

ORIGINAL ARTICLE

Occupational exposure to diesel engine exhaust and alterations in lymphocyte subsets

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ABSTRACT

Background The International Agency for Research on Cancer recently classified diesel engine exhaust (DEE) as a Group I carcinogen based largely on its association with lung cancer. However, the exposure–response relationship is still a subject of debate and the underlying mechanism by which DEE causes lung cancer in humans is not well understood.

Methods We conducted a cross-sectional molecular epidemiology study in a diesel engine truck testing facility of 54 workers exposed to a wide range of DEE (ie, elemental carbon air levels, median range: 49.7, 6.1–107.7 $\mu\text{g}/\text{m}^3$) and 55 unexposed comparable controls.

Results The total lymphocyte count ($p=0.00044$) and three of the four major lymphocyte subsets (ie, CD4+ T cells ($p=0.00019$), CD8+ T cells ($p=0.0058$) and B cells ($p=0.017$)) were higher in exposed versus control workers and findings were highly consistent when stratified by smoking status. In addition, there was evidence of an exposure–response relationship between elemental carbon and these end points ($p_{\text{trends}} < 0.05$), and CD4+ T cell levels were significantly higher in the lowest tertile of DEE exposed workers compared to controls ($p=0.012$).

Conclusions Our results suggest that DEE exposure is associated with higher levels of cells that play a key role in the inflammatory process, which is increasingly being recognised as contributing to the aetiology of lung cancer.

Impact This study provides new insights into the underlying mechanism of DEE carcinogenicity.

What this paper adds

- Although the International Agency for Research on Cancer recently classified diesel engine exhaust (DEE) as a Group I carcinogen based largely on its association with lung cancer, the exposure–response relationship is still an important subject of regulatory debate and the underlying mechanism by which diesel exhaust causes lung cancer in humans is not well understood.
- DEE exposure was associated with higher levels of lymphocytes and certain subsets that play a key role in the inflammatory process, which is increasingly being recognised as contributing to the aetiology of lung cancer.
- There was also evidence that certain lymphocyte subsets were higher among workers exposed to the lowest tertile of DEE.
- This study provides new insights into the potential underlying mechanism of DEE carcinogenicity.

subsets found some evidence of association with certain white blood cell types,⁵ but had several limitations. To evaluate the relationship between long-term, occupational DEE exposure and changes in peripheral blood cells, including the major lymphocyte subsets over a wide range of exposure, we conducted a cross-sectional molecular epidemiology study in a diesel engine testing facility in China.

INTRODUCTION

Recently, the International Agency for Research on Cancer (IARC) classified diesel engine exhaust (DEE) as a Group I carcinogen largely based on its association with lung cancer in epidemiological studies.¹ However, the exposure–response relationship is still an important subject of regulatory debate and the underlying mechanism by which diesel exhaust causes lung cancer in humans is not well understood. A few short-term human controlled DEE exposure studies^{2–4} provide some evidence that DEE can alter several types of white blood cells, but findings are not consistent. In the workplace setting, one study of the relationship between DEE exposure and peripheral lymphocyte

MATERIALS AND METHODS**Diesel engine facility**

Exposed workers were selected from an engine testing facility of a diesel engine manufacturing company that produces diesel engines for light and heavy trucks (production between 10 000 and 20 000 engines a month). All engines, after assembly and without any after treatment system, are tested and tuned by placing them on a diesel engine dynamometer in a ‘Dyno-room’. The facility has in total 44 Dyno-rooms that are semienclosed (open at the top); on average, 25–50% are in use at any point in time. Engine exhaust is ventilated to the outside by directly coupling the engine exhaust



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to a local exhaust system, and by general ventilation of the Dyno-rooms.

Control factories

Control workplaces were selected based on the absence of DEE and general dust exposure. Selected facilities included a bottling department of a brewery (n=24), a water treatment plant (n=18), a meat packing facility (n=8) and an administrative facility (n=5). Based on detailed walk-through surveys, no DEE sources were identified in any of these workplaces.

