Residing near allergenic trees can increase risk of allergies later in life: LISA Leipzig study

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Sources of funding

The LISA study was mainly supported by grants from the Federal Ministry for Education, Science, Research and Technology and in addition from Helmholtz Zentrum Munich (former GSF), Helmholtz Centre for Environmental Research—UFZ, Leipzig, Marien-Hospital Wesel, Pediatric Practice, Bad Honnef, for the first 2 years. The 4- year, 6-year, and 10-year follow-up examinations of the LISA study were covered from the respective budgets of the involved partners (Helmholtz Zentrum Munich [former GSF], Helmholtz Centre for Environmental Research—UFZ, Leipzig, Marien-Hospital Wesel, Pediatric Practice, Bad Honnef, IUF—Leibniz-Research Institute for Environmental Medicine) and in addition by a grant from the Federal Ministry for Environment (IUF, FKZ 20462296). The 15-year follow-up examination of the LISA study was supported by the Commission of the European Communities, the 7th Framework Program (Mechanisms of the Development of Allergy [MeDALL] project).

This analysis was supported by Förderprogramm für Forschung und Lehre (FöFoLe) granted by the Medical Faculty of Ludwig Maximilian University (Reg.-Nr. 1014).

Iana Markevych is currently supported from the “NeuroSmog: Determining the impact of air pollution on the developing brain” (Nr. POIR.04.04.00-1763/18-00) which is implemented as part of the TEAM-NET programme of the Foundation for Polish Science, co-financed from EU resources, obtained from the European Regional Development Fund under the Smart Growth Operational Programme.

Air pollutants were assessed under contract to the Health Effects Institute (HEI), an organization jointly funded by the United States Environmental Protection Agency (EPA) (Assistance Award No. R-82811201) and certain motor vehicle and engine manufacturers.

The aforementioned funding sources had no involvement in the design of the study, collection, analysis, and interpretation of data, writing of the report, and decision to submit the manuscript for publication.

Word count: 4,310

Abstract

Background: We investigated whether residing in places with higher greenness, more trees and more allergenic trees early in life increases the risk of allergic outcomes, and whether these associations differ depending on the concentration of air pollutants.

Methods: The analytic sample included 631 children from the German birth cohort LISA Leipzig. Asthma and allergic rhinitis, sensitization to aeroallergens and food allergens, as well as confounders, were collected prospectively up to 15 years. Greenness was assessed by Normalized Difference Vegetation Index (NDVI). A tree registry was used to derive information on trees, which were classified into allergenic and non-allergenic. Annual average concentrations of nitrogen dioxide (NO_2) and ozone were also used. Geographic exposures were assigned to home addresses at birth. Longitudinal associations were analysed using generalized estimating equations.

Results: Medium and high numbers (tertiles) of trees and allergenic trees in a 500 m buffer around birth addresses were associated with increased odds of allergic rhinitis up to 15 years regardless of NDVI. These exposures were also related to higher odds of sensitization to aeroallergens. Associations with asthma and sensitization to food allergens were less consistent. Effect estimates for allergic rhinitis were stronger in the high tertile of NO_2 compared to the low tertile, while an opposite tendency was observed for ozone.

Conclusion: We observed that early life residence in places with many trees, and allergenic trees specifically, may increase the prevalence of allergic rhinitis later in life. This association and its modification by air pollution should be pursued in further studies.

Capsule summary

Residing in places with many trees and many allergenic trees in early life may increase the risk of allergic rhinitis up to 15 years regardless of general level of vegetation.

Clinical implications

Planting tree species with low allergenic potential might reduce the risk of allergic rhinitis in urban residents.

Key words: allergic rhinitis; atopy; asthma; epidemiology; children; greenness; greenspace.

Abbreviations

CI	Confidence interval
ELAPSE	Effects of Low-Level Air Pollution: A Study in Europe
IgE	Immunoglobulin E
GAM	Generalized additive model
GEE	Generalized estimating equation
LISA	Influence of Life-Style Factors on the Development of the Immune System and Allergies in East and West Germany
LUR	Land use regression
NDVI	Normalized Difference Vegetation Index
NO_2	Nitrogen dioxide
OR	Odds ratio

Introduction

Greenspace is related to various beneficial health outcomes, including better mental health and well-being [1], reduced risk of all-cause mortality [2] and type 2 diabetes [3], improved pregnancy outcomes and decreased blood pressure [4], among others. These benefits are hypothesized to be due to the restorative potential of greenspace, reduced concomitant exposure to harmful environmental influences, as well as because greenspace provides settings for physical activity and social contacts which in turn improve physical and mental health. [5]

Mechanistic links between greenspace, asthma and allergic outcomes seem to be more complicated to resolve than those for other health indicators. On the one hand, greenspace was hypothesized to reduce the risk of allergic diseases by similar mechanisms as does living on a farm, [6] namely via increased contact with microbes and consequent modulating of immune responses and allergic inflammation. [7] On the other hand, greenspace is a source of allergenic pollen, short-term exposure to which can exacerbate asthma and allergic symptoms as studies based on pollen counts have demonstrated. [8]

It is not surprising then that greenspace research into allergic health outcomes stagnates. The published studies have typically reported associations, but those were in different directions depending on the study area. [9,10] One example for such diverging directions is our own study where higher greenness (i.e., vegetation degree) was associated with increased risk of allergic rhinoconjunctivitis and aeroallergen sensitization in the Munich study area, but with reduced risk of these outcomes in the Wesel area of the same multicentre birth cohort. [11] We failed to explain such inconsistency but managed to replicate it in five additional cohorts across three continents.[12]

Besides the complicated and potentially conflicting underlying mechanisms, the lack of consistency in greenspace-allergy studies was also blamed on metrics of greenspace that are too crude to differentiate between different types of vegetation. [12] Most of the existing research used either land use or land cover-derived distance to or amount of different types of green spaces, or satellite-derived vegetation indices capturing greenness. None of these metrics can distinguish trees from herbaceous vegetation, let alone allergenic from non-allergenic plant species.

Keeping all this in mind, we decided to explore whether residing in places with overall higher greenness, more trees and specifically more allergenic trees early in life can contribute to the onset of allergic diseases, or whether such exposures act as protective factors. To our knowledge, no study so far looked at the long-term exposure to allergenic trees and development of allergic diseases –

partially because detailed vegetation geodata are very scarce and partially because expert knowledge is needed to classify species into allergenic and non-allergenic. This study was possible thanks to the collaboration between environmental epidemiologists and plant biologists.

Since elevated air pollution levels are observed to increase the allergenicity of pollen, [13] as a secondary objective, we investigated whether the associations between greenness, trees and allergenic trees and allergic diseases differ depending on the concentration of several air pollutants. In addition, vegetation and air pollution are inversely spatially correlated and, depending on the spatial arrangement, composition and temperature, trees can remove, trap, or even emit air pollution. [14]

Methods

Study design and population

"Influence of **L**ife-Style Factors on the Development of the **I**mmune **S**ystem and **A**llergies in East and West Germany" (LISA) is an ongoing multicentre population-based German birth cohort. [15,16] Briefly, healthy full-term neonates with a normal birth weight were recruited at selected maternity hospitals in Munich, Leipzig, Wesel and Bad Honnef between 1997 and 1999. Parent-completed questionnaires were administered at birth and at ages 0.5, 1, 1.5, 2, 4, 6, 10 and 15. Blood samples were drawn from a subset of children at the ages of 2, 6, 10 and 15 years. For the purpose of this study, we utilized the data from 631 children from Leipzig (for details, see the flowchart, Figure E1). The LISA Leipzig cohort was approved by the local ethics committee at the University of Leipzig (560/1998; 206/2003; 345/2007) and the State Medical Council of Saxony (EK-BR-02/13-1), and informed consent was obtained from all families.

Allergic outcomes

Data on parent-reported doctor diagnosis of asthma and allergic rhinitis (AR) were collected at ages 0.5, 1, 1.5, 2, 4, 6, 10 and 15. If the time between follow-ups was longer than one year (e.g., 10 to 15 years), the presence of the diagnosis was asked separately for each year since the previous follow-up (e.g., 10, 11, 12, 13, 14 and 15). In line with our previous analyses, [17] only information collected from age 3 onwards was used due to difficulty of accurate diagnosis of these outcomes at younger ages. Asthma at ages Y = 3, 4, … 15 was defined as a positive response to "Was your child diagnosed with asthma by a doctor at the age of Y years?" AR at ages Y = 3, 4, … 15 was defined as a positive response to "Was your child diagnosed with hay fever/allergic rhinitis by a doctor at the age of Y years?"

