ORIGINAL ARTICLE

Endotoxin and gender modify lung function recovery after occupational organic dust exposure: a 30-year study

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ABSTRACT

Objectives The purpose of this study is to determine the trajectory of lung function change after exposure cessation to occupational organic dust exposure, and to identify factors that modify improvement.

Methods The Shanghai Textile Worker Study is a longitudinal study of 447 cotton workers exposed to endotoxin-containing dust and 472 silk workers exposed to non-endotoxin-containing dust. Spirometry was performed at 5-year intervals. Air sampling was performed to estimate individual cumulative exposures. The effect of work cessation on forced expiratory volume in 1 s (FEV_1) was modelled using generalised additive mixed effects models to identify the trajectory of FEV_1 recovery. Linear mixed effects models incorporating interaction terms were used to identify modifiers of FEV_1 recovery. Loss to follow-up was accounted for with inverse probability of censoring weights.

Results 74.2% of the original cohort still alive participated in 2011. Generalised additive mixed models identified a non-linear improvement in FEV_1 for all workers after exposure cessation, with no plateau noted 25 years after retirement. Linear mixed effects models incorporating interaction terms identified prior endotoxin exposure (p=0.01) and male gender (p=0.002) as risk factors for impaired FEV_1 improvement after exposure cessation. After adjusting for gender, smoking delayed the onset of FEV_1 gain but did not affect the overall magnitude of change.

Conclusions Lung function improvement after cessation of exposure to organic dust is sustained. Endotoxin exposure and male gender are risk factors for less FEV_1 improvement.

What this paper adds

- ▸ Long-term exposure to organic dust is associated with an accelerated decline in lung function.
- ▸ Few longitudinal studies are available to describe whether there is sustained improvement in lung function after exposure cessation to occupational organic dust. Whether there are factors that modify this improvement is unknown.
- ▸ This paper demonstrates that lung function continues to improve decades after work-related organic dust exposure. Men and those exposed to endotoxin improve less. Smoking delays the onset of lung function recovery but is not associated with the overall magnitude of recovery.

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INTRODUCTION

Chronic obstructive pulmonary disease (COPD) is projected to be the fourth leading cause of death worldwide.[1] Although tobacco smoke is most commonly identified as the main environmental exposure associated with COPD development, occupational exposures are thought to contribute 15% of the population attributable risk of COPD,[2] with estimates as high as 30% in non-smokers.[3] However, the vast majority of studies on occupational exposures focus on lung function changes during active exposure; few are available to describe lung function after exposure cessation due to worker retirement. Whether lung function recovers after removal from an occupational exposure is not well understood.

Exposure to endotoxin-containing cotton dust has been associated with the development of chronic lung disease. During early exposure, the disease is asthma-like, with airway hyper-reactivity and reversible airflow obstruction, while later disease resembles COPD, with fixed airflow obstruction and, more prominently, an accelerated decline in forced expiratory volume in 1 s(FEV_1). Autopsy series suggest airways disease and emphysema as the primary pathological lesions.[4] Human studies have affirmed that it is the amount of endotoxin rather than the amount of dust in cotton exposure that determines both acute[5] and chronic[6] declines in FEV_1. While endotoxin is a common occupational exposure,[7] it is also a common environmental exposure and is present in high concentrations in urban school,[8] homes burning biomass fuel[9] and tobacco smoke.[10]

The Shanghai Textile Worker Study is the longest active longitudinal study of cotton and silk textile workers. While cotton dust contains high levels of endotoxin, silk dust contains near-undetectable levels of endotoxin, creating a natural experiment in which to study the long-term effects of exposure to endotoxin-containing organic dust. Our primary goal was to evaluate whether the transient FEV_1 improvement noted after cessation of occupational dust exposure due to worker retirement in the 25-year follow-up of[11] this study was sustained given the further follow-up and use of more flexible modelling techniques. Our secondary goal was to determine whether prior occupational endotoxin exposure, smoking and gender modify FEV_1 improvement.

