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Research Paper

Genetic Risk Can Be Decreased: Quitting Smoking Decreases and Delays Lung Cancer for Smokers With High and Low *CHRNA5* Risk Genotypes — A Meta-Analysis

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A B S T R A C T

Background: Recent meta-analyses show that individuals with high risk variants in *CHRNA5* on chromosome 15q25 are likely to develop lung cancer earlier than those with low-risk genotypes. The same high-risk genetic variants also predict nicotine dependence and delayed smoking cessation. It is unclear whether smoking cessation confers the same benefits in terms of lung cancer risk reduction for those who possess *CHRNA5* risk variants versus those who do not.
Methods: Meta-analyses examined the association between smoking cessation and lung cancer risk in 15 studies of individuals with European ancestry who possessed varying rs16969968 genotypes ($N = 12,690$ ever smokers, including 6988 cases of lung cancer and 5702 controls) in the International Lung Cancer Consortium.
Results: Smoking cessation (former vs. current smokers) was associated with a lower likelihood of lung cancer ($OR = 0.48$, $95\%CI = 0.30–0.75$, $p = 0.0015$). Among lung cancer patients, smoking cessation was associated with a 7-year delay in median age of lung cancer diagnosis ($HR = 0.68$, $95\%CI = 0.61–0.77$, $p = 4.9 * 10^{-10}$). The *CHRNA5* rs16969968 risk genotype (AA) was associated with increased risk and earlier diagnosis for lung cancer, but the beneficial effects of smoking cessation were very similar in those with and without the risk genotype.
Conclusion: We demonstrate that quitting smoking is highly beneficial in reducing lung cancer risks for smokers regardless of their *CHRNA5* rs16969968 genetic risk status. Smokers with high-risk *CHRNA5* genotypes, on average, can largely eliminate their elevated genetic risk for lung cancer by quitting smoking- cutting their risk of lung cancer in half and delaying its onset by 7 years for those who develop it. These results: 1) underscore the potential value of smoking cessation for all smokers, 2) suggest that *CHRNA5* rs16969968 genotype affects lung cancer diagnosis through its effects on smoking, and 3) have potential value for framing preventive interventions for those who smoke.

1. Introduction

Cigarette smoking is the leading modifiable risk for cancer and multiple major chronic illnesses. Growing evidence identifies that genetic variation in the α5 nicotinic cholinergic receptor subunit *(CHRNA5)* gene plays a key role in both heavy smoking and nicotine dependence (Bierut et al., 2007; Bierut et al., 2008a). Multiple large meta-analyses demonstrated the association with smoking quantity, defined by number of cigarettes smoked per day (CPD) (Liu et al., 2010; Saccone et al., 2010; TAG, 2010; Thorgeirsson et al., 2010), with the most robust signal for two highly correlated genetic variants, rs16969968 and rs1051730 in subjects of European ancestry (TAG, 2010). The coding variant, rs16969968, results in an amino acid change in the α5 nicotinic cholinergic receptor subunit, and subsequently alters nicotinic receptor conductance in vitro (Bierut et al., 2008b; Kuryatov et al., 2011). Genetic variation in *CHRNA5* increases risk for smoking-related disorders such as lung cancer and chronic obstructive pulmonary disease (COPD) (Amos et al., 2008; Pillai et al., 2009; Thorgeirsson et al., 2008). Research suggests that the association between *CHRNA5* and lung cancer may be mediated by COPD (Young et al., 2011; Brennan et al., 2011; Wang et al., 2010). Further, *CHRNA5* has been reported to predict smoking cessation in both cessation trials (Baker et al., 2009; Bergen et al., 2013; Chen et al., 2012; Munafo et al., 2011; Sarginson et al., 2011) and general population studies (TAG, 2010; Chen et al., 2012; Chen et al., 2015a). The *CHRNA5* variant rs16969968 was recently shown to be a marker of delayed smoking cessation in a meta-analysis (Chen et al., 2015a). Smokers with the high-risk genotype had delayed age of smoking cessation compared with those with the low-risk genotype (age 56 versus age 52). Similarly, those with the high-risk genotype had a 4-year earlier age of lung cancer diagnosis compared to those with the low-risk genotypes (age 61 vs. age 65).

