Eur J Vasc Endovasc Surg (2014) 48, 703–710

Association Between Salivary Cotinine and Cardiovascular Biomarkers Among Nonsmokers and Current Smokers: Cross-sectional Study of 10,081 Participants

L. Lu [a], **D.F. Mackay** [a], **D.E. Newby** [b], **J.P. Pell** [a,*]

[a] Institute of Health and Wellbeing, University of Glasgow, Glasgow G12 8RZ, UK
[b] Centre for Cardiovascular Science, University of Edinburgh, Edinburgh EH16 4TJ, UK

WHAT THIS PAPER ADDS

This study adds to growing evidence that exposure to secondhand smoke is not only a risk factor for cardiovascular disease, but also carries a disproportionately higher risk than active smoking for a given level of smoke exposure. The main novelty of this study was the direct comparison, using several biomarkers, between nonsmokers with high levels of secondhand exposure, and light and moderate active smokers.

Objective: Both active smoking and exposure to secondhand smoke (SHS) are associated with cardiovascular disease, but sidestream smoke contains higher levels of small particles and toxic gases than mainstream smoke. The relationship between the concentration of cotinine and a number of cardiovascular biomarkers among nonsmokers and active smokers was examined.

Methods: A cross-sectional study using the Scottish Health Surveys conducted between 1998 and 2010 was undertaken. Inclusion was restricted to participants aged ≥16 years who had provided saliva and blood samples. Uni- and multivariate regression models were used to examine the relationships between the concentration of cotinine and C-reactive protein (CRP), high-density lipoprotein (HDL) cholesterol, and fibrinogen concentrations, as well as total:HDL cholesterol ratios.

Results: Of the 10,018 eligible participants, 7,345 (73.3%) were confirmed to be nonsmokers (cotinine <15.0 ng/mL) and 2,673 (26.7%) were confirmed to be current smokers (cotinine ≥15.0 ng/mL). CRP and total:HDL cholesterol increased, and HDL cholesterol decreased, with increasing cotinine concentration across nonsmokers and smokers (all $p < .001$). However, there were step changes at the interface, whereby nonsmokers with a high exposure to SHS had lower concentrations of cotinine than light active smokers but comparable concentrations of CRP ($p = .709$), HDL cholesterol ($p = .931$), and total:HDL cholesterol ($p = .405$). Fibrinogen concentrations were significantly raised in moderate and heavy active smokers only (both $p < .001$).

Conclusion: Exposure to SHS is associated with disproportionately higher biomarkers of cardiovascular risk compared with active smoking. Protection from exposure to SHS should be a public health priority.

Article history: Received 24 June 2014, Accepted 22 September 2014, Available online 23 October 2014

Keywords: Cardiovascular diseases, Cholesterol, C-reactive protein, Fibrinogen, Secondhand smoke, Tobacco smoke exposure

INTRODUCTION

Globally, cardiovascular disease (CVD) is the leading cause of death and is projected to cause 23 million deaths per annum by 2030.[1] The global prevalence of smoking is increasing, especially in large, developing countries such as China. A 2013 World Health Organization report has indicated that only 16% of the world's population is covered by comprehensive smoke-free legislations.[2] Active smoking is an established risk factor for coronary heart disease (CHD), stroke, and peripheral arterial disease (PAD).[3–5] There is growing evidence that exposure to secondhand smoke (SHS) is also a risk factor. Two meta-analyses reported relative risks of 1.25 [95% confidence interval (CI) 1.17–1.32] and 1.25 (95% CI 1.12–1.38) for CHD and stroke, respectively.[6,7] To date, four cross-sectional studies have examined the association with PAD,[8–11] with three reporting significant associations.[8,10,11] We previously demonstrated that nonsmokers with cotinine concentrations ≥2.7 ng/mL were significantly more likely to have intermittent claudication than those with cotinine concentrations <0.7 ng/mL (adjusted odds ratio 1.76, 95% CI 1.04–3.00).[11]

Active smoking is associated with higher concentrations of cardiovascular biomarkers, including C-reactive protein

* Corresponding author.
E-mail address: jill.pell@glasgow.ac.uk (J.P. Pell).

1078-5884/$ – see front matter

http://dx.doi.org/10.1016/j.ejvs.2014.09.008

(CRP),[12] fibrinogen,[13] and low-density lipoprotein (LDL) cholesterol.[14] SHS may contain both sidestream smoke, from burning cigarette tips, as well as exhaled mainstream smoke. Sidestream smoke contains higher concentrations of small respirable particles (<2.5 μm) and toxic gases than mainstream smoke inhaled by active smokers.[15–18] Brief exposure to SHS produces rapid changes in inflammatory markers,[19,20] resulting in concentrations comparable with active smokers.[21–23] Therefore, the sidestream smoke inhaled by nonsmokers exposed to SHS may convey a disproportionately higher risk of CVD. In the British Regional Heart Study, the risk of CHD events over 20 years of follow-up was comparable in nonsmokers exposed to high levels of SHS [adjusted hazard ratio (HR) 1.57, 95% CI 1.08–2.28] and light active smokers (adjusted HR 1.66, 95% CI 1.04–2.68) in spite of cotinine concentrations being nearly 30-fold higher in the latter group (mean 4.9 vs. 138 ng/mL).[24]

Scottish Health Surveys were used to explore the association between level of secondhand and active smoke exposure, measured by salivary cotinine concentration and a number of preclinical cardiovascular biomarkers [CRP, high-density lipoprotein (HDL) cholesterol, total:HDL cholesterol and fibrinogen.

