Assessment of the association between ACYP2 and laryngeal squamous cell carcinoma risk in Chinese males

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Abstract
Background: Laryngeal squamous cell carcinoma (LSCC) is one of the most prevalent malignant neoplasms of the upper respiratory tract. Studies have confirmed that an unstable chromosome constitution promotes the progress of laryngeal tumorigenesis, and ACYP2 has been confirmed as a telomere length-related gene. However, to date, the association between ACYP2 polymorphisms and LSCC susceptibility has not been investigated.

Methods: We performed this study to explore the effect of 11 single-nucleotide polymorphisms (SNPs) in ACYP2 on LSCC susceptibility in Chinese Han males. Unconditional logistic regression analysis adjusted for age was used to calculate the odds ratios and 95% confidence intervals.

Results: Based on allele and genotype models, our results showed that rs1682111 variant was significantly associated with a decreased LSCC susceptibility ($p < 0.05$). On the contrary, polymorphisms of rs10439478, rs11125529, rs12615793, rs843711, rs11896604, and rs17045754 were significantly associated with an increased LSCC risk ($p < 0.05$). The results of haplotype analysis indicated that haplotypes “TTCTCG” and “TTCTAA” in block 1 and “TG” in block 2 showed a risk factor for the development of LCSS ($p = 0.009$, $p < 0.001$, and $p = 0.001$, respectively). The results of Genotype-Tissue Expression analysis indicate that these significant SNPs were known to be associated with ACYP2 expression.

Conclusion: Our data demonstrated that ACYP2 polymorphisms may exert effects on LSCC susceptibility in Chinese Han males.

KEYWORDS
ACYP2, gene expression, laryngeal squamous cell carcinoma (LSCC), single-nucleotide polymorphisms (SNPs)
1 | BACKGROUND

Larynx, a mucosal organ located at the divergence of respiratory and digestive tracts, plays a vital role in sound generation and immunological decision-making. Laryngeal carcinoma is one of the most prevalent malignant neoplasms of the upper respiratory tract. Laryngeal squamous cell carcinoma (LSCC) is the major pathological type among laryngeal cancer, and is mainly detected in the epithelial lining of the larynx (Chu & Kim, 2008; Landry & Glastonbury, 2015). According to 2018 cancer statistics in United States, there was a remarkable prevalence of laryngeal carcinoma among males (of the 13,360 new cases [1% of total new cancers], 10,570 were in males and 2,790 females) and the deaths induced by laryngeal cancer among males was also greater than that in females (3,660 in males and 720 in females; Siegel, Miller, & Jemal, 2017). Although the incidence of laryngeal cancer is relatively modest compared to other major types of cancer, it has a low 5-year survival rate (Steuer, El-Deiry, Parks, Higgins, & Saba, 2017).

It is widely believed that genetic factors have an important effect on the occurrence of laryngeal cancer. Also, the occurrence and development of laryngeal cancer is a multistage and multilevel complex process as it involves more than one gene (Lawrence et al., 2015). The study by Veltman et al. (2000) indicated that an unstable chromosome constitution promote the progress of laryngeal tumorigenesis. Telomere is a region of repetitive hexamer (TTAGGGs) sequences at the end of a chromosome. Many research studies have proven that a shorter telomere length is associated with the development of many diseases (Blasco, 2005; Maguire, Neytchev, Talwar, McMillan, & Shiels, 2018), and telomere length has been regarded as a reliable biomarker or target in the medical treatment of diverse diseases (Gorenjak, Akbar, Stathopoulou, & Visvikis-Siest, 2018), including laryngeal cancer (Lei et al., 2015).

Acylphosphatase 2, encoded by \textit{ACYP2}, has been confirmed as a telomere length-related gene (Codd et al., 2013), and gene polymorphisms have been found to correlate with many cancers (Lin et al., 2017; Liu et al., 2017). However, to date, the association between \textit{ACYP2} variants and the LSCC susceptibility has not been investigated. In the present study, we selected 11 single-nucleotide polymorphisms (SNPs) of \textit{ACYP2} to explore the association between \textit{ACYP2} polymorphisms and the risk of laryngeal cancer in Chinese Han males.