Specific immunoglobulin E (IgE) against common allergens was assessed at ages 2, 6, 10 and 15 using the standardized CAP-RAST FEIA method (ThermoFischer, Freiburg, Germany). At 2 years, allergic sensitization was tested to tree and grass pollen (RX1), mold, cat and house dust mites. At 6, 10 and 15 years, allergic sensitization was tested by using inhalant mix SX1, which consists of house dust mite, cat, dog, mold, birch, rye, mugwort and timothy grass allergens. Allergic sensitization to aeroallergens was defined as IgE > 0.35 kU/L for at least one of the tested inhalant allergens at 2 years and for the SX1 mix at the other timepoints. Allergic sensitization to food allergens was defined as IgE > 0.35 kU/L for allergens in the FX5 screening test (milk, peanut, eggs, soya, cod and wheat flour) at all timepoints.

Greenness, trees and allergenic trees

The Normalized Difference Vegetation Index (NDVI) [18] was used as an indicator of greenness (i.e. vegetation degree). NDVI is constructed from the ratio of reflected light to absorbed light in two vegetation-informative bands of the electromagnetic spectrum: the red band (RED) and the near infrared band (NIR). NDVI is calculated as NDVI = (NIR − RED) / (NIR + RED). Thus, NDVI ranges from -1 (water) through 0 (barren areas) to +1 (areas completely covered by vegetation).

For our NDVI calculations, we used a single cloud-free Landsat 5 TM satellite image at a resolution of 30 by 30 m taken on June 6, 1997 (https://earthexplorer.usgs.gov/) which is about the time when recruitment for the study started. Summer images enable maximum spatial contrasts of greenness. [19]

Tree registry data of the city of Leipzig were utilized to derive information on trees. Briefly, data are available for street trees and trees in green spaces and include tree geocoordinates, tree species in binomial and German nomenclature and year of planting. Only trees planted before 1998 were considered for this analysis: 63,579 trees from at least 82 genera. Figure E2 shows the spatial distribution of trees superimposed over an NDVI map.

We used two definitions to classify tree genera into allergenic and non-allergenic. Both definitions are based on the same three criteria. For a tree genus to be classified as allergenic according to definition 1, it had to satisfy two out of the three criteria. The stricter definition 2 required that all three criteria be satisfied. The first criterion is satisfied by tree genera whose pollen are being routinely monitored in Germany (http://www.pollenstiftung.de/pollenvorhersage/pollenflug-kalender). The second and third criteria relate to tree species and are satisfied by a genus if among the trees of that genus that were planted in Leipzig before 1998 there is at least one species that satisfies the criterion. The second criterion is satisfied by a species if its pollen allergens were characterized in a published study, or if it was described by a study as causing allergenic reactions in people that came into contact with its pollen (e.g. measured by skin prick test), or if there are reports about its sensitization/allergenicity rates. Literature search was done via Google Scholar in combination with the library access service of Technical University Munich (TUM OPAC plus). The third criterion is satisfied by a species if www.allergen.org, the most complete database of allergens, had at least one aeroallergen listed for it as of September 2, 2019. The details on all tree genera of the city of Leipzig planted before 1998, together with their abundances, are provided in Table E1. According to definition 1, 21,324 trees from the following 16 genera (sorted by tree count in descending order) were classified as allergenic: *Fraxinus* (ash)*, Quercus* (oak)*, Platanus* (plane)*, Prunus* (prunus)*, Betula* (birch)*, Populus* (poplar)*, Carpinus* (hornbeam)*, Corylus* (hazel)*, Salix* (willow)*, Pinus* (pine)*, Fagus* (beech)*, Alnus* (alder)*, Sambucus* (elder)*, Castanea* (chestnut)*, Syringa* (lilac) *and Juniperus* (juniper). According to definition 2, there were 9,453 allergenic trees from 7 genera: *Fraxinus, Betula, Carpinus, Corylus, Fagus, Alnus* and *Syringa*.

Mean NDVI, total number of trees and number of allergenic trees according to the two definitions were calculated for circular buffers of 100, 500 and 1,000 m around each participant's home address at birth.

Geographic data pre-processing and calculations were conducted with ArcMap 10.4 and ArcGIS Pro 2.2 Geographical Information System (ESRI, Redlands, CA, USA).

Air pollution

The annual average concentrations of air pollutants at residential addresses at birth were assigned from raster surfaces at a resolution of 100 x 100 m. Estimates for nitrogen dioxide (NO_2) and warm season ozone were available for the year 2000 from the ELAPSE project. [20] Both air pollutants were modelled by hybrid land use regression models using data from AirBase, a database of routine air pollution measurements in the EU member states. The hybrid models for western Europe that used dispersion model estimates, land use and road data and for ozone also kriging on the residuals, were able to explain 60 % and 82 % of the variation in measured NO_2 and ozone concentrations, respectively.

Statistical analysis

Given the repeated assessment of allergic outcomes, generalized estimating equations (GEE) with logit link and exchangeable correlation structure were used to regress the prevalence of allergic outcomes up to 15 years on the exposures of interest at birth.

First, we used generalized additive models (GAM) [21] to test the non-linearity of the univariate relationships between each pair of exposure and "ever" outcome (e.g., ever-asthma is a constructed outcome variable that is 1 if at least one of the age-specific asthma variables is 1 and otherwise 0). We discovered that associations deviated from linearity in many cases. Therefore, we categorized NDVI variables and tree counts for the 500 m and 1000 m buffers into tertiles. Tree counts for the 100 m buffer were dichotomized (= 0 *vs* ≥ 1).

In line with our previous analyses on greenspace and allergic outcomes, [11,12] we considered the 500 m buffer variables our main exposures of interest. The main models were *a priori* adjusted for age, sex, family history of allergic diseases, parental education (based on the highest number of years of school education of either parent: <10 years, =10 years, >10 years, according to the German educational system) and season of birth (October to January, February to March, May to July, August to September, corresponding to next to no pollen, tree pollen season, grass pollen season, ragweed and mugwort pollen season, respectively). Season of birth was categorized according to a regional pollen calendar (http://www.pollenstiftung.de/pollenvorhersage/pollenflug-kalender).

A series of sensitivity analyses were performed. This included crude analyses, which were adjusted only for age, and additionally adjusted analyses with main models also controlled for maternal smoking during pregnancy and tobacco smoke exposure at home until age 4 (yes, likely no, no), presence of older siblings, exclusive breastfeeding during the first four months and birth weight (grams). We also checked the robustness of the results by using 100 m and 1,000 m buffers. Moreover, the models for total tree count and allergenic tree count according to definitions 1 and 2 were additionally adjusted for NDVI to account for vegetation not captured by the tree registry, e.g., trees on private grounds, herbs, bushes. Furthermore, we reran the analyses excluding participants with partially missing outcome data. Effect modification by age was tested by introducing an interaction term between the exposure variable and age. Additionally, we stratified our analyses by whether participants changed their place of residence between birth and 2 years of age. Finally, we checked effect modification by whether participants resided within 300 m from the nearest urban green space or forest of at least 1 ha [22]. Green spaces were derived using the Urban Atlas land use data for the year 2006 (http://www.eea.europa.eu/data-and-maps/data/urban-atlas).

To check whether air pollution modifies the association of interest, an interaction term between the air pollutant and the exposure variable was introduced. Additionally, models were stratified by tertiles of NO_2 and summer ozone levels. In all stratified analyses, we combined low and medium categories of parental education, as there were sometimes too few cases in the lowest educational category.

Data pre-processing and statistical analyses were done using the statistical software R 3.6.1 (Vienna, Austria). [23] GEE models were fitted by the *geeglm()* function from the *geepack* package. [24] GAM models were executed using the *gam()* function from the *mgcv* package. [25]

Results

Descriptive characteristics of the study population and exposure variables

Over half of the children (55.2 %) were from families with high level of parental education while only 2.4 % were from families with low level of parental education (Table 1). This was the only systematic difference between the analytic sample compared to the original LISA Leipzig population where the above-mentioned numbers were 48 % and 5 %, respectively.

The highest prevalence of doctor diagnosed asthma (4.5 %) was observed at 7 and at 11 years (Table 2). The highest prevalence of AR (12.2 %) was observed at 11 years. 47.4 % children at 15 years were sensitized to aeroallergens. The peak of sensitization to food allergens (16.5 %) was reached at 10 years.

Descriptions of the exposure variable categories for all three buffer sizes are provided in Table E2. Mean and standard deviations for NO_2 and summer ozone levels were 33.5±4.5 µg/m^3 and 84.7±3.9 µg/m^3, respectively.

