RESEARCH ARTICLE

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

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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

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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-

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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
Cortezzi 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	
	OR(95%CI)	I^2(%)	Egger	OR(95%CI)	I^2(%)	OR(95%CI)	I^2(%)	OR(95%CI)	I^2(%)
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	0.49	0.74(0.26-2.13)	45.17	4.01(1.00-16.07)	4.61	1.41(0.56-3.53)	0
ArgPro	1.25(0.91-1.72)	18.38	0.19	1.16(0.63-2.14)	0	0.60(0.10-3.50)	62.05	1.31(0.86-2.02)	32.50
ArgPro+ProPro	1.28(0.95-1.72)	32.72	0.53	0.99(0.56-1.75)	0	1.04(0.26-4.09)	69.57	1.40(0.93-2.10)	38.10

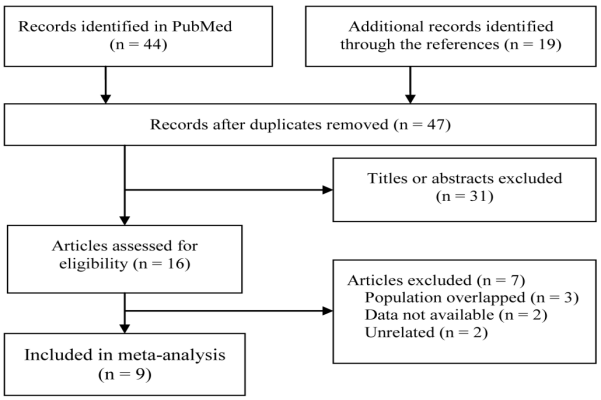

Figure 1. Flow Chart from Identification of Eligible Studies to Final Inclusion

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 by 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.

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. I^2 statistics was used to determine heterogeneity; $I^2 > 40\%$ 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 (Summersgi ll et al., 2000; Nagpal et al., 2002; Katiyar et al., 2003; Cortezzi 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

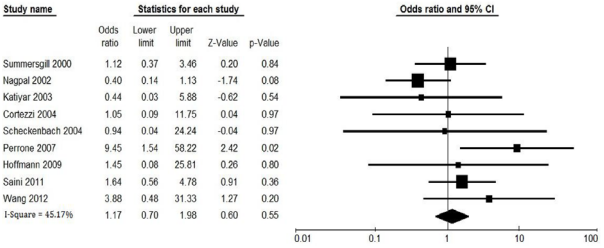

Figure 2. Forest Plot of Overall Meta-analysis Based on ProPro vs. ArgArg Genetic Model (fixed effect model)

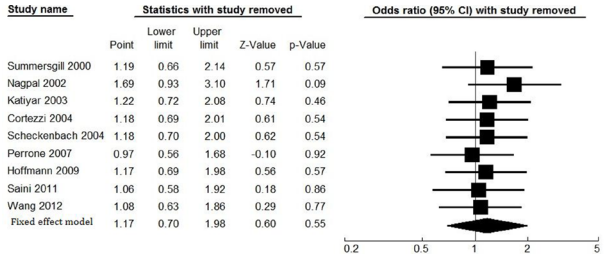

Figure 3. Forest Plot of Sensitivity Analysis Based on ProPro vs. ArgArg Genetic Model (fixed effect model)

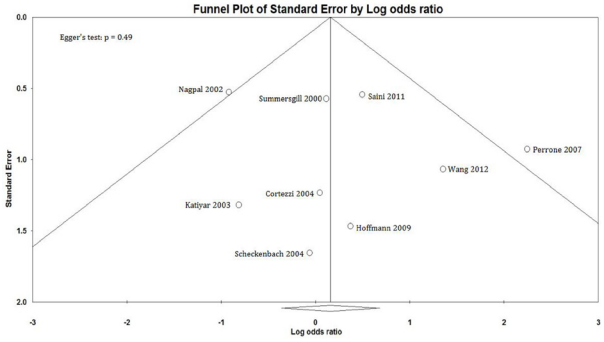

Figure 4. Funnel Plot for the Assessment of Publication Bias Based on ProPro vs. ArgArg Genetic Model (fixed effect model)

one was from Brazil (Cortezzi et al., 2004). The baseline characteristics of the patients are shown in Table 1.

Meta-analysis

Table 2 shows the overall and subgroup results. Overall, meta-analysis showed no association between p53 Arg72Pro polymorphism and the risk of HPV infection-related HNSCC [for ProPro vs. ArgArg, OR = 1.17, 95% CI = 0.70–1.98, Figure 2; for ArgPro vs. ArgArg, OR = 1.25, 95% CI = 0.97–1.72; for (ProPro+ArgPro) vs. ArgArg, OR = 1.28, 95% CI = 0.95–1.72]. The subgroup analysis results in terms of ethnicity were similar to the overall results.

Sensitivity and publication bias analysis

Sensitivity analysis showed that the results were robust, indicating the absence of influence from any particular included study (Figure 3). The funnel plot showed a relatively symmetrical distribution, suggesting the absence of publication bias (Figure 4). This result was also supported by Egger's test (Table 2).

Discussion

Main findings: The majority of head and neck cancers are HNSCC originating from the epithelium localized in 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.

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