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Levels of house dust mite allergen in cars

Howard J. Mason¹, Ian Smith¹, Siti Marwanis Anua^{2,3}, Nargiz Tagiyeva², Sean Semple², and
Graham Devereux²

Health & Safety Laboratory, Harpur Hill, Buxton¹, Respiratory Group, Division of Applied Health Sciences, School of
Medicine and Dentistry, University of Aberdeen, Foresterhill, Aberdeen², UK, School of Health Sciences, Health
Campus, Universiti Sains Malaysia, Kubang Kerian, Kelantan, Malaysia³

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This small study investigated house dust mite (HDM) allergen levels in cars and their owners' homes in north-east Scotland. Dust samples from twelve households and cars were collected in a standardised manner. The dust samples were extracted and measured for the *Dermatophagoides* group 2 allergens (Der p 2 and Der f 2) and total soluble protein. Allergen levels at homes tended to be higher than in the cars, but not significantly. However, they significantly correlated with paired car dust samples expressed either per unit weight of dust or soluble protein ($\rho=0.657$; $p=0.02$ and 0.769 ; $p=0.003$, respectively). This points to house-to-car allergen transfer, with the car allergen levels largely reflecting levels in the owner's home. Car HDM allergen levels were lower than those reported in Brazil and the USA. Twenty-five percent of the houses and none of the cars had allergen levels in dust greater than 2000 ng g^{-1} . This value is often quoted as a threshold for the risk of sensitisation, although a number of studies report increased risk of sensitisation at lower levels. This small study does not allow for characterisation of the distribution of HDM allergen in vehicles in this geographic area, or of the likely levels in other warmer and more humid areas of the UK. Cars and other vehicles are an under-investigated micro-environment for exposure to allergenic material.

KEY WORDS: *Der f 2*; *Dermatophagoides*; *Der p 2*; Scotland; vehicles

House dust mites (HDM) are almost ubiquitous in houses. Centrally heated, well-insulated homes with pets means that allergenic material related to HDM and cats and dogs is common (1, 2). Estimates from the Pet Food Manufacturers Association are that approximately 20 % of UK households keep dogs or cats. Sensitisation and subsequent allergic symptoms may be a relatively common outcome from such domiciliary indoor exposure scenarios. However, there may be other exposure scenarios for such allergens.

In Great Britain, over 77 % of households own one or more cars (3) and recent data suggest that every person in Great Britain, as a passenger or driver, travels on average about 14 miles and spends 35 minutes every day in a car (4). Employee car-sharing schemes and shared school-runs for children have become increasingly common in the UK. In contrast to the house indoor environment, we can find little data, and none from Europe (5, 6) on the levels of common allergenic material in cars.

In a recent study investigating why bakers' children are more likely to have asthma we clearly demonstrated work-to-home transfer of bakery allergens (7, 8). Subsequently we became interested in whether we could find evidence

of transfer of indoor household allergens, such as HDM, from houses to vehicles as an alternative exposure scenario for such allergens, and whether the levels found in vehicles could be of health concern.

Based on samples collected as part of the bakery-home study, we report the levels of HDM allergenic material using a *Dermatophagoides* species group 2 allergen assay in dust samples collected from a small number of matched homes and associated cars.

MATERIALS AND METHODS

The data reported here form part of an investigation in north-east Scotland on the transfer of allergens from bakeries to workers' homes (7). Dust samples had been collected from 34 houses and 13 cars. Twenty-four of the 34 house samples were collected in bakery workers' homes and 10 were control samples taken from the homes of university staff and students. The thirteen vehicles sampled belonged to bakery workers. The focus of this short report is the level of Der group 2 allergens in dust samples from the 12 matched households and cars.

These matched dust samples had been collected by vacuuming using Dustream collectors (Indoor Biotechnologies, Cardiff, UK) from twelve households in Scotland and their associated cars. Description of the

Correspondence to: Howard J. Mason, Health & Safety Laboratory, Harpur Hill, Buxton, SK17 9JN, UK, E-mail: howard.mason@hsl.gsi.gov.uk

sampling techniques has been fully reported (7), but essentially all household samples were obtained by vacuuming the seat of a well-used lounge chair and nearby floor area for approximately 5 minutes. Car dust samples were obtained by vacuuming the driver's seat and foot-well.

Dust samples were weighed and extracted for 2 hours by mixing them with 0.1 % Tween 20 (Sigma-Aldrich, Poole, UK) in phosphate buffered saline at 10 % weight/volume ratio. Extracts were centrifuged and the supernatant analysed for *Dermatophagoides* species group 2 allergens (Der p 2 + Der f 2) using the ELISA kit (Indoor Biotechnology, Cardiff, UK) and total soluble protein using the standard bicinchoninic acid colorimetric assay (Sigma-Aldrich, Poole, UK). The batch-to-batch method imprecision for the allergen and protein methods are 14 % and 10 % respectively. The lower limit of quantification calculated for the allergen assay using ProQuant software (QIVX Inc. Fort Collins, CO, USA) was 2 ng g⁻¹ dust. We measured Der group 2 allergens rather than the more usual Der p 1, as the latter can be underestimated in the presence of bakery/wheat components (9).

