



## Early-life residential exposure to soil components in rural areas and childhood respiratory health and allergy



Graham Devereux<sup>a,\*</sup>, Nara Tagiyeva<sup>a</sup>, Stephen W. Turner<sup>a</sup>, Jon G. Ayres<sup>b</sup>, Anthony Seaton<sup>a</sup>, Gordon Hudson<sup>c</sup>, Rupert L. Hough<sup>c</sup>, Colin D. Campbell<sup>c,d</sup>, Charles A. Shand<sup>c</sup>

<sup>a</sup> Division of Applied Health Sciences, School of Medicine and Dentistry, University of Aberdeen, Foresterhill, Aberdeen, AB25 2ZD, UK

<sup>b</sup> Institute of Occupational and Environmental Medicine, University of Birmingham, Edgbaston, Birmingham, B15 2TT, UK

<sup>c</sup> The James Hutton Institute, Craigiebuckler, Aberdeen, AB15 8QH, UK

<sup>d</sup> Department of Soil and Environment, Swedish University of Agricultural Sciences, Uppsala, Sweden

### HIGHLIGHTS

- We have linked a birth cohort with the Scottish Soils Database.
- We have looked for associations between asthma, allergy and soil content.
- No associations found for soil content at residence at age 5 years.
- Silt content at birth residence adversely linked with respiratory health up to age 5.
- Early life exposure to soil silt may adversely influence childhood respiratory health.

### ARTICLE INFO

#### Article history:

Received 1 March 2013

Received in revised form 28 June 2013

Accepted 30 June 2013

Available online xxxx

Editor: Simon James Pollard

#### Keywords:

Soil  
Silt  
Contamination  
Residential  
Children  
Asthma

### ABSTRACT

The increase in asthma and allergies has been attributed to declining exposure to environmental microorganisms. The main source of these is soil, the composition of which varies geographically and which is a major component (40–45%) of household dust. Our hypothesis-generating study aimed to investigate associations between soil components, respiratory health and allergy in a Scottish birth cohort. The cohort was recruited in utero in 1997/8, and followed up at one, two and five years for the development of wheezing, asthma and eczema. Lung function, exhaled nitric oxide and allergic sensitization were measured at age five in a subset. The Scottish Soils Database held at The James Hutton Institute was linked to the birth cohort data by the residential postcode at birth and five years. The soil database contained information on size separates, organic matter concentration, pH and a range of inorganic elements. Soil and clinical outcome data were available for 869, 790 and 727 children at one, two and five years. Three hundred and fifty nine (35%) of children had the same address at birth and five years. No associations were found between childhood outcomes and soil content in the residential area at age five. The soil silt content (2–20 µm particle size) of the residential area at birth was associated with childhood wheeze (adjusted OR 1.20, 95% CI [1.05; 1.37]), wheeze without a cold (1.41 [1.18; 1.69]), doctor-diagnosed asthma (1.54 [1.04; 2.28]), lung function (FEV<sub>1</sub>: beta −0.025 [−0.047; −0.001]) and airway inflammation (FE<sub>NO</sub>: beta 0.15 [0.03; 0.27]) at age five, but not with allergic status or eczema. Whilst residual confounding is the most likely explanation for the associations reported, the results of this study lead us to hypothesise that early life exposure to residential soil silt may adversely influence childhood respiratory health, possibly because of the organic components of silt.

© 2013 Elsevier B.V. All rights reserved.

**Abbreviations:** CI, confidence interval; FE<sub>NO</sub>, fractional exhaled nitric oxide; FEV<sub>1</sub>, forced expiratory volume in 1 s; FVC, forced vital capacity; GEEs, General Estimating Equations; IQR, interquartile range; NO, nitric oxide; OR, odds ratio; PEF, peak expiratory flow; PM, particulate matter; SIMD, Scottish Index of Multiple Deprivation; SSD, Scottish Soils Database; TIN, triangulated irregular network.

\* Corresponding author at: Child Health, Royal Aberdeen Children's Hospital, Aberdeen AB25 2ZG, UK. Tel.: +44 1224 558196; fax: +44 1224 551919.

**E-mail addresses:** [g.devereux@abdn.ac.uk](mailto:g.devereux@abdn.ac.uk) (G. Devereux), [n.tagiyeva-milne@abdn.ac.uk](mailto:n.tagiyeva-milne@abdn.ac.uk) (N. Tagiyeva), [s.w.turner@abdn.ac.uk](mailto:s.w.turner@abdn.ac.uk) (S.W. Turner), [j.g.ayres@bham.ac.uk](mailto:j.g.ayres@bham.ac.uk) (J.G. Ayres), [a.seaton@abdn.ac.uk](mailto:a.seaton@abdn.ac.uk) (A. Seaton), [gordon.hudson@hutton.ac.uk](mailto:gordon.hudson@hutton.ac.uk) (G. Hudson), [rupert.hough@hutton.ac.uk](mailto:rupert.hough@hutton.ac.uk) (R.L. Hough), [colin.campbell@hutton.ac.uk](mailto:colin.campbell@hutton.ac.uk) (C.D. Campbell), [charles.shand@hutton.ac.uk](mailto:charles.shand@hutton.ac.uk) (C.A. Shand).

## 1. Introduction

Soil is a prime source of the essential elements and potentially toxic substances, which can enter the human body indirectly through the consumption of food products. Soil can also enter the human body directly through inhalation, inadvertent ingestion, deliberate eating and skin lesions. There are several well known examples of the influences of deficiencies or excesses of essential or toxic elements in soil, and the effects of inhaling soil containing fibrous minerals on human health (Abrahams, 2002; Hough, 2007; Oliver, 1997).

The hygiene hypothesis (Strachan, 1989) has been proposed as an explanation for the increase in asthma and allergic disease in westernised countries in the latter decades of the 20th century. Humans have evolved in a pathogen rich environment and the hygiene hypothesis posits that a modern westernised, non-traditional lifestyle no longer exposes people to the diverse microbe-rich environment, in response to which the human immune system has evolved and most probably requires for 'normal' maturation. This concept is supported by the observation that children brought up on farms and exposed to a heavy burden and wide variety of micro-organisms are less likely to develop asthma and allergic disease (Braun-Fahrlander et al., 1999; Ege et al., 2006, 2011; Riedler et al., 2001). Early life exposure to a diverse, micro-organism-rich environment appears to be especially important in reducing the risk of asthma and allergic disease.