2 | MATERIALS AND METHODS

2.1 | Ethical compliance

Our experiment strictly followed the principles expressed in the declaration of Helsinki, and the use of human blood samples in this study was approved by Human Research Committee for Approval of Research Involving Human Subjects and Shaanxi Provincial Cancer Hospital. All subjects have perceived and provided their written informed consent for this study.

2.2 | Study participants

This study included 172 males who were diagnosed with LSCC using histopathological examination and had not been treated with chemotherapy prior to acquiring blood samples. The control group included 180 healthy males who had undergone physical examination during the same period at a physical examination center of the same hospital. The inclusion criteria were as follows: (a) all the participants were of Han ethnicity and there was no kinship with the other races; (b) the control had no history of tumor, no genetic family history of tumors, and no tumor was found during the physical examination; and (c) all subjects had no history of occupational exposure to carcinogenic substances, containing toxic gases and radiation.

2.3 | DNA extraction, SNP selection, and genotyping

Genomic DNA from peripheral blood leukocytes in whole blood samples was extracted using the GoldMag-mini full-blood genomic DNA purification kit (GoldMag. Co. Ltd., Xi’an, China) according to manufacturer’s instructions. A spectrophotometer (NanoDrop 2000; Thermo Fisher Scientific, Waltham, MA, USA) was used to measure the concentration and purity of DNA.

A total of 11 SNPs was selected for this study, and most of the 11 SNPs had been reported in the association study of other diseases. Single-nucleotide polymorphisms rs11125529, rs12615793, rs843711, rs11896604, and rs17045754 have been found to be associated with gastric cancer risk in Chinese (Li et al., 2017). Single-nucleotide polymorphisms rs6713088 and rs843752 have been found to be associated with the risk of high-altitude pulmonary edema (Zhu et al., 2017). Eleven SNPs in the \textit{ACYP2} were selected from DbSNP database (http://www.hapmap.org/index.html.en) and SNP Consortium database (http://snp.cshl.org/) for further genotyping.

We exploited Agena Bioscience Assay Design Suite V2.0 software (https://agenax.com/online-tools/) to design a multiplexed SNP MassEXTENDED assay. Single-nucleotide polymorphisms were genotyped using the standard protocol recommended by the MassARRAY Nanodispenser (Agena Bioscience, San Diego, CA, USA) and MassARRAY iPLEX platform (Agena Bioscience, San Diego, CA, USA), and data were analyzed using Agena Bioscience TYPER version 4.0 software.
2.4 Statistical analysis

Microsoft Excel (Microsoft Corporation, Redmond, WA, USA) and SPSS 18.0 (SPSS, Chicago, IL, USA) were used to perform statistical analyses. The Welch’s t test was used to assess the age difference between the two groups, and the exact test was used to determine the SNPs that departed from the Hardy–Weinberg equilibrium (HWE). Genotype models were used to assess the association between each genotype and the LSCC susceptibility (Yang et al., 2018). SHEsis software platform and Haploview software package (version 4.2) (Broad Institute, Cambridge, MA, USA) were used to construct the linkage disequilibrium block (Barrett, Fry, Maller, & Daly, 2005; Li et al., 2009). The Akaike information criterion and Bayesian information criterion were used to select the best-matched methods (Acquah, 2012). The effect of the polymorphisms on the LSCC risk were expressed as odds ratio (OR) and 95% confidence interval (CI) (Tian et al., 2018). All statistical tests were two-sided, and a value of \( p = 0.05 \) was considered the threshold of whether statistical significance was achieved or not.

### TABLE 1 Age distribution between the case and control groups

| Variants | Case | Control | \( p^* \) |
|----------|------|---------|----------|
| Number   | 172  | 180     |          |
| Age, mean ± SD | 60.78 ± 10.05 | 60.25 ± 5.49 | <0.001 |

\( p \) was calculated by the Welch’s t test.

