ORIGINAL RESEARCH

Interleukin-18 promoter genotype is associated with the risk of nasopharyngeal carcinoma in Taiwan

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Background: The incidence rate of nasopharyngeal carcinoma (NPC) has been documented to be high in Southeast Asia. Interleukin-18 (IL-18) is a multifunctional cytokine that augments interferon-γ production and acts as an important immunomediator in the development of several types of cancer.

Patients and methods: This case–control study evaluated the role of *IL-18* in NPC at the DNA level by genotyping its promoter polymorphisms at positions -656, -607, and -137 in a Taiwanese population. A total of 176 patients with NPC and age- and gender-matched 352 noncancer controls were included in this study.

Results: The CC genotype of the *IL-18*-607 polymorphism was found to be associated with significantly decreased risks of NPC compared to the AA genotype (crude OR =0.50, 95% CI =0.29–0.84, *P*=0.0093). This significant difference persisted even in the dominant and recessive models. A significantly lower C allele frequency at position -607 was detected in the NPC group (41.8% vs 50.3%; OR =0.77; 95% CI =0.63–1.04, *P*=0.0089). Regarding *IL-18*-656 and -137 polymorphisms, there were no differential distributions of their genotypes between the NPC and control groups. After substratification of the subjects according to their smoking, alcohol consumption, and areca chewing status, the genotype distribution of the *IL-18*-607 polymorphism was found to be different only among nonsmokers between the NPC and control subgroups.

Conclusion: This study suggests that *IL-18* plays an important role in the carcinogenesis of NPC in Taiwan and that the genotype–phenotype correlation of *IL-18*-607 polymorphism and its contribution to NPC need to be investigated further.

Keywords: genotype, *IL-18*, nasopharyngeal carcinoma (NPC), polymorphism, Taiwan

Introduction

Nasopharyngeal carcinoma (NPC) is a rapidly growing squamous cell carcinoma that is manifested by distant metastasis and affects both adults and children.[1] It has been reported that infection caused by the Epstein–Barr virus is a major risk factor for NPC.[2] Furthermore, studies have shown that cigarette smoking and alcohol consumption constitute the environmental risk factors for NPC.[3,4] Mounting evidence indicates that the variant genotypes of the genes related to inflammatory cytokines could modulate the susceptibility of individuals to cancers.[5–8] Polymorphisms of these genes may affect the cellular responses to inflammation and the secretion of cytokines by altering the expression and function of the genes. In NPC-endemic areas, although every individual is exposed to the same environment, only a few develop the disease, suggesting that genetic differences such as single-nucleotide polymorphisms (SNPs) contribute to the

carcinogenesis of NPC. However, the molecular mechanism by which these factors induce NPC still remains unknown. Increasing knowledge of NPC at the genomic level could indicate the novel biomarkers for the early detection and prediction of not only NPC susceptibility but also prognosis.

Interleukin-18 (IL-18), initially known as interferon-γ (IFN-γ)-inducing factor, is a cytokine with a proinflammatory biofunction and produced by activated macrophages, epithelial cells, osteoblasts, keratinocytes, and also cancer cells.[9,10] Studies using animal models have demonstrated that administration of additional IL-18 resulted in the suppression of tumor growth,[10,11] thus suggesting an important role of IL-18 in the host defense against cancer. Nevertheless, the role of IL-18 still remains incompletely understood. IL-18 is molecularly regulated and modulated under extremely complicated mechanisms in carcinogenesis, which is of great interest for oncologists. First, IL-18 may exert its tumor-suppressive effects by augmenting IFN-γ production, particularly in the presence of IL-12, promoting Th1 differentiation, enhancing the cytotoxic activities of natural killer cells and CD8+ lymphocytes,[10] inducing cancer cell apoptosis,[12] and inhibiting angiogenesis.[10] Second, the activities of IL-18 have been reported to be effected by the microenvironmental milieu status; for instance, IL-18 could enhance the differentiation of Th2 based on the presence of IL-4. Moreover, IL-18 has the ability to suppress the recognition of cancer cells by immune cells, thereby enhancing the ability of cancer cells to adhere to the walls of the vascular cells, inducing the production of angiogenic and growth factors, and promoting a prometastatic microenvironment.[9,13,14] IL-18 levels were found to be much higher in the blood of patients with metastatic cancer than in patients without metastasis and in healthy people, thus deducing the reasonable hypothesis that serum IL-18 concentrations may be used as a noninvasive marker for detecting metastasis in breast cancer.[15] All the abovementioned findings indicate that the pleiotropic cytokine IL-18 has the ability to biphasically perform its anticancerous and procancerous functions.[9,14]

