<th>Gln/Gln (AA) N (%)</th>
<th>G N (%)</th>
<th>A N (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total Controls</td>
<td>139 (92.7)</td>
<td>7 (4.7)</td>
<td>4 (2.7)</td>
<td>153 (51)</td>
<td>147 (49)</td>
</tr>
<tr>
<td>Cases</td>
<td>122 (93.1)</td>
<td>6 (4.6)</td>
<td>3 (2.3)</td>
<td>134 (51.1)</td>
<td>128 (48.9)</td>
</tr>
<tr>
<td>p-value</td>
<td>0.975</td>
<td>0.975</td>
<td>0.858</td>
<td>1</td>
<td>1</td>
</tr>
</tbody>
</table>

N= number; %= percentage; *p< 0.05 significant; **p<0.001 highly significant.

There were no significant differences in the frequencies of all genotypes and alleles in all lung cancer cases compared to all healthy controls as shown in table (4).

Table (5): Frequencies of XRCC1 Arg399Gln (28152 G→A) genotypes and allelic polymorphisms in lung cancer cases compared to healthy controls in the smoking status.

<table>
<thead>
<tr>
<th>Smoking status</th>
<th>XRCC1 genotypes</th>
<th>Individual Alleles</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Arg/Gln (GA) N (%)</td>
<td>Arg/Arg (GG) N (%)</td>
</tr>
<tr>
<td>Never Controls</td>
<td>7 (87.5)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Cases</td>
<td>36 (97.3)</td>
<td>1 (2.7)</td>
</tr>
<tr>
<td>p-value</td>
<td>0.471</td>
<td>0.100</td>
</tr>
<tr>
<td>Former Controls</td>
<td>15 (100)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>Cases</td>
<td>11 (100)</td>
<td>0 (0)</td>
</tr>
<tr>
<td>p-value</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Current Controls</td>
<td>117 (92.1)</td>
<td>7 (5.5)</td>
</tr>
<tr>
<td>Cases</td>
<td>75 (90.4)</td>
<td>5 (6)</td>
</tr>
<tr>
<td>p-value</td>
<td>0.899</td>
<td>0.882</td>
</tr>
</tbody>
</table>
In table (5) there were no significant differences in the frequencies of all genotypes and alleles in lung cancer cases compared to controls of all smoking status. While, highly significant differences were found in the frequency of the mutant AA genotype, in never smoking lung cancer cases compared to controls (p-value=0.0004).

**Discussion**

DNA repair systems act to maintain genomic integrity in the face of environmental insults, cumulative effects of age, and general DNA replication errors. Tobacco smoke contains an array of potent chemical carcinogens and reactive oxygen species that may produce DNA bulky adducts, cross-links, oxidative or base DNA damage, and DNA strand breaks. Among the several major DNA repair pathways that operate on specific types of damaged DNA by cigarette smoking, base-excision repair (BER) is involved in repair of DNA base damage and single strand breaks (Berwick and Vineis, 2000). APE1 is an essential enzyme in the base excision repair pathway, which is the primary mechanism for the repair of endogenous DNA damage resulting from cellular metabolisms, including those resulting from reactive oxygen species, methylation, deamination, and hydroxylation (Hoeijmakers, 2001). It is estimated that 2,000 to 10,000 apurinic/apyrimidinic sites arise per day in mammalian cell grown under normal physiologic conditions (Lindahl, 1993). XRCC1, one of more than twenty genes that participate in the base excision repair pathway, has multiple roles in repairing DNA base damage and single-strand DNA breaks (Thompson and West, 2000).

Inconsistent results have been reported regarding the associations between the Arg399Gln (exon 10) polymorphism of XRCC1 and either functional significance or the risk of tobacco associated cancers (Park et al., 2002). XRCC1 is thought to play a role in the multistep base excision repair pathway where “non-bulky” base adducts produced by methylation, oxidation, reduction, or fragmentation of bases by ionizing radiation or oxidative damage are removed (Yu et al., 1999). Although the specific function of XRCC1 has not been identified, it is believed that XRCC1 complexes with DNA ligase III via a BRCT domain in its COOH terminus and with DNA polymerase b in its NH2 terminus to repair gaps left during base excision repair (Lunn et al., 1999).