METHODS

Study population and study design

A total of 919 workers from two cotton mills and one silk textile mill in the same industrial sector in Shanghai, China were recruited in 1981 (study schema in online supplemental figure S1). The main inclusion criterion was at least 2 years of work in the identified mills in order to ensure a stable study population. The main exclusion criterion was a history of prior respiratory disease. The study population represented 90% of eligible workers in the yarn preparation areas of the three mills.[12] Cotton and silk workers were comparable in 1981 at the start of the study with respect to income, place of residence and other socioeconomic factors due to the hiring practices of the Shanghai Textile Bureau. Surveys were performed in 1981, 1986, 1992, 1996, 2001, 2006 and 2011, with eligibility for retesting based on presence in the baseline 1981 survey. Prebronchodilator spirometry, physical examination, modified American Thoracic Society symptom, work history and smoking questionnaires, and exposure assessment (in the period prior to worker retirement) was performed at each survey. Forced expiratory manoeuvres (up to seven trials to produce three acceptable curves) according to American Thoracic Society guidelines were performed under the direction of a trained technician on calibrated 8 L water-sealed field spirometers (W E Collins, Braintree, Massachusetts, USA) and spirometric curves were manually read by the same trained expert. The highest values for forced expiratory volume in 1 s (FEV_1) were used given that they were technically acceptable tests. A total of four spirometers, all of the same make and from the same manufacturer, were used for all field surveys from 1981 to 2011. Informed consent was obtained from all participants and the study was approved by the Institutional Review Boards at the Harvard School of Public Health and the Shanghai Putuo District People's Hospital.

Exposure assessment

Exposure assessment was performed as previously described.[13,14] Multiple area samples were collected from each of the six different work areas in the two cotton mills using vertical elutriators to collect respirable fractions of cotton dust, with sampling times ranging from 3 to 7 h. Filters were subsequently transported to a single laboratory at the National Institute of Occupational Safety and Health for endotoxin analysis. Endotoxin from collected filters was measured using a Limulus amoebocyte lysate gel test (Pyrostat-50), and values for each filter were summed and converted from ng/mL to $\mu g/m^3$ based on sampling time and air flow rates of each sampler. Exposure measurements collected in the first survey were used to estimate pre-1981 samples. Six full-shift samples in the silk mills had near-undetectable levels of endotoxin (0.001 EU/m^3) in vertical elutriator samples; thus, silk workers were considered unexposed to occupational endotoxin. Individual endotoxin exposure was calculated using geometric means of endotoxin measured in each work area multiplied by years of work in each work area, resulting in a lifetime cumulative index of occupational exposure measured in endotoxin units/meters^3-years (EU/m^3-yrs), with an interpretation analogous to that of pack-years for smoking. At each survey, a detailed work history was obtained to identify the date of textile work cessation as well as job descriptions postretirement.

Statistical analysis

The primary outcome of interest was change in FEV_1 associated with work cessation. However, in order to adjust for the potential bias from loss to follow-up using inverse probability of censoring weights, in our statistical models, FEV_1 rather than *change in* FEV_1 was used as the primary outcome measure. Covariates for the outcome models included age, gender, height, smoking status (defined as lifetime never, current or former) and cumulative pack-years. Exposure was modelled as either cotton versus silk textile work, or as log-transformed measured cumulative occupational endotoxin exposure.

We modelled FEV_1 using a generalised additive mixed effects model (GAMM)[15] with a penalised spline term for the number of years since work cessation. Such use of a GAMM allows the data to identify the functional form of the relationship between exposure cessation and FEV_1 change, rather than constraining the relationship based on modelling decisions. Our secondary research question focused on whether lung function recovery was modified by prior occupational endotoxin exposure, smoking or gender. The significance of an interaction between a categorical variable (ie, cotton vs silk) and a smoothed term (penalised spline term for work cessation-years) cannot be estimated in a generalised additive mixed model. Therefore, the final outcome model was a linear mixed model with both linear and quadratic terms for work cessation as suggested by the GAMM (see online supplement for details).

As mentioned, FEV_1 rather than *change in* FEV_1 was used as the outcome measure in our statistical models. Therefore, the main effect of the group represents baseline differences in FEV_1, whereas a *group×time interaction* represents the change in FEV_1 associated with that grouping variable in a longitudinal study.[16] Interaction terms between work cessation years and occupational exposure, smoking and gender were included in all models in order to determine whether *changes in* FEV_1 were modified by these variables. Models with random intercept and slope to account for within-participant correlation over time were used.

