Original research

Association between lung function of school age children and short-term exposure to air pollution and pollen: the PARIS cohort

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► Additional material is published online only. To view, please visit the journal online (http://dx.doi.org/10.1136/thoraxjnl-2020-215515).

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Received 10 June 2020
Revised 12 January 2021
Accepted 28 January 2021

ABSTRACT

Background Daily levels of ambient air pollution and pollen may affect lung function but have rarely been studied together. We investigated short-term exposure to pollen and air pollution in relation to lung function in school-age children from a French population-based birth cohort.

Methods This study included 1063 children from the PARIS (Pollution and Asthma Risk: an Infant Study) cohort whose lung function and FeNO measurements were performed at age 8 years old. Exposure data were collected up to 4 days before testing. We estimated daily total pollen concentration, daily allergenic risk indices for nine pollen taxa, as well as daily concentrations of three air pollutants (particulate matter less than 10 µm (PM_{10}), nitrogen dioxide (NO_2), ozone (O_3)). Children with similar pollen and air pollution exposure were grouped using multidimensional longitudinal cluster analysis. Associations between clusters of pollen and air pollution exposure and respiratory indices (FEV_1, FVC, FeNO) were studied using multivariable linear and logistic regression models adjusted for potential confounders.

Results Four clusters of exposure were identified: no pollen and low air pollution (Cluster 1), grass pollen (Cluster 2), PM_{10} (Cluster 3) and birch/plane-tree pollen with high total pollen count (Cluster 4). Compared with children in Cluster 1, children in Cluster 2 had significantly lower FEV_1 and FVC levels, and children from Cluster 3 had higher FeNO levels. For FEV_1 and FVC, the associations appeared stronger in children with current asthma. Additional analysis suggested a joint effect of grass pollen and air pollution on lung function.

Conclusion Daily ambient chemical and biological air quality could adversely influence lung function in children.

Key messages

What is the key question?

- Could lung function and FeNO measured in a school-age population-based birth cohort be influenced by both air pollution and pollen levels, in the days leading up to testing?

What is the bottom line?

- We found that children recently exposed to grass pollen had significantly lower FEV_1 and FVC levels, and children recently exposed to PM_{10} (particulate matter less than 10 µm) had higher FeNO levels, with a possible synergy between grass pollen and air pollution regarding lung function.

Why read on?

- Using an unsupervised approach to identify exposure profiles, we showed that, in a general population setting, children's lung function could be affected by recent air quality in terms of air pollutants and pollens.

INTRODUCTION

Lung function may be influenced by current ambient air quality. Most epidemiological studies have focussed on the effects of short-term exposure to ambient air pollutants such as ozone (O_3), nitrogen dioxide (NO_2), particulate matter with aerodynamic diameter <10 µm (PM_{10}),<2.5 µm ($PM_{2.5}$) and particularly traffic-related air pollution.[1–3] Concerning biological contaminants in ambient air, short-term exposure to pollen significantly increases the risk of allergic and asthmatic symptoms.[4] Pollen exposure has been little investigated in relation to lung function, especially in population-based studies, although exposure to pollen is a major public health concern, notably in the context of climate change.[5 6] Epidemiological studies have seldom examined the association of both chemical and biological contaminants on lung function simultaneously, although synergistic effects were suggested in experimental studies.[7 8]

Indeed, particulate and chemical pollutants can modulate pollen allergenic potency. Either airborne particles agglomerate onto pollen surfaces in heavy traffic areas[7] or exposure to different chemical pollutants such as O_3 or NO_2 induces the release of cytoplasmic granules by breaking the outer membrane of the pollen grain.[8] In their review, Sénéchal *et al*[9] showed how air pollution can affect the physical, chemical and biological properties of pollen grains. The authors used the concept of 'polluen' to describe the combination of pollen and air pollutants which constitutes a greater risk to respiratory health than pollen or air pollution separately.[10 11]

To cite: Amazouz H, Bougas N, Thibaudon M, *et al*. *Thorax* Epub ahead of print: [*please include* Day Month Year]. doi:10.1136/thoraxjnl-2020-215515

Associations between pollen, air pollution and lung function in children have been little studied, and results are heterogeneous and remains uncertain. As part of the BAMSE birth cohort a recent Swedish study suggested a deleterious effect of grass pollen exposure on lung function in children aged 8 after adjusting for air pollution.[12] Previously, Just *et al* found a significant interaction between O_3 and pollen count for asthma attacks in asthmatic children.[13] During the pollen season, Steerenberg *et al* reported higher levels of fractions of exhaled nitric oxide (FeNO) in pollen-sensitised children than in non-sensitised children, but no additional pro-inflammatory effects of air pollution were observed.[14] Multi-exposure to chemical and biological environmental pollutants can be difficult to study because of their correlation. These few studies suggest exposure to certain types of pollen have an effect on certain respiratory/ allergic morbidities.

