Association Between p53 Arg72Pro Polymorphism and the Risk of Human Papillomavirus-related Head and Neck Squamous Cell Carcinoma: A Meta-analysis

Ling-Yun Xia1,2, Xian-Tao Zeng2,3, Cheng Li1, Wei-Dong Leng2, Ming-Wen Fan1*

Abstract

This study aimed to investigate the association between p53 Arg72Pro polymorphism and the risk of human papillomavirus (HPV)-related head and neck squamous cell carcinoma (HNSCC) by conducting meta-analysis. The PubMed database was searched for relevant studies until May 30, 2013. Relevant studies were selected and data were extracted by two independent authors. Overall, subgroup, and sensitivity analyses were then conducted using the Comprehensive Meta-Analysis v2.2 software. Wild-genotype ArgArg was considered as reference [odds ratio (OR) = 1.00]. Nine studies involving 1071 HNSCC cases were obtained. Meta-analysis results indicated no association between p53 Arg72Pro polymorphism and the risk of HPV-related HNSCC: for Pro/Pro vs. Arg/Arg, OR = 1.17, 95% confidence interval (CI) = 0.70–1.98; for Arg/Pro vs. Arg/Arg, OR = 1.25, 95% CI = 0.97–1.72; and for (Pro/Pro + Arg/Pro) vs. Arg/Arg, OR = 1.28, 95% CI = 0.95–1.70. These meta-analysis results were supported by subgroup and sensitivity analysis results. In conclusions, p53 Arg72Pro polymorphism is a potential marker of HP infection-related HNSCC rather than a susceptibility gene polymorphism.

Keywords: p53 codon 72 - human papillomavirus - head and neck cancer - squamous cell carcinoma - polymorphism - meta-analysis

Asian Pac J Cancer Prev, 14 (10), 6127-6130

Introduction

Head and neck squamous cell carcinoma (HNSCC) is the sixth most common cancer worldwide (Moore et al., 2000; Kamangar et al., 2006; Warnakulasuriya, 2009). The classical risk factors of HNSCC include tobacco, alcohol, and human papillomavirus (HPV) (Farris et al., 2013; Galbiatti et al., 2013). Nevertheless, only some smokers, alcohol users, and HPV-infected individuals develop HNSCC, suggesting that the genetic susceptibility of individuals is a possible influencing factor (Liang et al., 2012).

p53 gene is located at chromosome 17p13 and considered as one of the most frequently mutated genes in human carcinogenesis (Tsui et al., 2009). rs1042522 polymorphism is located in exon 4 of p53 gene, in which an arginine (Arg)→proline (Pro) amino acid substitution is present at amino acid position 72; such polymorphism is commonly named Pro72Arg or codon polymorphism (Ara et al., 1990). Epidemiological studies have indicated that p53 Arg72Pro polymorphism increases the risk of many cancers, such as cutaneous melanoma (Oliveira et al., 2013), bladder cancer (Xu et al., 2012), and nasopharyngeal carcinoma (Zhuo et al., 2009). However, two meta-analyses have failed to identify a significant association between p53 Arg72Pro polymorphism and oral cancer (Zhuo et al., 2009; Jiang et al., 2013).

The incidence of HPV-caused HNSCC has increased, whereas the overall incidence of HNSCC has decreased (Farris et al., 2013). Other studies have investigated the association between p53 Arg72Pro polymorphism and HNSCC, but inconsistent results have been obtained. Hence, whether or not p53 Arg72Pro polymorphism can increase the risk of HNSCC with HPV infection remains unclear. Similarly, studies have not yet elucidated whether or not p53 Arg72Pro polymorphism merely functions as a marker of HPV-related HNSCC cases. For these reasons, we conducted meta-analysis to estimate the relationship between p53 Arg72Pro polymorphism and the risk of HPV-related HNSCC.

Materials and Methods

Inclusion Criteria

The following inclusion criteria were used: (1) the association of p53 Arg72Pro polymorphism with the risk of HPV-related HNSCC was evaluated; (2) HNSCC cases were diagnosed by histological, pathological, or cytological techniques; (3) the number of individual genotypes was provided in HPV-positive and HPV-
negative groups or could be calculated from provided data; or (4) odds ratio (OR) and 95% confidence interval (CI) were obtained.

**Search strategy**

The PubMed database was searched until May 30, 2013 using the following search terms: [(palatal OR tongue OR laryngeal OR hypopharyngeal OR pharynx OR oropharyngeal OR tonsillar OR oral OR mouth OR “head and neck”) AND (neoplasm OR cancer OR carcinoma) AND p53 AND polymorphism AND “human papillomavirus”]. In addition, the references of the included studies and previous relevant meta-analyses were manually searched.