METHODS

Data source

The Scottish Health Surveys are ongoing, repeated, cross-sectional studies used to monitor the health and health-related risk factors of the general population living in private households across Scotland (http://www.scotland.gov.uk/Topics/Statistics/Browse/Health/Scottish-health-survey). The surveys were undertaken in 1995, 1998, and 2003, and then annually from 2008 using a multistage, stratified sampling frame. Each survey recruited different households. The trained staff conducted face-to-face interviews and obtained measurements, including height and weight. All consenting individuals aged ≥16 years were visited by a nurse and invited to provide a salivary sample, for cotinine assay, and blood samples, for assays including lipids, CRP, and fibrinogen. Cholesterol concentrations were measured using cholesterol oxidase assays on an Olympus 640 analyser (Olympus, Canter Valley, PA, USA) prior to 2010 and, subsequently, a Roche Modular P analyser (Roche, Basel, Switzerland). CRP concentrations were determined using the N Latex CRP mono-immunoassay on the Behring Nephelometer II analyser (Behring, Milan, Italy). Fibrinogen concentrations were measured using the Organon Teknika MDA 180 analyser (Organon, Oss, the Netherlands). Cotinine was assayed using a Hewlett Packard hp5890 gas chromatograph (Hewlett Packard, Palo Alto, CA, USA).

Inclusion criteria and definitions

In this study, data from the 1998, 2003, 2008, 2009, and 2010 surveys were amalgamated as they collected consistent information on cotinine, CRP, fibrinogen, and lipid concentrations. Inclusion was restricted to participants aged ≥16 years who provided saliva and serum samples, and were not taking nicotine replacement products. Nonsmokers were defined as self-reported never or ex-smokers who had a salivary cotinine concentration <15.0 ng/mL. Current smokers were defined as self-reported current smokers who had a cotinine concentration ≥15.0 ng/mL.[25] Among nonsmokers, SHS exposure was classified into low (cotinine <0.7 ng/mL), moderate (cotinine 0.7–2.6 ng/mL), and high (cotinine ≥2.7 ng/mL). Current smokers were categorized into light (cotinine 15.0–100.0 ng/mL), moderate (cotinine 100.1–300.0 ng/mL), and heavy (cotinine >300.0 ng/mL). Body mass index (BMI) was categorized into normal weight (<25 kg/m^2), overweight (BMI 25–30 kg/m^2), and obese (≥30 kg/m^2).[26] Alcohol consumption was based on self-report and classified as never drinker, ex-drinker, low-risk drinker (men <28 units/week; women <21 units/week), increasing risk drinker (men <50 units/week; women <35 units/week), and high-risk drinker (men ≥50 units/week; women ≥35 units/week). Physically active was defined as self-report of any kind of physical activity for at least 3 hours per week.

Statistical analyses

The characteristics of nonsmokers and current smokers were summarized using frequencies and percentages for categorical data, medians, and interquartile ranges for nonparametric continuous data (CRP), and mean and SD for parametric continuous data (fibrinogen and lipids). The differences between exposure groups were assessed using chi-square tests for categorical variables and analysis of variance for continuous variables. Nonsmokers and current smokers were included in the same model in order to examine the effect of increasing cotinine concentration across the whole spectrum from nonsmokers protected from SHS exposure to heavy active smokers. Univariate and multivariate median regression models were used to examine the association between cotinine concentration and serum CRP using nonsmokers with low SHS exposure (cotinine <0.7 ng/mL) as the referent category. General linear regression models were used, in the same way, to examine the associations between cotinine concentration and fibrinogen and lipid concentrations. Three models were developed for each assay: unadjusted; partially adjusted (age and sex); and fully adjusted (age, sex, social class, BMI, alcohol consumption, and physical activity). Interactions with covariates were tested. Statistical significance was defined as a two-sided p-value <.001 for main effects and <.05 for interactions. All statistical analyses were undertaken using Stata 12.0 (Stata Corporation, College Station, TX, USA).

RESULTS

Of the 51,802 participants in the Scottish Health Surveys, 38,436 were aged ≥16 years. Of these, 180 were excluded because they were taking nicotine replacement therapy (nicotine chewing gum, patch, or nasal spray). Of the remaining 38,256, 10,512 provided saliva and blood