### TABLE 2 Basic information of candidate SNPs and minor allele frequencies distribution in case and control groups

| SNP rs#   | Chromosome | Position | Alleles A/B | Gene  | Minor allele frequency | \( p^{HWE} \) | OR (95% CI) | \( p^a \) |
|-----------|------------|----------|-------------|-------|------------------------|-------------|------------|--------|
| rs6713088 | 2          | 54,345,469 | G/C         | ACYP2 | Case 0.413, Control 0.400 | 1.05 (0.78–1.43) | 0.735 |
| rs12621038| 2          | 54,391,113 | T/C         | ACYP2 | Case 0.494, Control 0.416 | 1.37 (1.02–1.85) | 0.038b |
| rs1682111 | 2          | 54,427,979 | A/T         | ACYP2 | Case 0.262, Control 0.364 | 0.62 (0.45–0.86) | 0.004b |
| rs843752  | 2          | 54,446,587 | G/T         | ACYP2 | Case 0.251, Control 0.267 | 0.92 (0.66–1.30) | 0.647 |
| rs10439478| 2          | 54,459,450 | C/A         | ACYP2 | Case 0.497, Control 0.380 | 1.61 (1.19–2.18) | 0.002b |
| rs843645  | 2          | 54,474,664 | G/T         | ACYP2 | Case 0.241, Control 0.256 | 0.93 (0.66–1.31) | 0.660 |
| rs11125529| 2          | 54,475,866 | A/C         | ACYP2 | Case 0.268, Control 0.161 | 1.90 (1.32–2.76) | 0.001b |
| rs12615793| 2          | 54,475,914 | A/G         | ACYP2 | Case 0.269, Control 0.181 | 1.67 (1.17–2.40) | 0.005b |
| rs8437111 | 2          | 54,479,117 | T/C         | ACYP2 | Case 0.506, Control 0.428 | 1.37 (1.02–1.84) | 0.038b |
| rs11896604| 2          | 54,479,199 | G/C         | ACYP2 | Case 0.276, Control 0.169 | 1.87 (1.30–2.69) | 0.001b |
| rs17045754| 2          | 54,496,757 | C/G         | ACYP2 | Case 0.544, Control 0.158 | 6.33 (4.45–9.02) | < 0.001b |

Note: CIs, confidence intervals; HWE, Hardy–Weinberg equilibrium; OR, odds ratio; SNP, single nucleotide polymorphism; A/B stands for minor/major alleles on the control sample frequency.

Values of \( p^{HWE} \) were calculated by exact test.

\( p^* \) values were calculated by Pearson chi-square test.

\( p \) values of \( p < 0.05 \) indicates statistical significance.

2.5 Functional assessment of SNPs

In this study, Genotype Tissue Expression (GTEx) database of expression quantitative trait loci (eQTL) variants, a tissue bank for the scientific community to explore the relationship between genetic variation and gene expression in human tissues, was used to study the effect of LSCC-associated SNPs on ACYP2 expression. GTEx data are based on the database of genotypes and phenotypes (GTEx Consortium, 2015).

3 RESULTS

3.1 Characteristic of the study participates

Table 1 showed the distribution of mean age for the case and control groups, and there was a significant difference between the two groups (\( p < 0.001 \)). Therefore, in the subsequent statistical analysis, age-adjusted analysis was used to assess the association between SNPs and LSCC risk.

3.2 Genotype model analysis

We assumed that the minor allele of each SNP was a risk factor and analyzed the association between the polymorphism and LSCC risk under multiple inheritance models. Table 2 displays the information and allele frequencies of the 11 polymorphic loci. All SNPs were consistent with HWE in the controls (\( p > 0.05 \)). Using Pearson’s chi-square tests, we identified eight significant SNP variants associated with the risk of LSCC. Among them, only one minor allele “T” of
### Table 3: Association between polymorphisms of candidate SNPs and laryngeal cancer risk under four genotype models

