Gene 544 (2014) 123–127

Contents lists available at ScienceDirect

Gene

journal homepage: www.elsevier.com/locate/gene

Combined effect between CHRNB3–CHRNA6 region gene variant (rs6474412) and smoking in psoriasis vulgaris severity

Kun-Ju Zhu [a,*,1], Cheng Quan [b,1], Chi Zhang [c,1], Zhong Liu [a], Huan Liu [a], Ming Li [d], Shi-Jie Li [a], Cheng-Yao Zhu [a], Ge Shi [a], Ke-Shen Li [e,*], Yi-Ming Fan [a,*]

[a] Department of Dermatology, Affiliated Hospital of Guangdong Medical College, Zhanjiang, Guangdong, China
[b] Department of Dermatology, Affiliated Hospital of Xuzhou Medical College, Xuzhou, Jiangsu, China
[c] Department of Dermatology, Anhui Provincial Hospital, Hefei, Anhui, China
[d] Department of Dermatology, Xinhua Hospital, Shanghai Jiaotong University School of Medicine, Shanghai, China
[e] Guangdong Key Laboratory of Age-Related Cardiac and Cerebral Diseases, Affiliated Hospital of Guangdong Medical College, Zhanjiang, China

ARTICLE INFO

Article history:
Received 22 October 2013
Received in revised form 22 April 2014
Accepted 29 April 2014
Available online 30 April 2014

Keywords:
CHRNB3–CHRNA6 region gene variant
Smoking
Combined effect
Psoriasis vulgaris

ABSTRACT

Background: Many factors associated with causing psoriasis have been reported, such as the genetic and environmental factors. Smoking is one of the well-established environmental risk factors for psoriasis and also associated with the disease severity. In addition, several studies of psoriasis and psoriatic arthritis have documented gene–environment interactions involving smoking behavior. Although gene polymorphisms on nicotinic acetylcholine receptor subunits CHRNB3–CHRNA6 region gene have been found to correlate with smoking behavior and lung cancer susceptibility in Chinese Han population, the combined effect between the smoking-related genetic variants and smoking behavior on psoriasis vulgaris (PV) has been unreported.
Objective: To evaluate the combined effect of the smoking-related (rs6474412-C/T) polymorphism on CHRNB3–CHRNA6 region gene and smoking behavior on PV risk and clinic traits in Chinese Han population.
Methods: A hospital-based case–control study including 672 subjects (355 PV cases and 317 controls) was conducted. The variant of rs6474412 was typed by SNaPshot Multiplex Kit (Applied Biosystems Co., USA).
Results: The higher body mass index (BMI ≥ 25), smoking behavior and alcohol consumption were risk factors for PV, and the estimated ORs were 1.55 (95% CI, 1.09–2.29), 1.74 (95% CI, 1.22–2.49) and 1.81 (95% CI, 1.25–2.62) respectively. The smoking patients had more severe conditions than non-smokers (OR = 1.71, 95% CI, 1.08–2.70, $P = 0.020$). The alleles and genotypes of rs6474412 were not associated with risk of PV, but the combined effect of rs6474412 genotype (TT) and smoking behavior increased severity of PV (OR = 5.95; 95% CI, 1.39–25.31; $P < 0.05$; adjusted OR = 2.20; 95% CI, 1.55–3.14; $P < 0.001$).
Conclusions: Our results demonstrate that the combined effect of rs6474412-C/T polymorphism in smoking-related CHRNB3–CHRNA6 region gene and smoking behavior may not confer risk to PV, but may have impact on PV severity in Chinese Han population.

1. Introduction

Psoriasis is an etiologically complex dermatological disease that affects approximately 0.123% of Chinese Han population (Schon and Boehncke, 2005; Zhang et al., 2002), of which psoriasis vulgaris (PV) is the most common and accounts for about 90% of adult cases (Fry, 1988). Heritability of psoriasis in first- and second-degree relatives is 67.04% and 46.59%, respectively (Zhang et al., 2002). Recently, genome-wide association studies (GWAS) and resequencing for candidate loci have uncovered a number of common and rare psoriasis-associated gene variants (Chandran, 2013; Ellinghaus et al., 2010; Jordan et al., 2012a,b; Nair et al., 2009; Strange et al., 2010; Sun et al., 2010; Zhang et al., 2009). Furthermore, many studies have demonstrated associations between psoriasis and environmental factors (Armstrong et al., 2011; Lebwhol and Callen, 2006; Naldi and Mercuri, 2009; Zhu et al., 2011).