Exposure assessment

The exposure assessment survey in the diesel factory was conducted from October 2012 to March 2013 encompassing all workers in the testing facility. Repeated full-shift personal air samples for elemental and organic carbon (OC), and fine particulate matter were collected using a cyclone attached to the lapel near the breathing zone, with an aerodynamic cut-off of 2.5 µm (PM_{2.5}) at a flow rate of 3.5 L/min using quartz or Teflon filters, respectively. PM_{2.5} was assessed by preweighing and postweighing of the Teflon filters in an environmentally controlled weighing room using a microbalance at 1 µg accuracy. Each filter was preweighed and postweighed in duplicate. If the duplicate measurements differed by more than 5 µg, the filter was reweighed. Elemental carbon (EC) and OC were measured on the quartz filters using NIOSH Method 5040.⁶ Weights (PM_{2.5}, EC and OC) were divided by the volume of air drawn through the filters to provide exposure concentrations (µg/m³). We determined soot content of the Teflon and quartz filters using a smoke stain reflectometer (model M43D, diffusion Systems Ltd, London, UK) and converted the reflectance value into an absorption coefficient (10⁻⁵/m).

Normal probability plots indicated that PM_{2.5}, EC, OC and soot values followed essentially a log-normal distribution. Individual exposure was estimated using a mixed effect model using the natural logarithm of PM_{2.5} (n=71), EC (n=149), OC (n=149) and soot (n=237). Subjects were assigned as random effects. Fixed effects included job title based on position/task of the subjects in the testing department (testing workers and their location within the department, refuelling workers and inspection workers) and the season during which the measurement was taken (Autumn, Winter, Spring). Location of testing workers was included in the model as general ventilation efficiency differed within the testing department. For EC and OC, we additionally included a fixed effect for current smoking status of the subject, as smoking was allowed during breaks at the workplace and influenced these particular exposure proxies. For soot we additionally included a parameter for the filter type, as reflection was slightly different depending on the type of filter used (ie, Teflon, quartz).

The following formula was used to summarise the model:

$$y_{ij} = \mu + \beta_1 x_1 + \beta_2 x_2 + \dots + \beta_n x_n + bI_i + \varepsilon_{ij}$$

where y_{ij} represents the natural log transformed value of the PM_{2.5}, EC, OC, or soot exposure levels for person i , on day j . μ represents the intercept (ie, the 'background' level). β_1 through β_n represent fixed effect variable coefficients for variables x_1 through x_n . bI_i represents the random effect coefficient for subject i . ε_{ij} represents the error for subject i , on day j . Individual year average exposures were estimated by combining the estimates of the job-title and the estimate of the individual random effect corrected for season, filter type, and/or smoking status.

PM_{2.5} (n=5), EC (n=7), OC (n=7), and soot (n=12) exposure in controls was measured in a subset of the controls per factory except for the beer factory where no measurements could be obtained. Levels were averaged (geometric mean) by factory and assigned to all controls in that factory. Minimal variation between factories was observed and the average of all factories was assigned to the beer factory.

Subject enrolment

In March 2013, we conducted a cross-sectional molecular epidemiology study of 54 workers exposed to DEE in this engine-testing facility and 55 controls, which was integrated into a regular health exam administered by the local Center for Disease Control (CDC). These DEE exposed workers, all of whom are male, spend most of their shift in direct proximity to the engines being tested and, as a consequence, have the potential for exposure to substantial levels of DEE. Fifty-five unexposed male workers, frequency-matched to the exposed workers by age within 5 years and smoking status (ie, never, former, current), were identified in control workplaces in the same local region of China with work processes that do not involve exposure to DEE, other types of particulates, or any known or suspected genotoxic, hematotoxic or immunotoxic chemicals. The participation rates for DEE exposed workers and controls were approximately 90% and 80%, respectively. Peripheral blood samples were collected in an EDTA vacutainer tube for the complete blood cell count (CBC) and differential analysis by the Sysmex platform and in a heparin vacutainer for analysis of the major lymphocyte subsets by a FACSCalibur. Coefficients of variation for all cell counts from the CBC with differential and lymphocyte subsets were ≤5%, with the exception of basophils (13%) and eosinophils (11%). The study was approved by Institutional Review Boards at the US National Cancer Institute and the National Institute of Occupational Health and Poison Control, China CDC. Participation was voluntary and all subjects gave written informed consent.