Although it is clear that smoking cessation reduces cancer risk (Jha et al., 2013), and that *CHRNA5* risk variants affect both smoking heaviness and duration, it is unclear whether *CHRNA5* alters the health benefits of smoking cessation. For example, the *CHRNA5* high-risk variants may directly affect lung cancer risk or, instead, influence risk via effects on smoking heaviness and cessation (Amos et al., 2008; Thorgeirsson et al., 2008; Ji et al., 2015; Tseng et al., 2014; Wang et al., 2012; Hung et al., 2008a). In the first case, smoking cessation would have negligible effects on lung cancer latency or risk among those with the high-risk variants. Smokers with such risk variants may have smoked so

intensively prior to quitting, that cessation confers lesser benefit for them (in terms of lung cancer onset or likelihood). Conversely, smokers with low-risk genotypes may not benefit as much from cessation because they might have low genetic risk for lung cancer in addition to their lower risk for heavy smoking.

Large epidemiologic studies have shown the clear benefits of smoking cessation in reducing mortality and morbidity (Jha et al., 2013), but no studies have examined whether such benefits vary based on a smoker's genotype. Certainly, cessation has the potential to benefit almost any smoker, but discovering that some smokers may especially benefit from cessation could not only elucidate the nature of the mechanism(s) linking *CHRNA5* to lung cancer, but might, also, inform personalized prevention efforts. For instance, such findings might encourage directing some smokers to additional preventive interventions and might be used to support "gain framed" messaging for such smokers, an approach to prevention with especially strong supporting evidence (Gallagher and Updegraff, 2012).

The *CHRNA5* risk variants were chosen for this study because they are associated with lung cancer risk and onset, with exposure to a primary cancer etiologic factor (smoking duration and heaviness), and with the effects of a preventive action (smoking cessation). Therefore, they have the potential to elucidate the relations among genetic risk, etiologic factors, and preventive actions.

To address the gap in knowledge concerning *CHRNA5*, smoking cessation, and lung cancer risk, we meta-analyzed results from 15 European ancestry samples in the International Lung Cancer Consortium (ILCCO) and Transdisciplinary Research in Cancer of the Lung (TRICL). We explored two linked aims: 1) Do both *CHRNA5* genetic risk and smoking cessation independently affect the risk of lung cancer? 2) Does the effect of smoking cessation on the risk of lung cancer vary with variation in *CHRNA5*? i.e., does the effect of smoking cessation on lung cancer risk and onset differ as a function of *CHRNA5* rs16969968 genotype?

2. Methods

2.1. Samples

This is a collaborative meta-analysis based on the International Lung Cancer Consortium (ILCCO) and Transdisciplinary Research in Cancer of the Lung (TRICL) which were established with the aim of sharing

comparable data from ongoing case-control and cohort studies of lung cancer. (Hung et al., 2008b; Timofeeva et al., 2012). To examine *CHRNA5*, smoking cessation, and lung cancer, we invited all ILCCO and TRICL studies of individuals of European Ancestry, of which, 15 (out of 27 invited studies) participated in the collaborative meta-analysis and pooled analysis with shared individual-level data. Results from 15 case control studies of lung cancer ($N = 12{,}690$ unrelated smokers of European ancestry) contributed to the meta-analyses. Informed consent was obtained from participants, and all studies received approval from the appropriate institutional review boards. To be included in analyses, each subject was required to be an ever-smoker (>100 cigarettes in his or her lifetime). Tables S1, S2, and Text S1 provide additional details for each study.

With the aim of studying smoking cessation, *CHRNA5*, and lung cancer risk, we invited all ILCCO and TRICL studies of individuals of European Ancestry and 15 (out of 27 invited studies) participated in the collaborative meta-analysis and pooled analysis with shared individual-level data.

2.2. Outcomes

The primary outcomes of the analyses were: 1) case vs. control status for lung cancer in the combined sample of cases and controls, and 2) time (in years) from birth to lung cancer diagnosis among lung cancer cases.

2.3. Variants for Analyses

Because of its biological significance, we targeted the *CHRNA5* variant rs16969968 for association testing. The variant rs16969968 was available in all datasets except the Germany Study for which we used a proxy variant (rs1051730, $r^2 = 0.99$ estimated based on 1000 Genomes for the EUR samples) for analyses because the imputed genotype was not available (Altshuler et al., 2010; Durbin et al., 2010). In addition, we conducted the meta-analyses with and without the Germany Study and reached similar results.