Despite the high rate of participation at our 30-year survey, it is possible that loss to follow-up may lead to bias if missing data are not accounted for. For observations with a monotone pattern of missingness (ie, the participant never participated in another survey after the first missed survey), it was assumed that the missing data mechanism was missing at random (MAR). This mechanism implies that missingness can be explained by observed variables such as older age, presence of respiratory symptoms or occupational exposure. To adjust for the possibility that loss to follow-up differed by case history, the stabilised inverse probability of censoring weights[17] were used in the final models. The denominator of the weights was based on a logistic model predicting that the outcome was uncensored, that is, a technically acceptable FEV_1 measurement was present. Predictors were cotton versus silk exposure, age, gender, work cessation-years, years worked in the textile industry and both the presence of respiratory symptoms and FEV_1 at the preceding survey. The numerator of the weights was based on a logistic model for the same outcome, but included only exposure (cotton vs silk work) as the predictor.

Percent predicted FEV_1 was calculated based on prediction equations derived from Chinese populations.[18] Statistical analyses were performed using R 3.1.0 with the packages lme4,[19] mgcv[15] and ipw.[20]

RESULTS

A total of 919 workers (447 cotton and 472 control silk workers) were recruited in 1981 to participate in the Shanghai Textile Worker Study (see online supplemental figure S1). The median number of FEV_1 measurements obtained was 6 (IQR 4–7) per participant. The cotton and silk textile workers were quite comparable overall in 1981 and 2011 (table 1), although a higher proportion of cotton workers smoked compared to silk workers in 1981. Very few females smoked (one silk worker, 10 cotton workers in 1981; one silk worker, three cotton workers in 2011). In 1981, when all of the workers were actively working, cotton textile workers had more respiratory symptoms compared to silk textile workers. At the most recent survey in 2011, there were no significant differences in the proportion of cotton workers versus silk workers with respiratory symptoms. In 2011, the average duration of retirement was 18 years for both cotton and silk workers. Most of the textile workers retired between 1992 and 2001, with only three (two silk, one cotton) still active in textile work in 2011. There were no significant differences in follow-up rates between cotton and silk workers, with a similar average duration of follow-up in cotton workers compared to silk workers. Lifetime cumulative occupational endotoxin exposure was on average 38 928 (IQR 1730–65 204) EU/m^3-years for cotton workers in 2011, whereas it was assumed to be negligible for silk workers based on a limited number of full shift samples taken in silk mills which demonstrated near undetectable levels of endotoxin.

Individual unadjusted FEV_1 and per cent predicted FEV_1 trajectories with age are depicted in online supplemental figures S2 and S3. FEV_1 trajectories differ between each strata of smoking and occupational exposure, with cotton smokers having the steepest decline over time.

The adjusted effect of work cessation-years on FEV_1 based on the GAMM is depicted in figure 1 and table 2. Several observations can be made from these predictions. First, the effect of work cessation on FEV_1 is non-linear, with no plateau in FEV_1 improvement noted up to 25 years after work cessation in all strata. Second, the greatest improvements in FEV_1 after work cessation are seen in non-smoking silk >non-smoking cotton >smoking silk >smoking cotton textile workers over the observation period for both men and women. Third, for smokers, a gain in FEV_1 with work cessation as compared to active textile work was not seen immediately at the time of work cessation. In males, at 5 years of work cessation, a gain in FEV_1 for non-smokers was 28.7 (−14.5 to 71.9) mL for silk and 15.6 (−28.3 to 59.4) mL for cotton workers, whereas for smokers there was no gain in FEV_1 with average predicted changes being −2.3 (−35.5 to 30.9) mL for silk workers and −76.8 (−113.4 to −40.2) mL for cotton smokers (table 2). Given the low number of female smokers in our cohort, we cannot rule out that the same phenomenon may occur in women as well.