A major exposure source of micro-organisms is the soil and this has led to the concept that 'dirt' may be good in preventing asthma and allergic disease. Indeed it has been formally hypothesised that the disconnection of man and the soil might be one of the major contributors to the increase in asthma and allergic disease (von Hertzen and Haahtela, 2006). Soil is a complex, diverse mixture of mineral matter, organic matter, water, and air (Brady and Weil, 2008) whose composition varies widely geographically. The diversity of microbes in soil is vast and new molecular methods applied to landscape scale studies are starting to show that microbial community composition varies with soil type and land use (Singh et al., 2009). Whilst exposure to soil is conventionally considered to be an outdoor phenomenon, indoor exposure occurs as soil is transported indoors on shoes, clothes and in the air, comprising about 40–45% of household dust (Layton and Beamer, 2009; Trowbridge and Burmaster, 1997).

To investigate whether exposure to soil is protective against asthma and allergic disease, we conducted an exploratory study of possible associations between soil composition and early childhood asthma and allergic disease. We linked data from a local birth cohort study with data from the Scottish Soils Database (SSD). The soil database contained information on size separates (sand, silt, clay), organic matter concentration, pH values and a range of inorganic element concentrations. It was envisaged that the results of this investigation would generate novel hypotheses for further investigation. Outside occupational settings, few studies have investigated the influence of soil components on human health, although the specific effects of some soil minerals are well documented (Sing and Sing, 2010; Smith and Lee, 2003). To our knowledge there have been no studies on children's respiratory health in association with environmental exposure to soil components.

## 2. Methods

### 2.1. Data description

Two data sources were used: (1) a longitudinal birth cohort investigating associations between maternal diet and childhood asthma/allergic disease, and (2) the SSD held at The James Hutton Institute. The details of the birth cohort have been described elsewhere (Martindale et al., 2005). In brief, 2000 healthy pregnant women attending an antenatal clinic (Aberdeen Maternity Hospital,

AMH), at median 12 weeks gestation, were recruited in 1997/8. The AMH provides the antenatal care for the entire Grampian region (population ~ 550,000). There was no selection for asthma or allergic disease (Martindale et al., 2005). The ensuing cohort of 1924 live singleton births was followed up at one, two, and five years with response rates of 79% ( $n = 1511$ ), 71% ( $n = 1373$ ) and 65% ( $n = 1253$ ), respectively. At each follow-up a postal questionnaire was used to ascertain the presence of respiratory and allergic symptoms in the cohort children. The questions were those used in the International Study of Asthma and Allergies in Childhood (ISAAC) (International Study of Asthma and Allergies in Childhood Steering Committee, 1998): specifically 'wheeze ever', 'wheeze in the last 12 months', 'wheeze in the absence of colds', 'asthma ever' and doctor diagnoses of asthma, eczema and hay fever. A derived outcome of 'active asthma' was defined by positive responses to both 'doctor diagnosed asthma' and 'wheeze in the last 12 months'. At the five-year follow-up children were also invited to attend for a more detailed assessment. 755 children attended for measurement of lung function by spirometry (peak expiratory flow PEF, forced expiratory volume in 1 s FEV<sub>1</sub>, forced vital capacity FVC), exhaled nitric oxide (FE<sub>NO</sub>), and allergic status ( $\geq 1$  positive skin prick test to cat, timothy grass, egg, and house dust mite allergens, 3-mm cut off (ALK Abello, UK)). Other factors that could potentially confound any association with childhood outcomes were collected during pregnancy, at delivery and follow-up. The study was approved by the Grampian Research Ethics Committee and informed written consent was obtained from the mothers and assent from the children where possible. Data from the cohort study were selected for this study if the participants lived in a rural location. This selection was made because (1) the relationship of people with the soil is likely to be closer in rural settings, and (2) soil samples from built areas do not form part of the soil database. The cohort study data were linked to SSD soil composition data using the postcode of the child's mother's home at the date of birth and at five years.

The SSD has several components (Brown et al., 1987). We had the available soil profile descriptions of horizons (distinct soil layers approximately parallel to surface) and analytical data from approximately 13,000 sites throughout Scotland. The study area of soils was restricted to North East Scotland and included only cultivated top soils from the postcode areas of the cohort children. The soil used for the chemical analysis was taken from the central part of the cultivated surface horizon. From the available data (1439 out of 1587 samples) the average position of the soil sample was between 5 and 15 cm below the soil surface. The soils had been analysed according to the following protocols. The soils were air-dried and sieved through a screen with 2-mm openings. Particle size analysis according to the international classification (clay < 2, silt 2–20 and sand > 20–2000  $\mu\text{m}$ ) was determined using a hydrometer method after dispersion of the soil in 1 M sodium hydroxide (Bouyoucos, 1936). Exchangeable cations (Ca, Mg, Na, K) were measured by displacement with 1 M ammonium acetate (Chapman, 1965). The metals (Ba, Co, Cr, Cu, Ga, Mn, Mo, Ni, Pb, Sr, Ti, V) were measured by aqua regia extraction (Berrow and Stein, 1983). Total C and N were measured with an elemental analyser. Total P was measured by a fusion technique (Smith and Bain, 1982).

### 2.2. Data linkage

The postcode was used as the geographical link between the soil and health data. Postcode structure in the UK is hierarchical and consists of a series of alphanumeric characters (maximum of 7 characters). The full 7-character code defines a limited number of addresses or a single large mail-delivery point. For this study we used the 5-character postcode sector code. These spatial units for the cohort children data are arbitrary in terms of the soil pattern and may cross boundaries between soils having different properties.

This issue is described as the modifiable areal unit problem, which is known to yield higher correlations for larger spatial units (Openshaw, 1984). Soil composition for each 5-character postcode sector was derived using interpolation: first, a TIN was constructed from the soil attribute values at the profile point samples. In the second stage, the TIN was interpolated to provide a grid of soil attributes estimated values. The grid of estimated values was used to calculate a mean value for soil attributes within each post code district and sector. All interpolations and computations were carried out in ARC/INFO geographic information system.

### 2.3. Statistical analysis

Lung function outcomes were adjusted for age, sex and height. Univariate associations between childhood respiratory/allergic outcomes (symptoms, diagnoses, measurements) at one, two and five years and soil composition at birth were assessed using Mantel–Haenszel odds ratio (OR).