2.4. Statistical Analyses and Meta-analyses

In each dataset, we used logistic regression and Cox regression models to evaluate the association between rs16969968 and the two primary outcomes: i.e., lung cancer case vs. control, and age of lung cancer diagnosis among the cases, respectively. Age as a continuous variable and sex were included as covariates when appropriate. Additional covariates included smoking quantity and pack years. Smoking quantity when subjects smoked regularly was assessed with cigarettes smoked per day (CPD), defined as a 4-level ordered trait (CPD ≤ 10; 11 ≤ CPD ≤ 20; 21 ≤ CPD ≤ 30; CPD ≥ 31, coded as 0, 1, 2, 3, respectively). The variable "pack years" was defined by the product of smoking duration and CPD, and was modeled via quartiles.

Genotypes were coded additively as the number of minor alleles (A), where the reference allele was defined as the major allele (G) in the European ancestry population (Sherry et al., 2001). Consistency of allelic coding was confirmed by comparing allele labels and frequencies across datasets (Table S2). Analyses were performed at Washington University, as individual-level data were available for both meta-analyses and pooled analyses. Individual SNP analyses were performed using SAS (SAS Institute, Cary, NC).

The R package, rmeta, was used to generate meta-analysis plots (Lumley, 2012). We reported results from random effects models for all meta-analyses. Heterogeneity across studies was assessed with the Cochran Q test and resulting *P* values were reported for all analyses. There was evidence of heterogeneity across datasets for the analyses of the effect of smoking cessation on lung cancer, but not for the effect of genotypes on lung cancer. This heterogeneity may be due to the

varying study designs and ascertainment strategies. Heterogeneity results for all meta-analysis are listed in Table S3.

3. Results

We examined individuals who ever smoked at least 100 cigarettes (5833 current and 6857 former smokers) from 15 case-control studies of lung cancer. *CHRNA5* rs16969968 is known to predict risk for lung cancer and delayed smoking cessation based on previous reports of overlapping samples including these studies (Hung et al., 2008a; Chen et al., 2015a). We hypothesized that smoking cessation is associated with a decreased likelihood of lung cancer and delayed onset of lung cancer in this sample. Further, we investigated whether the association of smoking cessation and lung cancer risk varied with rs16969968 genotype risk levels.

3.1. As Previously Reported, CHRNA5 rs16969968 Predicts Lung Cancer and Earlier Diagnosis of Lung Cancer

First, we confirmed a consistent association as previously reported (Hung et al., 2008a; Chen et al., 2015a) between *CHRNA5* rs16969968 and 1) increased likelihood of lung cancer when comparing cases and controls of lung cancer (meta-analysis OR = 1.29, 95%CI = 1.21–1.38, $p = 3.5 * 10^{-13}$, $P_{Heterogeneity} = 0.196$, see Fig. S1; this genetic association does not vary significantly with smoking quantity), and 2) earlier diagnosis of lung cancer among cases of lung cancer (meta-analysis HR = 1.07, 95%CI = 1.03–1.11, $p = 1.1 * 10^{-4}$, $P_{Heterogeneity} = 0.561$, see Fig. S2).

3.2. Quitting Smoking Decreases Risk Of Lung Cancer - Effect does not Vary With rs16969968 Genotypes

In these case control studies, current smoking status was ascertained simultaneously with case or control status for lung cancer. Ever-smokers who reported having quit smoking >1 year prior to the cancer diagnosis were defined as former smokers; ever smokers who reported active smoking were defined as current smokers. The effect of smoking cessation was defined as the comparison of former vs. current smokers. Among ever smokers, smoking cessation was associated with a lower likelihood of lung cancer (OR = 0.48, 95%CI = 0.30–0.75, $p = 0.0015$, $P_{Heterogeneity} < 10^{-16}$). Both smoking cessation and rs16969968 were significantly associated with lung cancer risk (Fig. 1, Fig. S1).
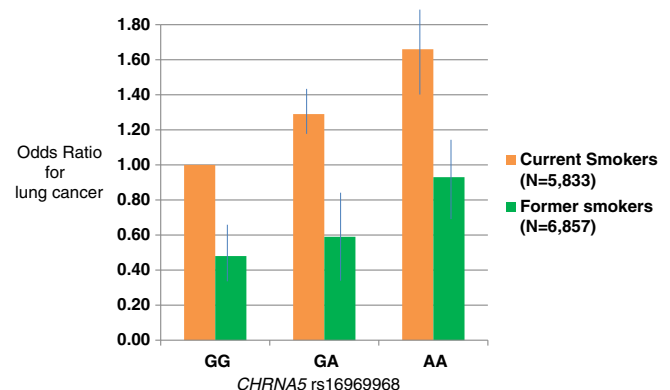
Quit: individuals who report having quit smoking when ascertained as cases of lung cancer or controls.
N=12,690 (6988 cases of lung cancer and 5702 controls)
All subjects have smoked at least 100 cigarettes (5833 active smokers, 6857 smokers who have quit)
Adjusted for age, sex and rs16969968.

