

## INVESTIGATIVE REPORT

# Effect of Tobacco Smoking and Alcohol Consumption on the Prevalence of Nickel Sensitization and Contact Sensitization

Jacob P. THYSSSEN<sup>1</sup>, Jeanne D. JOHANSEN<sup>1</sup>, Torkil MENNÉ<sup>2</sup>, Niels H. NIELSEN<sup>3</sup> and Allan LINNEBERG<sup>4</sup>

<sup>1</sup>Department of Dermato-Allergology, National Allergy Research Centre, <sup>2</sup>Department of Dermato-Allergology, Gentofte University Hospital, <sup>3</sup>Dermatology Clinic, Bagsværd, Denmark, and <sup>4</sup>Research Centre for Prevention and Health, Glostrup Hospital, University of Copenhagen, Copenhagen, Denmark

**There is evidence that stimulants such as alcohol and tobacco have an effect on the immune system, but little is known about how these lifestyle factors affect the prevalence of contact sensitization. This study investigated whether smoking and alcohol consumption were associated with contact sensitization and nickel sensitization. A random sample of adults ( $n=3460$ ) from the general population of Copenhagen was invited to participate in a general health examination including patch-testing. Alcohol consumption was not associated with nickel sensitization, whereas a significant trend ( $p<0.05$ ) was identified between smoking status and nickel sensitization in an adjusted model; i.e. nickel sensitization was higher among both previous smokers (odds ratio (OR)=1.19; confidence interval (CI)=0.81–1.76), current light smokers (OR=1.50; CI=0.94–2.37) and current heavy smokers (OR=1.56; CI=0.87–2.80) compared with never smokers. This study confirmed that smoking is associated with nickel sensitization, but rejected an association with alcohol consumption. *Key words: alcohol drinking; contact sensitization; general population; public health; tobacco smoking.***

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Jacob Pontoppidan Thyssen, National Allergy Research Centre, Department of Dermato-Allergology, Gentofte Hospital, University of Copenhagen, Niels Andersens Vej 65, DK-2900 Hellerup, Denmark. E-mail: jacpth01@geh.regionh.dk

Contact sensitization affects 10–20% of the general population (1, 2). It is caused mainly by exposure to nickel, fragrances, and preservatives, whereas genetic susceptibility seems to be of limited importance (3, 4). There is substantial evidence that alcohol and tobacco have an effect on the immune system (5–9), but little is known about how these lifestyle factors affect the prevalence of contact sensitization. Recently, a prospective Danish population-based study revealed contradictory results regarding a possible association between alcohol consumption and contact sensitization (10). Furthermore, three general population studies have examined whether tobacco smoking is associated with contact sensitization

(11–13): among 1056 adult Danes, a significant dose-response relationship was identified between smoking and contact sensitization (11). Furthermore, current smoking was significantly associated with contact sensitization among 690 Norwegian adult women (12), whereas no association was identified among 520 young Swedish young men doing military service (13). Thus, it remains to be determined convincingly whether alcohol consumption and tobacco smoking affect the prevalence and risk of contact sensitization.

The present study aimed to re-investigate a possible association between these lifestyle factors and contact sensitization. A random sample of adults from the general population in Copenhagen, Denmark, was invited to participate in a general health examination including patch-testing. The study focused on nickel sensitization as it is by far the most prevalent contact sensitization in the general population (1). Furthermore, data from previous cross-sectional studies have suggested that the association between tobacco smoking and nickel sensitization was slightly stronger than the association between smoking and contact sensitization to at least one allergen (11). The current study is of relevance, as smoking and drinking is prevalent in many countries, and as an association may have clinical implications (e.g. the interpretation of patch-test reactions in smokers).

## MATERIALS AND METHODS

### *Study population*

A cross-sectional study was performed in the general population in Copenhagen. A random sample of 7931 subjects aged 18–69 years was obtained from the Danish Central Personal Register. All were Danish adults with Danish citizenship and born in Denmark. A total of 3471 (43.7%) subjects participated in a general health examination and 3460 were patch-tested. The participation rate was higher among older age-groups than among younger age-groups in both genders (14). The study was approved by the ethics committee of Copenhagen County (KA-20060011). Written informed consent form was obtained from all participants prior to the beginning of the study.