From the viewpoints of the genome, the expression of IL-18 appears to be regulated by at least two SNPs at the -607 (A/C) and -137 (G/C) positions of its promoter sequences. The former SNP may cause a change from A to C at position -607, which disrupts a potential cAMP-responsive element-binding protein site, and the latter may cause a change at position -137 from G to C, which may block the human histone H4 gene-specific transcription factor-1 nuclear factor-binding site.[16] These differences in interaction between the sequences with related transcription factors are suggestive

of the mechanisms through which the two promoter polymorphisms affect the overall *IL-18* gene activity. Another well-known polymorphic site in the *IL-18* promoter region is -656 (A/C) whose biofunction has not yet been clearly elucidated. In other words, the promoter polymorphism of *IL-18* gene has been examined in autoimmune diseases[16] but not frequently among cancers.

The majority of genomic studies published in the literature have focused on investigating the association between *IL-18*-607 and -137 polymorphisms and various types of cancers. Some of them have reported positive results,[17–21] whereas others have reported negative findings.[22,23] For instance, Pratesi et al[24] showed that *IL-18*-607 or -137 polymorphism was not associated with NPC susceptibility. Similarly, Farhat et al[25] also demonstrated a negative finding. However, Nong et al[26] reported that the -137C/-607A haplotype of *IL-18* was associated with a significantly increased risk of NPC compared with its counterpart, the -137G/-607C haplotype. In 2013, a meta-analysis investigating the contribution of IL-18 genotypes to cancer risk proposed that the C-607A polymorphism is significantly associated with overall cancer risk, especially in NPC and gastrointestinal cancer, and the G-137C polymorphism is associated with an increased overall cancer risk in Asian populations and also significantly increases the risk of NPC.[27] In the present study, the SNPs at -656 (A/C, rs1946519), -607(A/C, rs1946518), and -137 (G/C, rs187238) in the promoter region of the *IL-18* gene were identified and analyzed in patients with NPC in Taiwan.

Patients and methods
Study subjects
During the period from 2003 to 2009, a total of 176 patients with NPC were included through the general surgery outpatient clinics in China Medical University Hospital. All the patients were asked to participate in the project freely and voluntarily and to complete a questionnaire, in addition to providing 5 mL of their peripheral blood samples. The well-designed questionnaire consisted of questions pertaining to basic characteristics and history of cancer. In addition, the frequency of alcohol consumption, areca chewing, and smoking behaviors were recorded, in which "ever" was defined as more than twice a week for at least 1 year and then reclassified during analysis. The histological status of NPC was identified and judged by expert surgeons according to the 1991 WHO classification system. The histologic types included keratinizing squamous cell carcinoma (WHO type I) in 5 (2.8%) and non-keratinizing carcinoma (WHO type II) in

171 (97.2%) cases. Furthermore, type II was categorized into non-keratinizing differentiated carcinoma (WHO type IIa) in 28 (15.9%) and non-keratinizing undifferentiated carcinoma (WHO type IIb) in 143 (81.3%) cases.

For each patient with NPC, twice the number of age- and gender-matched cancer-free healthy subjects were selected from the pool of individuals undergoing regular health examination (age-matching was done within <5 years of the patient's first diagnosis) and included in the final comparison and evaluation. Consequently, a total of 352 healthy individuals, exactly twice the number of patients with NPC, were subjected to genotyping and final analysis. All the patients and healthy volunteers provided written informed consent. The study was approved by the authority from the institutional review board of China Medical University Hospital (DMR101-IRB1-306).