In this context, as part of the PARIS (Pollution and Asthma Risk: an Infant Study) birth cohort follow-up, our aims were (1) to identify groups of children based on their recent exposure to pollen and air pollution before attending a health check-up at 8 years old, and (2) to investigate the short-term association between these groups and lung function, taking into account potential effect modification by current asthma, pollen-induced rhinitis, sex, history of allergy and sensitisation to pollen.

METHODS

Study design and population

The present study was based on the PARIS birth cohort follow-up. Between February 2003 and June 2006, 3840 healthy new-borns were recruited in five Parisian maternity hospitals.[15] Children were followed up using repeated self-administered questionnaires, and two health check-ups were offered at 18 months and 8 years old. Written informed consent was obtained from all parents and children. The present study deals with children who attended the health check-up at 8 years old.

Health data

Health check-ups of the 8 years olds took place in two reference asthma/allergy medical centres in Paris (Necker and Trousseau hospitals) in 2010 to 2015, and included spirometry and FeNO measurements, blood sampling and skin prick test (SPT). In addition, a paediatrician completed a standardised questionnaire including questions about respiratory and allergic symptoms, diagnoses and medications.

If applicable, parents were asked to stop any medication inhaled by the child 24 hours before spirometry tests. After checking that the children did not have any respiratory infections during the previous 4 weeks, FeNO measurement was performed using a NIOX MINO electrochemical analyser (Circassia Pharmaceuticals Inc, Chicago, USA). Following this, the best forced expiratory volume in 1 s (FEV_1) and forced vital capacity (FVC) among three reproducible spirometry tests were performed by trained technicians and recorded.[16] From these two indices, the FEV_1/FVC ratio was calculated.

Current asthma at 8 years old was defined according to the definition of the MeDALL (Mechanisms of the Development of ALLergy) consortium, by the presence of two of the following three criteria: ever doctor-diagnosed asthma, use of asthma medication in the last 12 months and wheezing in the last 12 months.

IgE sensitisation to a mixture of inhalant allergens was assessed using ImmunoCAP Phadiatop (Phadia, Thermo Fisher Scientific, Uppsala, Sweden). A positive result (>0.35 kUi/L) led to the measurement of specific IgE towards seven inhalant allergens including birch and timothy pollen.[17] In addition, SPTs were performed including birch, grass and mugwort pollen allergens. We considered children sensitised to pollen if birch or timothy specific IgE concentrations were >0.35 kUi/L or SPT with birch, grass or mugwort was positive (mean weal diameter ≥3 mm).

Table 1 Characteristics of the 1063 children of the PARIS cohort included in the pollen and air pollution cluster analysis, compared with those not included

	Non included n=2777	Included n=1063	P value*
Sex			0.29
Male	1410 (50.8)	560 (52.7)	
Female	1367 (49.2)	503 (47.3)	
Family socioeconomic status			**<0.001**
High	1681 (60.5)	713 (67.1)	
Medium	799 (28.8)	281 (26.4)	
Low	297 (10.7)	69 (6.5)	
Older siblings			**0.05**
None	1604 (57.8)	575 (54.1)	
One	911 (32.8)	365 (34.3)	
Two or more	262 (9.4)	123 (11.6)	
Place of residence at birth			0.34
Paris city	1772 (63.8)	661 (62.2)	
Paris suburbs	1005 (36.2)	402 (37.8)	
Maternal age at birth >35 years	637 (22.9)	308 (29.0)	**<0.001**
Parental history of asthma	553 (20.0)	211 (19.9)	0.96
Parental history of asthma, allergic rhinitis or eczema	1452 (52.3)	576 (54.2)	0.29
Maternal smoking during pregnancy	331 (11.9)	102 (9.6)	**0.04**
Exposure to ETS at home at birth	573 (21.5)	224 (21.4)	0.89

All data are n (%). Bold p values were used to highlight significant associations ($p<0.05$).
*χ^2 test.
ETS, environmental tobacco smoke.

Environmental exposures

Data on pollen and air pollution were collected for the 4 days prior to the health check-up. Pollen exposure estimates were assessed using data from the French aerobiological monitoring network (RNSA), which is located in central Paris on the roof of Institut Pasteur and was the only monitoring station available at the time of the study. We considered the daily allergenic risk (AR) indices (expressed from 0 to 5) based on the pollen grain count and allergenicity,[18] for each of the following nine taxa: *Alnus, Betula, Corylus, Cupressaceae-Taxaceae, Fraxinus, Platanus, Poaceae, Populus, Quercus*. In addition, we considered the daily total pollen concentration (expressed in pollen grains/ m^3) based on the following taxa: *Ambroisia, Artemisia, Alnus, Betula, Carpinus, Castanea, Quercus, Cupressaceæ-Taxaceæ, Fraxinus, Poaceae, Corylus, Olea, Rumex, Populus, Plantaginacæ, Platanus, Salix, Tilia, Urticacæ*.