### *Patch-tests*

Patch-testing was performed by using panel 1 and 2 from the standardized ready to apply TRUE-test® (Mekos Laboratories, Hillerød, Denmark). Directions to apply the patch-test panels to

the upper back 2 days before examination were posted together with the patch-tests. All testing was performed between June 2006 and May 2008. On the day of examination, they were read and photographed 1–1.5 h after removal by trained healthcare personnel (supervised by JPT and AL). The photographs were reviewed by TM, NHN, AL and JPT to ensure that the International Contact Dermatitis Research Group (ICRDG) criteria were used consistently over time. Contact sensitization was defined as a positive (at least grade 1+ according to ICRDG criteria) patch-test to at least one allergen or mixes of haptens. It has been estimated that approximately 18–29% of positive patch-test reactions to nickel are missed when patch-test readings are performed only on day 2 and not on day 4 (15). If the patch was found to have no skin contact upon patch-test removal, or if the subject had removed the patch prior to testing as a result of known contact sensitization, it was regarded as missing data.

#### Measurement of immunoglobulin E antibodies

Venous blood was taken on the day of examination and was left to coagulate for 2 h. The serum was then separated by centrifugation at 3000 rpm for 10 min and frozen immediately afterwards. The serum samples were analysed for immunoglobulin E (IgE) specific to birch, grass (timothy), cat, and mite (*Dermatophagoides pteronyssinus*) with the ADVIA Centaur

IgE antibody assay system (Bayer Corporation, Leverkusen, Germany) (16). Analysis for IgE antibodies was judged to be positive if the measurement was in excess of 0.35 kU/l. Specific IgE positivity was defined as a positive test to at least one of the four allergens tested.

#### Questionnaire

Table I lists the questions used for the present study. Participants were asked about smoking and drinking habits as well as about vocational training, social status and ear-piercing status. Occasional smokers (3.3%) were excluded from the analyses. The amount of tobacco in grams smoked per day among current smokers were calculated for cigarettes, cheroots, cigars and pipe tobacco by equating one cigarette or 1 g of pipe tobacco to 1 g tobacco, one cheroot to 3 g tobacco and one cigar to 5 g tobacco. The information was used to define smoking status (as “never smokers”, “previous smokers”, “current light smokers:  $\leq 15$  g/day”, or “current heavy smokers:  $> 15$  g/day”). It was assumed that one normal beer, one glass of wine, and one serving of spirits equalled one drink (each containing 12 g ethanol/15 ml) whereas one strong beer was assumed to equal 1.5 drinks (each contained 18 g ethanol/15 ml). The total weekly consumption was then calculated by adding the number of drinks of beer, wine, and spirits. The total alcohol consumption was categorized as 0, 1–7, 8–14,  $\geq 15$  drinks per week for the

Table I. Questions used in the questionnaire

| Question category   | Group questioned                   | Question                                                                                     | List of answers                                                                                                                                                                                                                                                                                                                                     |
|---------------------|------------------------------------|----------------------------------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Smoking             | All participants                   | Do you smoke?                                                                                | Yes, daily<br>Yes, occasionally (less than 1 cigarette, or 1 cheroot, or 1 pipe of tobacco per day)<br>No, but previously<br>No, never                                                                                                                                                                                                              |
|                     | Daily smokers only                 | Please indicate how much tobacco you smoke on average per day?                               | Number of cigarettes<br>Number of cheroots<br>Number of cigars<br>Grams of pipe tobacco                                                                                                                                                                                                                                                             |
| Alcohol consumption | All participants                   | Have you consumed any alcoholic drinks during the past 12 months?                            | Yes<br>No                                                                                                                                                                                                                                                                                                                                           |
|                     | Drinkers within the past 12 months | How many of the following drinks have you had on average per week during the past 12 months? | Number of normal beers<br>Number of strong beers<br>Number of glasses of wine (1 bottle of wine equals 6 glasses)<br>Number of glasses/units of spirits (standard drinks)                                                                                                                                                                           |
| Ear-piercing        | All participants                   | Have you ever had your ears pierced?                                                         | Yes<br>No                                                                                                                                                                                                                                                                                                                                           |
| Vocational training | All participants                   | Have you ever had vocational training?                                                       | Yes<br>No                                                                                                                                                                                                                                                                                                                                           |
|                     | All participants                   | What is your educational level?                                                              | Skilled or unskilled blue-collar workers<br>Short-cycle higher education ( $< 3$ years, e.g. dental technician and nursing assistants)<br>Medium higher education (3–4 years, e.g. nurse, school teacher, and physiotherapist)<br>Long-cycle higher education ( $> 4$ years, e.g. medical physician, psychologist, and engineer)<br>Other education |
| Social status       | All participants                   | What is your self-estimated social status based on education, job, income, etc.              | Very high<br>High<br>Middle<br>Below middle<br>Low                                                                                                                                                                                                                                                                                                  |
| Type of residence   | All participants                   | What kind of residence do you live in?                                                       | House<br>Apartment<br>Other                                                                                                                                                                                                                                                                                                                         |