DNA preparation
Genomic DNA isolated from the peripheral blood leukocytes of each study subject was prepared using the QIAamp Blood Mini Kit (Qiagen NV, Venlo, the Netherlands), stored for long term at −80°C, diluted, and aliquoted for genotyping as a working stock at −20°C, as we do frequently.[28,29]

IL-18 genotyping methodology
The genotyping discrimination of *IL-18*-607 (A/C, rs1946518) and -137 (G/C, rs187238) polymorphisms was assessed using the ABI StepOne™ Real-Time PCR System (Thermo Fisher Scientific, Waltham, MA, USA) and analyzed by the typical TaqMan assay. For genotyping the *IL-18*-656 (A/C, rs1946519) polymorphism, the PCR-restriction fragment length polymorphism technique was applied using the primers reported by Folwaczny et al.[30] The forward primer 5′-AGGTCAGTCTTTGCTATCATTC-CAGG-3′ and the reverse primer 5′-CTGCAACAGAAAG-TAAGCTTGCGGAGAGG-3′ were used to amplify a 120-bp fragment of the *IL-18*-656 (A/C) polymorphism. The PCR products were cut by the restriction enzyme *Mwo* I, and the DNA fragments were resolved in 3.0% agarose gel, stained by ethidium bromide, and visualized under UV (260 nm) light. For the A allele, a 120 bp PCR fragment was observed, whereas for the C allele, two 96/24 bp fragments were observed.[9]

Statistical analysis
The genotypic and clinical data of all the 352 controls and the 176 patients with NPC were statistically analyzed. To ensure that the control subjects were representative

of the Taiwan population and to prevent any possible genotyping errors, the deviation of the genotypic frequencies of the investigated SNPs in the control groups from those expected under the Hardy–Weinberg equilibrium was assessed using the goodness-of-fit test. In addition, Pearson's chi-squared test was used for assessing the statistical significance in the distribution of the investigated genotypes and Cochran–Armitage test was applied for the trend analysis. Student's *t*-test was used to examine the differences in age, which is a continuous index, between the two groups. The associations between NPC risk and *IL-18* genotypes were determined statistically by calculating the ORs and the corresponding 95% CIs using the logistic regression analysis, adjusted or unadjusted for these confounders. Any *P*-value <0.05 was considered to be statistically significant.

Results
Comparisons of basic characteristics of NPC and control groups
The frequency distributions of age, gender, and personal behavioral habits of the 176 patients with NPC and 352 non-cancer control subjects are summarized in Table 1. Because we matched the patients with twice the number of controls, there was no significant difference in age and gender between the case and control groups (*P*>0.05). There was also no significant difference between the case and control groups in terms of the distributions of personal behavioral habits, including smoking, alcohol consumption, and areca chewing status (*P*>0.05) (Table 1).

Association analysis of *IL-18* genotypes and NPC risk in the study subjects
The genotypes of -656 (A/C, rs1946519), -607(A/C, rs1946518), and -137 (G/C, rs187238) polymorphisms of *IL-18* were determined in all the patients and the age- and gender-matched healthy controls, and the results are summarized in Table 2. The genotype frequencies of the -656, -607, and -137 polymorphisms of *IL-18* in both the case and control groups fit the Hardy–Weinberg equilibrium.

The genotypes of the *IL-18* promoter -607(A/C) SNP were found to be differently distributed between patients and controls (P_{trend} =0.0090) (Table 2, middle panel). In detail, the *IL-18*-607 (A/C) homozygous CC variant genotypes were associated with decreased risks of NPC (OR =0.46, 95% CI =0.25–0.78, *P*=0.0093) (Table 2, middle panel). In both the dominant and recessive models, there were significant associations with the risk of NPC (Table 2, middle panel). These

Table 1 Demographic characteristics of the investigated NPC patients and noncancer healthy controls

Characteristics	Controls (n=352)			Cases (n=176)			P-value[a]
	n	%	Mean (SD)	n	%	Mean (SD)	
Age (years)			48.7 (10.8)			49.3 (9.4)	0.7138
Gender							1.0000
Male	256	72.7		128	72.7		
Female	96	27.3		48	27.3		
Behavioral habits							
Cigarette smoker	150	42.6		73	41.4		0.8519
Alcohol drinker	124	35.2		72	40.9		0.2150
Areca chewer	115	32.7		54	30.7		0.6926
Classification							
KSCC (WHO type I)				5	2.8		
NKC (WHO type II)				171	97.2		
NKDC (WHO type IIa)				28	15.9		
NKUC (WHO type IIb)				143	81.3		

Note: [a]Based on chi-squared test or Student's t-test.
Abbreviations: KSCC, keratinizing squamous cell carcinoma; NKC, non-keratinizing carcinoma; NKDC, non-keratinizing differentiated carcinoma; NKUC, non-keratinizing undifferentiated carcinoma; NPC, nasopharyngeal carcinoma.

significant results persisted even after adjusting for potential confounders, including age, gender, smoking, alcohol consumption, and areca chewing habits (Table 2, middle panel). Neither the genotypes nor the alleles at positions -656 and -137 showed any correlation with the risk of NPC in any of the groups (Table 2).