Exposure to air pollution was assessed using an aggregated index created by the air quality monitoring network for the Paris region (AIRPARIF). This ATMO index is based on daily ambient air concentrations of four pollutants subject to regulation (PM_{10}, NO_2, O_3 and sulphur dioxide (SO_2)), which are measured by

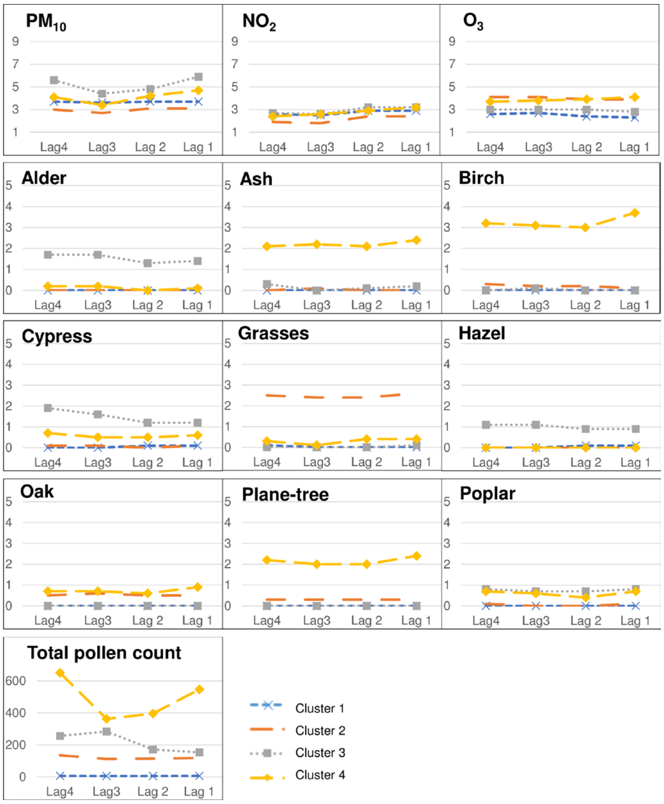

Figure 1 Clusters of recent exposure to air pollution and pollen identified in the 4 days preceding the health check-up at 8 years old in the PARIS birth cohort (n=1063). The x-axis indicates the four time points corresponding to the 4 days (lag4, lag3, lag2, lag1) preceding the health check-up, and the y-axis is the mean value at each time point in each cluster. The variables were coded as follows: PM_{10}, NO_2 and O_3 ATMO subindices from 1 (very good) to 10 (very bad), allergenic risk for each pollen species from 0 (nil) to 5 (very high) and total pollen concentration including 19 pollen taxa in grain/m^3. NO_2, nitrogen dioxide; O_3, ozone; PM_{10}, particulate matter with aerodynamic diameter <10 µm.

all background stations located in central Paris and suburbs (n=28). The daily ATMO index representative of both Paris and suburbs is calculated as the highest score among the four ATMO subindices for PM_{10}, NO_2, O_3 and SO_2, expressed from 1 (no or low level of pollution) to 10 (very high level of pollution). The PM_{10} subindex is based on the average of daily mean concentration while the NO_2, O_3 and SO_2 subindices are based on average of maximum hourly concentrations.[19]

In addition, the average ambient temperature and relative humidity were obtained from the French national meteorological service (Météo France) for the Paris-Montsouris station.

Covariates

Information such as sex, maternal smoking during pregnancy, place of residence (Paris, suburbs), and parental history of allergy, occupation and geographical origin was collected at maternity hospital during an interview with the mother. Family socioeconomic status (SES) was based on the highest position among both parents and divided into three categories. Exposure to environmental tobacco smoke (ETS) at home at 8 years old was documented using a parental questionnaire.

Statistical analysis

In order to group children with regards to their pollen and air pollution exposure in the 4 days before spirometry and FeNO testing, we used multidimensional, longitudinal cluster analysis including 3 variables for air pollution (daily PM_{10}, NO_2 and O_3 ATMO subindices) and 10 variables for pollen (daily total pollen count and AR indices for nine types of pollen). All 13 variables were standardised and considered at four time points corresponding to the previous 4 days (lag4, lag3, lag2, lag1). SO_2 was not considered in the analysis because it is no longer a major pollutant in the Paris region. Cluster analysis was performed using a version of K-means clustering adapted to the analysis of joint trajectories.[20] The optimal number of clusters was determined based on the Calinski-Harabasz criterion and the clusters considered relevant by experts in aerobiology and pollen monitoring. The analysis was performed on children with data available for ≥2 days for each variable (n=1063) and sensitivity analysis was conducted on children with complete data (n=1055). Each cluster was described by the distribution of average lag1-lag4 for pollen concentration and ATMO subindices.

In addition to the unsupervised analysis described above, and in order to test the possible joint effect of exposure to grass pollen and air pollution on lung function, four groups were created based on the maximum daily ATMO index and grass pollen concentration in the previous 4 days. Thresholds were chosen using a level of 6 for ATMO index (corresponding to poor air quality) and 10 grains/m^3 for grass pollen (corresponding to an intermediate AR index).