Statistical analysis

Unadjusted summary measures are presented for all end points. Linear regression using the natural logarithm (ln) of data

Table 1 Demographic characteristics among workers exposed to diesel engine exhaust and controls

Subjects	Controls (n=55)	Exposed (n=54)	p Value
Age, mean (SD)	42.1 (7.4)	42.0 (6.8)	0.99*
BMI, mean (SD)	25.2 (3.8)	24.7 (3.4)	0.57*
Smoking Status			
Current n (%)	35 (63.6)	34 (63.0)	0.95†
Former n (%)	12 (21.8)	11 (20.4)	
Never n (%)	8 (14.5)	9 (16.7)	
Current alcohol use			
No n (%)	8 (14.5)	15 (27.8)	0.091†
Yes n (%)	47 (85.5)	39 (72.2)	
Recent infection			
No n (%)	27 (49.1)	27 (50.0)	0.92†
Yes n (%)	28 (50.9)	27 (50.0)	
Work years in diesel factory			
Mean (SD)	–	19.6 (7.05)	–

*p Value of Wilcoxon test.

†p Value of χ^2 test.
BMI, body mass index.

Workplace

Table 2 Air concentration of DEE components among workers exposed to DEE and controls*

Exposure	Control (n=55)		Exposed (n=54)	
	Mean	(SD)	Mean	(SD)
Elemental carbon, $\mu\text{g}/\text{m}^3$				
Background adjusted	0	NA	48.5	(22.1)
Unadjusted	11.1	1.3	59.6	(22.1)
Organic carbon, $\mu\text{g}/\text{m}^3$				
Background adjusted	0	NA	70.2	(25.9)
Unadjusted	68.7	4.1	138.9	(25.9)
Soot (absorption coefficient in $10^{-5}/\text{m}$)				
Background adjusted	0	NA	43.9	(15.3)
Unadjusted	6.8	0.6	50.7	(15.3)
PM _{2.5} , mg/m^3				
Background adjusted	0	NA	0.1	(0.07)
Unadjusted	0.2	0.07	0.4	(0.07)

*Based on detailed walk-through surveys, no DEE sources were identified in any of these control factories. Adjusted values are adjusted for exposure levels in the control factories, which, it was assumed, had no DEE or particulate matter exposures, to reflect background outdoor levels in this region. Unadjusted values are original measurements.

DEE, diesel engine exhaust; NA, not applicable.

derived from the CBC, differential and lymphocyte subsets was used to test for differences between workers exposed to DEE and controls, and to conduct trend tests. All statistical models

were adjusted for the matching variables, age (as a continuous variable) and smoking status (current, former, never). Potential confounders that have been previously shown to influence one or more of the end points in this report were also included in the final models, that is, current alcohol consumption (yes/no), recent infection (flu or respiratory infections in the previous month) and body mass index (BMI). All analyses were carried out using SAS V.9.2 software (SAS Institute, Cary, North Carolina, USA).

RESULTS

Demographic characteristics of exposed and unexposed subjects are shown in [table 1](#) and were comparable with regard to age, BMI, current smoking status and recent infection ([table 1](#)). Unexposed subjects had a higher proportion of alcohol drinkers than exposed subjects but the difference was not statistically significant. Overall, study subjects were relatively light smokers (mean (SD) 13.2 (6.4) and 12.3 (7.3) cigarettes per day among current smokers in the DEE exposed and control groups, respectively).

There was a wide range of exposure to EC (median, range: 49.7, 6.1–107.7 $\mu\text{g}/\text{m}^3$) and other constituents of DEE including organic compounds, soot and PM_{2.5} ([table 2](#)). We present unadjusted exposure values and values that are adjusted for exposure levels in the control factories that were assumed, due to the absence of any DEE or particulate matter exposure sources, to reflect background outdoor levels in this region.