We also performed allelic analysis, and the results are shown in Table 3. Supporting the findings shown in Table 2, the results showed that the frequency of the variant allele C was 41.8% in the group of patients with NPC, which was significantly much lower than that (50.3%) in the control group (adjusted OR =0.77, 95% CI =0.63–1.04, P=0.0089). Again, the other two SNPs demonstrated no significant association with the risk of NPC (Table 3).

Stratified analysis of *IL-18* genotypes according to environmental factors

We further performed stratified analyses of the genotypic association between the *IL-18*-607 polymorphism and the risk of NPC according to potential environmental risk factors in Taiwan, including cigarette smoking, alcohol consumption, and areca chewing habits. The adjusted ORs for carriers with the genotypes AC and CC at *IL-18*-607 were 0.69 and 0.37 among nonsmokers (95% CI =0.42–1.02 and 0.22–0.73, P-values 0.0947 and 0.0091, respectively) and 0.81 and 0.62 among smokers (95% CI =0.41–1.42 and 0.33–1.37, P-values 0.6137 and 0.3385, respectively). The protective effects of the *IL-18*-607 genotype against the risk of NPC appeared to be more obvious among nonsmokers (Table 4). At the same time, there was also no interaction between the *IL-18*-656

or -137 genotype and any of the environmental risk factors (data not shown).

Discussion

Unlike other head and neck cancers, NPC is characterized by its specific multifactorial etiology, worldwide geographical distribution, and extremely high sensitivity to radiotherapy and chemotherapy induction during therapeutic processes.[31] For several years, the members of Terry Fox Cancer Research Laboratory have been working on genomic biomarkers for early detection and prediction in Taiwan, where NPC is prevalent and causes several cancer-related deaths.[32–36] Cytokines play a complex role in the initiation and progression of inflammation and tumorigenesis.[37] Studies have confirmed that the proinflammatory cytokine IL-18 possesses antitumor capacity against lung cancer in several murine models.[14,38] In addition, preclinical trials used the recombinant human IL-18 as an antidote and found it to be beneficial for the examined patients.[39] However, despite the conventional view that IL-18 is an anticancer agent, some reports have also suggested that this multifunctional cytokine exhibits a procancerous activity under some conditions.[14] Recently, an increasing number of studies have shown that various genotypes of cytokine genes may influence the production and serum levels of cytokines, which may be closely associated with susceptibility to certain human diseases.[16–21] Among the numerous SNPs, all the following three polymorphisms selected for this study are located in the promoter region of *IL-18*: -656 (G/T), -607 (C/A), and -137 (G/C). It has been reported that these polymorphisms cause differences in

Table 2 Distribution of *IL-18* genotypes among the NPC patients and noncancer healthy controls

	Controls		Patients		OR (95% CI)	OR (95% CI)[a]	*P*-value[b]
	n	%	n	%			
IL-18-656							
AA	134	38.0	64	36.4	1.00 (reference)	1.00 (reference)	
AC	150	42.6	80	45.4	1.12 (0.75–1.67)	1.02 (0.68–1.43)	0.5913
CC	68	19.3	32	18.2	0.99 (0.59–1.65)	1.03 (0.62–1.71)	0.9550
P_{trend}[c]							0.9326
Carrier comparison							
AA+AC	284	80.7	144	81.8	1.00 (reference)	1.00 (reference)	
CC	68	19.3	32	18.2	0.93 (0.58–1.48)	0.98 (0.62–1.30)	0.7534
AA	134	38.0	64	36.4	1.00 (reference)	1.00 (reference)	
AC+CC	218	62.0	112	63.6	1.07 (0.74–1.57)	1.03 (0.83–1.39)	0.7029
IL-18-607							
AA	86	24.4	59	33.5	1.00 (reference)	1.00 (reference)	
AC	178	50.6	87	49.4	0.71 (0.47–1.03)	0.73 (0.43–1.07)	0.1121
CC	88	25.0	30	17.1	**0.50 (0.29–0.84)**	**0.46 (0.25–0.78)**	**0.0093**
P_{trend}[c]							**0.0090**
Carrier comparison							
AA+AC	264	75.0	146	82.9	1.00 (reference)	1.00 (reference)	
CC	88	25.0	30	17.1	**0.62 (0.39–0.98)**	**0.59 (0.37–0.89)**	**0.0386**
AA	86	24.4	59	33.5	1.00 (reference)	1.00 (reference)	
AC+CC	266	75.6	117	66.7	**0.64 (0.43–0.95)**	**0.61 (0.51–0.86)**	**0.0274**
IL-18-137							
GG	281	79.8	133	75.6	1.00 (reference)	1.00 (reference)	
GC	65	18.5	38	21.6	1.24 (0.79–1.94)	1.17 (0.71–2.14)	0.3574
CC	6	1.7	5	2.8	1.76 (0.53–5.87)	1.67 (0.45–6.17)	0.3515
P_{trend}[c]							0.2150
Carrier comparison							
GG+GC	346	98.3	171	97.2	1.00 (reference)	1.00 (reference)	
CC	6	1.7	5	2.8	1.69 (0.51–5.60)	1.47 (0.42–4.78)	0.3888
GG	281	79.8	133	75.6	1.00 (reference)	1.00 (reference)	
GC+CC	71	20.2	43	24.4	1.28 (0.83–1.97)	1.25 (0.74–1.84)	0.2619