To determine whether occupational endotoxin exposure, smoking or gender modifies FEV_1 recovery after work cessation, interaction terms were added to the mixed effects models (table 3). Ten years of work cessation was associated with an average 168.5 mL improvement in FEV_1 in all textile workers. Cotton workers had 25.8 mL less improvement compared to silk workers ($p=0.02$), and men had 4.9 mL ($p=0.003$) less improvement compared to women. Current smokers had 15.4 mL less improvement compared to non-smokers ($p=0.63$), although the relationship was not statistically significant. Although few female textile workers were smokers, the effect of gender was not due solely to the effect of smoking. When the analysis was restricted to lifetime non-smokers, the gender×cessation interaction term remained statistically significant ($p=0.002$).

When occupational exposure was modelled as a log-transformed measure of cumulative endotoxin exposure (table 4), the interaction between work cessation-years and endotoxin remained significant ($p=0.01$), indicating a dose-dependent relationship between prior occupational endotoxin exposure and less FEV_1 improvement.

DISCUSSION

In this report, we demonstrate that retirement from work and therefore cessation of exposure to organic dust results in a sustained improvement in FEV_1. The effect of exposure cessation, however, had a complex, non-linear relationship with FEV_1. Importantly, recovery was adversely modified by prior occupational endotoxin exposure and male gender. After adjusting for gender, smoking was not associated with a statistically significant impact on FEV_1 change after work cessation, although it appeared to impact the trajectory of improvement. To the best of our knowledge, this is the first report to address whether lung function recovery is transient or sustained after occupational organic dust exposure, and also the first to identify prior occupational endotoxin exposure and gender as risk factors for decreased FEV_1 recovery.

Whether FEV_1 improves after exposure cessation in workers exposed to endotoxin-containing organic dust has been controversial. In the earliest longitudinal study to evaluate the effect of retirement, retired hemp workers had more respiratory symptoms and greater (but not statistically significant) annual declines in FEV_1 (53.3 vs 47.1 mL/year) at 9 year follow-up.[21] A subsequent 6-year study of cotton textile workers also found more respiratory symptoms and greater annual decline in FEV_1 in retired workers compared to active workers.[22] Studies in retired grain elevator workers, another group with occupational exposure to endotoxin-containing organic dust, found no improvement with retirement.[23] One study looking at removal from exposure to endotoxin in a bacterial single cell protein factory found that FEV_1 improved by 210 mL 1 year after exposure cessation in workers exposed to low levels of endotoxin, but there was no improvement in those exposed to high endotoxin levels.[24] Our own early studies on cessation did not find a significant association between work cessation and FEV_1 improvement,[6] and it was only at the 25-year follow-up[11] that we first reported FEV_1 improvement after cessation of textile work, although we reported that improvement plateaued, with a trend towards greatest improvement in smoking cotton workers. In the present analysis, we clarify that improvement is sustained, male cotton workers improve the least over time, and further identify prior occupational exposure and gender as important modifiers of FEV_1 recovery. There are several major differences between this work and our prior work, which may explain the apparent inconsistencies. First, we have an additional 5 years of follow-up. Second, our prior analysis did not allow for complex non-linearity. Third, we previously restricted our analysis to only participants who participated at every survey since 1981; here we used all available data from all participants while adjusting for loss to follow-up. Of particular interest is the observed lag between exposure cessation and an overall gain in FEV_1 in smokers. This delayed recovery may provide an explanation for

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Table 1 Characteristics of study participants at baseline (1981) and end of follow-up (2011)