| SNP     | Model    | Genotype | Genotype frequency | OR (95% CI) | p   | AIC   | BIC   |
|---------|----------|----------|--------------------|-------------|-----|-------|-------|
|         |          | Control  | Case               |             |     |       |       |
| rs1682111| Codominant| T/T      | 69 (38.3%)         | 1           | 0.011<sup>b</sup> | 480.5 | 495.9 |
|         |          | T/A      | 91 (50.6%)         | 0.55 (0.35–0.86) |     |       |       |
|         |          | A/A      | 20 (11.1%)         | 0.42 (0.19–0.94) |     |       |       |
|         | Dominant | T/T      | 69 (38.3%)         | 1           | 0.003<sup>b</sup> | 478.9 | 490.5 |
|         |          | T/A-A/A  | 111 (61.7%)        | 0.53 (0.34–0.81) |     |       |       |
|         | Recessive| T/T-T/A  | 160 (88.9%)        | 1           | 0.140 | 485.4 | 496.9 |
|         |          | A/A      | 20 (11.1%)         | 0.57 (0.26–1.22) |     |       |       |
|         | Log-additive| —       | —                  | 0.61 (0.43–0.85) |     | 0.003<sup>b</sup> | 478.9 | 490.4 |
| rs10439478| Codominant| A/A      | 69 (38.5%)         | 1           | 0.008<sup>b</sup> | 477.2 | 492.6 |
|         |          | C/A      | 84 (46.9%)         | 1.71 (1.05–2.80) |     |       |       |
|         |          | C/C      | 26 (14.5%)         | 2.57 (1.37–4.82) |     |       |       |
|         | Dominant | A/A      | 69 (38.5%)         | 1           | 0.006<sup>b</sup> | 477.1 | 488.7 |
|         |          | C/A-C/C  | 110 (61.5%)        | 1.92 (1.20–3.05) |     |       |       |
|         | Recessive| A/A-C/A  | 153 (85.5%)        | 1           | 0.027<sup>b</sup> | 479.9 | 491.5 |
|         |          | C/C      | 26 (14.5%)         | 1.84 (1.07–3.19) |     |       |       |
|         | Log-additive| —       | —                  | 1.62 (1.19–2.20) |     | 0.002<sup>b</sup> | 475.3 | 486.9 |
| rs11125529| Codominant| C/C      | 126 (70%)          | 1           | 0.002<sup>b</sup> | 477.5 | 492.9 |
|         |          | C/A      | 50 (27.8%)         | 1.76 (1.11–2.79) |     |       |       |
|         |          | A/A      | 4 (2.2%)           | 4.63 (1.46–14.70) |     |       |       |
|         | Dominant | C/C      | 126 (70%)          | 1           | 0.002<sup>b</sup> | 478.3 | 489.9 |
|         |          | C/A-A/A  | 54 (30%)           | 1.97 (1.27–3.06) |     |       |       |
|         | Recessive| C/C-C/C  | 176 (97.8%)        | 1           | 0.013<sup>b</sup> | 481.4 | 492.9 |
|         |          | A/A      | 4 (2.2%)           | 3.82 (1.21–11.99) |     |       |       |
|         | Log-additive| —       | —                  | 1.90 (1.31–2.77) |     | 0.001<sup>b</sup> | 475.8 | 487.3 |
| rs12615793| Codominant| G/G      | 120 (66.7%)        | 1           | 0.018<sup>b</sup> | 482.8 | 498.2 |
|         |          | A/G      | 55 (30.6%)         | 1.67 (1.07–2.62) |     |       |       |
|         |          | A/A      | 5 (2.8%)           | 3.04 (1.02–9.08) |     |       |       |
|         | Dominant | G/G      | 120 (66.7%)        | 1           | 0.009<sup>b</sup> | 481.9 | 493.5 |
|         |          | A/G-A/A  | 60 (33.3%)         | 1.78 (1.16–2.75) |     |       |       |
|         | Recessive| G/G-A/G  | 175 (97.2%)        | 1           | 0.081 | 485.8 | 497.4 |
|         |          | A/A      | 5 (2.8%)           | 2.52 (0.86–7.45) |     |       |       |
|         | Log-additive| —       | —                  | 1.70 (1.17–2.46) |     | 0.005<sup>b</sup> | 480.8 | 492.4 |
| rs843711 | Codominant| C/C      | 57 (31.7%)         | 1           | 0.1  | 490.9 | 506.3 |
|         |          | C/T      | 92 (51.1%)         | 1.27 (0.77–2.08) |     |       |       |
|         |          | T/T      | 31 (17.2%)         | 1.92 (1.05–3.53) |     |       |       |
|         | Dominant | C/C      | 57 (31.7%)         | 1           | 0.130 | 491.2 | 502.7 |
|         |          | C/T-T/T  | 123 (68.3%)        | 1.43 (0.90–2.29) |     |       |       |
|         | Recessive| C/C-C/T  | 149 (82.8%)        | 1           | 0.056 | 489.7 | 501.3 |
|         |          | T/T      | 31 (17.2%)         | 1.65 (0.98–2.77) |     |       |       |
|         | Log-additive| —       | —                  | 1.38 (1.02–1.86) |     | 0.037<sup>b</sup> | 489.1 | 500.6 |