Greenness, trees and allergenic trees and allergic outcomes: main results

Birth home addresses surrounded by medium greenness (second NDVI tertile) in a 500 m buffer were associated with increased prevalence of asthma up to 15 years (odds ratio (OR) = 1.46) (Table 3). Home addresses with high greenness (third NDVI tertile) showed decreased prevalence of asthma (OR = 0.44). Medium numbers of allergenic trees (second tertile) according to definition 1 were related to higher prevalence of asthma (OR = 1.92). This was not the case for definition 2. Medium and high numbers of trees and allergenic trees according to the both definitions were associated with higher prevalence of AR (Table 3). All exposures were related to higher allergic sensitization to aeroallergens, and high numbers of allergenic trees according to definition 1 were related to food sensitization.

Greenness, trees, allergenic trees and allergic outcomes: results from sensitivity analyses

Effect estimates were similar when we adjusted the three models with tree exposure variables additionally for NDVI (Table 3). Similar associations were observed for the 100 m (Table E3) and 1,000 m (Table E4) buffers, with some exceptions. In case of the 100 m buffer, associations between NDVI and asthma, the association between allergenic trees definition 1 and food sensitization, and the associations of total number of trees and NDVI with aeroallergen sensitization were not present. For the 1,000 m buffer, no associations with aeroallergen sensitization were observed and the association between medium NDVI and asthma was lost.

Effect estimates from crude and additionally adjusted models were very similar to those from the main models (Table E5). The same was true when children with partially missing outcome variables were excluded (data not shown). The associations between trees and asthma, allergenic trees definition 2 and asthma, and NDVI and aeroallergen sensitization got stronger with age. Effect modification by moving between birth and 2 years did not reveal a distinct pattern (Figure E3). Similarly, a mixed pattern was observed considering effect modification by residing close to structured green spaces (Figure E4).

Effect modification by air pollutants

The associations of trees and allergenic trees definition 1 with higher AR and asthma were stronger in the high tertile of NO_2 compared to the low tertile of NO_2 (Figure 1). The same holds for both definitions of allergenic trees and sensitization to aeroallergens. The associations of trees with aeroallergen sensitization and of allergenic trees definition 2 and food sensitization showed the opposite tendency. The associations of trees and allergenic trees and AR were stronger for the low ozone tertile compared to the high ozone tertile, the same was observed for the association between allergenic trees and aeroallergen sensitization (Figure E5).

Discussion

The results of our longitudinal analyses in 631 children from the city of Leipzig, Germany, show that early life residence in places with many trees and allergenic trees specifically, was consistently associated with increased prevalence of AR up to 15 years and, to a lesser extent, also with increased prevalence of allergic sensitization to aeroallergens. Associations with asthma and sensitization to food allergens were inconsistent. Associations with greenness, as measured by NDVI, were either inconclusive (for asthma) or mostly non-significant (for other outcomes). Effect estimates for trees and allergenic trees and AR tended to be stronger in high NO_2 and low ozone settings.

Comparison with previous findings and interpretations

We are not aware of previous studies on long-term exposure to allergenic trees and allergic outcomes. Many studies, however, considered NDVI in relation to asthma, AR and sometimes to allergic sensitization to aeroallergens. There are also several studies on trees and allergic outcomes.

Residential greenness and allergic outcomes

In line with our observations, the reported associations for NDVI and asthma were heterogeneous and, in some cases, non-linear. Like in our study, medium but not high NDVI increased the risk of asthma in 7,040 Taiwanese children. [26] Also in line with us, exposure to medium but not high greenness was associated with asthma in the Euro-Siberian climatic region of a Spanish multicentre study of 2,472 children. [27] Nevertheless, high greenness was protective against wheezing in the same region of the same study. [27] Higher NDVI in the 100 m buffer, but not in larger buffers, was related to increased asthma risk in 1,489 Kaunas children, and this association was stronger in children living further away from parks. [28] We observed opposite tendencies. Like our protective association between the high NDVI tertile and asthma, two other urban studies – one in 1,915 Chicago children [29] and another one in 187 Turin children [30] – reported protective greenness effects on asthma. Higher greenness was also related to lower asthma risk in a large New Zealand study of 49,956 children. [31] Nevertheless, an even larger study (n = 65,000) from the Vancouver Metropolitan Area failed to detect such an association. [32] Null findings between NDVI and asthma were also reported in 3,178 Spanish children [33] and 5,643 children from Suzhou, China. [34]

We did not observe any association between NDVI and AR and the association between NDVI and aeroallergen sensitization was inconsistent. This is in line with the aforementioned studies by Tischer et al., [27] Dadvand et al. [33] and Li et al. [34] In addition, Gernes et al. [35] reported null findings between NDVI and rhinitis and atopic sensitization in 425 children from the USA. Our two previous multi-centre studies, whose study populations were distinct from the one used in this analysis, reported differential associations with AR, nose and eye symptoms and aeroallergen sensitization, as is mentioned in the Introduction. [11,12] Interestingly, effect modification by moving did not reveal any consistent trends in Fuertes et al., [12] in line with our present analysis. Unlike our stratified results by air pollutants, no pattern was observed in analyses stratified by NO_2 subgroups in these two studies. [11,12]

Residential trees and allergic outcomes

Other studies on trees and allergic outcomes did not observe an association with AR. [35,36] This contrasts with our analysis where this association was the strongest and the most consistent. Yet, Lovasi et al. [36] reported increased risks of asthma and allergic sensitization in 549 7-year-old residents of New York, which was partially in line with us. Both studies used tree cover extracted from aerial images. Tree cover does not capture differences between trees.

We used data from a tree registry to distinguish between allergenic and non-allergenic trees. Our results for AR and to a lesser extent for aeroallergen sensitization suggest that in Leipzig having any trees close to home has about the same negative health effect as having allergenic trees close by. This might not be the case for other study areas. Future research might show that comparisons of findings concerning trees and allergic outcomes are only valid between cities with similar tree patterns, air quality and urban climate. [37]

No epidemiological study so far had enough information to adequately deal with the problem that pollen loads as well as allergenic potentials may vary between tree individuals and for the same tree individuum, also across years. [38] For most allergenic tree species this phenomenon has not yet been explored let alone been understood.

Effect modification by air pollutants

Trees and allergenic trees were associated with a higher prevalence of AR when NO_2 levels were comparatively high or ozone was comparatively low. Apart from the direct detrimental effect of nitric oxides on respiratory health, [39,40] these substances may have varying effects on the allergenicity and amount of pollen depending on plant species and environmental conditions. For example, a number of molecular studies reviewed by Frank and Ernst [41] found increasing allergenicity (tested by increased IgE reactivity) of NO_2-treated pollen of the common species *Acer negundo*, *Betula pendula*, *Ostrya carpinifolia* and *Carpinus betulus*, [42,43] while Beck et al.[13] did not find a correlation of birch (*Betula pendula*) pollen allergenicity with NO_2 levels. Divergent findings were also observed for ozone treatments. [41] In addition, NO_2 and ozone may have different effects on pollen allergenicity even within the same species. [44] The effect of these pollutants on a larger scale is further blurred by the chemical reactions between ozone and NO_2 that depend atmospheric conditions. [45,46] This makes it difficult for epidemiological studies like ours to investigate their influence on the associations of trees and allergenic trees with allergic outcomes.

Implications for city greening

Cities and their rural surroundings are often quite different environments in terms of their tree composition. For Leipzig, the overlap between tree genera planted in the city and those growing in regional forests is small. Unlike Leipzig, the surrounding forests consist mostly of spruce and pine (https://bwi.info, data for Saxony, accessed 2020-03-07), that is, taxa of no or minor importance as allergen producers (cf. Table E1). By planting trees in proportions found in natural habitats and eliminating highly allergenic taxa [47,48] city greening may become even more beneficial.

Attention to allergenic potential is also important when selecting tree species that are able to withstand the challenges posed by climate change. Resilient tree taxa [49-51] may prove detrimental for the development of allergenic diseases. Examples include *Alnus x spaethii* [52] and *Corylus colurna,* a close relative of the major allergen source *Corylus avellana,* which despite their sturdiness should be reconsidered as future city trees.