Many univariate tests of association were performed relating wheezing and asthma outcomes to soil constituents. Therefore, the Simes procedure was used, controlling the false discovery rate at 0.05 (Benjamini et al., 2001), in an attempt to discover if any of the associations could be considered statistically significant, given the multiple statistical tests carried out. As wheeze and asthma outcomes were highly correlated the Simes procedure was performed using the most highly significant association with a soil component. Logistic regression and General Estimating Equations (GEEs; to cope with the repeated measures structure of the several follow-up phases of the cohort study) with unstructured correlation matrices were used to relate soil composition data at birth to childhood respiratory/allergic outcomes at one, two and five years with adjustment for potentially confounding factors. Linear regression was similarly used for the lung function and log<sub>e</sub> transformed FE<sub>NO</sub> outcomes at five years. Given the clustering of cohort children by 5 digit postcodes, multilevel analysis with children as observational units at a basic level and 5 digit postcode at a superior level were also conducted on the cross-sectional data, these were similarly adjusted for potential confounding factors. Clustering of the longitudinal data within postcodes was addressed using GEE because GEE is able to handle many types of unmeasured dependence between outcomes and is robust to misspecification of the correlation structure/hierarchy. IBM SPSS Statistics for Windows, Version 20.0. (Armonk, NY) was used for data analyses.

The performance of the TIN approach to interpolation, for silt data only, was compared to ordinary kriging in a separate study of 5461 topsoil silt samples from cultivated soils taken across Scotland. In this comparison a validation set of 200 soil samples was selected at random and the TIN method or ordinary kriging used with the remaining 5261 samples to estimate the values at the validation sites. The outputs of the methods were compared using the package geoR (Diggle and Ribeiro, 2007) in the statistical computing environment provided by R 2.4.1 (The R Development Core Team, 2006).

## 3. Results

### 3.1. Study population and availability of data

A summary of the original birth cohort including socio-economic status and follow-up participation is presented in Table 1. Of the original cohort of 1924 live singleton births, 1015 (53%) had addresses which were considered rural and for which soil compositional data were available. Of these children, respiratory/allergic outcome data were available for 869 (86%), 790 (78%) and 727 (72%) at one, two and five years, respectively. Lung function and FE<sub>NO</sub> data at five years could be linked to soil composition data for 255 and 102 children respectively. Only 359 (49%) of the 727 subjects responding at five years had the same residential postcode at birth and five years.

### 3.2. Association between soil constituents and childhood outcomes

Univariate analyses relating the constituents of soil by postcode at birth to asthma and wheeze outcomes at one, two and five years revealed no significant associations with soil content of clay, or Ba, Co, Cr, Cu, Ga, Mn, Mo, Ni, Sr, Ti, V or exchangeable Ca concentrations. The soil concentration of Mg, Na, K, C, N, P, Pb and soil pH were significantly associated ( $0.01 < p < 0.05$ ) with one or two outcomes each. The strongest ( $0.001 < p < 0.05$ ) and most consistent associations (i.e.  $p < 0.05$  for 10 adverse associations with 14 outcomes) were observed between soil silt content (expressed as percentage dry weight of <2 mm sieved soil) for residential postcode of birth with asthma/wheeze at one and five years, and lung function at five years (Tables 2, 3). When the Simes procedure was carried out on the univariate associations with soil components, the association between PEF at age 5 and soil silt content remained significant. Within the study area there were 1587 soil profiles with silt (2–20 µm particle size) data for the topsoil horizon (Fig. 1). Soil silt content for postcode at birth was also associated with FE<sub>NO</sub> at five years (correlation coefficient  $\rho = -0.29$ ,  $p = 0.003$ ). There were no significant associations between constituents of soil for residential postcode at birth with eczema, hay fever or allergen skin prick test outcomes. Soil silt content for the residences at birth of the cohort approximated to a normal distribution ranging from 2.1% to 31.6% with a mean content of 14.5%, standard deviation (SD) 5.7. Associations with soil silt content were expressed as changes for 1 SD increase in soil silt content.

Multivariate and multilevel analyses relating soil silt content for postcode of birth with asthma/wheeze and lung function (at five years) demonstrated that the associations with silt were still present and consistent (Tables 2, 3). Log<sub>e</sub> transformed FE<sub>NO</sub> at five years was also associated with soil silt for residential postcode at birth (adjusted regression coefficient 0.15, (95% CI 0.03 – 0.27)  $p = 0.017$  for a 1SD increase in soil silt, with multilevel modelling OR and 95% CI were unchanged but  $p = 0.045$ ).

General Estimating Equations were used to model the longitudinal 'wheeze in the last 12 months' and 'wheeze in the absence of a cold in the last 12 months' data collected at one, two and five years. These analyses demonstrated that soil silt content for postcode of birth

**Table 1**

Comparison of data for children for whom both childhood outcome and soil data were available with the rest of the cohort.

| Group                                                                   | Soil data                |                         | P      |
|-------------------------------------------------------------------------|--------------------------|-------------------------|--------|
|                                                                         | Unavailable<br>(n = 909) | Available<br>(n = 1015) |        |
| Maternal age of leaving full time education, median (IQR <sup>a</sup> ) | 17 (16–20)               | 17 (16–21)              | 0.029  |
| Mother obtained a degree                                                | 22.5%                    | 25.2%                   | 0.215  |
| Paternal non-manual social status                                       | 49.7%                    | 62.3%                   | <0.001 |
| Scottish Index Of Multiple Deprivation, median (IQR <sup>a</sup> )      | 13.46 (6.13–27.36)       | 7.18 (4.83–10.20)       | <0.001 |
| Maternal smoking during pregnancy                                       | 37.6%                    | 22.1%                   | <0.001 |
| Maternal asthma                                                         | 17.8%                    | 14.4%                   | 0.041  |
| Maternal eczema                                                         | 16.2%                    | 16.5%                   | 0.860  |
| Maternal hay fever                                                      | 23.8%                    | 25.6%                   | 0.341  |
| Participation in 1 year follow-up                                       | 70.6%                    | 85.6%                   | <0.001 |
| Participation in 2 year follow-up                                       | 64.1%                    | 77.8%                   | <0.001 |
| Participation in 5 year follow-up                                       | 57.8%                    | 71.6%                   | <0.001 |
| Child wheezed during first year (n = 1512)                              | 31.8%                    | 27.9%                   | 0.099  |
| Child wheezed during second year (n = 1374)                             | 15.4%                    | 15.2%                   | 0.900  |
| Child wheezed during fifth year (n = 1253)                              | 14.9%                    | 11.6%                   | 0.086  |
| Doctor diagnosed asthma at 5 years                                      | 13.7%                    | 10.3%                   | 0.064  |

<sup>a</sup> IQR = interquartile range.