(Continues)
rs1682111 was found to decrease the LSCC risk (OR = 0.62, 95% CI = 0.45–0.86, \( p = 0.004 \)). As for others, SNP variants were found to play a harmful role in LSCC risk. Other variants were minor allele “T” of rs12621038 (OR = 1.37, 95% CI = 1.02–1.85, \( p = 0.038 \)), minor allele “C” of rs10439478 (OR = 1.61, 95% CI = 1.19–2.18, \( p = 0.002 \)), minor allele “A” of rs11125529 (OR = 1.9, 95% CI = 1.32–2.76, \( p = 0.001 \)), minor allele “A” of rs12615793 (OR = 1.67, 95% CI = 1.17–2.4, \( p = 0.005 \)), minor allele “T” of rs843711 (OR = 1.37, 95% CI = 1.02–1.84, \( p = 0.038 \)), minor allele “G” of rs11896604 (OR = 1.87, 95% CI = 1.3–2.69, \( p = 0.001 \)), and minor allele “C” of rs17045754 (OR = 6.33, 95% CI = 4.45–9.02, \( p < 0.001 \)).

In Table 3, we performed the logistic test to analyze further model association. We found that six SNPs loci had a significant association with LSCC risk under genotype model. The minor allele “A” of rs1682111 was associated with a decreased risk of LSCC based on codominant model (\( p = 0.011 \)), dominant model (\( p = 0.003 \)), and log-additive model (\( p = 0.003 \)). The remaining five loci had shown a risk factor for the development of LSCC. The minor allele “C” of rs10439478 and the minor allele “A” of rs11125529 were found play a harmful role in LSCC risk based on codominant model (\( p = 0.008 \), \( p = 0.002 \) respectively), dominant model (\( p = 0.006 \), \( p = 0.002 \) respectively), recessive model (\( p = 0.027 \), \( p = 0.013 \) respectively), and log-additive model (\( p = 0.002 \), \( p = 0.001 \) respectively). Moreover, the minor allele “A” of rs12615793 and the minor allele “G” of rs11896604 were associated with an increased risk of LSCC based on codominant model (\( p = 0.018 \), \( p = 0.002 \) respectively), dominant model (\( p = 0.009 \), \( p < 0.001 \) respectively), and log-additive model (\( p = 0.005 \), \( p < 0.001 \) respectively).

As for rs843711, the risk of LSCC was increased only in a log-additive model (OR = 1.38, 95% CI = 1.02–1.86, \( p = 0.037 \)). The result of crude analysis is shown in Table S1.

### 3.3 | Haplotype analysis

Two blocks were detected in ACYP2 SNPs by haplotype analyses (Figure 1). Block 1 contained six SNPs (rs1682111, rs843752, rs10439478, rs843645, rs11125529, and rs12615793) and block 2 contained two SNPs (rs843711 and rs11896604). The results of the connection between the ACYP2 haplotypes and the risk of LSCC are listed in Table 4. We only listed the haplotypes with frequency more than 1%. In block 1, haplotype “TTCTCG” and haplotype “TTCTAA” were more prevalent in the case group than in the control group and were significantly associated with an increased risk of LSCC (“TTCTCG” OR = 1.88, 95% CI = 1.17–3.01, \( p = 0.009 \); “TTCTAA” OR = 2.33, 95% CI = 1.46–3.71, \( p < 0.001 \)). In block 2, haplotype “TG” had an increased effect on the risk of LSCC (OR = 1.93, 95% CI = 1.3–2.88, \( p = 0.001 \)).

### 3.4 | Function annotations

In Table 5, we explored the association between SNPs (rs12621038, rs1682111, rs10439478, rs11125529, rs12615793, rs843711, rs11896604, and rs17045754) and the ACYP2 expression. The single-nucleotide polymorphism rs1682111 was identified as cis-eQTLs in the muscle, testis, small intestine, and colon, and the SNP rs843711 was identified as cis-eQTLs in the muscle, esophagus, testis, small intestine, and thyroid tissue. For others, SNPs were diagnosed as cis-eQTLs in the muscle only. The results indicated that these SNPs variants might influence the expression of ACYP2.