Table 3 Peripheral blood cell counts in workers exposed to diesel engine exhaust and controls

Subject category	Controls (n=55)	Exposed (n=54)	Difference in mean levels (%)	P _{crude} *	P _{adjusted} †
<i>Peripheral blood cell counts‡</i>					
White blood cell counts, cells/ μL	7808 (1796) 7630	7701 (1449) 7600	−1.38	0.89	0.94
Neutrophils, cells/ μL	4633 (1530) 4370	4146 (1164) 4195	−10.5	0.18	0.074
Basophils, cells/ μL	24.2 (15.1) 20.0	30.6 (19.9) 25.0	26.7	0.023	0.038
Eosinophils, cells/ μL	130 (116) 105	143 (103) 113	10.0	0.29	0.14
Lymphocytes, cells/ μL	2514 (650) 2570	2905 (647) 2807	15.5	0.0021	0.00044
T cells, cells/ μL	1729 (472) 1712	2120 (569) 1990	22.6	0.00027	4.4E-5
CD4+ T cells, cells/ μL	950 (288) 937	1183 (386) 1142	24.6	0.00073	0.00019
CD8+ T cells, cells/ μL	705 (261) 665	827 (261) 771	17.3	0.013	0.0058
B cells, cells/ μL	352 (160) 345	425 (192) 385	20.5	0.051	0.017
Natural killer cells, cells/ μL	414 (211) 376	421 (200) 380	1.84	0.70	0.79
Monocytes, cells/ μL	507 (147) 495	476 (107) 470	−6.27	0.31	0.43
Haemoglobin, g/L	157 (9.8) 158	154 (10.4) 154	−1.90	0.10	0.18
Mean corpuscular volume, fL	91.5 (4.6) 91.5	90.7 (3.8) 90.2	−0.85	0.22	0.29
Platelets, $10^3/\mu\text{L}$	234 (46.7) 234	232 (52.5) 231	−0.71	0.62	0.98

*p Value of Wilcoxon test, control versus exposed workers.

†p Value using the ln transformation of the dependent variable and category of diesel engine exhaust (control vs exposed workers), adjusted for age, BMI, smoking status, current alcohol use and recent infection.

‡Unadjusted mean (\pm SD) and median.
BMI, body mass index.

Among the DEE exposed workers, the correlation was very high between EC and OC (Spearman $R=0.86$, $p<0.0001$) and soot ($R=0.91$, $p<0.0001$). We use EC as the primary exposure proxy for DEE for analyses of biological end points since EC is considered a specific marker for DEE in occupational settings and has been used in the most recent epidemiological investigations of lung cancer.^{7 8} There was no correlation between EC and $PM_{2.5}$ among the DEE exposed subjects ($R=0.09$, $p=0.53$).

The total lymphocyte count and three of four major lymphocyte subsets including $CD4^+$ T cells, $CD8^+$ T cells and B cell counts were statistically significantly increased in workers exposed to DEE compared to controls (table 3), with 16–25% higher cell counts in exposed versus control workers. In contrast, there was no evidence that the fourth major lymphocyte subset, natural killer (NK) cells, was elevated among DEE exposed workers. Also, total T cells were highly significantly elevated in DEE exposed workers compared to controls ($p=0.00044$). In addition, the basophil count was elevated in DEE exposed workers compared to controls. More extensive adjustment in the analysis for tobacco intensity, duration and pack-years, and for alcohol intensity, did not alter the findings (not shown). Further, we conducted sensitivity analyses by excluding one group of control workers at a time and comparing the remaining controls to DEE exposed subjects; findings were minimally altered (not shown).

To evaluate the consistency of these findings by smoking status, we conducted analyses among current, former and never smokers. Although these subgroups were relatively small, especially never smokers, we found that the above patterns were highly consistent across the three groups (see figure 1 and online supplementary tables S1a–c). However, statistically

significant differences were present only for effects among current and former smokers, not among never-smokers.

We observed statistically significant exposure–response relationships with lymphocytes and lymphocyte subsets (with the exception of NK cells) with air levels of EC (table 4) comparing controls and DEE exposed workers categorised into tertiles of exposure based on personal air monitoring for EC. Similar results were obtained using DEE as a continuous exposure variable. In analyses conducted among the DEE exposed workers only, there was a significant exposure–response relationship for $CD4^+$ T cells (table 4). Further adjustment in the analysis for duration of employment did not alter the findings (not shown). There was no association between air levels of $PM_{2.5}$ and other peripheral blood counts among the DEE exposed workers.