Fig. 1. *CHRNA5* rs16969968 predicts risk of lung cancer. Smoking cessation decreases probability of lung cancer regardless of *CHRNA5* rs16969968 genotype. Quit: individuals who report having quit smoking when ascertained as cases of lung cancer or controls. N = 12,690 (6988 cases of lung cancer and 5702 controls). All participants have smoked at least 100 cigarettes (5833 active smokers, 6857 smokers who have quit). Adjusted for age, sex and rs16969968 genotype.

The effect of smoking cessation on lung cancer risk remained after additionally adjusting for smoking quantity or pack years, and smoking cessation remained a predictor of lower risk of lung cancer diagnosis (OR = 0.46, 95%CI = 0.29–0.74, p = 0.0013 adjusted for smoking quantity) When adjusting for pack years, the effect size point estimate was similar (OR = 0.47, 95%CI = 0.14–1.52, p = 0.21) although it did not approach statistical significance as data on pack years were available for only a subset of the studies).

Further, we examined whether this association varied in individuals with different rs16969968 genotypes and found that smoking cessation was significantly associated with lower lung cancer risks for all genotypes (GG: OR = 0.48, 95%CI = 0.30–0.77, p = 0.0024, $P_{Heterogeneity} < 10^{-16}$; GA: OR = 0.46, 95%CI = 0.28–0.76, p = 0.0025, $P_{Heterogeneity} < 10^{-16}$; AA: OR = 0.56, 95%CI = 0.34–0.91, p = 0.019, $P_{Heterogeneity} = 3.36 * 10^{-6}$) (Fig. S1). Genotype did not modify the effect of smoking cessation on decreased risk of lung cancer among ever smokers, and there was no interaction (OR = 1.01, 95%CI = 0.86–1.18, p = 0.93, $P_{Heterogeneity}$ = 0.139, Fig. S1). The finding remained robust in the sensitivity analyses when all possible sets of studies (up to $N = 10$) were excluded from the meta-analysis.

In addition, we obtained similar results in pooled analysis using available individual-level data adjusted for age, sex, rs16969968 genotype, and study, and found that smoking cessation was associated with decreased risk of lung cancer ($N = 12,690$, OR = 0.78, 95%CI = 0.75–0.91, $P = 5.65 * 10^{-31}$), and that genotype did not interact with smoking cessation in predicting lung cancer likelihood (i.e., the beneficial effect of smoking cessation did not vary with rs16969968 genotype (OR = 0.99, 95%CI = 0.94–1.05, P = 0.887)).

3.3. Quitting Smoking Delays Lung Cancer and This Effect Does not Vary With CHRNA5 rs16969968 Genotype

Among ever smokers who were diagnosed with lung cancer, smoking cessation was associated with delayed age of diagnosis, adjusted for sex and rs16969968 (HR = 0.68, 95%CI = 0.61–0.77, $p = 4.9 * 10^{-10}$, $P_{Heterogeneity} = 2.87 * 10^{-8}$, see Fig. S2). Both smoking cessation and rs16969968 were independent predictors of age of lung cancer diagnosis. The median age at lung cancer diagnosis was 59 years for current smokers, which was a 7-year earlier onset compared to former smokers, who had a median age of diagnosis of 66 years (Fig. 2).