RESULTS AND DISCUSSION

The levels of HDM in the 12 bakery households matching the cars were not different from the larger data set of 24 bakery homes.

Measurable levels of the Der group 2 allergens were detected in all dust samples collected from the twelve houses and the matching cars. Their medians (and ranges) were 55 (5-12,863) ng g⁻¹ and 23 (3-454) ng g⁻¹ dust, respectively, or 6.48 (0.28-740.1) and 2.93 (0.14-65.11) when expressed as ng mg⁻¹ soluble protein (Figure 1). Although allergen levels were higher in house samples

expressed either way, these differences did not achieve statistical significance (Friedman test; $p=0.082$ and 0.586 , respectively). However, allergen levels correlated significantly between the paired dust samples from cars and houses; Spearman's coefficients of correlation (ρ) were 0.657 , $p=0.02$ for the results expressed per gram of dust and 0.769 , $p=0.0034$ when expressed per mg of soluble protein (Figure 2).

For the reason noted earlier, we measured Der group 2 rather than the more usual Der p 1. Attempting to convert our results to likely Der p 1 levels can only yield approximate values. Custovic (10) showed good correlation in UK field samples between Der p 1 and p 2, whereby 1 ng of Der p 2 was equivalent to 1.9 ng of Der p 1. Our group 2 measurement also theoretically includes any Der f 2 present, but in the UK *D. pteronyssinus* is predominant over *D. farina*. Thus we assume that likely Der p 1 levels are somewhere between the same as, or require doubling of, our measurements.

Justino et al. (6) measured Der group 1 allergens in car dust samples from Brazilian university staff and students, reporting a geometric mean [Der p 1 + Der f 1] of 540 ng g⁻¹ dust. Car allergen levels in our study are lower than these. Neal et al. (5) conducted a study of paired vehicles and homes in Ohio, USA. Their overall means of Der group 1 in houses and vehicles were 25,600 and 1,300 ng g⁻¹ dust, respectively. While Neal et al. (5) presents arithmetic means without any indication of the distribution of allergen data, which were very non-normal in both Justino's and our study, their results suggest much higher vehicle and household levels of HDM allergens than we report. Interestingly, Neal et al. (5) also found a positive association between house and vehicle allergen levels.