Implications for future research

We argue that NDVI and other vegetation indices are not just unable to differentiate vegetation types but are also easily confounded by urbanicity and thus air pollution. When investigating associations where knowing the amount of greenspace or the distance to greenspace is not enough, like with allergic outcomes, we suggest that more detailed vegetation measures be considered. In our study, number of trees appeared to be a good proxy for number of allergenic trees, but this unlikely to always be the case across study areas. Digital tree registry data are scarce but available for some cities, including New York, Frankfurt am Main, Vienna, Hamburg and Melbourne. Similarly, rich datasets are available for different areas, like the maps of 12 key allergenic taxa in the UK for the year 2012 in 1 x 1 km resolution produced by McInnes et al. [53]

Strengths and limitations

Besides the modest sample size and somewhat limited external validity due to loss to follow-up, primarily of families with low school education, the largest limitation of our study is that it is based on a single study area. Therefore, our findings cannot be generalized to other areas. Future studies should further investigate our observed associations in urban, suburban and rural contexts across different climates and countries. Moreover, we cannot exclude residual confounding of our findings by allergenic shrubs and herbs and even by trees on private grounds, as these were not part of the tree registry. We co-adjusted our models with tree exposures for general vegetation degree, but this is not sufficient to rule out potential impacts of other allergenic vegetation. Limited sample size together with relatively low prevalence of asthma and AR precluded us from testing more effect modifiers, in particular, socioeconomic status. Nevertheless, LISA is an established cohort study meant to explore various risk factors of allergic diseases. All allergic outcomes were collected by using state-of-the-art methods at many timepoints. Repeated measurements analysis provides enough power to detect even small associations. The impact of confounding was reduced by controlling for many perinatal and early postnatal allergy risk factors. Finally, we were able to classify all tree genera into allergenic *vs* non-allergenic by several different criteria, something that was never done before in epidemiological analyses.

Conclusions

We observed that early life residence in places with many trees and specifically many allergenic trees may increase prevalence of AR later in life regardless of general level of vegetation. This association, as well as its modification by nitrogen dioxide and ozone, needs replication in further studies.

Conflict of interest

The authors declare that they have no conflict of interest.

Acknowledgements

We thank all children and parents for their cooperation, and all technical and administrative support staff and medical and field work teams. We are also grateful to all members of the LISA Study Group. The LISA Study group consists of the following: Helmholtz Zentrum München, German Research Center for Environmental Health, Institute of Epidemiology, Munich (Heinrich J, Schnappinger M, Brüske I, Ferland M, Schulz H, Zeller C, Standl M, Thiering E, Tiesler C, Flexeder C); Department of Pediatrics, Municipal Hospital "St. Georg", Leipzig (Borte M, Diez U, Dorn C, Braun E); Marien Hospital Wesel, Department of Pediatrics, Wesel (von Berg A, Berdel D, Stiers G, Maas B); Pediatric Practice, Bad Honnef (Schaaf B); Helmholtz Centre of Environmental Research – UFZ, Department of Environmental Immunology, Leipzig (Lehmann I, Bauer M, Röder S, Schilde M, Nowak M, Herberth G, Müller J); Technical University Munich, Department of Pediatrics, Munich (Hoffmann U, Paschke M, Marra S); Clinical Research Group Molecular Dermatology, Department of Dermatology and Allergy, Technische Universität München (TUM), Munich (Ollert M, Grosch J).

We would also like to thank Sebastian Fried, Sachgebiet Stadtbäume, Amt für Stadtgrün und Gewässer, Stadt Leipzig, Germany, for providing us Leipzig tree registry ("Baumkataster") data without which this analysis would not have been possible.

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Table 1. Descriptive characteristics of the analytic sample

Characteristic	**Category**	**n/N**	**%**
Season of birth	October to January	224/631	35.5
	February to April	169/631	26.8
	May to July	145/631	23.0
	August to September	93/631	14.7
Sex	Female	324/631	51.3
	Male	307/631	48.7
Highest parental education	Low (<10 years)	15/631	2.4
	Medium (=10 years)	268/631	42.5
	High (>10 years)	348/631	55.2
Parental atopy	No	342/631	54.2
	Yes	289/631	45.8
Older siblings	No	416/631	65.9
	Yes	215/631	34.1
Smoking during pregnancy	No	514/612	84.0
	Yes	98/612	16.0
Smoking at home between birth and 4 years	Never	318/629	50.6
	Likely never	56/629	8.9
	Ever	255/629	40.5
Exclusive breastfeeding first 4 months	No	262/619	42.3
	Yes	357/619	57.7
Birth weight (grams)	Mean and SD	3509.0	453.9
Changed place of residence between birth and 2 years	No	471/631	74.6
	Yes	160/631	25.4

SD – standard deviation; n – number of participants in the category; N – number of participants with available data.

Table 2. Prevalence of allergic outcomes in the analytic sample at all ages

Age, years	Asthma		Allergic rhinitis		Aeroallergen sensitization		Food sensitization	
	n/N	%	n/N	%	n/N	%	n/N	%
2					26/533	4.9	59/534	11.0
3	3/534	0.6	8/535	1.5				
4	7/524	1.3	13/520	2.5				
5	11/459	2.4	19/462	4.1				
6	12/461	2.6	29/461	6.3	76/262	29.0	37/262	14.1
7	16/356	4.5	24/353	6.8				
8	11/356	3.1	27/355	7.6				
9	13/356	3.7	31/354	8.8				
10	10/356	2.8	40/353	11.3	104/242	43.0	40/242	16.5
11	16/355	4.5	43/353	12.2				
12	12/355	3.4	28/351	8.0				
13	14/355	3.9	29/351	8.3				
14	11/355	3.1	31/351	8.8				
15	10/355	2.8	30/351	8.5	117/247	47.4	17/247	6.9

n – number of participants in the category; N – number of participants with available data.

Table 3. Associations between greenness, trees and allergenic trees in 500 m buffers around birth residential addresses with allergic outcomes up to 15 years, estimated by GEE models

		Asthma			Allergic rhinitis			Allergic sensitization to aeroallergens			Allergic sensitization to food allergens		
	Exposure	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**
Single exposure models	NDVI 500 m (T1 ref.)												
	T2	**1.46**	**1.02**	**2.10**	1.26	0.96	1.65	**1.52**	**1.05**	**2.18**	1.11	0.73	1.70
	T3	**0.44**	**0.27**	**0.72**	0.90	0.68	1.19	1.10	0.77	1.59	0.91	0.59	1.39
	Trees 500 m (T1 ref.)												
	T2	1.06	0.72	1.55	**1.62**	**1.21**	**2.17**	1.28	0.90	1.82	1.09	0.71	1.68
	T3	0.77	0.50	1.17	**2.13**	**1.60**	**2.82**	**1.52**	**1.07**	**2.17**	1.29	0.85	1.95
	Allergenic trees (def. 1) 500 m (T1 ref.)												
	T2	**1.92**	**1.29**	**2.84**	**2.09**	**1.58**	**2.78**	1.36	0.95	1.95	1.50	0.97	2.32
	T3	0.99	0.63	1.58	**1.70**	**1.27**	**2.27**	**1.71**	**1.20**	**2.43**	**1.59**	**1.05**	**2.42**
	Allergenic trees (def. 2) 500 m (T1 ref.)												
	T2	1.39	0.94	2.06	**2.06**	**1.53**	**2.76**	1.36	0.96	1.94	1.03	0.67	1.58
	T3	0.89	0.58	1.38	**2.06**	**1.53**	**2.76**	**1.48**	**1.03**	**2.13**	1.35	0.90	2.04
Models adjusted	Trees 500 m (T1 ref.)												
	T2	0.78	0.53	1.15	**1.62**	**1.19**	**2.21**	1.30	0.90	1.89	1.06	0.68	1.66
	T3	**0.54**	**0.35**	**0.84**	**2.13**	**1.59**	**2.85**	**1.55**	**1.07**	**2.22**	1.24	0.81	1.91
	NDVI 500 m (T1 ref.)												