**Table 2**

Associations between soil silt content based on 5-digit postcode at birth and childhood outcomes at one, two and five years.

| Outcome N 1048<br>n (%)                               | Unadjusted OR <sup>b</sup><br>[95% CI] | P     | Adjusted OR <sup>b</sup><br>[95% CI] <sup>b</sup> | P     | Adjusted OR <sup>b</sup><br>[95% CI] <sup>c,d</sup> | P     | OR maximum vs.<br>minimum silt |
|-------------------------------------------------------|----------------------------------------|-------|---------------------------------------------------|-------|-----------------------------------------------------|-------|--------------------------------|
| Wheeze 0–12 months, n = 241 (27%)                     | 1.21 [1.05–1.41]                       | 0.011 | 1.23 [1.04–1.46]                                  | 0.017 | 1.18 [1.00–1.40]                                    | 0.052 | 2.40                           |
| Wheeze without a cold 0–12 months, n = 73 (8.6%)      | 1.50 [1.18–1.91]                       | 0.001 | 1.43 [1.09–1.86]                                  | 0.009 | 1.36 [1.04–1.77]                                    | 0.023 | 5.10                           |
| Wheeze 12–24 months, n = 120 (15.1%)                  | 1.11 [0.92–1.35]                       | 0.286 | 1.20 [0.96–1.50]                                  | 0.115 | 1.22 [0.93–1.59]                                    | 0.149 |                                |
| Wheeze without a cold 12–24 months, n = 40 (5.1%)     | 1.22 [0.89–1.67]                       | 0.214 | 1.34 [0.94–1.92]                                  | 0.110 | 1.19 [0.86–1.65]                                    | 0.294 |                                |
| Wheeze 0–5 years (reported at 5 years), n = 148 (20%) | 1.09 [0.91–1.30]                       | 0.357 | 1.14 [0.93–1.40]                                  | 0.218 | 1.17 [0.91–1.49]                                    | 0.225 |                                |
| Wheeze 48–60 months, n = 87 (12%)                     | 1.27 [1.01–1.59]                       | 0.040 | 1.31 [1.02–1.70]                                  | 0.038 | 1.52 [1.04–2.21]                                    | 0.029 | 9.20                           |
| Wheeze without a cold 48–60 months, n = 46 (6%)       | 1.46 [1.08–1.97]                       | 0.013 | 1.58 [1.09–2.28]                                  | 0.015 | 1.32 [0.94–1.84]                                    | 0.105 |                                |
| Doctor diagnosed asthma at 5 years, n = 77 (10%)      | 1.37 [1.08–1.75]                       | 0.010 | 1.48 [1.12–1.97]                                  | 0.006 | 1.54 [1.04–2.28]                                    | 0.031 | 9.86                           |
| Active asthma at 5 years, n = 53 (7.4%)               | 1.37 [1.00–1.81]                       | 0.026 | 1.51 [1.09–2.11]                                  | 0.014 | 1.38 [1.01–1.88]                                    | 0.043 | 5.51                           |
| Allergic at 5 years, n = 86 (21.5%) <sup>a</sup>      | 1.09 [0.86–1.37]                       | 0.480 | 1.17 [0.89–1.54]                                  | 0.248 | 1.14 [0.84–1.53]                                    | 0.400 |                                |

<sup>a</sup> Assessed by skin prick testing of 700 children at 5 years.<sup>b</sup> Odds ratio expressed for 1 SD change in silt content expressed as percentage dry weight of <2 mm sieved soil.<sup>c</sup> Adjusted for maternal: atopy, smoking during pregnancy, age of leaving full time education, educational status; child's: sex, birth crown–heel length, birth head circumference, birth weight, antibiotics in the first year, birth order; paternal social class.<sup>d</sup> Multilevel analysis with children as observational units at a basic level and 5 digit postcode at a superior level.

was associated with an increased likelihood of wheezing with an odds ratio (per SD increase) of 1.20 (95% CI 1.05–1.37;  $p = 0.006$ ) and for wheeze in the absence of a cold with an odds ratio of 1.41 (1.18–1.69;  $p < 0.001$ ) in the first five years of life. These results suggest that children resident in the highest silt areas at birth were 2.6 times more likely to wheeze in the first five years of life when compared with children resident in the lowest silt areas; similarly, they were 6.2 times more likely to wheeze in the absence of a cold.

Soil silt content for postcode of birth was not associated with any of the potential confounders included in the multivariate models. Further analyses were conducted using soil composition based on 4 digit postcodes, using differing parameters of socio-economic status (Scottish Index of Multiple Deprivation [SIMD] based on postcode, highest educational qualifications), and replacing maternal allergic status based on skin prick test results with maternal and/or paternal history of asthma. None of these analyses substantially affected the associations reported above between soil silt and childhood asthma/wheeze and lung function outcomes. A further analysis using soil silt composition for residential postcode at five years revealed no associations between silt and five year outcomes. Restricting the analyses to those children remaining at the same postcode up to the age of 5 years made little difference to the magnitudes of the odds ratio and regression coefficients. Wheezing/asthma outcomes were not associated with children changing postcodes.

### 3.3. TIN vs. ordinary kriging

The TIN interpolation and ordinary kriging results at the validation set of 200 samples were compared using the difference between the measured value and estimated values at the 200 validation sites; there was strong correlation between the two methods (Spearman's rho 0.89,  $p < 0.001$ ). Boxplots, computed and plotted in R (The R Development Core Team, 2006), (Fig. 2) show little difference in the errors between using the TIN and ordinary kriging procedures.

The layout of the R boxplot is fully explained in the R reference manual (The R Development Core Team, 2006, pp. 615–620). The notches in the sides of the boxes in Fig. 2 overlap indicating that there is no significant difference in the median errors (95% level).