	1981		2011	
	Silk (n=472)	Cotton (n=447)	Silk (n=291)	Cotton (n=296)
Male	200 (42.4%)	214 (47.9%)	112 (38.5%)	122 (41.2%)*
Age, years	36.7±10.7	37.8±10.6	65.8±9.7	65.6±9.9
Follow-up time, years	–	–	29.4±0.07	29.6±0.06
Height, cm	162.5±7.3	163.9±7.5*	160.6±7.6	162.1±7.9
Current smoking	118 (25.0%)	159 (35.6%)*	60 (20.6%)	66 (22.3%)
Male	117 (58.5% of males)	149 (69.6% of males)	59 (52.7% of males)	63 (51.6% of males)
Female	1 (0.9% of females)	10 (2.2% of females)	1 (0.6% of females)	3 (1.7% of females)
Pack-years†	9.2±10.0	8.5±9.8	28.2±19.8)	28.2±19.0
Lifetime non-smoker	348 (73.7%)	284 (65.5%)*	182 (66.9%)	178 (62.9%)
Male	77 (38.5% of males)	61 (28.5% of males)	22 (20.2% of males)	25 (20.8% of males)
Female	271 (99.6% of females)	223 (95.7% of females)	160 (98.2% of females)	153 (93.9% of females)
Active textile work	472 (100%)	447 (100%)	2 (0.34%)	1 (0.17%)
Years employed	16.2±11.3	16.2±10.3	27.3±9.2	26.3±8.0
Years since retirement‡	–	–	18.5±4.5	17.8±5.2
Cumulative endotoxin exposure, EU/m^3-years§	–	11 540 (3442–32 111)	–	38 928 (17 030–65 204)
FEV_1, litres	2.84±0.67	2.90±0.72	2.29±0.58	2.28±0.64
Per cent predicted FEV_1¶	99.6±13.1	100.2±13.9	107.7±18.9	104.4±18.9
FEV_1/FVC ratio	0.84±0.09	0.83±0.09	0.77±0.08	0.76±0.08
FEV_1/FVC<0.7**	20 (4.2%)	27 (6.0%)	39 (13.4%)	40 (13.5%)
FEV_1 ≥80% predicted	12	16	31	30
50% ≤FEV_1 <80% predicted	7	10	7	7
30% ≤FEV_1 <50% predicted	1	1	1	2
FEV_1 <30% predicted	0	0	0	1
Unadjusted annual FEV_1 change (mL/year)	–	–	−19.6±12.1	−23.0±12.9*
Chronic bronchitis	36 (7.6%)	96 (21.5%)*	22 (2.6%)	29 (4.9%)
Chronic cough	33 (7.0%)	87 (19.5%)*	7 (1.2%)	11 (1.9%)
Dyspnoea on exertion	18 (3.8%)	67 (15.0%)*	71 (12.1%)	67 (11.4%)
Byssinosis	0 (0%)	34 (7.6%)*	0 (0%)	0 (0%)

Continuous variables are presented as mean±SD or median (IQR). Categorical variables are presented as n (%).
*p<0.05.
†Calculated among ever-smokers only.
‡Calculated among retired workers only.
§A limited number of full shift samples taken in silk mills were found to have undetectable levels of endotoxin by the limulus amoebocyte lysate assay. Thus, silk workers were considered unexposed to endotoxin.
¶Prediction equations derived from the Chinese reference population.[18]
**Pre-bronchodilator spirometry.
FEV_1, forced expiratory volume in 1 s; FVC, forced vital capacity.

the negative results of studies with less than 10 years of follow-up, where both smokers and men comprise a significant proportion of the population studied, and highlights the importance of both long-term follow-up and use of advanced regression techniques when studying the recovery of lung function after exposure to a toxic environmental exposure.

The mechanism of lung function improvement after occupational dust exposure is unclear. Animal studies of repeated endotoxin exposure suggest that there are exposure-related structural changes such as epithelial and mesenchymal fibroproliferation[25] along with emphysema.[26] This implies that some component of endotoxin-related chronic lung disease is irreversible. Other studies demonstrate that repeated endotoxin exposure is associated with an expansion in the proinflammatory dendritic cell subsets in the lung.[27] It is possible that with exposure cessation, there is slow resolution of the inflammatory process. In preliminary studies, we have noted persistent changes in lung density on high-resolution chest imaging in cotton workers that may represent ongoing inflammation decades after exposure cessation.[28] Future biomarker studies to identify the underlying basis for persistent effects of occupational endotoxin exposure decades after exposure cessation may be informative.

Furthermore, it is not clear why there are gender differences in recovery. A meta-analysis of person-level data pooled from 12 cross-sectional studies of workers exposed to organic dust found that women were less likely than men to experience lower respiratory symptoms within the same industry, although this study did not include exposure assessment and so was unable to exclude differences in gender-specific workplace exposures as the explanation.[29] We have previously reported in our cohort that endotoxin-exposed men are at higher risk than women of developing reduced lung function and mortality due to all causes of death combined.[30] Animal studies demonstrating an augmented response to endotoxin related to male sex hormones[31] provide a potential biological explanation. However, gender differences in FEV_1 recovery were also noted among silk workers. Job descriptions after retirement were manually reviewed to determine whether workers remained in an environment with high-endotoxin exposure; however, beyond job descriptions, further exposure assessment was not performed. Additional gender-specific differences in exposure may have existed after worker retirement, or alternatively, gender may have a biological effect on FEV_1 recovery.