As a well established environmental risk factor for psoriasis, smoking is also associated with psoriasis severity (Fortes et al., 2005), but the underlying mechanism is unclear. Several studies found that smoking can modify risk estimates for genetic markers in psoriasis, cancer, peripheral arterial disease and coronary artery disease (Duffin et al., 2009;

Abbreviations: PV, psoriasis vulgaris; GWAS, genome-wide association studies; PASI, Psoriasis Area and Severity Index; IL13, interleukin-13; nAChR, nicotinic acetylcholine receptor; BMI, body mass index; WHO, World Health Organization; OR, odds ratios; CI, confidence interval; SPSS, Statistical Package for Social Scientists; CHRNB3, the β3 nAChR subunit gene; CHRNA6, the α6 nAChR subunit gene; LD, Linkage Disequilibrium.

* Corresponding authors at: No. 57, Renmin Avenue, Xiashan District, Zhanjiang, Guangdong, 524001, China.
E-mail address: zhukunju1111@163.com (K.-J. Zhu).

[1] These authors contributed equally to this work.

http://dx.doi.org/10.1016/j.gene.2014.04.070
0378-1119/

Jaworowska et al., 2011; Jin et al., 2009; Kramer and Esser, 2006; Lewis et al., 2011; Li et al., 2012; Yin et al., 2013). Psoriatic subjects with a non-variant CYP1A1 (Allele *1A) genotype and smoking behavior may be more susceptible to psoriasis (Kramer and Esser, 2006). A minor allele of interleukin-13 (IL13) polymorphism rs1800925*T may protect carriers from developing psoriatic arthritis, but this effect can be weakened by smoking (Duffin et al., 2009). The psoriasis risk of smokers with HLA-Cw6 increased by about 11-fold compared with nonsmokers without HLA-Cw6 in Chinese (Jin et al., 2009). The interaction between CSMD1 (rs7007032 and rs10088247) and smoking was evident in another Chinese study (Yin et al., 2013). In principle, these studies provide possible interactions between smoking and psoriasis.

However, variations of the smoking-related genes have been not mentioned in most of these studies. To our knowledge, smoking behavior is also under genetic influence (Agrawal et al., 2012). Previous GWA studies identified numerous smoking-related susceptibility loci (Agrawal et al., 2012; Tobacco-and-Genetics-Consortium, 2010). A GWA meta-analysis including more than 77,000 samples depicted nicotinic acetylcholine receptor (nAChR) subunits (CHRNB3 and CHRNA6, 8p11.2) within regions harboring nAChR gene sequence variant (rs6474412-T/C) was correlated with smoking behavior (Thorgeirsson et al., 2010). CHRNB3–CHRNA6 region gene polymorphisms were related to smoking behavior and lung cancer susceptibility in Chinese Han population (Wei et al., 2012; Zhang et al., 2011b). Therefore, genetic factors may not only affect smoking behavior, but also play a role in the development of smoking-related diseases. We speculate that the combined effect between smoking behavior and smoking-related genetic variant may be existent in PV. In the present study, the association between (rs6474412-C/T) polymorphism among CHRNB3–CHRNA6 region gene PV risk and clinic traits was investigated in 355 PV cases and 317 controls.

2. Materials and methods

2.1. Recruitment and clinical evaluation of patients and controls

All subjects of this study were recruited from December 2009 to May 2013 from southern China. The clinical diagnoses of all cases were confirmed by at least two dermatologists. PV patients at above 15 years of age were eligible. The 355 cases were consecutively admitted to the outpatients. Clinical data including age, gender, smoking behavior, alcohol consumption, body mass index (BMI), age of onset (early onset ≤ 40; later onset > 40), family history, and severity of psoriasis lesions (Psoriasis Area and Severity Index; PASI score) were collected. The 317 non-psoriatic controls at above 15 years of age were recruited simultaneously in a health examination center and free from history of other autoimmune diseases. This study was approved by the ethical committee of our hospital, and the informed consents were obtained from all participants.