The effect of smoking cessation on age of lung cancer diagnosis remained after additionally adjusting for cigarettes smoked per day, or pack years, and smoking cessation remained a significant predictor of a delayed age of lung cancer diagnosis (HR = 0.68, 95%CI = 0.60–0.76, $p = 1.4 * 10^{-10}$, adjusted for cigarettes smoked per day; HR = 0.62, 95%CI = 0.46–0.85, p = 0.0032, adjusted for pack years for studies with the information).

We examined whether this association varies by genotype and found that smoking cessation was associated with delayed age of lung cancer diagnosis for all genotypes (GG: HR = 0.70, 95%CI = 0.61–0.82, $p = 5.2 * 10^{-6}$, $P_{Heterogeneity}$ = 0.002; GA: HR = 0.63, 95%CI = 0.55–0.72, $p = 7.8 * 10^{-12}$, $P_{Heterogeneity}$ = 0.001; AA: HR = 0.66, 95%CI = 0.57–0.76, $p = 1.3 * 10^{-8}$, $P_{Heterogeneity}$ = 0.344) (Fig. S2). Even though CHRNA5 rs16969968 risk genotypes were associated with delayed quitting and earlier age of lung cancer diagnosis as previously reported (Chen et al., 2015a), genotypes did not modify the effect of smoking cessation on the delayed age of lung cancer diagnosis and
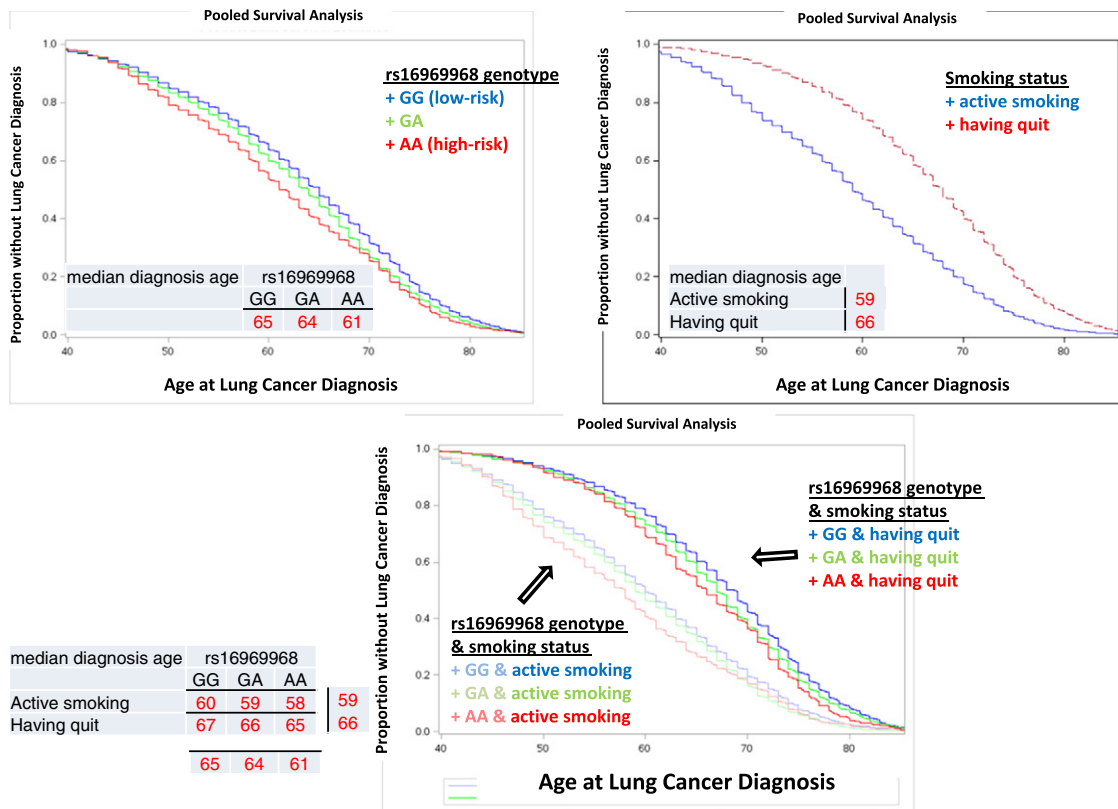
Quit: individuals who report having quit smoking when ascertained as cases of lung cancer.
N=6,988 cases of lung cancer who are ever smokers. (3,471 current smokers, 3,517 former smokers who have quit.)
Adjusted for sex, study, and rs16969968.