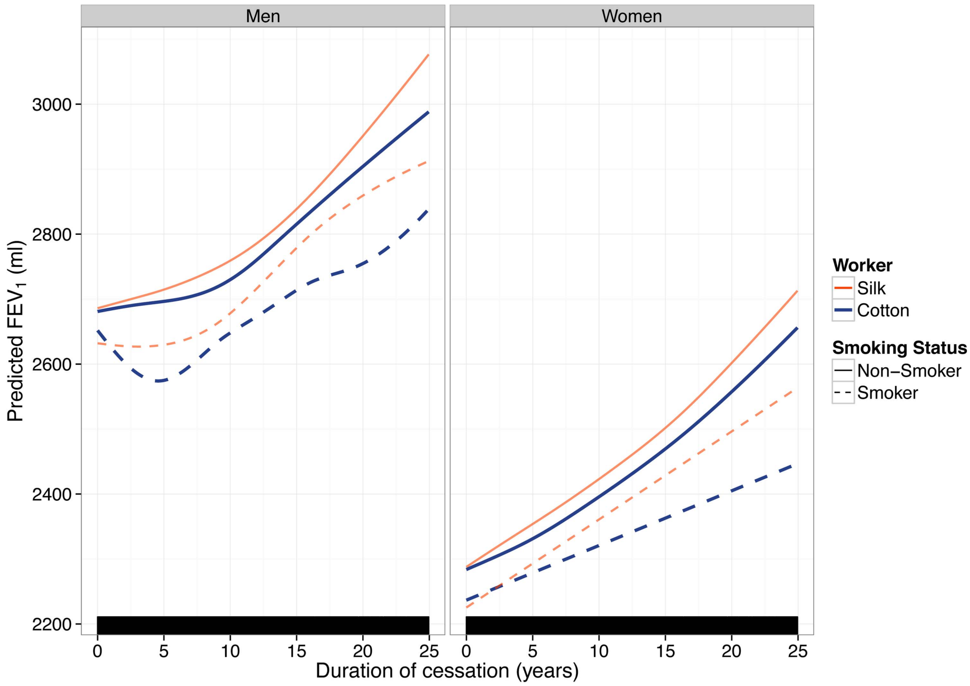

Figure 1 Adjusted effect of work cessation on forced expiratory volume in 1 s (FEV_1), stratified by gender, occupational exposure and smoking status based on a generalised additive mixed model. Predictions are for eight hypothetical workers of different gender, smoking and occupational (cotton vs silk) exposures. Predictions assume that these workers have the same age, height, pack-year history (zero if non-smokers, the average number of pack-years if smokers) at each value of work cessation-years. Rug plot (bottom) indicates values of cessation-years for which an observation was present. No plateau is seen in FEV_1 improvement after work cessation. For both men and women, FEV_1 improvement is greatest in non-smoking silk >non-smoking cotton >smoking silk >smoking cotton workers.

Our study has several strengths. First, most studies on the effect of exposure cessation are cross-sectional with matched population controls, or are longitudinal studies of shorter duration. Our study spans 30 years, with little loss to follow-up, high participation and a large number of FEV_1 measurements per participant. Second, while a long duration of follow-up is desirable, an improvement in lung function over time might be attributed to a survivor bias if loss to follow-up was not accounted for. Third, exposure assessment was performed[13 14] rather than reliance on a surrogate such as number of years worked. Finally, a large number of non-smoking men participated, allowing us to demonstrate that gender has effects on FEV_1 recovery independent from smoking.