DISCUSSION

We found that occupational exposure to DEE was associated with an elevation in the total peripheral lymphocyte count and in three of the four major lymphocyte subsets (ie, $CD4^+$ T cells, $CD8^+$ T cells and B cells) and was consistent across current, former and never smokers. However, statistically significant differences were present only for effects among current and former smokers, not among never-smokers, which were due to the very small sample size of the latter group. Further, there was evidence of exposure-dependent increases with air levels of EC and these end points overall, and for $CD4^+$ T cell counts among the exposed workers only. This is the first molecular epidemiology study, to the best of our knowledge, that has systematically evaluated the major lymphocyte subsets, and compared effects in workers exposed to DEE and unexposed controls in an occupational setting.

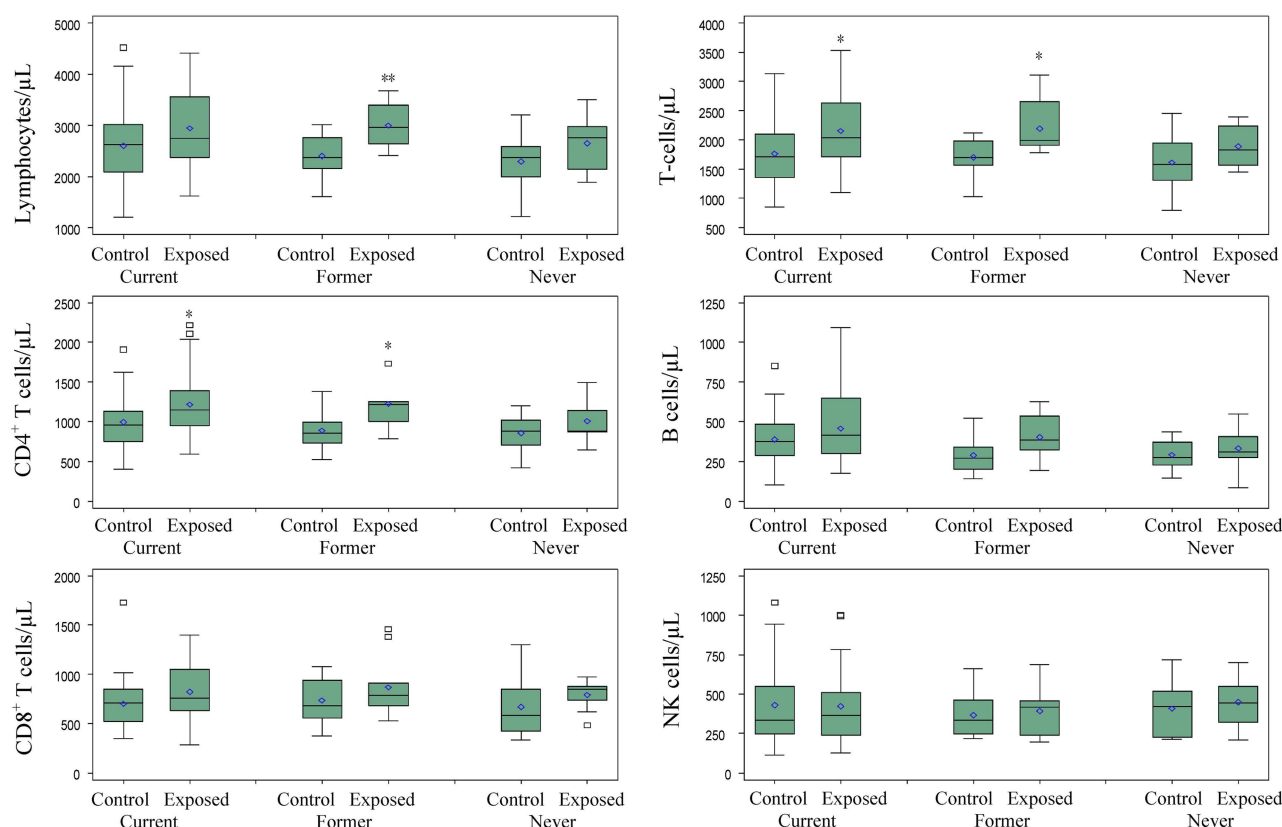


Figure 1 Peripheral blood lymphocyte cell counts in workers exposed to diesel engine exhaust and controls, by smoking status. Cell counts are shown by box-and-whiskers plots. Differences in cell counts were tested using a linear regression analysis of ln-transformed end points. The p values are indicated as: * $p<0.05$; ** $p<0.01$.