Fig. 2. CHRNA5 rs16969968 predicts earlier lung cancer. Smoking cessation delays lung cancer, regardless of CHRNA5 rs16969968 genotype. Quit: individuals who report having quit smoking when ascertained as cases of lung cancer. N = 6,988 cases of lung cancer who are ever smokers. (3,471 current smokers, 3,517 former Smokers. Adjusted for sex, study, and rs16969968 genotype.

there was no interaction (HR = 0.98, 95%CI = 0.91–1.05, p = 0.57, $P_{Heterogeneity}$ = 0.542, Fig. S2). The finding remained robust in the sensitivity analyses when all possible sets (up to N = 10) of studies were excluded from the meta-analysis.

The median ages of lung cancer diagnosis for current and former smokers of different genotypes are in Fig. 3. Smokers with the high-risk AA genotypes, compared with those with the low-risk GG genotypes, have earlier age of lung cancer diagnosis, but this risk is greatly reduced by successful smoking cessation. Individuals with the high-risk genotype who have quit smoking have a mean age of lung cancer diagnosis of 65 vs. a mean age of 58 for those who continue to smoke. Individuals with the low-risk genotype who have quit smoking have a mean age of lung cancer diagnosis of 67 vs. a mean age of 60 for who continue smoking.

In addition, we reached similar conclusions in pooled analysis using available individual-level data adjusted for sex, rs16969968, and study; we found that smoking cessation was associated with delayed lung cancer diagnosis (N = 6988,HR = 0.62, 95%CI = 0.59–0.65, P = $1.26 * 10^{-77}$), and this association did not vary with rs16969968 genotype (HR = 1.00, 95%CI = 0.94–1·07, P = 0.958).

4. Discussion

Genetic information is increasingly used by both patients and providers to determine health risks. Multiple meta-analyses have shown that the nicotinic receptor variant, CHRNA5 rs16969968, predicts heavy smoking (Liu et al., 2010; Saccone et al., 2010; TAG, 2010; Thorgeirsson et al., 2010), delayed quitting (TAG, 2010; Baker et al., 2009; Bergen et al., 2013; Chen et al., 2012; Munafo et al., 2011; Sarginson et al., 2011; Chen et al., 2015a), and earlier onset and higher probability of lung cancer (Amos et al., 2008; Thorgeirsson et al., 2008; Chen et al., 2015a; Hung et al., 2008a; Lips et al., 2010; Spitz et al., 2008). It is important to determine how malleable these genetic risks are to preventive or treatment efforts. For example, will individuals with high-risk genetic variants for lung cancer benefit from known effective preventive measures such as smoking cessation? In essence, do genetically high-risk and low-risk individuals experience different levels of benefit from cessation? Evidence related to this issue could both further elucidate the influences on cancer, and also serve as a basis for genetically informed preventive intervention. This is a large meta-analysis to examine the benefit of smoking cessation for lung cancer in individuals with different CHRNA5 rs16969968 genotypes. Obviously, these results cannot be extrapolated to the effects of other genotypes or other preventive actions (i.e., different from smoking cessation).

We found that smoking cessation is beneficial in reducing and delaying lung cancer and this beneficial effect does not vary with CHRNA5 rs16969968, which is a genetic marker for nicotine dependence (Liu et al., 2010; Saccone et al., 2010; TAG, 2010; Thorgeirsson et al., 2010), lung cancer (Amos et al., 2008; Thorgeirsson et al., 2008), COPD (Pillai et al., 2009), and delayed quitting (Chen et al., 2015a). Even though the CHRNA5 variant, rs16969968, predicts a 4-year earlier diagnosis of lung cancer among smokers with the high-risk genotypes vs. the low-risk genotypes (Chen et al., 2015a), quitting smoking is a highly effective preventive measure, cutting lung cancer risk approximately in half for individuals with all genotypes. Furthermore, among those who developed lung cancer, quitting smoking delayed diagnosis by 7 years (from 59 years of age for active smokers to 66 years of age, Fig. 2), with the delay not differing by genotype.