T2	**1.44**	**1.01**	**2.07**	1.30	0.99	1.70	**1.54**	**1.07**	**2.22**	1.12	0.74	1.71
T3	**0.37**	**0.23**	**0.61**	1.12	0.84	1.50	1.28	0.87	1.89	0.96	0.62	1.49
Allergenic trees (def. 1) 500 m (T1 ref.)												
T2	1.49	0.97	2.30	**2.03**	**1.52**	**2.71**	1.32	0.92	1.92	1.49	0.95	2.32
T3	0.82	0.50	1.33	**1.68**	**1.25**	**2.26**	**1.75**	**1.22**	**2.50**	**1.55**	**1.00**	**2.38**
NDVI 500 m (T1 ref.)												
T2	1.38	0.96	1.97	1.23	0.93	1.61	**1.56**	**1.08**	**2.26**	1.11	0.73	1.69
T3	**0.45**	**0.27**	**0.74**	1.03	0.78	1.36	1.25	0.86	1.82	1.00	0.65	1.53
Allergenic trees (def. 2) 500 m (T1 ref.)												
T2	1.29	0.86	1.93	**2.03**	**1.52**	**2.72**	1.31	0.92	1.87	1.27	0.83	1.96
T3	0.72	0.45	1.15	**1.98**	**1.46**	**2.67**	1.41	0.97	2.03	1.21	0.78	1.87
NDVI 500 m (T1 ref.)												
T2	**1.46**	**1.00**	**2.12**	1.11	0.84	1.46	**1.46**	**1.01**	**2.11**	1.08	0.71	1.65
T3	**0.41**	**0.25**	**0.68**	0.87	0.65	1.15	1.11	0.77	1.61	0.90	0.59	1.38

def. – definition; GEE – generalized estimating equations; NDVI – Normalized Difference Vegetation Index; LCI – lower 95% confidence interval; OR – odds ratio; T1 – 1st tertile, T2 – 2nd tertile, T3 – 3rd tertile (see Table 3); UCI – upper confidence interval.

Boldface identifies significant associations ($p < 0.05$).

All models were adjusted for age, sex, season of birth, parental atopy and parental education.

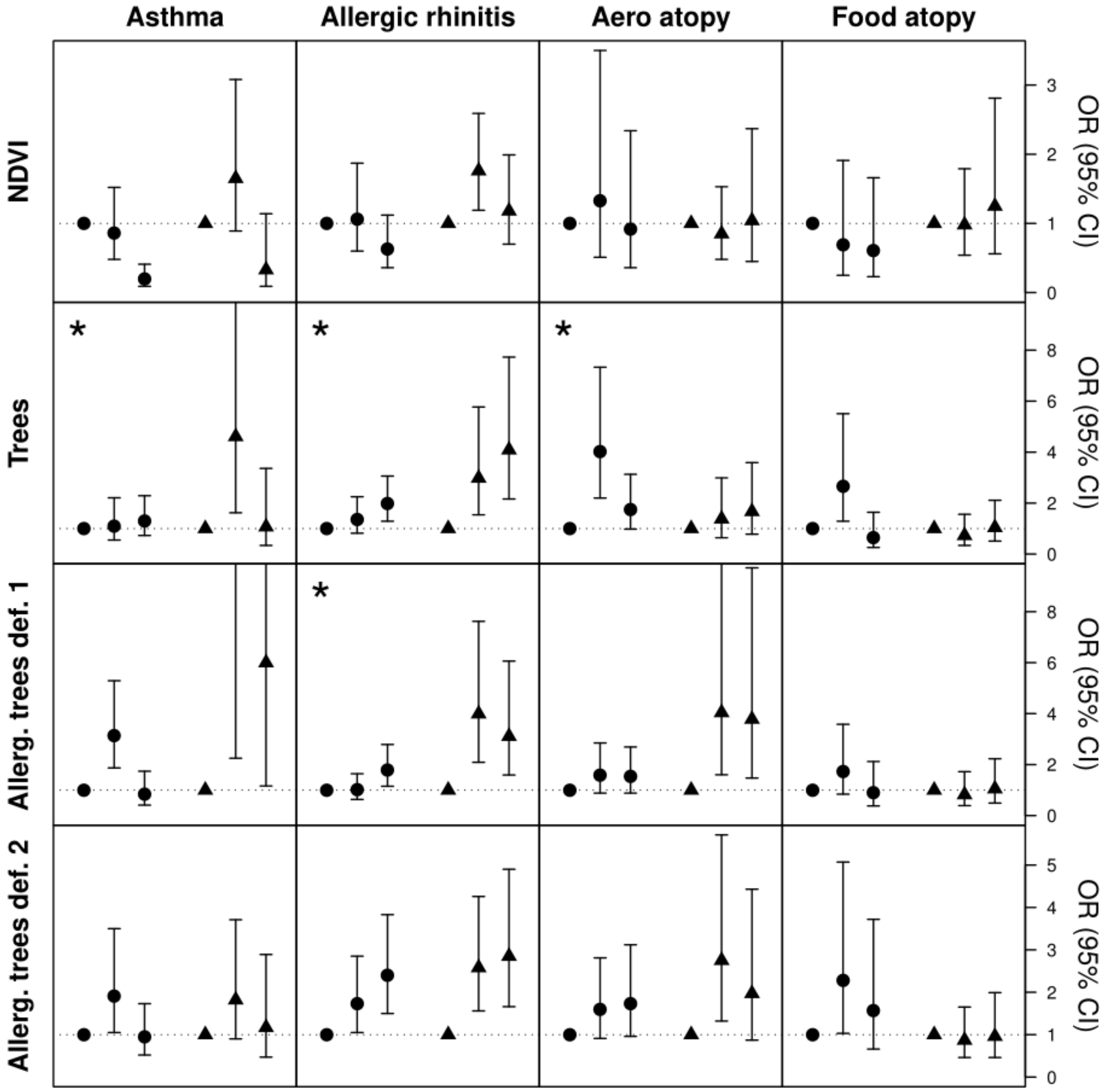

Figure 1. Associations between greenness, trees and allergenic trees in 500 m buffers around birth residential addresses with allergic outcomes up to 15 years, estimated by GEE models and stratified by NO_2 levels. Circles represent the low tertile of NO_2 while triangles represent the high tertile. An asterisk identifies a significant interaction term between the exposure variable and the continuous NO_2 variables.

All models were adjusted for age, sex, season of birth, parental atopy and parental education.

Allerg. – allergenic; CI – confidence interval; def. – definition; OR – odds ratio; NDVI – Normalized Difference Vegetation Index; NO_2 – nitrogen dioxide.

Online repository

Residing near allergenic trees can increase risk of allergies later in life: LISA Leipzig study

Iana Markevych, Romina Ludwig, Clemens Baumbach, Marie Standl, Joachim Heinrich, Gunda Herberth, Kees de Hoogh, Karin Pritsch, Fabian Weikl

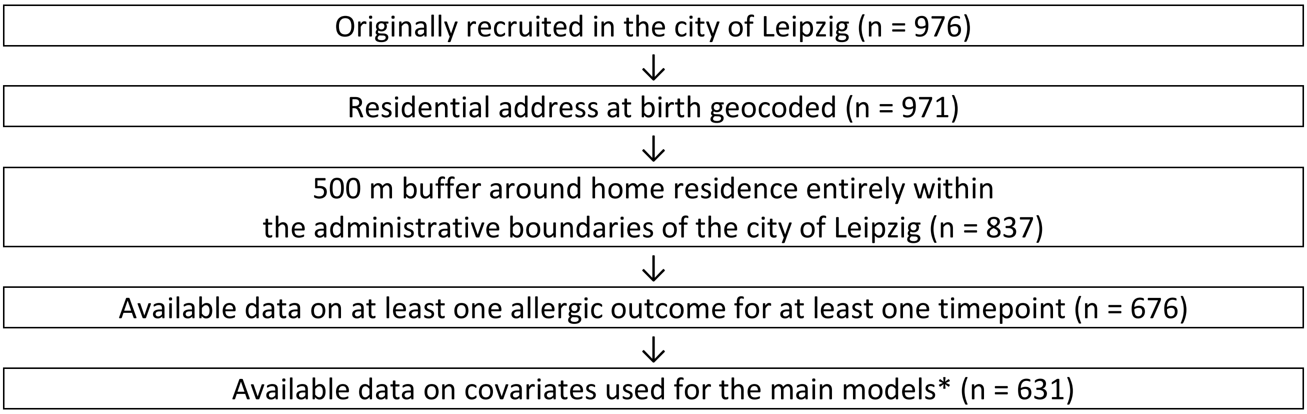

Figure E1. Flowchart of the study participants

* Main covariates are the following: age, sex, season of birth, parental atopy and parental education.

Table E2. Description of exposure variable categories

	Category 1		**Category 2**		**Category 3**	
	Min	**Max**	**Min**	**Max**	**Min**	**Max**
NDVI 100 m	0.1867	0.3606	0.3613	0.4586	0.4593	0.7616
NDVI 500 m	0.2498	0.3853	0.3858	0.4497	0.4499	0.7497
NDVI 1000 m	0.2737	0.4007	0.4008	0.4588	0.4591	0.7563
Trees 100 m	0	0	1	176		
Trees 500 m	1	221	222	453	455	2,221
Trees 1000 m	64	964	965	1,892	1,893	6,509
Allergenic trees (def. 1) 100 m	0	0	1	77		
Allergenic trees (def. 1) 500 m	0	44	45	134	135	1,068
Allergenic trees (def. 1) 1000 m	3	264	266	540	541	2,499
Allergenic trees (def. 2) 100 m	0	0	1	48		
Allergenic trees (def. 2) 500 m	0	16	17	61	62	393
Allergenic trees (def. 2) 1000 m	4	92	93	258	260	1,076

def. – definition; Min – minimum; Max – maximum; NDVI – Normalized Difference Vegetation Index.