## 4. Discussion

This exploratory study arose from the realisation that by combining data from the SSD with a local prospective cohort study we could explore the hypothesis that early life exposure to soil components, especially particulate matter of different sizes influences the development of childhood asthma and allergic disease (von Hertzen and Haahtela, 2006). We envisaged that the results of this investigation could generate novel hypotheses for further investigation. If the hygiene hypothesis is true, we would expect associations between soil components and reduced likelihood of childhood asthma, wheeze and allergic outcomes. We found that these outcomes in the first five years of life were not associated with the soil composition and acidity (pH) in the area of residence at five years or with elemental soil composition or its acidity at birth. However, a high silt content in soil at the mother's (and presumably the child's) residential address at birth (but not at five years) was associated with an increased likelihood of childhood wheeze up to the age of five years and at that age with an increased likelihood of asthma, reduced lung function and increased FE<sub>NO</sub> levels. These associations were not attenuated by adjustment for major potential confounding factors, including socio-economic status. This fact, and the fact that the associations with silt are consistent across a number of outcomes both self-reported and physiological measures, adds weight to the inference that these results are highly unlikely to be due to chance alone. The associations with silt are not easily ignored, since children resident at birth in areas of the highest silt composition were nearly 10 times more likely to have active asthma at age five and had a 14% reduction in lung function when compared with children from low silt areas.

**Table 3**

Associations between soil silt content based on 5-digit postcode at birth and lung function outcomes at five years.

| 5 year outcome   | Regression coefficient $\beta^a$<br>[95% CI] | P     | Adjusted regression coefficient $\beta^b$<br>[95% CI] | P     | Adjusted regression coefficient $\beta^{b,c}$<br>[95% CI] | P     | % Difference maximum vs.<br>minimum silt |
|------------------|----------------------------------------------|-------|-------------------------------------------------------|-------|-----------------------------------------------------------|-------|------------------------------------------|
| PEF              | −4.31 [−6.86–−1.76]                          | 0.001 | −4.37 [−7.35–−1.39]                                   | 0.004 | −4.75 [−7.91–−1.58]                                       | 0.003 | 18%                                      |
| FEV <sub>1</sub> | −0.020 [−0.037–−0.004]                       | 0.016 | −0.024 [−0.045–−0.003]                                | 0.023 | −0.025 [−0.047–−0.001]                                    | 0.038 | 14%                                      |
| FVC              | −0.025 [−0.043–−0.008]                       | 0.004 | −0.024 [−0.046–−0.003]                                | 0.028 | −0.028 [−0.047–−0.008]                                    | 0.006 | 15%                                      |

All values of  $\beta$  are for 1 SD increase in soil silt content expressed as percentage dry weight of <2 mm sieved soil.<sup>a</sup> Adjusted for age, height and sex at 5 years.<sup>b</sup> Adjusted for maternal: atopy, smoking during pregnancy, age of leaving full time education, educational status; child's: sex, height, birth crown–heel length, birth head circumference, birth weight, antibiotics in the first year, birth order; paternal social class.<sup>c</sup> Multilevel analysis with children as observational units at a basic level and 5 digit postcode at a superior level.

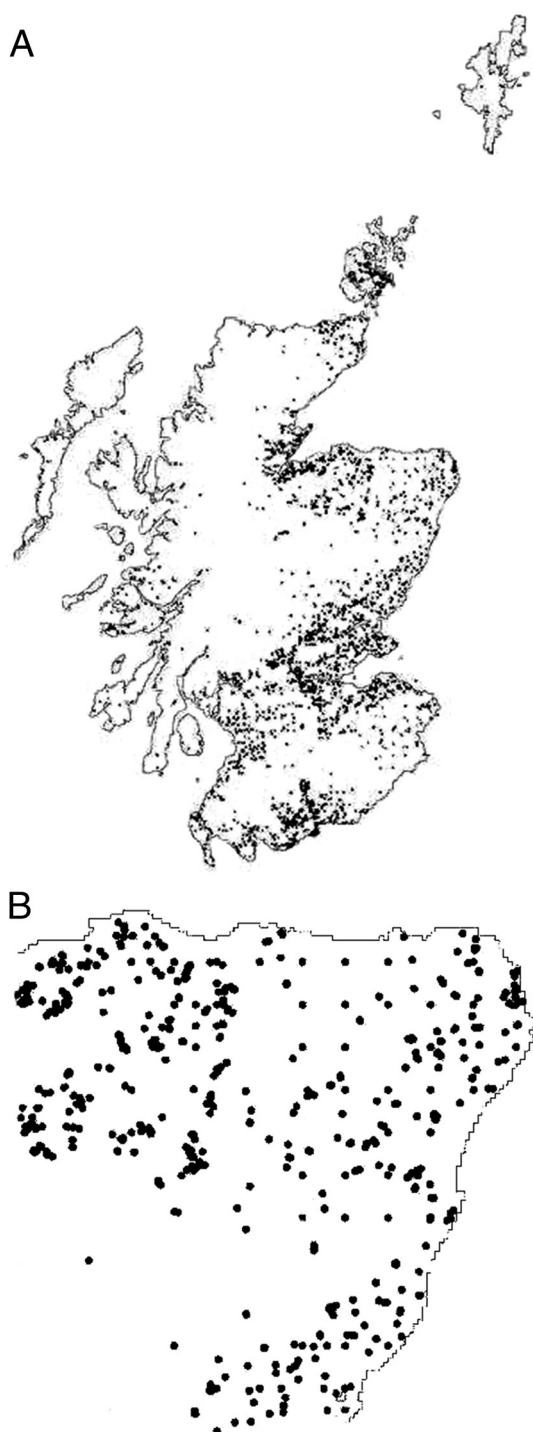


**Fig. 1.** Location of topsoil samples for Scotland a) for which silt content (2–20  $\mu\text{m}$  particle size) was available; b) more detailed locations of topsoil samples in North East Scotland used in this study.

Although several methods are available for the interpolation of spatial data each has its virtues and deficiencies. We chose the TIN method to estimate the silt concentration in soil at the postcode location at birth. Initially, this choice was made due to the large number of soil variables in our initial investigations. Once we identified silt as a possible factor, our comparison with ordinary kriging (Fig. 2) showed that there was little practical difference in the estimated values between the methods.