Associations between clusters of recent exposure or groups of joint exposure and respiratory indices were studied using multivariable, linear regression models for FEV_1 (mL), FVC (mL) and FEV_1/FVC ratio (%), and logistic regression model for FeNO >20 ppb[21] adjusted for potential confounders identified in the literature and using a directed acyclic graph (DAG).[22] All models were adjusted for sex, ethnicity, family SES (based on the highest SES position among parents), age and height at health check-up, exposure to environmental tobacco smoke at home at 8 years old, current asthma and average ambient temperature and relative humidity to the day before the health check-up. FeNO was considered with a threshold as the distribution was not normal. Results were expressed as adjusted regression coefficients (aβ) or adjusted ORs (aORs) with their 95% CIs. When possible, potential effect modification by current asthma, pollen-induced rhinitis, sex and sensitisation to pollen at 8 years old was explored by testing multiplicative interaction terms using a significance level of 0.10.

Cluster analyses were conducted using R software (V.3.5.2, R foundation for Statistical Computing, Vienna, Austria). Regression analyses were performed using Stata/SE 15.1 (StataCorp, College Station, Texas, USA). We followed the STROBE (Strengthening the Reporting of Observational Studies in Epidemiology recommendations for reporting observational cohort studies.[23]

RESULTS

Participants

Compared with children from the original PARIS cohort not included in the present study, children participating (n=1063) were more likely to live in a family with high SES, to have older siblings, a mother aged >35 years at birth and less likely to have a mother who smoked during pregnancy (table 1). However, the participants did not differ regarding sex, place of residence at birth, parental history of asthma or allergy and exposure to ETS

Table 2 Characteristics of clusters of recent exposure to pollen and air pollution identified at 8 years of age in the PARIS cohort: distribution of exposure levels in the previous 4 days and season of health check-up

	Cluster 1 ('low exposure')		Cluster 2 ('grass pollen')		Cluster 3 ('PM_{10}')		Cluster 4 ('tree pollen')	
n (%)	550 (51.7)		327 (30.8)		104 (9.8)		82 (7.7)	
	Median	**P25-P75**	**Median**	**P25-P75**	**Median**	**P25-P75**	**Median**	**P25-P75**
Average lag1-lag4 daily air pollution ATMO subindices (from 1 to 10)*								
PM_{10}	4	3–5	3	3–3	6	4–7	4	3–6
NO_2	3	3–3	2	2–3	3	3–3	3	3–3
O_3	3	2–3	4	3–5	3	3–3	4	4–4
Average lag1-lag4 daily pollen concentration (in grain/m^3)								
Alder	0	0–0	0	0–0	23.4	16.8–41.1	1.6	0.8–2.5
Ash	0	0–0	0	0–1.0	1.6	0.4–2.8	33.9	26.7–126.6
Birch	0	0–0	0.2	0–1.6	0.2	0–1.0	174.5	36.1–269.1
Cypress	0	0–0	4.9	3.1–7.6	70.6	34.9–131.4	15.2	8.4–32.9
Grasses	0	0–0	19.9	6.8–38.6	0	0–0.2	0.8	0.4–1
Hazel	0	0–0	0	0–0	12.1	4.5–20.5	0	0–0.4
Oak	0	0–0	2.7	0–17.2	0	0–0	2.1	0.6–48.7
Plane-tree	0	0–0	0.2	0–3.7	0	0–0	153.9	1.0–252.3
Poplar	0	0–0	0	0–0.4	5.6	0.8–14.5	6.6	4.3–13.7
Daily total concentration	0	0–1.4	113.5	60.6–176.1	154.4	92.5–250.2	509.5	428.2–666.4
Season of health check-up, n (%)								
Spring	29 (5.3)		233 (71.3)		15 (14.4)		82 (100.0)	
Summer	41 (7.4)		94 (28.7)		0 (0.0)		0 (0.0)	
Autumn	287 (52.2)		0 (0.0)		0 (0.0)		0 (0.0)	
Winter	193 (35.1)		0 (0.0)		89 (85.6)		0 (0.0)	

P25: 25th percentile, P75: 75th percentile.
*1: no or low level of pollution, 10: very high level of pollution.
NO_2, nitrogen dioxide; O_3, ozone; PM_{10}, particulate matter with aerodynamic diameter <10 µm.

at birth (table 1). The selection of study participants is shown in flowchart (online supplemental figure S1). Of the children, 47.3% were a women, 8.8% of children suffered from asthma, 6.8% suffered from pollen-induced rhinitis and 18.0% were sensitised to pollen. The description of pollen and air pollution for participants is given in online supplemental table S1.