Table 4 Exposure–response relationship between air concentrations of EC among workers exposed to DEE and controls, and peripheral blood cell counts*

Subject category	Elemental carbon exposure category†				P _{trend} (all subjects)‡	P _{trend} (exposed only)§
	Control factory workers (n=55)	Diesel engine manufacturing workers				
		1st tertile (n=18)	2nd tertile (n=17)	3rd tertile (n=19)		
Peripheral blood cell counts¶						
White cell count, cells/μL	7808 (1796) 7630	7906 (1688) 7508	7182 (1312) 7340	7971 (1259) 8050	1.00	0.83
Neutrophils, cells/μL	4633 (1530) 4370	4420 (1510) 4325	3860 (889)** 3890	4143 (993) 4195	0.078	0.68
Basophils, cells/μL	24.2 (15.1) 20.0	32 (17) 30	27 (14) 25	32 (26) 25	0.070	0.87
Eosinophils, cells/μL	130 (116) 105	154 (109) 133	138 (121) 75	137 (84) 115	0.23	0.83
Lymphocytes, cells/μL	2514 (650) 2570	2817 (524)** 2763	2729 (639) 2635	3146 (714)** 3305	0.00021	0.086
T cell, cells/μL	1729 (472) 1712	2071 (525)** 2013	1963 (505) 1856	2307 (637)** 2378	3.9E-05	0.16
CD4+ T cells, cells/μL	950 (288) 937	1149 (361)** 1145	1013 (335) 996	1368 (388)** 1351	3.4E-05	0.034
CD8+ T cells, cells/μL	705 (261) 665	828 (258)** 746	826 (258) 770	826 (282) 830	0.022	0.90
B cells, cells/μL	352 (160) 345	381 (114) 365	376 (157) 406	510 (249)** 523	0.0073	0.093
Natural killer cells, cells/μL	414 (211) 376	395 (145) 423	450 (244) 379	420 (210) 368	0.76	0.84
Monocytes, cells/μL	507 (147) 495	482 (95) 495	429 (95)** 410	512 (117) 545	0.70	0.56
Haemoglobin, g/L	157 (9.8) 158	151 (11) 153	155 (12) 155	156 (8) 157	0.67	0.22
Mean corpuscular volume, fL	91.5 (4.6) 91.5	92 (3) 91	89 (3) 89	91 (5) 90	0.25	0.42
Platelets, 10 ³ /μL	234 (46.7) 234	247 (50) 235	218 (46) 225	231 (59) 219	0.65	0.46

*Workers classified into lower, medium and higher DEE (diesel engine exhaust) exposure groups based on workplace evaluation as described in the methods.

†Adjusted concentrations are presented to characterise EC (elemental carbon) levels in tertiles of DEE exposed workers above background levels (also, see [table 2](#) and text). The median (range) EC (μg/m³) levels for the three categories: 1st tertile, 23.8 (6.1–39.0), 2nd tertile, 49.7 (39.1–54.5), 3rd tertile, 69.4 (54.6–107.7).

‡P value using the ln transformation of the dependent variable and category of diesel exposure as an ordinal variable taking on values of 0, 1, 2 and 3 (0: controls, 1: lower exposed, 2: medium exposed and 3: higher exposed), adjusted for age, BMI (body mass index), smoking status, current alcohol use and recent infection.

§P value using the ln transformation of the dependent variable and category of diesel exposure as an ordinal variable taking on values of 1, 2 and 3 (1: lower exposed, 2: medium exposed and 3: higher exposed), adjusted for age, BMI, smoking status, current alcohol use and recent infection.

¶Unadjusted mean (±SD) and median.

**p Value <0.05 compared to controls, adjusted for age, BMI, smoking status, current alcohol use and recent infection.

One previous study evaluated the same end points in an occupational setting among salt miners but did not include an unexposed control group. They found evidence of non-significant increased levels of lymphocytes and T cells when looking at relationships with surrogates of DEE exposure.⁵ Overall, our results are consistent with these findings of increased lymphocyte and lymphocyte subsets among DEE exposed workers in our study.