prevalence calculations. The questions used for assessment of alcohol consumption had been validated previously against increased levels ( $\geq 80$  IU/l) of serum  $\gamma$ -glutamyl transferase (GGT), a marker of alcohol exposure (17). The results revealed that self-reported total alcohol intake (total number of drinks/week) was significantly and positively associated with increased levels of GGT (18).

### Statistical analysis

Characteristics of participants were compared using the  $\chi^2$  test. A logistic regression model was performed with nickel sensitization as the dependent variable, and sex, age-group ("18–35 years", "36–55 years", "56–69 years"), and smoking status ("never smokers", "previous smokers", "current light smokers:  $\leq 15$  g/day", "current heavy smokers:  $> 15$  g/day") as the independent variables. In this model, a test for interaction between sex and smoking status was performed by using a log-likelihood ratio test. In order to examine the potential confounding effects of selected variables, we performed several logistic regression models adding one variable at a time while observing changes in the risk estimates for the exposure variables (smoking and alcohol consumption). These analyses were performed with nickel sensitization as the dependent variable and with sex, age-group ("18–35 years", "36–55 years", "56–69 years"), smoking status ("never smokers", "previous smokers", "current light smokers:  $\leq 15$  g/day", "current heavy smokers:  $> 15$  g/day"), ear-piercing ("yes", "no"), alcohol consumption ("0", "1–7", "8–14", " $\geq 15$ "), and educational level ("skilled or unskilled blue-collar workers", "short-cycle higher education", "medium higher education", "long-cycle higher education", "other education") as the explanatory variables. In further analyses, possible confounding by other socio-economic variables, such as "self-estimated social status", "vocational training" and "type of residence", were investigated and revealed essentially similar results as adjustment with the variable for educational

level. In fact, an analysis adjusted for "self-estimated social status" instead of educational level revealed a much stronger association between tobacco smoking and nickel sensitization. Also, adjustment for the occurrence of IgE antibodies was performed, but this did not change the results. Finally, similar logistic regression analyses were performed with "contact sensitization to at least one allergen" and "contact sensitization to at least one allergen but not nickel", respectively, as the dependent variables and with the explanatory variables listed in Table III. Associations were expressed as odds ratios (ORs) with 95% confidence intervals (95% CI). Data analyses were performed using the Statistical Products and Service Solutions package (SPSS Inc., Chicago, IL, USA) for Windows (release 15.0).

## RESULTS

Characteristics of the study population according to gender are presented in Table II. The prevalence of contact sensitization to at least one allergen, nickel contact sensitization, and ear-piercing was markedly higher among women than men, whereas men consumed significantly more alcohol than women. The prevalence of never smokers and previous smokers was nearly identical among women and men, whereas the prevalence of current light smokers ( $\leq 15$  g/day) was higher among women than men (16.3% vs. 9.5%) and the prevalence of current heavy smokers ( $> 15$  g/day) was higher among men than women (12.6% vs. 7.9%).