In the current study, the genotype and allele distributions of the APE1 Asp148Glu (2197 T→G) polymorphism in lung cancer cases and controls were shown in table (2). Between cases and control groups displayed no significant differences in the frequencies of all genotypes and alleles (p-value>0.05). These results were in consistent with that of Taiwanese (Yen et al., 2009); Japanese populations (Ito et al., 2004); African-Americans populations (Chang et al., 2009); Chinese populations (Shen et al., 2005); Taiwanese populations (Lo et al., 2009); Latinos populations (Chang et al., 2009); Caucasian populations (Popanda et al., 2004);
and Caucasian populations (Zienolddiny et al., 2006).

In the present study, we found the frequency of the mutant G allele (rare-allele) of the APE1 Asp148Glu (2197 T→G) polymorphism in the Egyptian total lung cancer cases was (30%), this result is consistent with that of Caucasians populations (31%) (Zienolddiny et al., 2006). However, the frequencies of the same allele were reported in other studies on several populations, as following, Latinos populations (36%) (Chang et al., 2009); Afro-Americans populations (39%) (Chang et al., 2009); Taiwanese populations (39%) (Lo et al., 2009); Caucasian populations (45%) (Popanda et al., 2004); Chinese populations (49%) (Shen et al., 2005) and Japanese populations (42%) (Ito et al., 2004).

The frequency of the mutant G allele (rare-allele) of the APE1 Asp148Glu (2197 T→G) polymorphism in the Egyptian total healthy controls was (26%), while, in Caucasians populations was (37%) (Zienolddiny et al., 2006); in Afro-Americans populations was (38%) (Chang et al., 2009); in Taiwanese populations was (39%) (Lo et al., 2009); in Japanese populations was (39%) (Ito et al., 2004); in Chinese populations was (40%) (Shen et al., 2005); in Latinos populations was (42%) (Chang et al., 2009) and in Caucasian populations was (49%) (Popanda et al., 2004).

We found the frequency of the mutant GG genotype (rare-alleles) of the APE1 Asp148Glu (2197 T→G) polymorphism in the Egyptian total lung cancer cases was (11.5%), this result is consistent with that of Latinos populations (12.4%) (Chang et al., 2009) and lower than that of Japanese populations (18%) (Ito et al., 2004); Caucasian populations (19.4%) (Popanda et al., 2004); Taiwanese populations (16.32%) (Lo et al., 2009). Afro-Americans populations (15.7%) (Chang et al., 2009); Chinese populations (21.8%) (Shen et al., 2005) and Caucasian populations (23.3%) (Zienolddiny et al., 2006).

And the frequency of the mutant GG genotype (rare-alleles) of the APE1 Asp148Glu (2197 T→G) polymorphism in the Egyptian total healthy controls of this study was (14%), which agree with that of Japanese populations (14.3%) (Ito et al., 2004); Afro-Americans populations (14.6%) (Chang et al., 2009) and Chinese populations (13.3%) (Shen et al., 2005); but lower than that of Taiwanese populations (16.34%) (Lo et al., 2009); Latinos populations (18.7%) (Chang et al., 2009); Caucasian populations (23.2%) (Popanda et al., 2004); and Caucasian populations (29.5%) (Zienolddiny et al., 2006).

Generally, the differences in allele or genotype frequencies detected among these studies might be due to ethnic variation, heterogeneity of studied populations and different sample sizes.

The present results were in consistent with that of White Americans (Cote et al., 2009); Afro-Americans (Cote et al., 2009); Chinese populations (Ratnasinghe et al., 2001); Korean populations (Park et al., 2002); Japanese populations (Ito et al., 2004); Chinese populations (Zhang et al., 2005); Caucasian populations (Zhou et al., 2003); Chinese populations (Chen et al., 2002); Caucasian
populations (Hung et al., 2005); Latinos (Chang et al., 2009); Afro-Americans (Chang et al., 2009); Chinese populations (Shen et al., 2005); Caucasian populations (Popanda et al., 2004) and Caucasian populations (Zienolddiny et al., 2006).