Most data in the literature regarding the impact that DEE has on peripheral blood and immune cells come from controlled human studies with exposure to DEE lasting 1–3 h. Results from these studies have supported the hypothesis that DEE is associated with pulmonary and systemic inflammation.⁹ These short-term exposure studies found evidence of inflammatory changes in bronchoalveolar lavage and bronchial biopsies, but results in peripheral blood were less consistent.^{2–4 10–12} After DEE exposure in controlled chambers, there were higher levels of neutrophils, eosinophils, mast cells and lymphocytes, including total T cells, CD4+ T cells and CD8+ T cells^{2–4 10–12} in bronchoscopy samples. These findings indicate that short-term DEE exposure has the potential to induce airway inflammatory

responses. Our study, which evaluated workers who had been employed for many years in a factory that produces diesel engines, found that multiple lymphocyte subsets were associated with DEE exposure, especially T cells. The strongest evidence for association was for CD4+ T cells, which showed a statistically significant exposure–response relationship among the exposed workers ([table 4](#)), and it is well established that certain subgroups of CD4+ T cells (eg, those involved with the Th1 and Th17 pathways) play an important role in the inflammatory response.¹³ Although the physiological significance of the magnitude of change we observed for lymphocyte subsets counts in the DEE exposed workers versus controls is not clear with regard to future risk of lung cancer, an increase in multiple lymphocyte subsets has been found in the airways and lung parenchyma of individuals with chronic obstructive pulmonary disease, which is itself a risk factor for lung cancer.¹⁴ In addition, a recent report from a prospective cohort study has shown that higher levels of several serum markers of inflammation that stimulate T lymphocyte proliferation or are produced in part by lymphocyte subsets associated with DEE exposure in our study

were associated with future risk of lung cancer, providing a potential link between our findings and lung cancer.¹⁵

We did not find evidence that smoking interacted with DEE exposure using the end points analysed in this study. A previous nested case-control study of lung cancer in a cohort of non-metal miners with detailed DEE exposure data found that the effect of DEE exposure and risk of lung cancer was attenuated among current heavy smokers (ie, >2 packs per day).⁷ Smokers in our study population were relatively light smokers (ie, <1 pack per day) and as such we could not explore this previously observed interaction.

A strength of this study was the detailed exposure assessment among the diesel exposed workers. Exposure assessment among the controls was more limited and covered a random subsample of the control workers. We did detect relatively high levels of PM_{2.5}, EC and OC in the air samples from the control workers (though far lower than levels measured in the diesel engine testing facility). These were assumed to be due to high outdoor air pollution levels because of the absence of any indoor combustion sources in these factories. As this introduced some uncertainty in the exposure assessment, we present adjusted as well as unadjusted results for exposure levels in the control factories. Results of these analyses, however, were not materially different.

In summary, our study showed that lymphocyte counts and three of the major lymphocyte subsets including CD4+ T cells, CD8+ T cells and B cells were significantly increased among workers exposed to DEE compared to controls, with evidence of an exposure-response relationship for these end points. Further, these patterns were consistent among subgroups of the study population defined by smoking status. Our findings suggest that DEE exposure is associated with higher levels of peripheral blood cells that play a key role in the inflammatory process, which is increasingly being recognised as playing an important role in the aetiology of lung cancer.¹⁵ However, additional work is needed to explore other immunological and inflammatory changes that occur in the presence of DEE, to evaluate other potential mechanisms of DEE biological action, and to further explore exposure-response relationships. Finally, larger studies are needed to replicate and extend these findings over a wide range of DEE exposures and to explore potential interactions between constituents of DEE and tobacco exposure.

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Contributors QL, RV, DS, YZ and NR conceived of and designed the study. QL, RV, YD, DR, WH, HD, YN, JX, WF, KM, BZ, JY, MY, XJ, TM, PB, HDH, DS and NR participated in data collection. QL, RV, WH, DS and NR conducted the analyses, and were primarily responsible for drafting the paper. All authors reviewed and approved the manuscript.

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Competing interests None.

Patient consent Obtained.

Ethics approval US National Cancer Institute and the National Institute for Occupational Health and Poison Control, China CDC.

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