**Data extraction**

Studies were selected and the data of these included studies were obtained independently by two authors; disagreements were resolved by discussion. The subjects of four previous studies were considered partly overlapping (Chen et al., 2008; Ji et al., 2008; Wang et al., 2012). Therefore, we selected one study containing the most comprehensive information for our meta-analysis (Wang et al., 2012). The pertinent data obtained were listed as follows: the last name of the first author; publication year; countries of origin and ethnicity; HPV status of HNSCC cases; number and genotyping distribution of HPV-positive and negative cases; and genotyping method.

**Table 1. Characteristics of Included Studies in the Meta-analysis**

| Reference       | Country (Ethnicity)      | Sample (+/-) | ArgArg (+/-) | ArgPro (+/-) | ProPro (+/-) | Genotype method |
|-----------------|--------------------------|--------------|--------------|--------------|--------------|-----------------|
| Summersgill 2000 | USA (Mixed)              | 46/144       | 26/76        | 15/55        | 5/13         | PCR             |
| Nagpal 2002     | India (Asians)           | 41/69        | 14/17        | 20/38        | 7/14         | PCR             |
| Katiyar 2003    | India (Asians)           | 13/31        | 2/8          | 10/14        | 1/9          | PCR             |
| Cortezi 2004    | Brazil (Mixed)           | 8/42         | 4/22         | 3/13         | 1/7          | PCR             |
| Scheckenbach 2004 | Germany (Caucasians)     | 37/85        | 17/49        | 20/35        | 0/1          | PCR             |
| Perrone 2007    | Italy (Caucasians)       | 16/61        | 11/52        | 1/7          | 4/2          | PCR             |
| Hoffmann 2009   | Germany (Caucasians)     | 12/30        | 11/16        | 0/13         | 1/1          | PCR             |
| Saini 2011      | Malaysia (Asians)        | 52/47        | 11/11        | 18/22        | 23/14        | PCR             |
| Wang 2012       | USA (Mixed)              | 230/79       | 130/56       | 91/22        | 9/1          | PCR             |

**Table 2. Overall and Subgroup Analysis and Publication Bias Results of Meta-analysis**

|                   | Overall                  | Asians                  | Caucasians              | Mixed Ethnicity          |
|-------------------|--------------------------|-------------------------|--------------------------|--------------------------|
| No. of study      | 9                        | 3                       | 3                        | 3                        |
| ArgArg            | 1.0                      | 1.0                     | 1.0                      | 1.0                      |
| ProPro            | 1.17 (0.70-1.98)         | 30.15                   | 45.17                    | 4.01 (1.00-16.07)         | 4.61                   | 1.41 (0.56-3.53) |
| ArgPro            | 1.25 (0.91-1.72)         | 18.38                   | 0.19                     | 0.60 (0.10-3.50)         | 62.05                   | 1.31 (0.86-2.02) |
| ArgPro+ProPro     | 1.28 (0.95-1.72)         | 32.72                   | 0.53                     | 0.99 (0.56-1.75)         | 69.57                   | 1.40 (0.93-2.10) |

**Figure 1. Flow Chart from Identification of Eligible Studies to Final Inclusion**

**Statistical analysis**

ORs and 95% CIs were used to pool the data from the included studies. We estimated the OR of an HNSCC associated with ProPro genotype, ArgPro genotype, and (ArgPro + ProPro) genotype and then compared with wild-type ArgArg. In all of the models, ArgArg was considered as reference (OR = 1.00). The fixed model was initially used to summarize ORs. I² statistics was used to determine heterogeneity; I² > 40% indicated the presence of heterogeneity. The model was then changed to a random-effect model. The subgroups were analyzed based on ethnicity. Sensitivity was also analyzed by omitting any study. Publication bias was detected by funnel plot and Egger’s test. These analyses were conducted using the Comprehensive Meta-Analysis v2.2 software.

**Results**

**Characteristics of the included studies**

A total of 44 studies were searched from the PubMed database and 19 studies were obtained by manual search. Nine studies involving 1071 HNSCC cases were included in the meta-analysis (Summersgill et al., 2000; Nagpal et al., 2002; Katiyar et al., 2003; Cortezi et al., 2004; Scheckenbach et al., 2004; Perrone et al., 2007; Hoffmann et al., 2009; Saini et al., 2011; Wang et al., 2012). Figure 1 shows the screening process.