By simultaneously studying both genetic risk and preventive action (cessation), this study provides an informative perspective on comparative risk. For instance, it is clear that the effect of smoking cessation is larger than the effect of CHRNA5 rs16969968 genotypes on the risk of lung cancer. Smokers with the high-risk genotype, compared with those with the low-risk genotype, do have an increased risk for lung cancer, but this risk can be greatly reduced if they successfully quit smoking. In fact, smokers with the high-risk genotype who quit have lower risk than those with the low-risk genotypes who continue smoking (i.e., individuals with the high-risk genotype who have quit smoking have a mean age of lung cancer diagnosis of 65 vs. a mean age of 60 for smokers with a low-risk genotype who continue smoking).

The mechanisms through which CHRNA5 increases the risk for, and accelerates the onset age of, lung cancer likely involve multiple direct or indirect pathways (mediated through cigarette smoking (Liu et al., 2010; Saccone et al., 2010; TAG, 2010; Thorgeirsson et al., 2010), deeper inhalation of cigarettes leading to higher carcinogen exposure (Bloom et al., 2014; Le Marchand et al., 2008), and a delay in smoking cessation (Chen et al., 2015a). There has been inconsistent evidence on whether

Genotype	Quit Smoking Age	Smoking Status	Lung Cancer Diagnosis Age
		Continue smoking →	**Median lung cancer diagnosis age = 58**
	High Risk AA **Median quit age*= 56**	*Quit smoking* →	**Median lung cancer diagnosis age = 65**
***CHRNA5* rs16969968 Genotype**			
	Low Risk GG **Median quit age*= 52**	*Continue smoking* →	**Median lung cancer diagnosis age = 60**
		Quit smoking →	**Median lung cancer diagnosis age = 67**
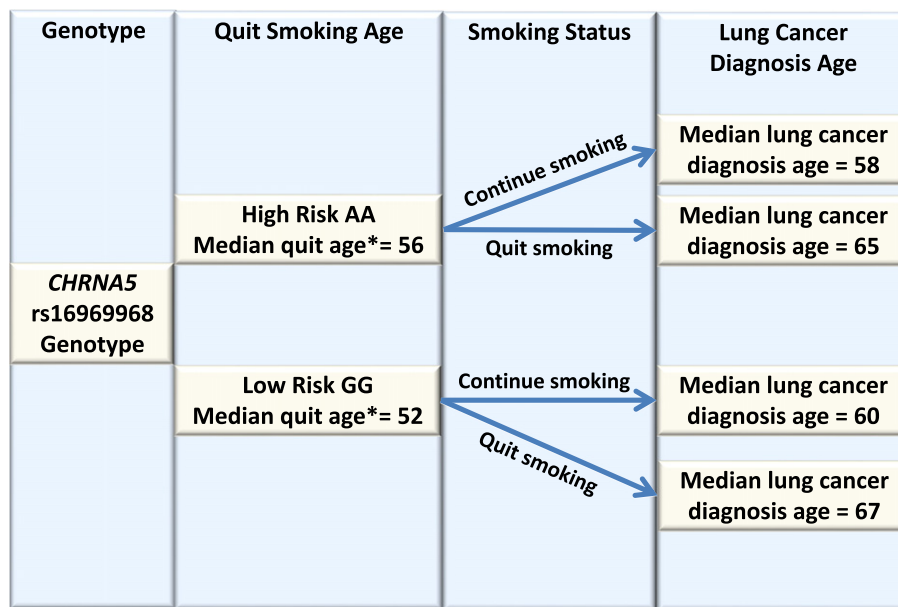
* These associations are based on existing meta-analysis evidence (Chen et al, 2015).

Fig. 3. Summary diagram of CHRNA5 rs16969968 genotypes, smoking cessation, and lung cancer risks. *These associations are based on existing evidence (Chen et al., 2015a, b).

or not the genetic association of *CHRNA5* and lung cancer is largely mediated through smoking) (Amos et al., 2008; Thorgeirsson et al., 2008; Ji et al., 2015; Tseng et al., 2014; Wang et al., 2012; Hung et al., 2008a).