Table III shows the baseline characteristics of participants stratified by smoking status. The proportion of current light smokers ( $\leq 15$  g/day) was higher among subjects who were ear-pierced or were nickel sensitized

Table II. Gender-specific characteristics regarding contact sensitization (to at least one of 24 allergens), nickel contact sensitization, a history of ear-piercing, specific immunoglobulin (Ig)E status, alcohol consumption, smoking status, and educational level. Data was based on a general health examination including patch-testing among 3471 18–69-year-old participants from a cross-sectional study performed in Copenhagen, Denmark between 2006 and 2008

|                                                             | Men<br>% (n/total) | Women<br>% (n/total) | p-value <sup>a</sup> |
|-------------------------------------------------------------|--------------------|----------------------|----------------------|
| Contact sensitization to at least one allergen              | 4.7 (73/1547)      | 14.2 (272/1913)      | 0.001                |
| Nickel contact sensitization                                | 1.0 (15/1495)      | 10.3 (189/1913)      | 0.001                |
| Ear-piercing                                                | 17.0 (261/1538)    | 82.2 (1564/1902)     | 0.001                |
| Specific IgE <sup>b</sup>                                   | 27.3 (418/1531)    | 20.0 (378/1889)      | 0.001                |
| Alcohol consumption (drinks per week within past 12 months) |                    |                      |                      |
| 0                                                           | 9.0 (138/1532)     | 19.2 (367/1912)      | <0.001               |
| 1–7                                                         | 33.7 (516/1532)    | 53.0 (1013/1912)     |                      |
| 8–14                                                        | 24.3 (372/1532)    | 17.7 (338/1912)      |                      |
| $\geq 15$                                                   | 33.0 (506/1532)    | 10.1 (194/1912)      |                      |
| Smoking status                                              |                    |                      |                      |
| Never smokers                                               | 43.3 (640/1478)    | 43.1 (795/1846)      | <0.001               |
| Previous smokers                                            | 34.6 (512/1478)    | 32.7 (604/1846)      |                      |
| Current light smokers $\leq 15$ g/day                       | 9.5 (140/1478)     | 16.3 (301/1846)      |                      |
| Current heavy smokers $> 15$ g/day                          | 12.6 (186/1478)    | 7.9 (146/1846)       |                      |
| Educational level                                           |                    |                      |                      |
| Skilled or unskilled blue-collar workers                    | 44.8 (602/1345)    | 37.3 (609/1633)      | <0.001               |
| Short-cycle higher education                                | 14.1 (189/1345)    | 20.1 (328/1633)      |                      |
| Medium higher education                                     | 17.4 (234/1345)    | 26.1 (426/1633)      |                      |
| Long-cycle higher education                                 | 13.4 (180/1345)    | 7.5 (122/1633)       |                      |
| Other                                                       | 10.4 (140/1345)    | 9.1 (148/1633)       |                      |

<sup>a</sup>p-value of  $\chi^2$  test for the comparison of women and men.

<sup>b</sup>Analysis for IgE specific to birch, grass (timothy), cat, and mite (*Dermatophagoides pteronyssinus*). The analysis was judged to be positive if the measurement was in excess of 0.35 kU/l.

Table III. Characteristics of 3471 participants from a cross-sectional study performed in Copenhagen grouped by smoking status