Clusters of recent exposure to pollen and air pollution

The four-cluster classification provided the second best Calinski-Harabasz criterion (online supplemental table S2) and optimal relevance according to pollen experts, leading to the following clusters (figure 1 and online supplemental figure S2):

- Cluster 1 ('low exposure') showed no pollen exposure and low exposure to air pollution and was considered as reference (51.7% of children).
- Cluster 2 ('grass pollen') was characterised by moderate exposure to grass pollen and low exposure to air pollution (30.8% of children).
- Cluster 3 ('PM_{10}') experienced the highest exposure to PM_{10} and low exposure to pollen (9.8% of children).
- Cluster 4 ('tree pollen') showed the highest total pollen count, high exposure to birch pollen, moderate exposure to plane-tree pollen and low exposure to other pollens and air pollution (7.7% of children).

These clusters clearly depended on the season of the health check-up (table 2). Indeed, the health check-ups of Cluster 1 were mainly carried out in autumn, Cluster 2 at the end of spring/beginning of summer, Cluster 3 in winter and children from Cluster 4 attended the health check-up at the beginning of spring. The sensitivity analyses performed in children without any missing data led to similar clusters (online supplemental figure S3 and table S3).

Clusters of recent exposure and lung function in children aged 8 years

Associations between the exposure clusters previously identified and respiratory indices are shown in table 3. Lower FEV_1 and FVC values were observed in Clusters 2, 3 and 4 when compared with the reference group. Compared with children in the 'low exposure' cluster, children in the 'grass pollen' cluster had significant lower FEV_1 and FVC (aβ=−40.3 mL, 95% CI: –75.2 to –5.4 and aβ=−57.2 mL, 95% CI: –99.2 to –15.2, respectively). Children in the 'PM_{10}' cluster were more likely to have a FeNO >20 ppb (aOR=1.85, 95% CI: 1.01 to 3.37). No significant association was found in the 'tree pollen' cluster.

Effect modification

Significant interactions between clusters of recent exposure and those who had current asthma were found for FEV_1 and FVC. The deleterious association between the 'grass pollen' and 'PM_{10}' clusters and lung function was stronger in children suffering from current asthma (figure 2).

Table 3 Associations between clusters of recent exposure to pollen and air pollution and respiratory indices in children aged 8 years in the PARIS birth cohort: multivariable analysis

Exposure clusters	FEV_1 (mL) n=973	FVC (mL) n=973	
	aβ (95% CI)	aβ (95% CI)	
Cluster 1	Reference	Reference	
Cluster 2	−40.3 (−75.2 to −5.4)	−57.2 (−99.2 to −15.2)	
Cluster 3	−13.8 (−55.9 to 28.3)	−31.5 (−82.1 to 19.1)	
Cluster 4	−21.5 (−74.3 to 31.3)	−32.7 (−96.2 to 30.8)	
	FEV_1/FVC (%) n=973	FeNO>20 ppb n=866	
	aβ (95% CI)	n/N	aOR (95% CI)
Cluster 1	Reference	84/449	Reference
Cluster 2	0.5 (−0.5 to 1.5)	52/260	0.91 (0.53 to 1.55)
Cluster 3	0.6 (−0.6 to 1.8)	24/84	1.85 (1.01 to 3.37)
Cluster 4	0.3 (−1.2 to 1.9)	15/73	0.83 (0.38 to 1.83)

Cluster 1: 'low exposure', Cluster 2: 'grass pollen exposure', Cluster 3: 'PM_{10} exposure', Cluster 4: 'tree pollen exposure'.
All models were adjusted for sex, age and height at the health check-up, ethnicity, family socioeconomic status, exposure to environmental tobacco smoke at home at 8 years of age, current asthma and average ambient temperature and relative humidity the day before the health check-up. The estimates in bold differ significantly from the reference (p<0.05). Total N for each model may vary due to missing information.
aβ, adjusted regression coefficient; aOR, adjusted OR; FeNO, fraction of exhaled nitric oxide; FEV_1, forced expiratory volume in 1 s; FVC, forced vital capacity.

Among children who had current asthma, compared with children in the 'low exposure' cluster, those in the 'grass pollen' cluster had a significant lower FEV_1 (aβ=−163.0 mL, 95% CI: −292.9 to −33.1) and FVC (aβ=−165.4 mL, 95% CI: −325.1 to −5.6). Those in the 'PM_{10}' cluster had a lower FVC (aβ=−247.8 mL, 95% CI: −445.4 to −50.1). Lung function indices did not differ according to clusters of recent exposure among children who did not have current asthma. No effect modification was found with pollen-induced rhinitis, sex and sensitisation to pollen.