Notes: [a]Adjusted with age, gender, smoking, alcohol drinking, and areca chewing habits. [b]Based on chi-squared test without Yates' correction. [c]Cochran–Armitage test was applied for the trend analysis. Bold values are statistically significant.
Abbreviations: IL-18, interleukin-18; NPC, nasopharyngeal carcinoma.

Table 3 Allelic frequency analysis for *IL-18* polymorphisms and NPC

Allele	Controls, n (%)	Patients, n (%)	OR (95% CI)[a]	*P*-value[b]
IL-18-656				
G	418 (59.4)	208 (59.1)	1.00 (reference)	
T	286 (40.6)	144 (40.9)	1.05 (0.84–1.23)	0.9294
IL-18-607				
A	350 (49.7)	205 (58.2)	1.00 (reference)	
C	354 (50.3)	147 (41.8)	0.77 (0.63–1.04)	**0.0089***
IL-18-137				
G	627 (89.1)	304 (86.4)	1.00 (reference)	
C	77 (10.9)	48 (13.6)	1.15 (0.71–1.95)	0.2006

Notes: [a]OR was adjusted with age, gender, smoking, alcohol drinking, and areca chewing. [b]Based on chi-squared test without Yates' correction. Bold values are statistically significant.
Abbreviations: IL-18, interleukin-18; NPC, nasopharyngeal carcinoma.

their binding capacity with transcription factors.[16] The two *IL-18* promoter polymorphisms -607 and -137 were previously shown to be positively associated with several types of cancers, including esophageal squamous cell carcinoma[17] and prostate cancer[18] in Chinese subjects, colorectal cancer[19] in Greek subjects, ovarian cancer[20] in US subjects (Hawaiians), and breast cancer[21] in Iranian subjects. In contrast, studies have also reported negative findings regarding the

Table 4 OR for *IL-18* promoter -607 genotype and NPC after stratified by smoking status

Genotypes	Nonsmokers		OR (95% CI)[a]	*P*-value[b]	Smokers		OR (95% CI)[a]	*P*-value[b]
	Controls	Cases			Controls	Cases		
AA	46	36	1.00 (reference)		40	23	1.00 (reference)	
AC	104	51	0.69 (0.42–1.02)	0.0947	74	36	0.81 (0.41–1.42)	0.6137
CC	52	16	**0.37 (0.22–0.73)**	**0.0091**	36	14	0.62 (0.33–1.37)	0.3385
P$_{trend}$[c]				**0.0080**				0.3399

Notes: [a]ORs were adjusted with age, gender, alcohol drinking, and areca chewing. [b]Based on chi-squared test without Yates' correction. [c]Cochran–Armitage test was applied for the trend analysis. Bold values are statistically significant.
Abbreviations: IL-18, interleukin-18; NPC; nasopharyngeal carcinoma.

association between *IL-18* polymorphisms and squamous cell carcinoma of the head and neck cancers in Iranian subjects[23] or oral cancer in Greek subjects.[22] These discrepant results may be explained by the following three possibilities: the dual effects of IL-18 on the tumor immune response,[14] the different types of tumors, and the various populations that were investigated.[23]