The strengths of this study come from the use of a cohort study recruited in utero and followed up at one, two and five years using

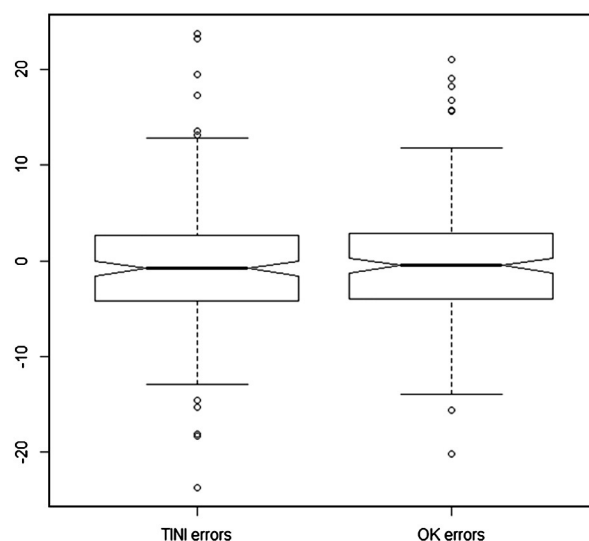


**Fig. 2.** Box and whisker plots of summary statistics for the estimated minus the predicted values for silt (2–20  $\mu\text{m}$  particle size) determined by TIN interpolation and ordinary kriging.

standard respiratory outcome questionnaires based on the ISAAC questionnaire and objective measurements of lung function at age five. As repeated questionnaires were used to collect information on wheezing symptoms, the effect of exposure to soil silt was further analysed using GEE methodology in order to control the within-child correlated observations between repeated measurements and provide a summary estimate over time, which produced similar results to support the cross-sectional associations between soil silt content in the residential area at birth and childhood wheeze and asthma. There are some weaknesses: as with most cohort studies there has been loss to follow-up that amounted to 35% at five years and those mothers participating in the study at five years differed from the original cohort in being from higher socio-economic strata, less likely to smoke and older (Devereux et al., 2007). However it is unlikely that continued participation is influenced by soil composition in the postcode area five years beforehand. A limitation of the soil database used is that because of the agricultural origins of the database we were only able to derive soil composition data for cultivated and hence predominantly rural areas. Furthermore because within the local major population centre of Aberdeen City the 5-character postcode districts are very small geographically, it was not possible to extrapolate meaningful soil composition data from database of soil samples collected outside the built up city area. Variations in the composition of rural, peri-urban and urban soils are however most likely to differ in elemental concentration and nutrient content and in one study in this region the mineralogy and particle sizes were similar across the urban–rural gradient with soils likely originating from common soil typologies (Paterson et al., 1996; Yang et al., 2006). It is possible that the adverse associations with silt reported in our study could be a chance finding consequent upon many tests of association between clinical outcomes and many soil constituents. The numerous univariate analyses did reveal some (one or two) associations between respiratory/allergic outcomes and some soil components with  $p$  values  $>0.01$ . These associations were considered most likely to be chance associations and taken no further. However, the associations with silt had lower  $p$  values ( $0.001 < p < 0.05$ ) and in addition were consistent, sizeable and biologically plausible. In an attempt to discover if any of the many univariate associations between soil constituents and clinical outcomes could be considered statistically significant, the Simes procedure was used, controlling the false discovery rate at 0.05. This indicated that the association between PEF at age 5 and soil silt content remained significant. Although we attempted to minimise residual

confounding by adjusting for many potentially confounding exposures, including socio-economic indices not based on postcode, as with all observational studies we are unable to exclude the possibility that the observed associations with silt are a consequence of residual confounding. Clearly before further detailed investigation on the possible association between asthma and silt is conducted, the adverse association with silt reported here needs to be replicated in other parts of the world where soil composition databases and respiratory based birth cohorts co-exist.

Respiratory responses to airborne dust may be altered by the duration of exposure, its concentration, or the presence of additional contaminants. This study did not consider exposure to outdoor or indoor air pollutants, e.g. traffic density outside the house, residential proximity to the main road, type of home heating system, etc. These factors are major sources of ambient PM<sub>2.5</sub> and PM<sub>10</sub>, exposure which contributes to respiratory morbidity (Boezen et al., 1999; McConnell et al., 1999; O'Connell et al., 2008). Neither did we consider time spent at home or outside the house or hygiene practises in the households. Besides mineral and elemental characteristics of soil, we did not consider potential respiratory health effects of exposure to a variety of organic compounds or microbial toxins derived from soil as these were not measured.

In the current study the observed associations between silt and respiratory symptoms, lung function and FE<sub>NO</sub> are consistent with a pro-inflammatory mechanism. However the absence of association with allergic outcomes, particularly eczema and allergic sensitisation suggests that any pro-inflammatory effect is limited to the airways. Our demonstration of associations with silt in the area of residence at the time of birth and not at five years, by which time about 50% of the children had moved from their residential postcode at birth, suggests that early rather than later life exposure to silt is relevant. This observation raises the possibility of silt contributing to the programming of airway disease during a critical time period of susceptibility during infancy, whereas exposure after this critical window of susceptibility in later childhood is less detrimental. Some animal studies have linked PM related airway inflammation with PM chemical composition, particularly the presence of metals (e.g. vanadium, nickel, zinc) (Batalha et al., 2002; Clarke et al., 2000; Saldiva et al., 2002). In the present study neither airway inflammation nor asthma symptoms nor lung function was associated with elemental (metal or non-metal) soil composition in the area of residence at birth.

Soils vary enormously in their microbial community composition, soil typology (Girvan et al., 2003) and land use (Yao et al., 2000) being major determining factors. However, currently the SSD does not include microbial composition data. Soils in Scotland are geologically diverse, comprising 140 different associations (i.e. parent materials) (Soil Survey Staff, 1984) and have a greater proportion of organic matter (median 26% organic carbon) than soils in England and Wales (median 3.6% organic carbon) (Paterson, 2011). Organic components of soil (low density vegetative residue and humic substances associated with small amounts of mineral matter) may preferentially become airborne (Delany and Zenchelsky, 1976). The physics of dust generation from soil are complex and inter-related (Chepil and Woodruff, 1963). The three main factors which alter the amount of dust generated from soil and suspended in the air by wind erosion are: (1) features of the surface material, (2) energy expended onto the surface, and (3) the atmospheric conditions. Particles of silt have similar shape to particles of sand but the pores between particles of silt are smaller and more numerous. Although silt retains water, silt does not have much stickiness or plasticity and soils with high silt content are susceptible to erosion by the forces of wind and water. The small degree of plasticity, cohesion and adsorptive capacity exhibited by some silt fractions is due to a film of adhering clay (Brady and Weil, 2008).