NDVI variables and tree counts for the 500 m and 1000 m buffers were categorized into tertiles. Tree counts for the 100 m buffer were dichotomized (= 0 vs ≥ 1).

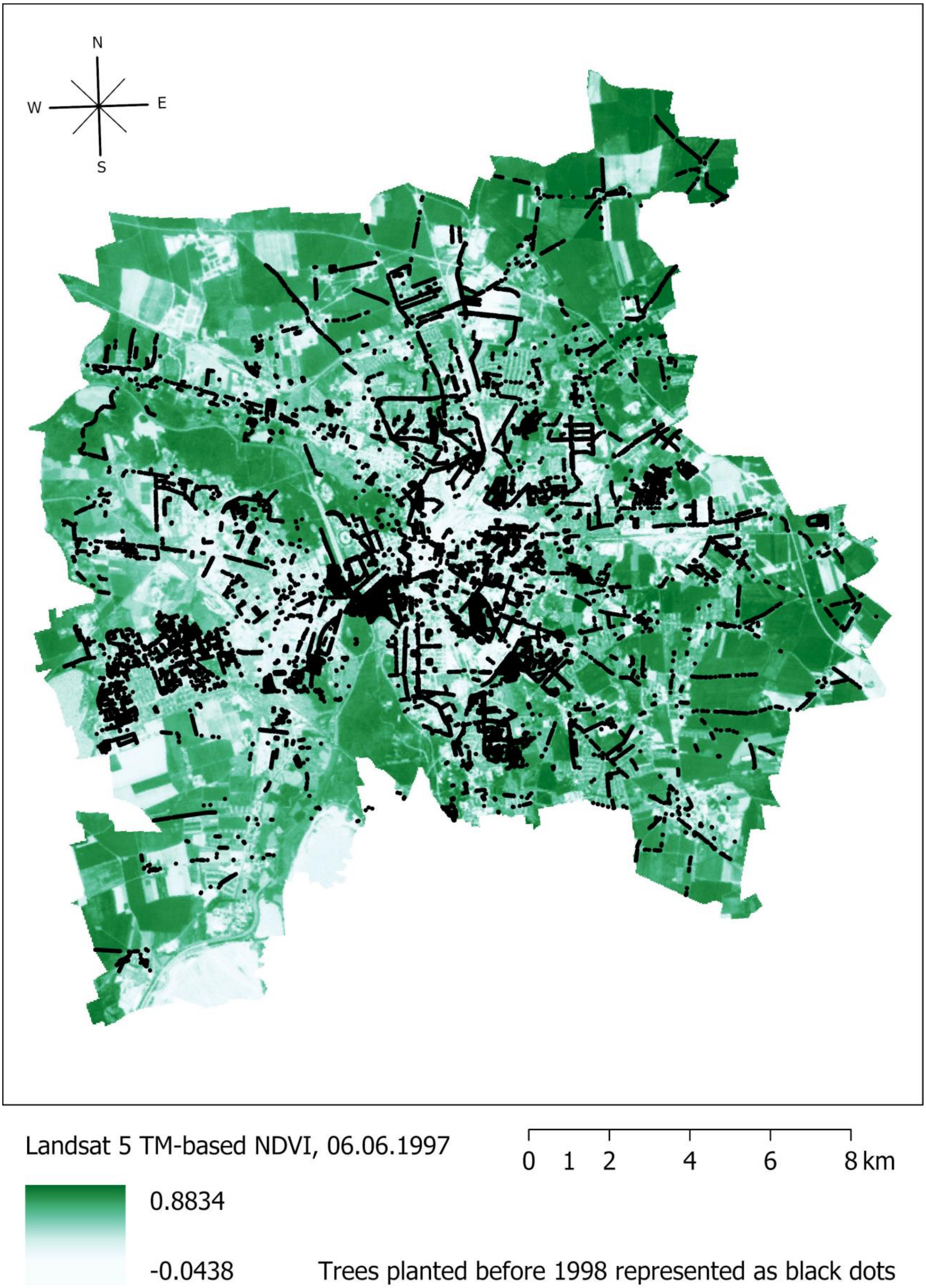

Figure E2. Trees of the city of Leipzig planted before 1998 superimposed over Normalized Difference Vegetation Index (NDVI) layer from 1997

Table E3. Associations between greenness, trees and allergenic trees in 100 m buffers around birth residential addresses with allergic outcomes up to 15 years, estimated by GEE models

	Asthma			**Allergic rhinitis**			**Allergic sensitization to aeroallergens**			**Allergic sensitization to food allergens**		
Exposure	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**
NDVI 100 m (T1 ref.)												
T2	1.10	0.74	1.64	1.04	0.79	1.36	0.90	0.64	1.28	0.77	0.50	1.17
T3	0.77	0.50	1.20	0.92	0.70	1.21	0.83	0.59	1.19	0.85	0.57	1.29
Trees 100 m (0 ref.)												
≥1	**3.12**	**1.99**	**4.90**	**2.14**	**1.64**	**2.78**	1.30	0.96	1.76	1.10	0.76	1.58
Allergenic trees (def. 1) 100 m (0 ref.)												
≥1	**1.45**	**1.05**	**2.01**	**1.40**	**1.12**	**1.74**	**1.50**	**1.12**	**2.00**	1.05	0.74	1.48
Allergenic trees (def. 2) 100 m (0 ref.)												
≥1	0.70	0.48	1.02	**1.48**	**1.19**	**1.85**	**1.51**	**1.12**	**2.04**	1.05	0.73	1.50

def. – definition; GEE – generalized estimating equations; NDVI – Normalized Difference Vegetation Index; LCI – lower 95% confidence interval; OR – odds ratio; T1 – 1st tertile, T2 – 2nd tertile, T3 – 3rd tertile (see Table 3); UCI – upper confidence interval.

Boldface identifies significant associations ($p < 0.05$).

All models were adjusted for age, sex, season of birth, parental atopy and parental education.

Table E4. Associations between greenness, trees and allergenic trees in 1,000 m buffers around birth residential addresses with allergic outcomes up to 15 years, estimated by GEE models

	Asthma			**Allergic rhinitis**			**Allergic sensitization to aeroallergens**			**Allergic sensitization to food allergens**		
Exposure	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**
NDVI 1,000 m (T1 ref.)												
T2	0.74	0.50	1.11	1.11	0.84	1.46	1.06	0.73	1.55	1.46	0.94	2.25
T3	**0.50**	**0.33**	**0.75**	0.84	0.64	1.10	1.27	0.89	1.81	1.16	0.75	1.79
Trees 1,000 m (T1 ref.)												
T2	1.16	0.74	1.80	**1.46**	**1.09**	**1.96**	1.08	0.77	1.53	1.42	0.92	2.18
T3	**1.53**	**1.03**	**2.27**	**1.95**	**1.48**	**2.57**	1.16	0.82	1.65	**1.70**	**1.12**	**2.60**
Allergenic trees (def. 1) 1,000 m (T1 ref.)												
T2	**1.29**	**0.84**	**1.98**	**1.43**	**1.07**	**1.90**	1.37	0.96	1.96	1.50	0.97	2.32
T3	1.46	0.98	2.17	**1.71**	**1.30**	**2.25**	1.37	0.97	1.94	**1.59**	**1.05**	**2.42**
Allergenic trees (def. 2) 1,000 m (T1 ref.)												
T2	1.07	0.71	1.62	**1.77**	**1.33**	**2.37**	1.35	0.95	1.92	1.03	0.67	1.58
T3	1.19	0.80	1.77	**2.11**	**1.59**	**2.81**	1.39	0.97	1.99	1.35	0.90	2.04

def. – definition; GEE – generalized estimating equations; NDVI – Normalized Difference Vegetation Index; LCI – lower 95% confidence interval; OR – odds ratio; T1 – 1st tertile, T2 – 2nd tertile, T3 – 3rd tertile (see Table 3); UCI – upper confidence interval.

Boldface identifies significant associations ($p < 0.05$).

All models were adjusted for age, sex, season of birth, parental atopy and parental education.