We acknowledge limitations to our study. First, although control silk workers were not exposed to endotoxin in the workplace, they were exposed to other organic dust and this

Table 2 Predicted effect of work cessation on FEV_1 by gender, occupational exposure, and smoking status based on a generalised additive mixed model

	Change in FEV_1, mL (95% CI)							
Cessation years	**Silk non-smokers**		**Silk smokers**		**Cotton non-smokers**		**Cotton smokers**	
Men								
5	28.7	(−14.5 to 71.9)	−2.3	(−35.5 to 30.9)	15.6	(−28.3 to 59.4)	−76.8	(−113.4 to −40.2)
10	73.2	(11.8 to 134.6)	46.2	(4.2 to 88.3)	49.0	(−11.8 to 109.8)	−3.7	(−48.4 to 41.0)
15	152.7	(74.8 to 230.6)	146.8	(95.7 to 197.8)	134.4	(59.1 to 209.7)	61.9	(7.1 to 116.7)
20	265.2	(154.1 to 376.2)	227.3	(155.1 to 299.6)	223.2	(124.3 to 322.2)	103.0	(26.1 to 179.8)
25	391.8	(214.2 to 569.4)	280.8	(161.3 to 400.4)	308.0	(159.5 to 456.6)	187.8	(76.8 to 298.7)
Women								
5	66.1	(43.8 to 88.4)	67.8	(−21.8 to 157.3)	47.4	(24.4 to 70.4)	42.1	(−1.0 to 85.1)
10	135.2	(103.8 to 166.7)	135.5	(−43.6 to 314.6)	112.0	(79.6 to 144.3)	84.1	(−1.9 to 170.2)
15	213.9	(175.0 to 252.8)	203.3	(−65.4 to 472.0)	185.7	(145.5 to 226.0)	126.2	(−2.9 to 255.3)
20	313.9	(264.1 to 363.8)	271.0	(−87.2 to 629.3)	273.7	(221.3 to 326.0)	168.3	(−3.8 to 340.4)
25	426.1	(358.7 to 493.4)	338.8	(−109 to 786.6)	373.2	(302.3 to 444.2)	210.4	(−4.8 to 425.5)

Predictions are for eight hypothetical workers of different gender, smoking and occupational (cotton vs silk) exposures. Predictions assume that these workers have the same age, height, pack-year history (zero if non-smokers, the average number of pack-years if smokers) at each value of work cessation-years. Values represent the adjusted change in FEV_1 (95% CI), in mL, for that value of work cessation-year, as compared to active work.
FEV_1, forced expiratory volume in 1 s.

Table 3 Factors modifying FEV_1 change (in mL) after work cessation based on a linear mixed effects model, with exposure modelled as cotton versus silk

	FEV_1, mL (95% CI)			
	All		**Non-smokers**	
Cessation-years	14.2***	(12.0 to 16.5)	9.4***	(7.1 to 11.7)
Cessation-years^2	0.3***	(0.1 to 0.4)	0.3***	(0.2 to 0.5)
Cotton×cessation-years	−2.7*	(−5.0 to −0.5)	−1.7†	(−4.2 to 0.7)
Current smoker×cessation-years	1.1	(−2.6 to 4.9)	–	–
Former smoker×cessation-years	0.4	(−4.4 to 5.2)	–	–
Male×cessation-years	−5.7**	(−9.1 to −2.3)	−4.5**	(−7.8 to −1.3)
Observations	4702		3068	

Model adjusted for age, height, gender, exposure (cotton vs silk), smoking (never, current, former), cumulative pack-years and interaction terms between cessation-years and occupational exposure, cessation-years and smoking and cessation-years and gender. Loss to follow-up accounted for by using inverse probability of censoring weights. Note: as the modelled outcome was FEV_1 and not change in FEV_1, the interaction terms with cessation-years represent the associations between occupational exposure, smoking, gender and *change in* FEV_1 after leaving work.
Quadratic interaction terms were significant only for the male×cessation-years^2 term, with β=0.5 (0.2, 0.8), p=0.0009 for all workers and β=0.4 (0.1, 0.7), p=0.02 for the analysis restricted to non-smokers.
*p<0.05; **p<0.01; ***p<0.001.
†Cotton×cessation-years interaction p=0.08 in analysis restricted to non-smokers.
FEV_1, forced expiratory volume in 1 s.