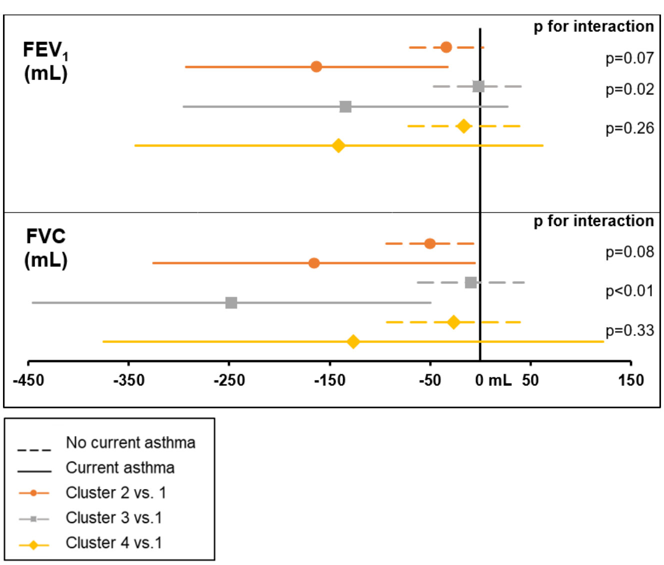

Figure 2 Associations between clusters of recent exposure to air pollution and pollen and respiratory indices at 8 years old according to current asthma status: multivariable analysis in the PARIS birth cohort. Squares represent the adjusted β regression coefficients, and horizontal lines the 95% CIs. Models were adjusted for the same variables as in table 3.

Joint exposure to air pollution and grass pollen and lung function

Half of the children (51%) had low exposure to both grass pollen and air pollution in the previous 4 days before health check-ups (Group 1), 25% had low exposure to grass pollen and high exposure to air pollution (Group 2), 19% had high exposure to grass pollen and low exposure to air pollution (Group 3) and 5% (Group 4) had high exposure to both grass pollen and air pollution.

Associations between these groups and respiratory indices are shown in table 4. Groups 3 and 4 had a lower FEV_1 and FVC. Compared with Group 1, children from Group 4 had a significant lower FEV_1 (aβ=−70.2 mL, 95% CI: −129.7 to −10.7) and FVC (aβ=−92.5 mL, 95% CI: −164.1 to −20.9). Group 3 tended to have a lower FVC (aβ=−41.0 mL, 95% CI: −84.1 to 2.1). No significant association was observed for Groups 2 and 3. No effect modification was found with pollen-induced rhinitis, sex and sensitisation to pollen. Due to small numbers, effect modification by current asthma was not tested.

DISCUSSION

Key results

In the population-based PARIS birth cohort, we applied an unsupervised approach to identify the exposure of school aged children to pollen and air pollution a few days before spirometry and FeNO measurements. We showed that recent exposure to grass pollen with low air pollution levels was associated with lower lung function, compared with children exposed to low air pollution and pollen levels. This effect appeared higher in children with current asthma. In addition, the group with high PM_{10} levels and low pollen exposure was associated with increased bronchial inflammation as assessed by FeNO. Additional analyses suggested a synergy between exposure to grass pollen and poor air quality regarding lung function indices.

Strengths and limitations

One of the strengths of this study is its reliance on a prospective birth cohort, whose population is well documented from a health, sociodemographic, behavioural and environmental point of view. The objective measurements of lung function were performed during a standardised medical examination in two specialised paediatric pulmonology departments, ensuring the quality of clinical data, thereby reducing measurement and classification bias.

Exposure during the 4 days before the health check-up precisely measured by two French networks specialising in air pollution and pollen monitoring. Other studies have dealt with exposure to pollen using various indicators, mostly surrogate variables such as pollen season[14 24–27] or pollen concentration for a single type[12 28 29] or total count.[13 30] The interest of the indices we used to describe children's exposure to pollen relies on their integrative nature. These indices which combined concentrations (pollen counts) and potential health risk (allergenicity differing according to pollen type) were used for the first time in an epidemiological study on children's lung function. Similarly, the ATMO indices allowed us to assess exposure to air pollution according to pollutant concentration and the potential health risks associated with air quality.

Another strength is the identification of multi-pollutant exposure clusters, jointly considering exposure to air pollution and

Table 4 Associations between groups of combined exposure based on maximum daily levels of grass pollen and air pollution exposure in the previous 4 days and respiratory indices in the PARIS birth cohort: multivariable analysis

	Grass pollen >10 grains/m^3	Poor air quality*	FEV_1 (mL) n=973 aβ (95% CI)	FVC (mL) n=973 aβ (95% CI)	
Group 1	No	No	Reference	Reference	
Group 2	No	Yes	7.7 (−20.9 to 36.3)	8.3 (−26.1 to 42.6)	
Group 3	Yes	No	−19.9 (−55.7 to 15.9)	−41.0 (−84.1 to 2.1)	
Group 4	Yes	Yes	−70.2 (−129.7 to −10.7)	−92.5 (−164.1 to −20.9)	
			FEV_1/FVC (%) n=973	**FeNO>20 ppb n=866**	
	Grass pollen >10 grains/m^3	**Poor air quality***	**aβ (95% CI)**	**n/N**	**aOR (95% CI)**
Group 1	No	No	Reference	86/446	Reference
Group 2	No	Yes	−0.1 (−1.0 to 0.7)	55/227	1.38 (0.90 to 2.11)
Group 3	Yes	No	0.8 (−0.3 to 1.8)	26/153	0.70 (0.40 to 1.24)
Group 4	Yes	Yes	0.9 (−0.9 to 2.6)	8/40	0.81 (0.33 to 2.03)

All models were adjusted for sex, age and height at the health check-up, ethnicity, family SES, exposure to environmental tobacco smoke at home at 8 years of age, current asthma and average ambient temperature and relative humidity the day before the health check-up. The estimates in bold differ significantly from the reference (p<0.05). Total N for each model may vary due to missing information.