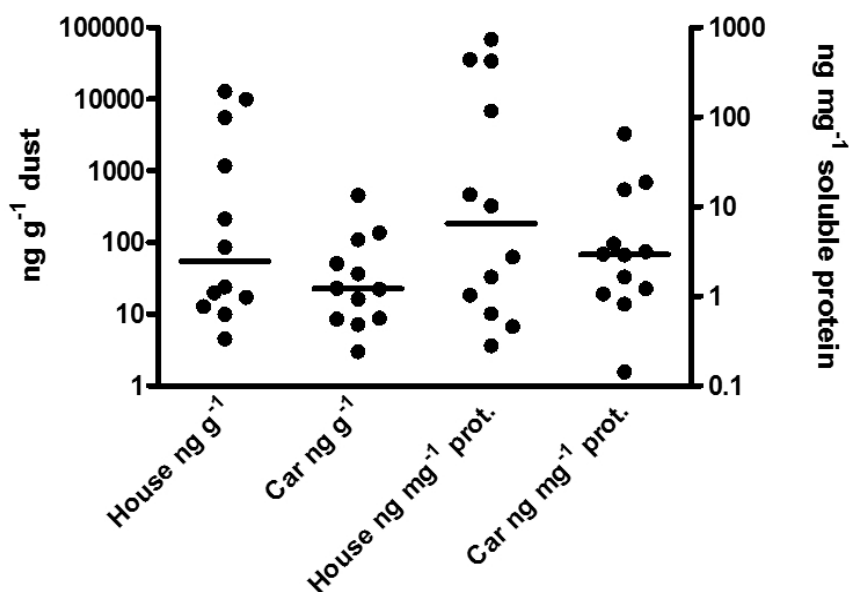


Figure 1 Levels of Der group 2 (Der p 2 + Der f 2) found in houses and cars

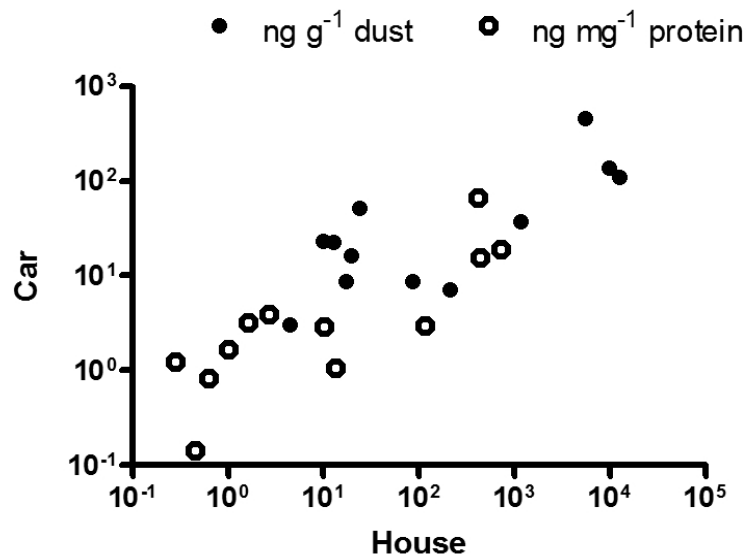


Figure 2 Correlation between car and house levels of HDM allergens

The value of 2000 ng g⁻¹ of Der p 1 in settled dust is often quoted as the threshold for the risk of sensitisation (11). Our data suggest that 25 % of the matched households would exceed this. No allergen levels in the cars exceed it, and therefore suggest no risk of sensitisation. However, a number of studies (12-14) show that exposure to allergen levels of less than 2000 ng g⁻¹ can still be a significant sensitisation risk. Dose-response relationships for other outcomes (rhinitis, upper and lower respiratory symptoms) in sensitised individuals are unclear. Therefore, while the health risk in this small vehicle group is probably small, their HDM levels deserve further investigation in a larger UK cohort, particularly if focussed on households that are likely to have high HDM levels. What also deserves further investigation is the use of allergen levels in settled dust as the inhalation exposure metrics for health outcomes (15). Such metrics have been employed for HDM allergens, as air measurements have often failed to detect airborne levels of Der p 1 unless there is significant dust disturbance or human activity. This possibly relates to the Der p 1 particle size. Analysis of airborne HDM allergens after normal human activity in both the house and the car may help clarify exposure-risk relationships.

Our household allergen levels are comparable with those reported in a Dutch study of living room carpets (16). The group 2 measurement reflects mite body proteins and is more heat-stable than the Der p 1 allergen associated with a faecal protein (11), and Der p 2 has been shown to correlate with mite numbers indoors (17). However, we do not know whether the vehicle results reflect the transfer of the living mites from home or only the transfer of the allergenic protein.

The significant association between the allergen levels in matched household and car dust samples suggests transfer from home to car. We have already established transfer of

allergens between workplaces and homes through contaminated clothes, hair, skin, with increased bakery allergen levels found in bakers' cars used for commuting (7). This study also suggests that the car could be a relatively neglected micro-environment for secondary exposure to indoor and occupational allergenic material.

Levels of house dust allergen in a car may reflect the levels of the allergen in the house and the extent of house-to-car transfer. While HDM allergen levels measured in this study in north-east Scotland indicate low risk (18), the small sample size can hardly serve to predict the distribution of HDM allergen, especially the levels that might constitute a health risk. Further investigations of HDM levels (including Der p 1 measurements) in UK households and vehicle dust are warranted, especially where high household HDM levels are likely to be found (i.e. higher ambient temperatures and humidity). Given that the major health risks are due to inhalation rather than indirectly from settled dust content and that airborne HDM allergen derives largely from human activity and disturbance of settled dust, we suggest measuring vehicle airborne allergen levels during real-life use of vehicles. We also hope this study will raise further interest in exposure to allergens and their possible health risks in all forms of transport.

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Razine alergena prašinskih grinja u automobilima

U ovome smo preliminarnom istraživanju izmjerili razine alergena prašinskih grinja u automobilima i domovima njihovih vlasnika u sjevernoistočnoj Škotskoj. Uzorci prašine uzeti su na standardizirani način iz dvanaest domova i dvanaest odgovarajućih automobila. Nakon ekstrakcije izmjerene su razine 2. skupine alergena grinja roda *Dermatophagoides* (Der p 2 i Der f 2) te njihove ukupne topljive bjelančevine. Razine alergena u kućama bile su mahom više nego u automobilima, ali ne značajno. Uočena je međutim značajna korelacija njihovih razina s razinama u automobilima, bez obzira na to jesu li iskazane udjelom težine po gramu prašine ili topljivim bjelančevinama ($\rho=0,657$; $p=0,02$ odnosno $0,769$; $p=0,003$). To upućuje na prijenos alergena iz kuće u auto tj. pokazuje da razine alergena u autima odražavaju razine u domovima njihovih vlasnika. Razine alergena kućnih prašinskih grinja u automobilima u Škotskoj bile su niže od onih izmjerenih u Brazilu i SAD-u. U 25 % domova razina alergena bila je viša od 2000 ng g⁻¹, a ni u jednom uzorku nije izmjerena razina viša od te. Ta se vrijednost često smatra graničnom za rizik od senzitivizacije, premda neka istraživanja govore o povišenom riziku od senzitivizacije i pri nižim razinama alergena. Rezultati ovog istraživanja ne daju uvida u stvarnu prisutnost alergena kućnih prašinskih grinja u vozilima u sjeveroistočnoj Škotskoj niti upućuju na to kolike bi mogle biti njihove razine u toplijim i vlažnijim krajevima Ujedinjenog Kraljevstva. Automobili i ostala vozila zanemareni su kao izvori izloženosti alergenima.

KLJUČNE RIJEČI: Der f 2; Dermatophagoides; Der p 2; Škotska; vozila