### 4 | DISCUSSION

In this study, we aimed to investigate the association between ACYP2 polymorphisms and laryngeal cancer risk in
Chinese Han males. The results showed that rs1682111 was associated with a decreased risk of LSCC, while rs10439478, rs11125529, rs12615793, rs843711, rs11896604, and rs17045754 play a harmful role in the development of LSCC. Furthermore, the results of GTEx analysis indicate that these significant SNPs are known to be associated with ACYP2 gene expression. Our findings indicate that ACYP2 may play a crucial role in the process of laryngeal canceration.

**FIGURE 1** Linkage disequilibrium (LD) analysis of candidate single-nucleotide polymorphisms (SNPs) in ACYP2. LD plots containing 11 SNPs in ACYP2, and standard color frame is used to show LD pattern. Two blocks had been found. Block 1 contained six SNPs (rs1682111, rs843752, rs10439478, rs843645, rs11125529, and rs12615793). Block 2 contained two SNPs (rs843711 and rs11896604).

**TABLE 4** Haplotype analysis

| Block | Haplotype | Haplotype frequency | OR (95% CI) | \( p^a \) |
|-------|-----------|---------------------|-------------|-----|
| Block 1 | ATATCG | 0.233 | 0.347 | 1 | — |
| | TGAGCG | 0.216 | 0.251 | 1.38 (0.90 – 2.11) | 0.140 |
| | TTCTCG | 0.221 | 0.193 | 1.88 (1.17 – 3.01) | 0.009<sup>b</sup> |
| | TTCTAA | 0.234 | 0.159 | 2.33 (1.46 – 3.71) | <0.001<sup>b</sup> |
| | TTCTCA | 0.021 | 0.02 | 1.55 (0.50 – 4.77) | 0.450 |
| | TGATCG | 0.017 | 0.011 | 2.62 (0.63 – 10.79) | 0.180 |
| Block 2 | CC | 0.487 | 0.571 | 1 | — |
| | TC | 0.237 | 0.259 | 1.11 (0.77 – 1.59) | 0.580 |
| | TG | 0.269 | 0.169 | 1.93 (1.30 – 2.88) | 0.001<sup>b</sup> |

Note: CIs, confidence intervals; OR, odds ratio.

<sup>a</sup>\( p \) values were calculated by unconditional logistic regression analysis adjusted for age.

<sup>b</sup>Value of \( p < 0.05 \) indicates statistical significance.

**TABLE 5** The effect of LSCC-associated SNPs on ACYP2 expression

| SNP     | Effect size | \( p \)-value | Tissue          |
|---------|-------------|---------------|-----------------|
| rs12621038 | 0.24        | 5.8 \times 10^{-12} | Muscle—skeletal |
| rs1682111  | 0.17        | 8.6 \times 10^{-10} | Muscle—skeletal |
| rs1682111  | −0.35       | 4.7 \times 10^{-6}  | Testis          |
| rs1682111  | −0.34       | 9.3 \times 10^{-6}  | Small intestine—terminal ileum |
| rs1682111  | −0.3        | 2.5 \times 10^{-5}  | Colon—sigmoid   |
| rs10439478 | 0.14        | 1.9 \times 10^{-5}  | Muscle—skeletal |
| rs11125529 | 0.19        | 8.9 \times 10^{-6}  | Muscle—skeletal |
| rs12615793 | 0.19        | 6.4 \times 10^{-6}  | Muscle—skeletal |
| rs843711   | −0.28       | 2.1 \times 10^{-7}  | Esophagus—mucosa |
| rs843711   | −0.35       | 1.8 \times 10^{-6}  | Testis          |
| rs843711   | 0.13        | 1.8 \times 10^{-6}  | Muscle—skeletal |
| rs843711   | −0.32       | 8.6 \times 10^{-6}  | Small intestine—terminal ileum |
| rs843711   | −0.17       | 1.6 \times 10^{-5}  | Thyroid         |
| rs843711   | −0.19       | 5.2 \times 10^{-5}  | Esophagus—muscularis |
| rs11896604 | 0.18        | 1.5 \times 10^{-5}  | Muscle—skeletal |
| rs17045754 | 0.18        | 4.7 \times 10^{-6}  | Muscle—skeletal |

Note: LSCC: laryngeal squamous cell carcinoma; SNP, single-nucleotide polymorphism.