It is well recognised that soil is an important contributor not only to outdoor but also to indoor air pollution/air quality, and that the composition of house dust depends on the composition of soil outside

the house (Trowbridge and Burmaster, 1997). However, relatively little attention has been paid to the role of outdoor soil (Layton and Beamer, 2009; Trowbridge and Burmaster, 1997) in asthma and allergic disease. One study that did, the 1970 British Cohort Study, reported that exposure to soil in early life (playing in the garden) did not reduce the risk of developing asthma, eczema, or hay fever by the age of 26 years (Montgomery et al., 1999). With a particle size diameter of 2–20 µm, silt can contribute to the respirable particulate matter fraction of indoor and outdoor air (PM<sub>10</sub>). The adverse health effects of PM are well documented (Chen et al., 2008) with day-to-day exposures to particulates and ozone being associated with exacerbations of asthma. This is supported by mechanistic studies which show that PM<sub>10</sub> has a pro-inflammatory effect in the respiratory tract of murine models, enhances neutrophil infiltration and epithelial injury and causes oxidant stress (Li et al., 1997). The epidemiological evidence relating air pollution to the development of asthma is generally unresponsive despite mechanistic hypotheses that are very plausible (Committee on the Medical Effects of Air Pollutants, 2010).

The silt-size fraction itself consists of various microbiological constituents as well as organic matter and mineral particles and the associations reported in this study may potentially be mediated by bacterial and fungal components that are capable of interacting with the innate immune system. Given the microbial contribution to the organic component of silt it is tempting to speculate that some of the observed associations with silt could be the consequence of endotoxin within the silt organic fraction. Braun-Fahrlander et al. (2002) reported that endotoxin exposure was associated with an increased likelihood of non-allergic wheeze in children with a mean age of 9.5 years. Our reported adverse association between silt and wheeze would be consistent with an endotoxin mediated mechanism suggested by Braun-Fahrlander et al. (2002). Our reported association with silt provides an alternative explanation for the observation that stringent antenatal and early life dust mite eradication measures that reduce the level of house dust, whilst having minimal, if any effect on allergic sensitisation, has been reported to improve lung function (specific airway conductance) at age 3 years (Woodcock et al., 2004). Our findings raise the possibility that the beneficial effects of early life dust mite eradication may be a consequence of reductions in household silt.

## 5. Conclusions

The study has revealed for the first time that the silt content (2–20 µm particle size) of cultivated soils in the area of the residential home at birth is adversely associated with childhood asthma symptoms, lung function and airway inflammation at the age of five years. The study has highlighted a potentially important environmental factor in the aetiology of asthma and wheezing disease but further studies are needed to replicate the novel association reported here. If these findings are confirmed further research will be needed to analyse silt in more detail, make more accurate exposure measurements, identify factors which may lead to changes in silt content over time/space and identify “windows of exposure”, i.e. more detailed, individual-level analyses before the impact from exposure to soil silt on respiratory health can be established conclusively.

## Conflict of interest

None of the authors have conflict of interest.

## Acknowledgement

The work was supported by Asthma UK, Grant 02/017 and Scottish Government's Rural and Environment Science and Analytical Services (RESAS). We thank Mark Jones of the former Macaulay Land Use

Research Institute, Aberdeen for assistance with the preliminary numerical analysis of soil data.