most likely explains why there was FEV_1 recovery after leaving the workplace. Others have demonstrated that silk dust can have adverse respiratory effects.[32] However, measured endotoxin and dust levels in the cotton mills were not well correlated (correlation 0.38), and endotoxin levels in the cotton mills were significantly higher than in silk mills (836 vs 0.001 EU/m^3); thus, it is unlikely that the exposure–response relationship we observed with endotoxin is spurious. While the effect of occupational endotoxin exposure on recovery may appear small, the magnitude is comparable to that observed for tobacco smoke in our study. Second, there may be limited generalisability. Our study population is composed entirely of Han Chinese participants; population differences in the prevalence of genetic polymorphisms known to confer differences in risk[33] from endotoxin may exist. Common in occupational studies is the presence of the healthy worker survivor effect, which may be more prominent in cotton textile workers.[34 35] We selected cotton and silk workers who were free of respiratory disease after 2 years of work in the textile workforce; thus, the most susceptible workers were probably excluded from this study and this may explain why few participants ultimately met the criteria for COPD. It is likely that in a less healthy population, the effect of endotoxin exposure on lung function would be larger than that seen in our study. While exposure misclassification is possible as we used area samplers in combination with job histories to estimate individual exposures, misclassification would be expected to be non-differential, and use of area samplers to estimate individual endotoxin exposure has been shown to be a reasonable surrogate.[36] Finally, while we used endotoxin to estimate exposure to Gram-negative bacteria, it is clear that this surrogate of microbial load does not capture the complexity of exposures present in organic dust. Cotton dust contains a diverse variety of bacteria[37] as well as fungi.[38] Recent studies of environmental microbial exposures using high-throughput sequencing to identify microbial type have shown that it may be the presence of specific microbes,[39] or the diversity of microbial exposure,[40] that determines whether the ultimate effect on health is protective or harmful. We do not have residual organic dust to further refine the specific microbial exposures (of which endotoxin may be a marker of) that is associated with decreased lung function recovery. The identification of specific microbes that are harmful using sequencing techniques may represent exciting future areas of research.

In summary, this study is the first to demonstrate that FEV_1 improvement is sustained long after cessation of workplace exposure to organic textile dust. However, the FEV_1 improvement with exposure cessation is delayed in some subgroups, suggesting that studies of recovery of lung function after cessation of an environmental exposure needs to be of a sufficiently long duration. While lung function recovery is sustained, male gender adversely affects recovery, suggesting that men may represent a subpopulation that would benefit from early screening for respiratory disease. Despite exposure cessation, past exposure to occupational endotoxin has an exposure-dependent relationship with decreased FEV_1 recovery, affirming the importance of workplace limits on not just the amount of organic dust, but also the amount of endotoxin.

Table 4 Factors modifying FEV_1 change (in mL) after work cessation based on a linear mixed effects model, with exposure modelled as cumulative occupational endotoxin exposure

	FEV_1, mL (95% CI)			
	All		**Non-smokers**	
Cessation-years	11.8***	(9.7 to 13.9)	7.9***	(5.7 to 10.1)
Cessation-years^2	0.3***	(0.1 to 0.4)	0.3***	(0.2 to 0.5)
Endotoxin×cessation-years	−0.1*	(−0.2 to −0.02)	−0.1[a]	(−0.1 to 0.03)
Current smoker×cessation-years	1.1	(−2.6 to 4.9)	–	–
Former smoker×cessation-years	0.4	(−4.4 to 5.2)	–	–
Male×cessation-years	−5.7**	(−9.1 to −2.3)	−4.5**	(−7.8 to −1.3)
Observations	4702		3068	

Model adjusted for age, height, gender, exposure (cotton vs silk), smoking (never, current, former), cumulative pack-years and interaction terms between cessation-years and occupational exposure, cessation-years and smoking, and cessation-years and gender. Loss to follow-up accounted for by using inverse probability of censoring weights. Note: as the modelled outcome was FEV_1 and not change in FEV_1, the interaction terms with cessation-years represent the associations between occupational exposure, smoking, gender, and *change in* FEV_1 after leaving work.
*p<0.05; **p<0.01; ***p<0.001
Quadratic interaction terms were significant only for the male×cessation-years^2 term, with β=0.5 (0.2, 0.8), p=0.001 for all workers and β=0.4 (0.1, 0.7), p=0.02 for the analysis restricted to non-smokers.
†Cotton×cessation-years interaction p=0.08 in analysis restricted to non-smokers.
FEV_1, forced expiratory volume in 1 s.

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Endotoxin and gender modify lung function recovery after occupational organic dust exposure: a 30-year study

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