*Poor air quality was defined as an air quality index ATMO ≥6 (on a scale of 1 to 10) based on the daily average air concentrations of PM_{10}, NO_2, O_3 and SO_2.

aβ, adjusted regression coefficient; aOR, adjusted OR; FeNO, fraction of exhaled nitric oxide; FEV_1, forced expiratory volume in 1 s; FVC, forced vital capacity; NO_2, nitrogen dioxide; O_3, ozone; PM_{10}, particulate matter with aerodynamic diameter <10 µm; SO_2, sulphur dioxide.

pollen using a multidimensional classification method. This method better reflects the reality of multiple exposures and avoids possible correlations between air pollution and pollen exposure, whereas most previous studies of lung function focussed on a single exposure, either to pollen or air pollution.[25 26 31] Moreover, regression models were adjusted for ambient air temperature and relative humidity, which could be associated with both exposure to pollen/air pollution and lung function indices.[32] Meteorological parameters are not always considered as possible confounding factors of the relations between pollution and respiratory health in individual observational studies,[12 25 26] limiting the interpretation of the results. In addition, our models were not adjusted for season: it was not necessary to do so because season reflects meteorological parameters and respiratory infectious diseases and these variables are controlled in the multivariable models.

As frequently observed in cohort follow-up, our study sample shows an over-representation of high SES families but adjusting for family SES should avoid confounding bias regarding the association between exposure and lung function. Although based on a prospective cohort study, the present research is limited in its cross-sectional approach as it was not possible to study the temporal co-variations between exposure and lung function indices. A panel study or cohort study with repeated measures could explore lung function in relation to daily variation in pollen and air pollution levels. We were not able to investigate the risk of having lung function below a clinical threshold due to the small number of children with such outcomes in our study sample from the general population.

Lastly, pollen exposure was assessed using the only monitoring station thought the children from the PARIS cohort lived in central Paris or its suburbs. In the same way, in the Swedish BAMSE cohort composed of children living in the Stockholm region, pollen exposure was derived from a roof-level urban background monitoring station within central Stockholm.[12] Concerning air pollution exposure, we used air quality indices which were less sophisticated than concentrations measured from the closest background monitoring station but which are relevant in distinguishing days with 'good' and 'poor' air quality. This can lead to non-differential misclassification bias which would underestimate the real associations. However, the use of these indices could allow to include the possible mobility of children within the Paris region in the few days before health check-up.

Interpretation

The present study suggests a deleterious effect of grass pollen on FEV_1 and FVC. In the literature, few studies have examined associations between grass pollen and lung function and results are inconsistent. Two previous studies found no significant association between grass pollen season and FEV_1 in children with asthma[27] and in children with seasonal allergic asthma.[26] Conversely, in children with seasonal allergic rhinitis, FEV_1 was lower than in the control group during the pollen season (including grass season).[25] Our results are consistent with findings from the BAMSE birth cohort,[12] where exposure to grass pollen up to 6 days before lung function testing was associated with a lower FEV_1 in children of 8 years old (aβ=−32.4 mL, 95% CI: −50.6 to −14.2) with an increase in 3 counts/m^3. Similar associations were shown with FVC. We did not find any association between grass pollen exposure and FeNO. In allergic children, Steerenberg *et al*[14] showed significant positive associations between levels of grass pollen in the 4 days before testing and FeNO. Similarly, in asthmatic children, two studies[26 27] showed an increased level of FeNO during grass pollen season. However, contrary to our study conducted on the general population, these two studies included children more susceptible to suffering adverse health effects from pollen exposure.

In our study, the association between grass pollen exposure and lung function appeared stronger in children with current asthma. No evidence of an effect modification by sensitisation to pollen was found but the number of children sensitised to pollen in the 'grass pollen' cluster was very small. In the BAMSE study, associations between grass pollen and lung function in children were modified by sensitisation to pollen allergens.[12] A recent meta-analysis of panel studies concluded that pollen exposure during the same day increases respiratory symptoms on the same day, as well as specifically upper respiratory and ocular symptoms among allergic and asthmatic participants but there was no detected effect on lung function.[4]

As with the BAMSE study, we did not find any association between tree pollen and children's respiratory indices.[12] This might be explained because children from the 'grass pollen' cluster, who were exposed in late spring-summer, spent more time outside than children from the 'tree pollen' cluster, who were exposed in early spring at the beginning of the pollen season.