## References

- Abrahams PW. Soils: their implications to human health. *Sci Total Environ* 2002;291:1–32.
- Batalha JR, Saldiva PH, Clarke RW, Coull BA, Stearns RC, Lawrence J, et al. Concentrated ambient air particles induce vasoconstriction of small pulmonary arteries in rats. *Environ Health Perspect* 2002;110:1191–7.
- Benjamini Y, Drai D, Elmer G, Kafkafi N, Golani I. Controlling the false discovery rate in behavior genetics research. *Behav Brain Res* 2001;125:279–84.
- Berrow ML, Stein WM. Extraction of metals from soils and sewage sludges by refluxing with aqua regia. *Analyst* 1983;108:277–85.
- Boezen HM, van der Zee SC, Postma DS, Vonk JM, Gerritsen J, Hoek G, et al. Effects of ambient air pollution on upper and lower respiratory symptoms and peak expiratory flow in children. *Lancet* 1999;353:874–8.
- Bouyoucos GJ. Directions for making mechanical analysis of soils by the hydrometer method. *Soil Sci* 1936;42:225–8.
- Brady NC, Weil RR. The nature and properties of soils. Upper Saddle River, NJ, USA: Pearson-Prentice Hall; 2008.
- Braun-Fahrlander C, Gassner M, Grize L, Neu U, Sennhauser FH, Varonier HS, et al. Prevalence of hay fever and allergic sensitization in farmer's children and their peers living in the same rural community. SCARPOL team. Swiss Study on Childhood Allergy and Respiratory Symptoms with Respect to Air Pollution. *Clin Exp Allergy* 1999;29:28–34.
- Braun-Fahrlander C, Riedler J, Herz U, Eder W, Waser M, Grize L, et al. Environmental exposure to endotoxin and its relation to asthma in school-age children. *N Engl J Med* 2002;347:869–77.
- Brown KWM, Gauld JH, Smith BFL, Bain DC, Burridge JC, Inkson RHE. Design of a database for Scottish soils. *J Soil Sci* 1987;38:267–77.
- Chapman HD. Cation-exchange capacity. In: Black CA, editor. *Methods of soil analysis. Part 2: chemical and microbiological properties*. Madison, Wisconsin, USA: American Society of Agronomy; 1965. p. 891–901.
- Chen H, Goldberg MS, Villeneuve PJ. A systematic review of the relation between long-term exposure to ambient air pollution and chronic diseases. *Rev Environ Health* 2008;23:243–97.
- Chepil WS, Woodruff NP. The physics of wind erosion and its control. *Adv Agron* 1963;15:211–302.
- Clarke RW, Coull B, Reinisch U, Catalano P, Killingsworth CR, Koutrakis P, et al. Inhaled concentrated ambient particles are associated with hematologic and bronchoalveolar lavage changes in canines. *Environ Health Perspect* 2000;108:1179–87.
- Committee on the Medical Effects of Air Pollutants (COMEAP). Does outdoor air pollution cause asthma? Statement COMEAP; 2010 [1–11–2011].
- Delany AC, Zenchelsky S. The organic component of wind-erosion-generated soil-derived aerosol. *Soil Sci* 1976;121:146–55.
- Devereux G, McNeill G, Newman G, Turner S, Craig L, Martindale S, et al. Early childhood wheezing symptoms in relation to plasma selenium in pregnant mothers and neonates. *Clin Exp Allergy* 2007;37:1000–8.
- Diggle PJ, Ribeiro Jr PJ. *Model-based geostatistics*. New York, USA: Springer; 2007.
- Ege MJ, Bieli C, Frei R, van Strien RT, Riedler J, Ublagger E, et al. Prenatal farm exposure is related to the expression of receptors of the innate immunity and to atopic sensitization in school-age children. *J Allergy Clin Immunol* 2006;117:817–23.
- Ege MJ, Mayer M, Normand AC, Genuneit J, Cookson WOCM, Braun-Fahrlander C, et al. Exposure to environmental microorganisms and childhood asthma. *N Engl J Med* 2011;364:701–9.
- Girvan MS, Bullimore J, Pretty JN, Osborn AM, Ball AS. Soil type is the primary determinant of the composition of the total and active bacterial communities in arable soils. *Appl Environ Microbiol* 2003;69:1800–9.
- Hough RL. Soil and human health: an epidemiological review. *Eur J Soil Sci* 2007;58:1200–12.
- International study of asthma and allergies in childhood steering committee. Worldwide variations in the prevalence of asthma symptoms: the International Study of Asthma and Allergies in Childhood (ISAAC). *Eur Respir J* 1998;12:315–35.
- Layton DW, Beamer PI. Migration of contaminated soil and airborne particulates to indoor dust. *Environ Sci Technol* 2009;43:8199–205.
- Li XY, Gilmour PS, Donaldson K, MacNee W. In vivo and in vitro proinflammatory effects of particulate air pollution (PM<sub>10</sub>). *Environ Health Perspect* 1997;105:S1279–83.
- Martindale S, McNeill G, Devereux G, Campbell D, Russell G, Seaton A. Antioxidant intake in pregnancy in relation to wheeze and eczema in the first two years of life. *Am J Respir Crit Care Med* 2005;171:121–8.
- McConnell R, Berhane K, Gilliland F, London SJ, Vora H, Avol E, et al. Air pollution and bronchitic symptoms in Southern California children with asthma. *Environ Health Perspect* 1999;107:757–60.
- Montgomery SM, Twamley SI, Murch SH, Pounder RE, Wakefield AJ. Soil exposure no protection against atopy. *Immunol Today* 1999;20:289–90.
- O'Connell S, Au-Yeung HK, Gregory CJ, Matthews IP. Outdoor and indoor respirable air particulate concentrations in differing urban traffic microenvironments. *J Toxicol Environ Health A* 2008;71:1069–72.
- Oliver MA. Soil and human health: a review. *Eur J Soil Sci* 1997;48:573–92.
- Openshaw S. The modifiable areal unit problem. Concepts and techniques in modern geography. Norwich, UK: Geo Books; 1984.
- Paterson E. In: Campbell CD, Coull M, Shand C, editors. *Geochemical atlas for Scottish topsoils*. Aberdeen, UK: The Macaulay Land Use Research Institute; 2011.
- Paterson E, Sanka M, Clark L. Urban soils as pollutant sinks - A case study from Aberdeen, Scotland. *Appl Geochem* 1996;11:129–31.
- Riedler J, Braun-Fahrlander C, Eder W, Schreuer M, Waser M, Maisch S, et al. Exposure to farming in early life and development of asthma and allergy: a cross-sectional survey. *Lancet* 2001;358:1129–33.
- Saldiva PHN, Clarke RW, Coull BA, Stearns RC, Lawrence J, Murthy GKG, et al. Lung inflammation induced by concentrated ambient air particles is related to particle composition. *Am J Respir Crit Care Med* 2002;165:1610–7.
- Sing D, Sing CF. Impact of direct soil exposures from airborne dust and geophagy on human health. *Int J Environ Res Public Health* 2010;7:1205–23.
- Singh BK, Campbell CD, Sorenson SJ, Zhou J. Soil genomics. *Nat Rev Microbiol* 2009;7:756.
- Smith BFL, Bain DC. A sodium hydroxide fusion method for the determination of total phosphate in soils. *Commun Soil Sci Plant Anal* 1982;13:185–90.
- Smith JL, Lee K. Soil as a source of dust and implications for human health. *Adv Agron* 2003;80:1–32.
- Soil Survey Staff. Organisation and methods of the 1:250 000 soil survey of Scotland. Aberdeen, UK: Macaulay Institute for Soil Research; 1984.
- Strachan DP. Hay fever, hygiene, and household size. *Br Med J* 1989;299:1259–60.
- The R Development Core Team. R: a language and environment for statistical computing reference index. Vienna, Austria: R Foundation for Statistical Computing; 2006.
- Trowbridge PR, Burmaster DE. A parametric distribution for the fraction of outdoor soil in indoor dust. *J Soil Contam* 1997;6:161–8.
- von Hertzen L, Haahtela T. Disconnection of man and the soil: reason for the asthma and atopy epidemic? *J Allergy Clin Immunol* 2006;117:334–44.
- Woodcock A, Lowe LA, Murray CS, Simpson BM, Phipps SD, Kissen P, et al. Early life environmental control: effect on symptoms, sensitization, and lung function at age 3 years. *Am J Respir Crit Care Med* 2004;170:433–9.
- Yang YG, Campbell CD, Clark L, Cameron CM, Paterson E. Microbial indicators of heavy metal contamination in urban and rural soils. *Chemosphere* 2006;63:1942–52.
- Yao H, He Z, Wilson MJ, Campbell CD. Microbial biomass and community structure in a sequence of soils with increasing fertility and changing land use. *Microb Ecol* 2000;40:223–37.