|                                                         | Smoking status         |                           |                                             |                                              | <i>p</i> -value <sup>a</sup> |
|---------------------------------------------------------|------------------------|---------------------------|---------------------------------------------|----------------------------------------------|------------------------------|
|                                                         | Never smokers<br>% (n) | Previous smokers<br>% (n) | Current light smokers<br>≤15 g/day<br>% (n) | Current heavy smokers<br>> 15 g/day<br>% (n) |                              |
| Age (years)                                             |                        |                           |                                             |                                              |                              |
| 18–35 (n=593)                                           | 57.8 (343)             | 20.4 (121)                | 14.2 (84)                                   | 7.6 (45)                                     | 0.001                        |
| 36–55 (n=1613)                                          | 39.7 (641)             | 35.0 (565)                | 13.9 (224)                                  | 11.3 (183)                                   |                              |
| 56–69 (n=1118)                                          | 40.3 (451)             | 38.5 (430)                | 11.9 (133)                                  | 9.3 (104)                                    |                              |
| Ear-piercing                                            |                        |                           |                                             |                                              |                              |
| Yes (n=1752)                                            | 38.7 (678)             | 34.1 (598)                | 17.1 (300)                                  | 10.0 (176)                                   | 0.001                        |
| No (n=1563)                                             | 48.0 (751)             | 33.0 (516)                | 9.0 (140)                                   | 10.0 (156)                                   |                              |
| Nickel sensitization                                    |                        |                           |                                             |                                              |                              |
| Yes (n=1752)                                            | 31.4 (678)             | 36.6 (598)                | 21.1 (300)                                  | 10.8 (176)                                   | 0.001                        |
| No (n=1563)                                             | 44.4 (751)             | 33.2 (516)                | 12.6 (140)                                  | 9.7 (156)                                    |                              |
| Alcohol consumption (drinks/week within past 12 months) |                        |                           |                                             |                                              |                              |
| 0 (n=472)                                               | 43.2 (204)             | 31.8 (150)                | 12.5 (59)                                   | 12.5 (59)                                    | 0.001                        |
| 1–7 (n=1484)                                            | 50.1 (743)             | 30.1 (447)                | 12.7 (188)                                  | 7.1 (106)                                    |                              |
| 8–14 (n=673)                                            | 39.5 (266)             | 36.4 (245)                | 15.5 (104)                                  | 8.6 (58)                                     |                              |
| ≥15 (n=668)                                             | 31.0 (207)             | 40.0 (267)                | 13.0 (87)                                   | 16.0 (107)                                   |                              |
| Educational level                                       |                        |                           |                                             |                                              |                              |
| Skilled or unskilled blue-collar worker (n=1174)        | 38.4 (451)             | 35.2 (413)                | 14.1 (166)                                  | 12.3 (144)                                   | 0.001                        |
| Short cycle higher education (n=499)                    | 37.7 (188)             | 36.5 (182)                | 14.0 (70)                                   | 11.8 (59)                                    |                              |
| Medium cycle higher education (n=631)                   | 46.0 (290)             | 37.1 (234)                | 11.6 (73)                                   | 5.4 (34)                                     |                              |
| Long cycle higher education (n=289)                     | 64.7 (187)             | 24.9 (72)                 | 6.2 (18)                                    | 4.2 (12)                                     |                              |
| Other education (n=275)                                 | 44.4 (122)             | 35.3 (97)                 | 12.7 (35)                                   | 7.6 (21)                                     |                              |

<sup>a</sup>*p*-value of  $\chi^2$  test for the comparison of different categories of smoking status.

in comparison with subject who were not ear-pierced and who were not nickel sensitized. Alcohol consumption tended to increase with smoking status and the proportion of current heavy smokers (> 15 g/day) was higher among subjects with a short education.

Crude data analyses without adjustment for potential confounders showed that nickel sensitization was significantly associated with female sex, ear-piercing, alcohol consumption (≥15 drinks per week), and tobacco smoking (Table IV). The relationship between nickel sensitization and educational level revealed no clear pattern except a higher prevalence of nickel sensitization among subjects with a short-cycle higher education. We evaluated whether it could be assumed that the effects of smoking were independent of gender. Thus, a logistic regression model was performed with nickel sensitization as the dependent variable, and with sex, age-group (“18–35 years”, “36–55 years”, “56–69 years”), smoking status (“never smokers”, “previous smokers”, “current light smokers ≤15 g/day”, “current heavy smokers > 15 g/day”), and an interaction term between sex and smoking status as the independent variables. No significant interaction was found between sex and smoking status ( $p=0.97$ ), which means that the possible effect of smoking status on the prevalence of nickel sensitization did not differ between men and women. In order to examine possible confounding, several logistic regression models were performed in which one variable was added at a time while observing changes in the risk estimates for the exposure variables (smoking and alcohol consumption) (Table IV). The regression analyses revealed that ear-piercing was an important

confounder, which indicates that nickel sensitization to a high degree is an environmental disorder. Furthermore, the analyses showed that alcohol consumption was not associated with nickel sensitization, whereas a significant trend ( $p<0.05$ ) was identified between smoking status and nickel sensitization in the fully adjusted model (Table IV). Finally, similar logistic regression analyses were performed with “contact sensitization to at least one allergen” and “contact sensitization to at least one allergen but not nickel”, respectively, as the independent variable and with the explanatory variables listed in Table IV. These analyses did not show any significant associations between contact sensitization on the one hand and alcohol consumption or smoking status on the other hand. Thus, the fully adjusted regression analysis, with contact sensitization to at least one allergen as the dependent variable, revealed a non-significant trend test for smoking status ( $p<0.6$ ) (data not shown).