Some study suggests that *CHRNA5* confers risk of lung and cardiovascular diseases largely through an effect on behavior (Thorgeirsson et al., 2008), while other studies suggest that an increased risk with *CHRNA5* and lung cancer in non-smokers would indicate that the disease mechanism with lung cancer is unlikely to be explained by an association with tobacco addiction. (Ji et al., 2015; Hung et al., 2008a) Most studies suggest that *CHRNA5* had both a direct effect on overall lung cancer risk and an indirect effect through smoking. (Amos et al., 2008; Tseng et al., 2014; Wang et al., 2012) Our finding that smoking cessation decreases the elevated genetic risk for lung cancer supports the view that smoking at least partially mediates the effect of this genetic risk. This evidence suggests that the increased risk associated with high-risk genotypes for lung cancer is at least partially attributed to the smoker's inability to quit smoking relatively early (Chen et al., 2015a). For those who succeed in quitting, the elevated genetic risk for lung cancer conferred by rs16969968 (AA genotypes: 30% more likely to get lung cancer and 4 year earlier diagnosis) can be reduced drastically, resulting in halving the risk for lung cancer and delaying the age of diagnosis by 7 years.

These results should be interpreted in the context of multiple limitations. First, smoking cessation in the samples was self-reported and not assessed using biochemical confirmation. However, research shows that self-report is a valid indicator of current smoking, especially when there are no strong incentives to deceive (Subcommittee on Biochemical Verification SRNT, 2002). Second, we compared current smokers and former smokers based on the self-reported smoking status when cases and controls for lung cancer were recruited. Caution is needed for results based on the cross-sectional nature of the data. For a small portion of smokers (5.7%) who receive the lung cancer diagnosis and quit smoking in the same year, it is unknown whether they quit before the manifestation of disease or their diagnosis. We have repeated the same analyses by including, excluding, or reclassifying their smoking status and reached similar results. Third, this work analyzed only one genetic variant, and it is clear that multiple genes or variants contribute to lung cancer and smoking behaviors (Uhl et al., 2012). Fourth, we found no significant interaction between smoking cessation and rs16969968 genotype on lung cancer risk, despite the non-linear trend in point estimates of the effect of quitting across genotypes on lung cancer risk or age at diagnosis. With our sample size of 12,690 smokers and the allele frequency of rs16969968, we have sufficient power (0·8) to detect an interaction effect size of 1.12. Clearly a larger sample size would be needed to detect a smaller interaction. Fifth, this work analyzed the association between *CHRNA5* and lung cancer, and it is possible that this association is mediated by other factors such as chronic obstructive pulmonary disease (COPD) (Young et al., 2008). Finally, this study included only subjects of European ancestry.

Lung cancer is the most common cancer in US and worldwide (Centers for Disease Control and Prevention and National Center for Health Statistics, 2013; American Cancer Society, 2014), and the survival rate is low (Howlader et al., 2013). Most cases (90%) of lung cancer are attributable to smoking (General, 2014). We demonstrate that quitting smoking is highly beneficial in reducing lung cancer risks for smokers regardless of their *CHRNA5* rs16969968 genetic risk status. Evidence suggest that relative to individuals with *CHRNA5* rs16969968 low-risk genotypes, those with high-risk genotypes are likely to smoke greater quantities, inhale more deeply, have more difficulty quitting, and have higher risk for lung cancer (Bierut et al., 2007; Saccone et al., 2010; TAG, 2010; Thorgeirsson et al., 2010; Thorgeirsson et al., 2008; Chen et al., 2012; Chen et al., 2015a; Hung et al., 2008a; Bloom et al., 2014). We now extend this evidence to show that quitting smoking produces essentially equivalent benefit regardless of genotype for this genetic risk factor. These results have potential value for preventive counseling with smokers; smokers with high-risk *CHRNA5* genotypes can be informed that, on average, smokers with their genetic risk can largely eliminate their elevated genetic risk for lung cancer by quitting smoking. They can cut their risk of lung cancer in half and delay its onset by 7 years, if they develop it. These results underscore the potential value of smoking cessation for all smokers, they elucidate the causal path from *CHRNA5* risk to lung cancer diagnosis, and they have potential value for framing preventive interventions for those who smoke.

Conflict of interest

LJ Bierut is listed as an inventor on Issued U.S. Patent 8,080,371,"Markers for Addiction" covering the use of certain SNPs in determining the diagnosis, prognosis, and treatment of addiction.

All other authors have no conflict of interest to disclose**.**

Contribution to Literature Search

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Contribution to Figures

Li-Shiun Chen, Amy Horton.

Contribution to Study Design

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Appendix A. Supplementary data

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