All of the cases in the included studies were HNSCC. Two studies were from the USA [ethnicity was mixed (white, black, and others; or non-Hispanic white and others)] (Summersgill et al., 2000; Wang et al., 2012), two were from India (Nagpal et al., 2002; Katiyar et al., 2003), one was from Malaysia (Saini et al., 2011), two were from Germany (Scheckenbach et al., 2004; Hoffmann et al., 2009), one was from Italy (Perrone et al., 2007), and
the upper aerodigestive tract (comprising the oral cavity, pharynx, and larynx) (Funk et al., 2002). Molecular epidemiological studies have indicated that high-risk HPV genotypes are possibly involved in HNSCC (Gillison et al., 2000). The p53 codon 72 polymorphism is a nucleotide polymorphism that encodes either Arg or Pro (Summersgill et al., 2000), and this polymorphism was first considered as an important factor in HPV-related cancer development in 1998 (Storey et al., 1998). In 2000, Summersgill et al. (Summersgill et al., 2000) reported that p53 codon Arg72Pro is not associated with HPV infection; p53 polymorphism is also not associated with the risk of oral cancer. Since then, numerous studies have been published on this topic but provided inconsistent results.

Meta-analysis is a method used to combine relevant studies worldwide and resolve the statistical power and discrepancy of genetic-association studies (Munafo et al., 2004). Therefore, we performed meta-analysis based on nine relevant studies. The results indicated no significant association between p53 Arg72Pro polymorphism and the risk of HPV-related HNSCC regardless of ethnicity. Conducting sensitivity analysis, we found that the results were robust. Publication bias was not observed.

**Strengths and limitations:** Two relevant meta-analysis studies have been published. For instance, Zhou et al. (Zhuo et al., 2009) investigated HPV-related HNSCC as a subgroup; however, only three studies were included in the previous meta-analysis (Summersgill et al., 2000; Nagpal et al., 2002; Katiyar et al., 2003). Jiang et al. (2013) included four relevant studies (Summersgill et al., 2000; Nagpal et al., 2002; Katiyar et al., 2003; Saini et al., 2011), but no subgroup analysis was performed. In contrast to these previous meta-analyses (Zhuo et al., 2009; Jiang et al., 2013), the present meta-analysis only focused on HPV-related HNSCC and nine studies were considered. Our sample sizes were more extensive compared with the two previous studies and our results were more reliable.

Some limitations of our meta-analysis were demonstrated. First, the sample sizes of the studies included in the present meta-analysis were relatively small except two studies (Summersgill et al., 2000; Wang et al., 2012). Small sample sizes can decrease statistical power. Furthermore, the overall sample sizes of our meta-analysis were not sufficiently large. Second, the heterogeneity of Caucasians was high and found in the overall population. Subgroup analysis results indicated that heterogeneity was possibly caused by ethnicity. Although heterogeneity is extremely common in genetic-association meta-analysis, this factor should not be ignored. Third, studies beyond the pseudo-95% CI were still included, although publication bias was not detected. As a language limitation, studies published only in English were searched. Fourth, subgroup analysis in terms of location was not performed because of a limited number of included studies and reported information. Therefore, differences in SCC sites remain unclear. Fifth, we could not perform analysis on adjusted data (e.g., adjusting for smoking and alcohol consumption) because of limited reported information from the included studies.

In conclusion, considering evidence obtained in the present meta-analysis as well as other vertical
and horizontal evidence, we found that p53 Arg72Pro polymorphism is not associated with HPV-related HNSCC. Considering the limited objectives of this meta-analysis, we recommend that further studies should be conducted using larger sample sizes and nested case-control or prospective cohort designs.

Acknowledgements

This research was supported (in part) by the Nature Science Foundation of Hubei Province (2012FFB03902), the Natural Science Foundation of Hubei Ministry of Education (D20122405), the Intramural Research Program of the Hubei University of Medicine (2011CZX01), and Evidence-based Medicine Nursery Fund of Taihe Hospital (EBM2013038), without commercial or not-for-profit sectors. The author(s) declare that they have no competing interests.