The frequency of the mutant A allele rare-allele XRCC1 Arg399Gln (28152 G→A) polymorphism in the Egyptian total lung cancer cases was (48.9%), however that of Caucasian populations was (36.7%) (Popanda et al., 2004); in White Americans was (35.05%) (Cote et al., 2009); in Caucasian populations was (35.75%) (Zhou et al., 2003); in Caucasian populations was (35.6%) (Hung et al., 2005); in Chinese populations was (28.35%) (Zhang et al., 2005); in African-Americans was (15.2%) (Chang et al., 2009); in Chinese populations was (26.2%) (Ratnasinghe et al., 2001); in Korean populations was (28.35%) (Park et al., 2002); in Japanese populations was (26.45%) (Ito et al., 2004); in Latinos was (31.4%) (Chang et al., 2009); in Chinese populations was (24.3%) (Chen et al., 2002); in African-Americans was (15.15%) (Chang et al., 2009); in Chinese populations was (20.2%) (Shen et al., 2005) and in Caucasian populations was (33.95%) (Zienolddiny et al., 2006).

The frequency of the mutant AA genotype (rare-alleles) of the XRCC1 Arg399Gln (28152 G→A) polymorphism in the Egyptian total lung cancer cases was (2.3%), this result was consistent with that of Afro-Americans (1.6%) (Chang et al., 2009) and that of Chinese populations (3.4%) (Shen et al., 2005); however, that of Korean populations was (8.8%) (Park et al., 2002), which was agree with that of Caucasian populations (9%) (Zienolddiny et al., 2006).

The frequency of the mutant AA genotype (rare-alleles) of the XRCC1 Arg399Gln (28152 G→A) polymorphism in the Egyptian total healthy controls was (2.7%), this result was consistent with that of African-Americans (2.1%) (Chang et al., 2009) and that of Chinese populations (3.5%) (Shen et al., 2005); while, that of Japanese populations (5.8%) (Ito et al., 2004); that of Korean populations was (4.4%) (Park et al., 2002), which was agree with that of Black Americans.

(4.1%) (Cote et al., 2009); that of Caucasian populations was (13.1%) (Zienolddiny et al., 2006), which was agree with that of Caucasian populations (12.9%) (Hung et al., 2005); that of Caucasian populations was (11.5%) (Zhou et al., 2003), which was agree with that of White Americans (11.3%) (Cote et al., 2009).

The differences in allele or genotype frequencies detected among these studies might be due to ethnic variation, heterogeneity of studied populations and different sample sizes.

The risk for lung cancer among smokers is thought to increase with cumulative tobacco exposure (Ruano-Ravina et al., 2003), and genetic susceptibility to lung cancer may depend on the level of exposure to tobacco smoke (Nakachi et al., 1991 and Vineis, 1997). Therefore, we examined further gene-environment interaction between tobacco smoke exposure and the polymorphisms of APE1 and XRCC1.

When the subjects were divided into three groups according to the smoking status, we found that, the frequency of the mutant GG genotype of the APE1 Asp148Glu (2197 T→G) polymorphism in the former smokers had a statistically significant difference (p-value=0.003) regarding decreased risk of lung cancer, where the frequency of the lung cancer cases was lower than that of the healthy controls, while, current and never smokers did not, table (3). While, in Japanese populations the frequency of the mutant GG genotype of the APE1 Asp148Glu (2197 T→G) polymorphism in the current smokers had a statistically significant difference (p-value=0.044) regarding risk of lung cancer, while, former and never smokers did not (Ito et al., 2004); in Taiwanese populations the frequency of the mutant GG genotype in the heavy smokers had a statistically significant difference (p-value=0.026) regarding decreased risk of lung cancer, where the frequency of the lung cancer cases was lower than that of the healthy controls, while, never and light smokers did not (Lo et al., 2009) and Chinese populations the frequency of the mutant GG genotype in the male heavy smokers had a statistically highly significant difference (p-value=0.0003) regarding highly decreased risk of lung cancer, where the frequency of the lung cancer cases was lower than that of the healthy controls, while, male light smokers did not (Shen et al., 2005).

On the other hand, the frequency of the mutant AA genotype of the XRCC1 Arg399Gln (28152 G→A) polymorphism in the never smokers had a statistically highly significant difference (p-value=0.0004) regarding highly decreased risk of lung cancer, where the frequency of the lung cancer cases was lower than that of the healthy controls, while, current and former smokers did not, table (5). While, there were no significant differences in frequencies of the mutant AA genotype of the XRCC1 Arg399Gln (28152 G→A) polymorphism of all smoking status in Caucasian populations (Zhou et al., 2003); in Japanese populations (Ito et al., 2004) and in Caucasian populations (Hung et al., 2005).