To our knowledge, this is the first study to investigate the SNPs at the positions -656 (G/T), -607 (C/A), and -137 (G/C) of the *IL-18* promoter region in patients with NPC and non-cancer healthy control individuals in a Taiwanese population and evaluate their contribution to the prediction of NPC risk. The results showed significant protective effects of the CC homo-variant genotypes and the C allele at position -607 of the *IL-18* gene against NPC susceptibility (Tables 2 and 3). Regarding -656 and -137 genotypes of *IL-18*, there was no significant association between any genotype or allelic type and the risk of NPC (Tables 2 and 3). The positive finding indicating that the genotype of the *IL-18*-607 polymorphism was the determinant of NPC susceptibility and was found to be inconsistent with all the previous findings investigating the contribution of *IL-18* genotypes to the risk of NPC, which were reported by Pratesi et al,[24] Farhat et al,[25] and Nong et al.[26] In 2013, the meta-analysis reported by Guo and Xia[40] combined all the available limited and updated literature and demonstrated that the C allele of *IL-18*-607 had a protective effect against the risk of NPC, especially for Asians, but not for Africans or Europeans. This is consistent with another meta-analysis reported in 2013.[27] The first positive finding of this study, while uniquely consistent with the abovementioned overall meta-analysis, may be due to the fact that Taiwan is located in East Asia and is an island with conserved genetic, cultural, and environmental background. Although the sample size of this study is relatively larger than that of other studies, the contribution of *IL-18* genotypes, especially those at *IL-18*-607, must be validated using larger samples and other populations across the world. All the results pertaining

to the contribution of *IL-18* genotypes to the risk of NPC are concisely summarized in Table 5. Among the NPC susceptibility genes investigated in this study, the roles of human leukocyte antigen (HLA) genes have been most intensively examined and claimed to be associated with the outcome of NPC by different research groups across the world.[41–47] However, the findings are still inconclusive,[48] and the detailed mechanisms underlying the effect have not been completely elucidated. Similar to the case of *HLA* genes, further clinical studies investigating the contribution of *IL-18* genotypes to NPC using larger sample sizes from various ethnic groups are still needed to fulfill the possibility of enhancing the early diagnosis and personalized treatment of NPC.

Substratification of the patients with NPC and noncancer subjects according to their personal behavioral habits revealed that the *IL-18*-607 genotype may play a significantly important role in the determination of the susceptibility to NPC among nonsmokers, but not smokers (Table 4). In 2017, Long et al reported that cigarette smoking was associated with an increased risk for NPC, especially for subjects who are young, male, and had early-onset, long-term, and undifferentiated type of NPC.[4] However, the authors did not examine the effect of interaction between genotypes (such as *IL-18*-607) and smoking on the risk of NPC. Therefore, further investigations are needed to elucidate the detailed mechanisms underlying the interaction between IL-18 and other molecules and environmental factors related to the etiology of NPC.

Conclusion
To our knowledge, this is the first study indicating a significant association between *IL-18*-607 polymorphism and the risk of NPC in Taiwan. This is also the first study to investigate the interaction between *IL-18* genotypes and environmental factors in terms of the risk of NPC. The significant association between *IL-18*-607 polymorphism and the risk of NPC was particularly observed in patients without

Table 5 Summary of the characteristics of studies investigating *IL-18-607* genotypes and NPC

Study	Year	Population	Ethnicity	Design	Sample size (case/control)	Matching criteria	Major findings	Reference
Huang et al (current study)	2018	Taiwan	Asian	Case–control	176/352	Age, gender	C allele is protective, especially for nonsmokers	–
Nong et al[26]	2009	China	Asian	Case–control	250/270	Age, gender	–607C alone is not protective, but –137C/–607A haplotype is risk	26
Farhat et al[25]	2008	Tunisia	African	Case–control	163/164	Age, gender	C allele has no effect	25
Pratesi et al[24]	2006	Italy	European	Case–control	89/130	Age, gender	C allele has no effect on risk, but on late stages	24

Abbreviations: IL-18, interleukin-18; NPC, nasopharyngeal carcinoma.

smoking behavior. Additional data from a larger number of subjects and other populations are required to validate the positive findings of *IL-18* genotypes as a biomarker for early detection and prediction of NPC. These results suggest that *IL-18* plays an important role in the carcinogenesis of NPC in Taiwan and that the interaction between *IL-18*-607 polymorphism and smoking in NPC etiology is worth further research.

Acknowledgments

Disclosure

The authors report no conflicts of interest in this work.

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