Table E5. Crude and additionally adjusted associations between greenness, trees and allergenic trees in 500 m buffers around birth residential addresses with allergic outcomes up to 15 years, estimated by GEE models

		Asthma			Allergic rhinitis			Allergic sensitization to aeroallergens			Allergic sensitization to food allergens		
	Exposure	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**	**OR**	**LCI**	**UCI**
Crude models	NDVI 500 m (T1 ref.)												
	T2	1.35	0.93	1.95	1.18	0.90	1.53	**1.58**	**1.11**	**2.24**	1.06	0.70	1.59
	T3	**0.42**	**0.26**	**0.69**	0.86	0.65	1.14	1.12	0.78	1.61	0.88	0.58	1.35
	Trees 500 m (T1 ref.)												
	T2	1.00	0.68	1.47	**1.54**	**1.16**	**2.05**	1.25	0.89	1.76	1.05	0.69	1.59
	T3	0.76	0.50	1.15	**2.00**	**1.52**	**2.64**	**1.46**	**1.03**	**2.06**	1.23	0.82	1.85
	Allergenic trees (def. 1) 500 m (T1 ref.)												
	T2	**1.82**	**1.23**	**2.70**	**2.03**	**1.53**	**2.68**	1.36	0.96	1.92	1.47	0.97	2.25
	T3	0.96	0.61	1.50	**1.65**	**1.24**	**2.20**	**1.66**	**1.17**	**2.35**	1.49	0.98	2.28
	Allergenic trees (def. 2) 500 m (T1 ref.)												
	T2	1.23	0.84	1.81	**2.06**	**1.55**	**2.73**	1.37	0.97	1.92	1.34	0.88	2.02
	T3	0.88	0.57	1.35	**1.97**	**1.48**	**2.64**	**1.48**	**1.04**	**2.11**	1.23	0.80	1.88
Additionally adjusted models	NDVI 500 m (T1 ref.)												
	T2	**1.64**	**1.12**	**2.38**	1.27	0.96	1.67	**1.49**	**1.02**	**2.18**	1.14	0.74	1.77
	T3	**0.43**	**0.26**	**0.72**	0.90	0.67	1.20	1.00	0.68	1.47	0.90	0.58	1.40
	Trees 500 m (T1 ref.)												
	T2	1.04	0.69	1.57	**1.57**	**1.16**	**2.11**	1.22	0.85	1.76	1.03	0.66	1.60
	T3	0.84	0.53	1.32	**2.09**	**1.56**	**2.80**	**1.60**	**1.12**	**2.29**	1.30	0.85	1.98
	Allergenic trees (def. 1) 500 m (T1 ref.)												
	T2	**1.83**	**1.21**	**2.77**	**2.11**	**1.58**	**2.82**	1.08	0.75	1.55	1.40	0.90	2.19
	T3	1.04	0.64	1.70	**1.64**	**1.20**	**2.22**	**1.36**	**0.95**	**1.94**	1.48	0.95	2.31
	Allergenic trees (def. 2) 500 m (T1 ref.)												
	T2	1.31	0.86	1.98	**2.05**	**1.52**	**2.76**	1.32	0.92	1.91	1.20	0.78	1.85
	T3	0.88	0.56	1.38	**1.93**	**1.42**	**2.63**	**1.67**	**1.17**	**2.40**	1.19	0.77	1.85

def. – definition; GEE – generalized estimating equations; NDVI – Normalized Difference Vegetation Index; LCI – lower 95% confidence interval; OR – odds ratio; T1 – 1st tertile, T2 – 2nd tertile, T3 – 3rd tertile (see Table 3); UCI – upper confidence interval.

Boldface identifies significant associations ($p < 0.05$).

Crude models were adjusted for age.

Additionally adjusted models were adjusted for age, sex, season of birth, parental atopy, parental education, maternal smoking during pregnancy and smoking at home, older siblings, breastfeeding and birth weight.

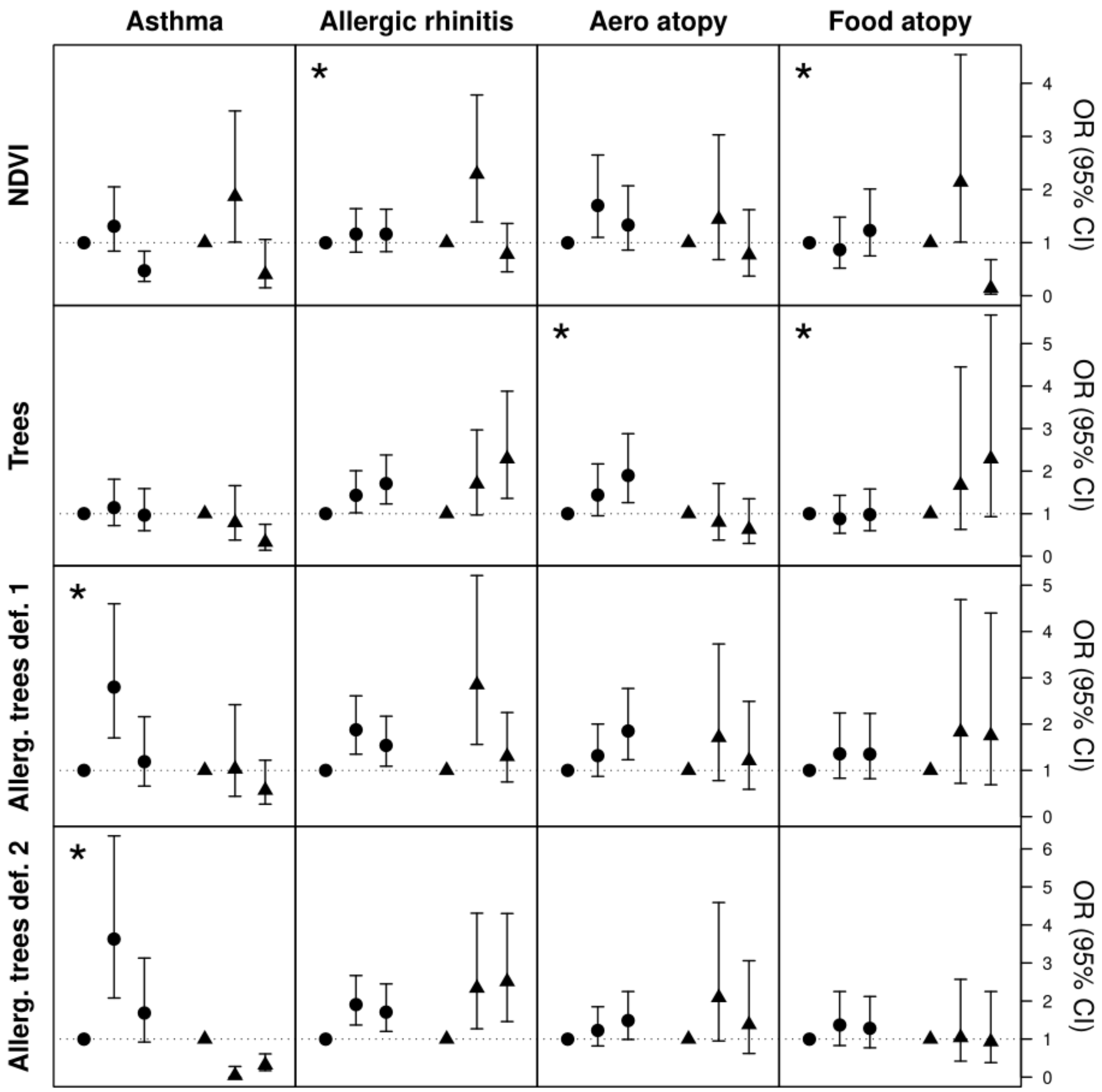

Figure E3. Associations between greenness, trees and allergenic trees in 500 m buffers around birth residential addresses with allergic outcomes up to 15 years, estimated by GEE models and stratified by whether a participant changed the place of residence between birth and 2 years. Circles represent non-movers and triangles movers. An asterisk identifies a significant interaction term between the exposure and the mover variables.

All models were adjusted for age, sex, season of birth, parental atopy and parental education.

Allerg. – allergenic; CI – confidence interval; def. – definition; OR – odds ratio; NDVI – Normalized Difference Vegetation Index.