In conclusion, the frequency of the mutant GG genotype of the APE1 Asp148Glu (2197 T→G) polymorphism in the former smokers had a statistically significant difference regarding decreased risk of lung cancer, where the
frequency of the lung cancer cases was lower than that of the healthy controls, while, current and never smokers did not, whereas, the frequency of the mutant AA genotype of the XRCC1 Arg399Gln (28152 G→A) polymorphism in the never smokers had a statistically highly significant difference regarding highly decreased risk of lung cancer, where the frequency of the lung cancer cases was lower than that of the healthy controls, while, current and former smokers did not.

References


الملخص العربي
تعدد أشكال الجين لجينات إصلاح الحمض النووي APE1 and XRCC1 بين المدخنين المصابين بسرطان الرئة من المصريين

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يعتبر سرطان الرئة من الأسباب المؤدية الى الوفاة ولذلك فهو يمثل مشكلة صحية رئيسية عامة. تلف الحمض النووي الناجم عن عوامل داخلية وخارجية يحدث باستمرار بنسب مرتفعة في الخلايا البشرية. ازالة أو إصلاح القواعد التالفة من الاليات الهامة للحفاظ على سلامة الجينوم البشري وذلك من خلال مسارات مختلفة منها مسار الإصلاح باستئصال القاعدة (BER), الذي يضمن قواعد مغذية غير صحيحة أو تالفة. APE1 وتعدد أشكال الجين XRCC1 هما من بروتينات إصلاح الحمض النووي التي تلعب دوراً هاماً في طريقة الإصلاح باستئصال القاعدة (BER).

في هذه الدراسة قمنا بدراسة العلاقة بين التغير الجيني لجينات الحمض النووي في طريقة الإصلاح باستئصال القاعدة (APE1 and XRCC1) عند المدخنين وسرطان الرئة. وقد شملت هذه الدراسة 131 حالة مصابة بسرطان الرئة تشمل 33 حالة سرطان الخلايا الصغيرة (25.2٪) و89 حالة سرطان الخلايا غير الصغيرة (74.8٪). تم تجميعهم من مركز الأورام، جامعة المنصورة، مصر في الفترة ما بين إبريل 2008 إلى مارس 2010.

للمقارنة، تم اختيار مجموعة ضابطة سلبية، وتتضمن 150 من الأشخاص الأصحاء عشوائياً من المتبرعين بالدم. هذه المجموعة الضابطة تم اختيارها بناءً على خلو هؤلاء الأشخاص من السرطان، إلى جانب عدم وجود تاريخ مرضي عائلي للسرطان لديهم. من زاروا مستشفيات جامعة المنصورة في الفترة ما بين إبريل 2008 إلى مارس 2010.

تم استخلاص الحمض النووي DNA من عينات الدم الطريقة الكامل باستخدام عمود التقاط، وتنقية جيل الحمض النووي (Gentra DNA system, USA) kit. تم استخدام تفاعل بلمرة التسلسلية في وجود زوجين من البادئة المتقابلة (PCR-CTPP). وقد تم تحليل البيانات والنتائج إحصائياً باستخدام برنامج الإحصائي الإصدار 10. وكانت النتائج بالنسبة لـ APE1 Asp148Glu (2197 T→G) وكانت النتائج بالنسبة لـ XRCC1 Arg399Gln (28152 G→A).

لم يكن هناك اختلاف كبير في جميع الترددات لكل الطرز والأليلات في مجموعة الحالات المصابة بسرطان الرئة مقارنة بالأصحاء. في جميع الحالات المصابة بسرطان الرئة مقارنة بالأصحاء، لم يكن هناك اختلاف كبير في جميع الترددات لكل الطرز والأليلات.

ومع ذلك، كانت هناك اختلافات كبيرة في الترددات من جميع الابعاد الجينية وأليلات في حالات الإصابة بسرطان الرئة مقارنة مع الضوابط في كل حالات التدخين. بينما في الأفراد الغير مدخنين لاحظنا وجود اختلاف كبير جداً في النمط الجيني المتحول GG في حالات الاصابة بسرطان الرئة، حيث كانت (p-value=0.003).

وكانت هناك اختلاف كبير في جميع الترددات لكل الطرز والأليلات في حالة الإصابة بسرطان الرئة مقارنة مع الضوابط حيث كانت (p-value=0.0004).