## DISCUSSION

The results of this study show that nickel sensitization is significantly associated with tobacco smoking. The association was dose-dependent and independent of gender. The results are in line with those from another cross-sectional population-based study performed in 1056 Danish adults (11) and are supported by a Norwegian patch-test study in which a significant association was identified in adult women (12).

It is important to evaluate to what extent confounding by other factors could explain the positive association

Table IV. The relationship of different potential risk factors to the prevalence of nickel sensitization

|                                                         | Nickel sensitization<br>% (n/total) | Crude OR<br>(95% CI) | Adjusted OR <sup>a</sup><br>(95% CI) | Adjusted OR <sup>b</sup><br>(95% CI) | Adjusted OR <sup>c</sup><br>(95% CI) | Adjusted OR <sup>d</sup><br>(95% CI) |
|---------------------------------------------------------|-------------------------------------|----------------------|--------------------------------------|--------------------------------------|--------------------------------------|--------------------------------------|
| Smoking status                                          |                                     |                      |                                      |                                      |                                      |                                      |
| Never smokers                                           | 4.4 (61/1397)                       | 1.00                 | 1.00, <sup>c</sup> <i>p</i> <0.001   | 1.00, <sup>c</sup> <i>p</i> <0.005   | 1.00, <sup>c</sup> <i>p</i> <0.009   | 1.00, <sup>c</sup> <i>p</i> <0.05    |
| Previous smokers                                        | 6.6 (71/1071)                       | 1.56 (1.09–2.21)     | 1.60 (1.11–2.91)                     | 1.45 (1.00–2.09)                     | 1.41 (0.98–2.05)                     | 1.19 (0.81–1.76)                     |
| Current light smokers ≤15 g/day                         | 9.7 (41/421)                        | 2.36 (1.57–3.57)     | 1.91 (1.25–2.31)                     | 1.72 (1.13–2.63)                     | 1.65 (1.08–2.53)                     | 1.50 (0.94–2.37)                     |
| Current heavy smokers >15 g/day                         | 6.7 (21/313)                        | 1.58 (0.94–2.63)     | 1.97 (1.15–3.35)                     | 1.78 (1.04–3.05)                     | 1.73 (1.01–2.98)                     | 1.56 (0.87–2.80)                     |
| Sex                                                     |                                     |                      |                                      |                                      |                                      |                                      |
| Men                                                     | 1.0 (15/1495)                       | 1.00                 | 1.00                                 | 1.00                                 | 1.00                                 | 1.00                                 |
| Women                                                   | 10.3 (189/1843)                     | 11.27 (6.63–19.16)   | 11.03 (6.36–19.16)                   | 5.50 (2.95–10.2)                     | 5.83 (3.10–10.97)                    | 5.55 (2.85–10.81)                    |
| Age (years)                                             |                                     |                      |                                      |                                      |                                      |                                      |
| 18–35                                                   | 7.2 (45/622)                        | 1.00                 | 1.00                                 | 1.00                                 | 1.00                                 | 1.00                                 |
| 36–55                                                   | 7.9 (128/1625)                      | 1.10 (0.77–1.56)     | 1.04 (0.71–1.52)                     | 1.09 (0.75–1.60)                     | 1.03 (0.70–1.52)                     | 0.99 (0.65–1.51)                     |
| 56–69                                                   | 2.8 (31/1091)                       | 0.38 (0.24–0.60)     | 0.41 (0.25–0.64)                     | 0.48 (0.29–0.79)                     | 0.45 (0.27–0.74)                     | 0.41 (0.24–0.73)                     |
| Ear-piercing                                            |                                     |                      |                                      |                                      |                                      |                                      |
| No                                                      | 1.2 (19/1567)                       | 1                    | –                                    | 1.00                                 | 1.00                                 | 1.00                                 |
| Yes                                                     | 10.6 (184/1741)                     | 9.63 (5.97–15.52)    | –                                    | 3.35 (1.89–5.96)                     | 3.44 (1.93–6.13)                     | 3.01 (1.66–5.46)                     |
| Alcohol consumption (drinks/week within past 12 months) |                                     |                      |                                      |                                      |                                      |                                      |
| 0                                                       | 7.6 (36/475)                        | 1.00                 | –                                    | –                                    | 1.00                                 | 1.00                                 |
| 1–7                                                     | 6.7 (98/1472)                       | 0.87 (0.59–1.29)     | –                                    | –                                    | 1.02 (0.67–1.56)                     | 0.96 (0.61–1.52)                     |
| 8–14                                                    | 6.4 (44/683)                        | 0.84 (0.53–1.32)     | –                                    | –                                    | 1.42 (0.86–2.34)                     | 1.33 (0.77–2.29)                     |
| ≥15                                                     | 3.8 (26/682)                        | 0.48 (0.29–0.81)     | –                                    | –                                    | 1.34 (0.77–2.37)                     | 1.05 (0.56–1.97)                     |
| Educational level                                       |                                     |                      |                                      |                                      |                                      |                                      |
| Skilled or unskilled blue-collar worker                 | 5.8 (67/1151)                       | 1.00                 | –                                    | –                                    | –                                    | 1.00                                 |
| Short cycle higher education                            | 9.3 (46/495)                        | 1.66 (1.12–2.45)     | –                                    | –                                    | –                                    | 1.16 (0.76–1.76)                     |
| Medium cycle higher education                           | 6.4 (41/638)                        | 1.11 (0.74–1.66)     | –                                    | –                                    | –                                    | 0.87 (0.57–1.33)                     |
| Long cycle higher education                             | 4.0 (12/297)                        | 0.68 (0.36–1.28)     | –                                    | –                                    | –                                    | 0.71 (0.33–1.49)                     |
| Other education                                         | 5.3 (15/281)                        | 0.91 (0.51–1.62)     | –                                    | –                                    | –                                    | 0.99 (0.54–1.84)                     |