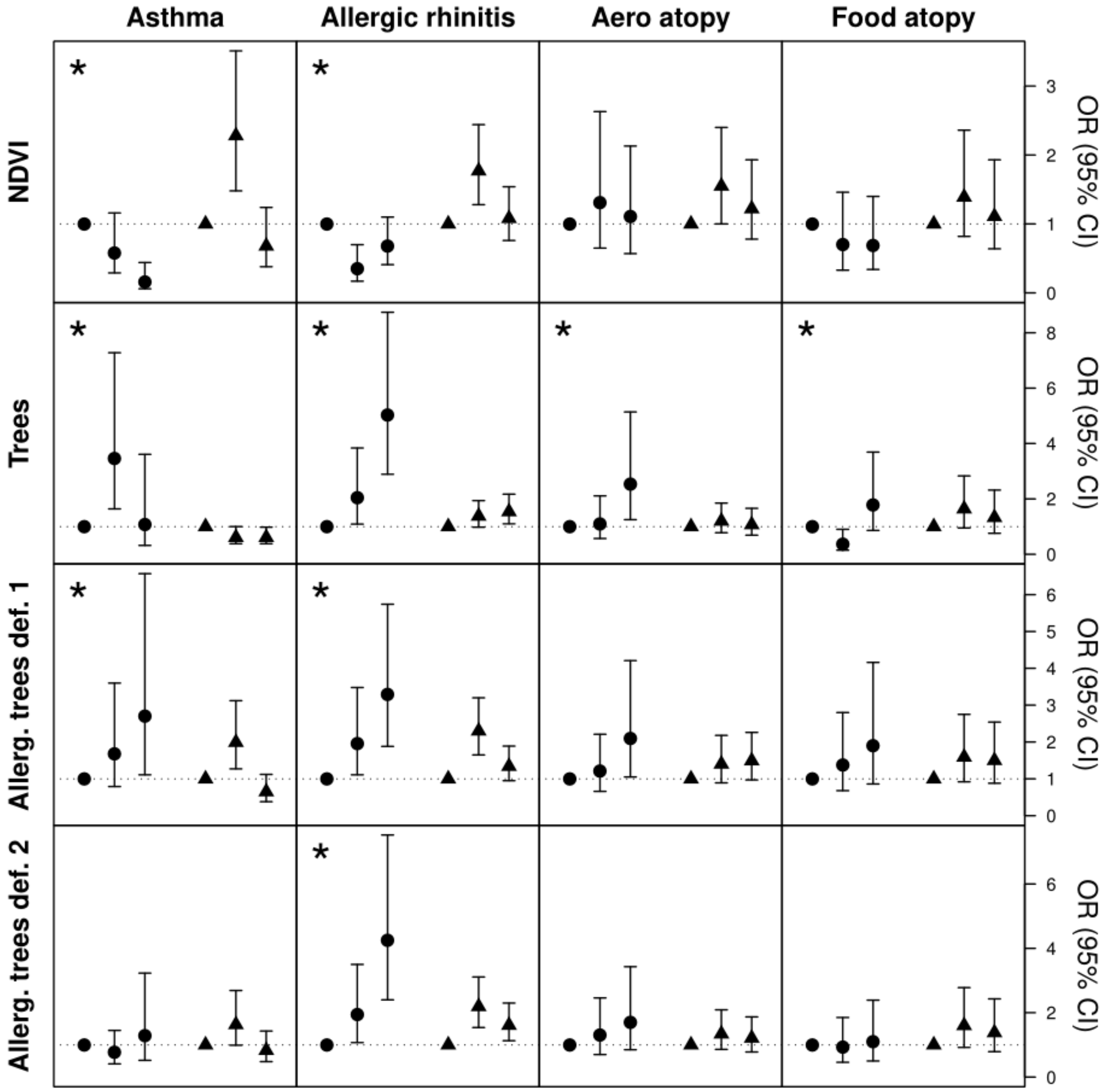

Figure E4. Associations between greenness, trees and allergenic trees in 500 m buffers around birth residential addresses with allergic outcomes up to 15 years, estimated by GEE models and stratified by whether a participant resided within 300 m from green space of at least 1 ha. Circles represent green spaces further away than 300 m while triangles represent green spaces within 300 m. An asterisk identifies a significant interaction term between the exposure and the green spaces variables.

All models were adjusted for age, sex, season of birth, parental atopy and parental education.
Allerg. – allergenic; CI – confidence interval; def. – definition; OR – odds ratio; NDVI – Normalized Difference Vegetation Index.

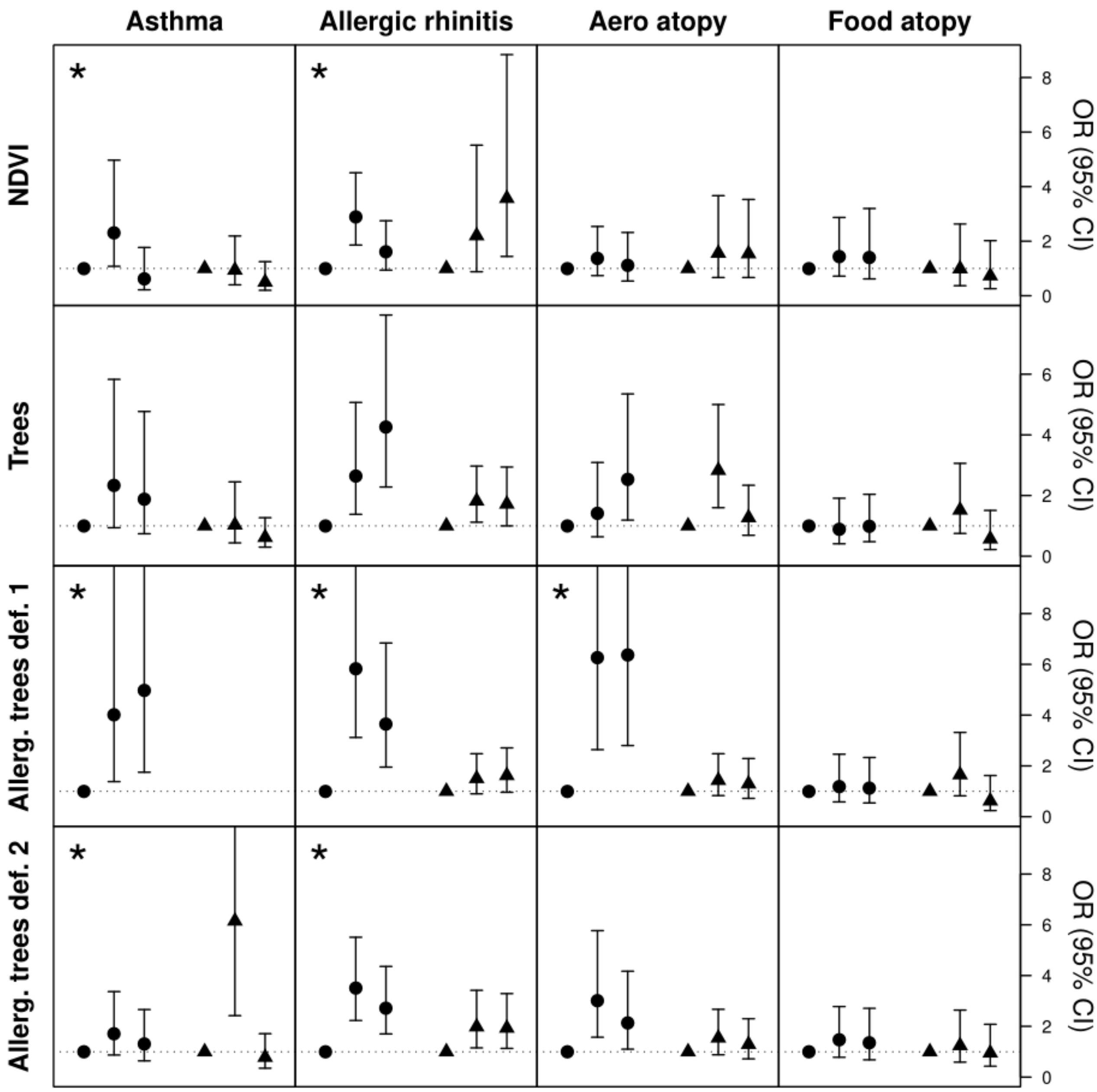

Figure E5. Associations between greenness, trees and allergenic trees in 500 m buffers around birth residential addresses with allergic outcomes up to 15 years, estimated by GEE models and stratified by ozone levels. Circles stand for the low tertile of ozone while triangles stand for the high tertile. An asterisk identifies a significant interaction term between the exposure and the continuous ozone variables.

All models were adjusted for age, sex, season of birth, parental atopy and parental education.

There are no odds ratios for allergenic trees definition 1 and the high tertile of ozone because the GEE model failed to return meaningful results in this case.

Allerg. – allergenic; CI – confidence interval; def. – definition; OR – odds ratio; NDVI – Normalized Difference Vegetation Index.

Repository E Table - Excel file

20200421_Markevych_Allergenic_trees_allergies_Table_E1.xlsx