Data Source: GTEx Analysis Release V7 (dbGaP Accession phs000424.v7.p2).

The gene ACYP2 is located in 2p16.2. Acylphosphatase 2 can hydrolyze the phosphoenzyme intermediate of different membrane pumps, particularly the Ca<sup>2+</sup>/Mg<sup>2+</sup>-ATPase from the sarcoplasmic reticulum of skeletal muscle (Nassi, Nediani, Liguri, Taddei, & Ramponi, 1991). Studies have revealed that...
ACYP2 plays a crucial role in pyruvate metabolism whose activation is an important factor for malignant transformation (Conde et al., 2015; Szlosarek, Lee, & Pollard, 2014; Won et al., 2012). Furthermore, ACYP2 has been confirmed as a telomere length-related gene (Codd et al., 2013). As we all know, telomere can protect chromosome against deteriorating and fusing during DNA replication. Differences in telomere length between individuals have a close connection with the etiology of cancer and age-related diseases (Aubert & Lansdorp, 2008; L. Xu, Li, & Stohr, 2013). In addition, the research reported that the downregulation of ACYP2 inhibits the proliferation of breast cancer cells (Yu et al., 2011). Although the detail mechanism of ACYP2 on the laryngeal cancer is not well understood, our results demonstrated that ACYP2 polymorphisms had a close connection with the risk of LSCC. ACYP2 SNP loci may be associated with LSCC susceptibility via regulating the pyruvate metabolic pathway or affecting telomere length.

Previous association studies have found ACYP2 variants associated with the risk of many diseases. For example, variants of rs1682111 and rs10439478 and its interaction were found associated with an increased risk of breast cancer (Wu, Wang, Liu, & Li, 2017). Single-nucleotide polymorphisms rs1125529, rs12615793, rs843711, rs11896604, and rs17045754 were related with increased ischemic stroke susceptibility (Liang et al., 2017). Furthermore, rs11896604 and rs17045754 variants were found to be associated with a decreased risk of high-altitude pulmonary edema, but rs843752 minor allele “G” was a harmful factor for the risk of high-altitude pulmonary edema (He et al., 2016). In addition, ACYP2 variants affected susceptibility to cisplatin-induced hearing loss (H. Xu et al., 2015; Zhu et al., 2017). We found that genome-wide association studies (GWAS) on LSCC risk are limited and only few research studies are available. Genome-wide association study has identified three new susceptibility loci for LSCC in the Chinese population, and they are rs174549 at 11q12, rs2857595 at 6q21, and rs10492336 at 12q24 (Wei et al., 2014). Moreover, these LSCC-associated SNPs variants are of functional importance for the expression of ACYP2. Although the information about ACYP2 as a molecular marker of laryngeal cancer is very little in both Chinese and other ethnic groups, ACYP2 polymorphisms might play an influential role in the development of LSCC.

There are some limitations in our study. First, due to the strict principles on the sample selection, the sample size was relatively small and all the samples were restricted to Chinese Han males. Second, we were short of replication and mechanism studies. The studies on ACYP2 polymorphisms and the risk of cancers are relatively more, but they all lack replication studies to confirm their findings. Therefore, the results we found here should be further confirmed using the replication study with bigger sample size and different populations, and cell and molecular biology methods should be used to explore the association between SNP loci in ACYP2 and the etiology of laryngeal cancer.

5 | CONCLUSION

This study provided fundamental evidence that SNPs in ACYP2 were related with LSCC susceptibility in Chinese Han males. It is possible that these variants are important factors for the development of laryngeal cancer, and these findings need to be further confirmed. We served a theoretical foundation for other scholars to explore the connection between ACYP2 and laryngeal cancer risk in Chinese or other ethnic groups.

CONSENT FOR PUBLICATION

All authors approved the publication of this study.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

DATA AVAILABILITY STATEMENT

The datasets generated during and/or analyzed during this study are available from the corresponding author on reasonable request.

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**SUPPORTING INFORMATION**

Additional supporting information may be found online in the Supporting Information section at the end of the article.