References

Ara S, Lee PS, Hansen MF, et al (1990). Codon 72 polymorphism of the TP53 gene. Nucleic Acids Res, 18, 4961.
Chen X, Sturgis EM, El-Naggar AK, et al (2008). Combined effects of the p53 codon 72 and p73 G4C14-to-A4T14 polymorphisms on the risk of HPV16-associated oral cancer in never-smokers. Carcinogenesis, 29, 2120-25.
Cortezzi SS, Provazzi PJ, Sobrinho JS, et al (2004). Analysis of human papillomavirus prevalence and TP53 polymorphism in head and neck squamous cell carcinomas. Cancer Genet Cytogenet, 150, 44-9.
Farris C, Petitte DM (2013). Head, neck, and oral cancer update. Home Healthc Nurse, 31, 322-8.
Funk GF, Karnell LH, Robinson RA, et al (2002). Presentation, treatment, and outcome of oral cavity cancer: a National Cancer Data Base report. Head Neck, 24, 165-80.
Galbiatti AL, Padovani-Junior JA, Maniglia JV, et al (2013). Head and neck cancer: causes, prevention and treatment. Braz J Otorhinolaryngol, 79, 239-47.
Gillison ML, Koch WM, Capone RB, et al (2000). Evidence for a causal association between human papillomavirus and a subset of head and neck cancers. J Natl Cancer Inst, 92, 709-20.
Hoffmann M, Scheunemann D, Fazel A, et al (2009). Human papillomavirus and p53 polymorphism in codon 72 in head and neck squamous cell carcinoma. Oncol Rep, 21, 809-14.
Ji X, Neumann AS, Sturgis EM, et al (2008). p53 codon 72 polymorphism associated with risk of human papillomavirus-associated squamous cell carcinoma of the oropharynx in never-smokers. Carcinogenesis, 29, 875-9.
Jiang N, Pan J, Wang L, et al (2013). No significant association between p53 codon 72 Arg/Pro polymorphism and risk of oral cancer. Tumour Biol, 34, 587-96.
Kamangar F, Dores GM, Anderson WF (2006). Patterns of cancer incidence, mortality, and prevalence across five continents: defining priorities to reduce cancer disparities in different geographic regions of the world. J Clin Oncol, 24, 2137-50.
Katyar S, Thelma BK, Murthy NS, et al (2003). Polymorphism of the p53 codon 72 Arg/Pro and the risk of HPV type 16/18-associated cervical and oral cancer in India. Mol Cell Biochem, 252, 117-24.
Liang C, Marsit CJ, Houseman EA, et al (2012). Gene-environment interactions of novel variants associated with head and neck cancer. Head Neck, 34, 1111-8.
Moore SR, Johnson NW, Pierce AM, et al (2000). The epidemiology of mouth cancer: a review of global incidence. Oral Dis, 6, 65-74.
Munafo MR, Flint J (2004). Meta-analysis of genetic association studies. Trends Genet, 20, 439-44.
Nagpal JK, Patnaik S, Das BR (2002). Prevalence of high-risk human papilloma virus types and its association with P53 codon 72 polymorphism in tobacco addicted oral squamous cell carcinoma (OSCC) patients of Eastern India. Int J Cancer, 97, 649-53.
Oliveira C, Rinck-Junior JA, Lourenco GJ, et al (2013). Assessment of the XPC (A2920C), XPF (T30028C), TP53 (Arg72Pro) and GSTP1 (Ile105Val) polymorphisms in the risk of cutaneous melanoma. J Cancer Res Clin Oncol, 139, 1199-206.
Perrone F, Mariani L, Pastore E, et al (2007). p53 codon 72 polymorphisms in human papillomavirus-negative and human papillomavirus-positive squamous cell carcinomas of the oropharynx. Cancer, 109, 2461-65.
Saini R, Tang TH, Zain RB, et al (2011). Significant association of high-risk human papillomavirus (HPV) but not of p53 polymorphisms with oral squamous cell carcinomas in Malaysia. J Cancer Res Clin Oncol, 137, 311-20.
Scheckenbach K, Lieven O, Gotte K, et al (2004). p53 codon 72 polymorphic variants, loss of allele-specific transcription, and human papilloma virus 16 and/or 18 E6 messenger RNA expression in squamous cell carcinomas of the head and neck. Cancer Epidemiol Biomarkers Prev, 13, 1805-9.
Storey A, Thomas M, Kalita A, et al (1998). Role of a p53 polymorphism in the development of human papillomavirus-associated cancer. Nature, 393, 229-34.
Summersgill KF, Smith EM, Kirchner HL, et al (2000). p53 polymorphism, human papillomavirus infection in the oral cavity, and oral cancer. Oral Surg Oral Med Oral Pathol Oral Radiol Endod, 90, 334-9.
Tsui IF, Poh CF, Garnis C, et al (2009). Multiple pathways in the FGF signaling network are frequently deregulated by gene amplification in oral dysplasias. Int J Cancer, 125, 2219-28.
Wang Z, Sturgis EM, Zhang Y, et al (2012). Combined p53-related genetic variants together with HPV infection increase oral cancer risk. Int J Cancer, 131, E251-8.
Wang Z, Sturgis EM, Guo W, et al (2012). Association of combined p73 and p53 genetic variants with tumor HPV16-positive oropharyngeal cancer. PLoS One, 7, e55522.
Warnakulasuriya S (2009). Global epidemiology of oral and oropharyngeal cancer. Oral Oncol, 45, 309-16.
Xu T, Xu ZC, Zou Q, et al (2012). P53 Arg72Pro polymorphism and bladder cancer risk—meta-analysis evidence for a link in Asians but not Caucasians. Asian Pac J Cancer Prev, 13, 2349-54.
Zhuo XL, Cai L, Xiang ZL, et al (2009). TP53 codon 72 polymorphism contributes to nasopharyngeal cancer susceptibility: a meta-analysis. Arch Med Res, 40, 299-305.
Zhuo XL, Li Q, Zhou Y, et al (2009). Study on TP53 codon 72 polymorphisms with oral carcinoma susceptibility. Arch Med Res, 40, 625-34.