In our study, the cluster represented by PM_{10} exposure was associated with higher levels of FeNO, a marker for airway inflammation. Interestingly, this association was observed in all children and was not modified by sex, sensitisation or current asthma. In the literature, Berhane *et al* found that short-term increase in PM_{10} was associated with FeNO independently of asthma and allergy status in a Southern California cohort.[31] Similarly, Carlsen *et al*[28] found that PM_{coarse} (2.5–10 µm in aerodynamic diameter) was associated with increased FeNO in non-asthmatic children aged 11 years.

Experimental studies showed that particles aggregate on the surface of pollen grains in urban areas.[7] Moreover, Motta *et al*[8] suggested an effect of traffic-related pollutants on pollen cytoplasmic granules (PCG), which contain allergens. After exposure to NO_2 or O_3 the grass pollen grain is damaged and emits PCG. In the present epidemiological study, the issue of the combined effect of air pollution and pollen was not resolved in the cluster analysis. Consequently, we performed additional analyses which showed a joint deleterious effect of poor air quality and grass pollen level ≥10 grains/m^3 during the previous days on the lung function (FEV_1 and FVC) of school-age children.

Very few epidemiological studies have investigated the combined effect of air pollution and pollen on lung function, especially in children. In these studies, exposure to pollen and pollution is most often accounted for by mutual adjustment in the statistical models,[12] or by testing the interaction between two exposure variables.[13 14 28–30] In the study by Scarlett *et al*,[30] pollen exposure did not modify the association of PM_{10} levels with lung function. In Just *et al*,[13] interactions were demonstrated between O_3 and pollen count for asthma attacks. Previous epidemiological studies have suggested an interaction between air pollution and pollen exposure, regarding respiratory and allergic morbidity. Recently, Bédard *et al*[24] showed that associations between uncontrolled rhinitis and pollutants were stronger during grass pollen season, but not during birch pollen season.

Lastly, in our study, significant but slightly lower respiratory parameters were observed in association with grass pollen and air pollution, which may have little or no impact on respiratory health at the individual level, especially in non-asthmatic children. Nevertheless, from a comprehensive perspective, small and clinically unimportant changes in lung function at the individual level may have a substantial impact at the population level, by increasing the proportion of children with clinically low lung function.

CONCLUSION

This study showed a deleterious effect of short-term exposure to both grass pollen and air pollution on the lung function of children from the general population and suggested a synergy between chemical and biological air contaminants.

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Acknowledgements The authors are grateful to all the children and parents of the PARIS birth cohort and to the administrative staff (Dominique Viguier, Marianne Bijou and Bruno Metivier) for their involvement in the PARIS cohort follow-up. We thank the physicians (Dr Michèle Boulé, Dr Bernard Boutin, Dr Mathieu Pellan, Dr Rym Belmir, Dr Michaela Semeraro, Dr Isabelle Haegy, Dr Wajed Aljundi, Dr Candice Meyzer, Dr Eric Daireaux, Dr Anne-Marie Le Marec, Dr Sofia Kalaboka), the nurses (Eve Thioux, Patricia Laskowsky, Dorothée Nguyen Van Suong, Charlotte Pellerin), the lung function test technicians, Dr Stephan Gabet for IgE measurements at age 8 years old and the administrative staff members for their invaluable collaboration during the 8 year health check-up. The authors thank RNSA, Airparif and Météo-France for having provided them with the air pollution, pollen and meteorological data for the Paris area. The authors would like to thank Dr Jean-Pierre Besancenot for his critical review of the manuscript.

Contributors IM contributed to the implementation of the PARIS birth cohort. IM and FR coordinated the PARIS cohort follow-up. JJ, NBe and MBH supervised the medical examination and lung function tests of the children at 8 years old at Trousseau Hospital. GL supervised the medical examination and lung function tests of the child participants at 8 years old at Necker Hospital. MT contributed to the interpretation of pollen data. HA performed the statistical analysis and drafted the manuscript. NBo contributed to the statistical analysis. IM and FR supervised data analysis and manuscript preparation. All authors were involved in the interpretation of the results, critically revised the manuscript and approved the final version as submitted.

Funding This work was supported by the Paris Municipal Department of Social Action, Childhood, and Health (DASES), the European collaborative programme MeDALL (FP7-261357) and Paris Descartes University.

Competing interests HA, NB, MT, NBe, IM and FR have nothing to disclose. GL reports personal fees from Novartis Pharma, personal fees from AstraZeneca, personal fees from YSSUP research, during the conduct of the study; personal fees from DBV technologies, personal fees from Aimune therapeutics, outside the submitted work. MB-H reports personal fees from Stallergene, personal fees from ALK, outside the submitted work; personal fees and non-financial support from Stallergene, personal fees and non-financial support from ALK, non-financial support from Astra Zeneca, outside the submitted work. JJ reports grants and personal fees from Novartis, grants and personal fees from AstraZeneca, personal fees from ALK, personal fees from Thermofischer, outside the submitted work.

Patient consent for publication Not required.

Ethics approval The French ethics committees approved the PARIS study (permission nos. 031153, 051289, and ID-RCB 2009-A00824-53).

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement Data are available upon reasonable request.

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