<sup>a</sup>Logistic regression analysis adjusted for sex, age and smoking.<sup>b</sup>Logistic regression analysis adjusted for sex, age, smoking, and ear-piercing.<sup>c</sup>Logistic regression analysis adjusted for sex, age, smoking, ear-piercing, and alcohol consumption.<sup>d</sup>Logistic regression analysis adjusted for sex, age, smoking, alcohol consumption, ear-piercing and educational level.<sup>e</sup>Trend test.

OR: odds ratio; CI: confidence interval.

observed between smoking and nickel sensitization (Table III). The association remained relatively unchanged after adjustment for confounders by multivariable regression analyses (Table IV) although it cannot be ruled out that residual confounding (insufficient adjustment) or confounding by factors not included in this study could play a role. When the logistic regression analysis was adjusted for educational level, the association between smoking and nickel sensitization was weakened. Thus, it is possible that we were not able to sufficiently adjust for social status in our analyses as an association between nickel sensitization and socio-economic status has been suggested previously (19). In Malmö, Sweden, the prevalence of nickel sensitization was significantly higher among immigrants, unemployed, and patients on social security than among patients from higher socio-economic groups (19). Furthermore, a German study showed that the prevalence of nickel sensitization was higher among nurses (24.9%) and receptionist (29.3%) than among physicians (12.1%), indicating that nickel sensitization may be less prevalent in high-income groups (20). Despite the suggested association between nickel sensitization and socio-economic status, no association was identified between education

level and nickel sensitization in both an adjusted and an unadjusted analysis in this study (Table IV). We cannot exclude that the association between nickel allergy and tobacco smoking to some degree was explained by ear-piercing as it was more frequently reported among current light smokers (Table III).