2.2. Anthropometric measurements

All subjects underwent physical examinations. Body height and weight were measured to the nearest 0.1 cm and 0.1 kg, respectively. BMI was classified according to the World Health Organization (WHO): normal body weight (BMI < 25 kg/m^2) and abnormal body weight (BMI ≥ 25 kg/m^2; including overweight and obesity). Smoking status was coded as nonsmokers and smokers (including both current and former smokers who took 15 or more cigarettes per day for 1 year or longer), while alcohol consumption was divided into drinkers and non-drinkers (including individuals who drank alcohol less than 170 g/week for 1 year or longer). A positive family history was determined if one or more first- and second-degree relatives of the proband had psoriasis. The severity of the psoriasis was assessed using PASI score (Langley and Ellis, 2004), and then classified into mild (PASI ≤ 6.3) and severe (PASI > 6.3) based on median distribution.

2.3. DNA isolation and genotyping

Genomic DNA was extracted from peripheral blood leukocytes using Flexi Gene DNA kits (QIAGEN, Germany) according to the manufacturer's instruction. The polymorphism was analyzed by SNaPshot Multiplex Kit (Applied Biosystems Co., USA). The primers used were as follows: rs6474412_F: AGCAAGACAGCCTCTTTTCC, rs6474412_R: CATCCCACTGTAGAAACCTC.

2.4. Statistical analyses

Comparisons of characteristics between PV patients and controls were made using t-test, the Pearson's chi-square test and odds ratios (OR) by using the Statistical Package for Social Scientists (SPSS) version 11.5. The SNP was analyzed for an association with the disease by means of comparison of the minor allele frequency in patients and controls using PLINK v1.07 (Purcell et al., 2007), and it was tested for significant deviation from Hardy–Weinberg equilibrium in controls and passed the test with P values > 0.05. In addition to the allelic test of association, the additive genetic model was calculated (dominant, additive or negative).

A logistic regression analysis was performed to obtain OR and P-values to explore predictive ability of combined effect smoking and variant on the risk of PV, adjusted for age, gender, BMI and alcohol consumption. We also performed case-only analyses to examine PASI score that is conferred by combined effect smoking and variant on PASI score of PV by logistic regression analysis adjusted for age, gender, BMI, alcohol consumption, family history and age of onset. P value, OR, and 95% CI were determined for each kind of PASI score. All tests were two-sided and the level of statistical significance was set at $P < 0.05$.

3. Results

3.1. Baseline characteristics

A total of 355 PV patients (range from 15 to 81 years) and 317 controls (range from 15 to 83 years) were investigated. The baseline characteristics of PV patients and controls are shown in Table 1. The age and sex differences were insignificant between cases and controls, but BMI, smoking behavior and alcohol consumption showed significant differences ($P < 0.05$). Compared with the control group, BMI, smoking behavior and alcohol consumption were risk factors for PV, and the estimated ORs were 1.55 (95% CI, 1.09–2.29), 1.74 (95% CI, 1.22–2.49) and 1.81 (95% CI, 1.25–2.62) respectively (Table 1).

3.2. No association of rs6474412 alleles and genotypes with smoking in controls and PV cases

We did not find a significant association between the SNP and smoking (for allele the $P = 0.17$, and for genotypes the $P = 0.99$ (CC VS. CT) and $P = 0.33$ (CC VS. TT)). The genotype and allele frequencies of rs6474412 polymorphism in cases and controls are shown in Table 2. The chi-squared test revealed no statistical association between rs6474412-C/T polymorphism and PV risk ($P > 0.05$), and so did in dominant (CC + CT vs. TT) and recessive (CC vs. CT + TT) models ($P > 0.05$).

3.3. Combined effect of smoking and rs6474412 genotypes on PV risk and severity

Based on the above data, we further analyzed the combined effect of rs6474412 genotypes and smoking behavior on PV. The combined OR of rs6474412 and smoking increased from 1.27 to 2.80, although the test did not achieve significance between cases and controls. Even if adjusting the confounder factors such as age, sex, BMI and alcohol consumption, the combined effect was still not associated with risk of PV (combined ORs being from 1.04 to 1.67, $P > 0.05$).