This study did not identify any significant associations between smoking status and “contact sensitization to at least one allergen but not nickel” and “contact sensitization to at least one allergen”, respectively. It should be emphasized that the prevalence of contact sensitization to contact allergens other than nickel was low in this general population (Table II). Also, since patch-test readings were performed only on day 2 in this study, a lower prevalence of contact sensitization was expected (15, 21). The low prevalence estimates will necessarily lead to reduced statistical power in the regression analyses, which may hide associations. However, a previous Danish study also showed that nickel sensitization had a slightly stronger association with smoking than contact sensitization to at least one allergen (11). The stronger association observed for nickel sensitization may be explained by the fact that nickel is found in tobacco plants as a result of absorption from soil, fertilizing

products or pesticides. Furthermore, the nickel content in cigarettes and tobacco is high regardless of its kind and origin (22). One study examined the nickel concentration in 123 blood samples and 147 urine samples from smokers and non-smokers. It revealed a significantly higher concentration of nickel in the urine but not in the blood of smokers in comparison to non-smokers (22). It is therefore possible that T-cells in smokers are exposed to nickel in concentrations that may lead to nickel sensitization. However, nickel exposure from cigarettes is probably of minor importance in terms of inducing nickel contact sensitization as the prevalence of nickel sensitization in men was approximately 1%, whereas almost 50% of men reported current or previous smoking. Finally, the findings in this study (i.e. a stronger association for nickel sensitization than contact sensitization) could be coincidental or a result of confounding, as nickel sensitization may have a stronger association with lower social groups than, for example, fragrance and preservative sensitization.

Contact sensitization and autoimmune conditions have traditionally been regarded as T-helper 1 (Th1)-mediated immune responses, whereas sensitization to aeroallergens, as observed in allergic asthma and rhinitis, has been regarded as a Th2 mediated condition (23, 24). The Th1/Th2 dichotomy was for many years the cornerstone of immunological thinking and dictated that Th1 cells were down-regulated by cytokines released from Th2 cells and vice versa. As it was recently demonstrated that a subgroup of T cells, "T-regulatory" (Treg), may suppress both Th1- and Th2-mediated immune responses, the dichotomy may only partially explain the development of various immune responses (25, 26). However, tobacco smoking has been causally linked to the development of autoimmune diseases, such as systemic lupus erythematosus, multiple sclerosis, Grave's hyperthyroidism, rheumatoid arthritis (27), and contact sensitization (11, 12) whereas prospective population-based studies have suggested that tobacco smoking may decrease the risk of IgE-mediated allergic sensitization to aeroallergens (28, 29). Also, cross-sectional population based studies have demonstrated a lower prevalence of sensitization to common aeroallergens among smokers and ex-smokers than among non-smokers (30, 31). Thus, it seems plausible that tobacco smoking favours Th1-mediated immune responses and suppresses Th2-mediated immune responses. These immunological perspectives support the findings from this study, although it should be recognized that humane immune responses are very complex as demonstrated by contact sensitization being inversely related to type I diabetes and inflammatory bowel diseases (32, 33).

This study did not identify any association between alcohol consumption and the prevalence of nickel sensitization (or contact sensitization), although nickel allergy appeared to be lower for individuals who reported

alcohol abstinence in the adjusted analysis (Table IV). However, as participants were asked only about alcohol consumption within the past 12 months, it is possible that we did not accurately assess the cumulated alcohol exposure. Also, the limitations of day two patch-test readings reduced statistical power in our analysis, which may hide an association (15, 21). We are aware of only one previous study that also addressed the association between contact sensitization and alcohol consumption (10). This study did not identify any association between the prevalence of alcohol consumption and contact sensitization, whereas it suggested that the 8-year incidence of contact sensitization was significantly higher among non-drinking women (10). In general, a prospective incidence-based analysis is considered more reliable than cross-sectional studies when determining the cause-effect relationship. A follow-up of the present study population would be of interest to investigate this issue further. Furthermore, it may be of interest to take into account genetic variations in alcohol metabolism, as certain genetic variations may influence both alcohol drinking behaviour and susceptibility to the immunological effects of alcohol (34). Such genetic influence would tend to bias associations between alcohol and immune effects.

In conclusion, this general population study confirmed our previous finding that smoking is associated with nickel sensitization. The possible biological mechanisms underlying this association remain to be elucidated. We could not confirm the previously reported negative association between alcohol consumption and the development of contact sensitization. In future prospective studies, it could be of interest to investigate whether tobacco-smoking leads to a poor prognosis of allergic nickel contact dermatitis in comparison with non-smokers.

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