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RESEARCH ARTICLE

A study of pneumoproteins in crystalline silica exposed rock drillers

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ABSTRACT

Objective: The objective was to assess serum concentrations of club cell protein 16 (CC-16) and the surfactant proteins A (SPs-A) and D (SP-D) in male rock drillers ($N = 123$) exposed to crystalline silica and in 48 occupationally non-exposed. **Methods:** The arithmetic mean (AM) duration of exposure was 10.7 years. The geometric mean (GM) crystalline silica exposure was 36 μg/m^3 at the time of the study. The GM cumulative exposure was 239 μg/m^3. **Results:** The concentrations of SP-D (GM 12.7 vs. 8.8 μg/L, $p < 0.001$) and SP-A (AM 1847 vs. 1378 ng/L, $p = 0.051$) were higher among rock drillers than among occupationally non-exposed. A positive significant association was observed between cumulative crystalline silica exposure and the SP-D concentrations ($\beta = 0.07$; $p < 0.05$). Rock drillers with small airway obstruction with maximal mid-expiratory flow % (MMEF%) <70% ($N = 29$) had higher SP-D concentrations than rock drillers with MMEF% ≥ 70% ($N = 91$) (GM 17.3 *vs.* 11.4 μg/L, $p = 0.001$). Rock drillers with MMEF% ≥70% ($N = 91$) had higher concentrations of SP-A (1957 vs. 1287 ng/L, $p = 0.01$) and SP-D (11.4 vs. 9.0 μg/L, $p = 0.007$) than non-exposed with MMEF% ≥70% ($N = 39$). Rock drillers with airway obstruction (FEV1/FVC < 0.70, $N = 11$) had significantly lower CC-16 concentrations than rock drillers with FEV1/FVC ≥0.70 ($N = 109$) after adjusting for relevant potential confounders ($p = 0.02$). **Conclusion:** The results indicate that pulmonary surfactant is a target for crystalline silica toxicity. The alterations appear to be driven by pulmonary alterations in the small airways and by exposure itself. Further studies on pneumoproteins and pulmonary function in other groups of workers exposed to crystalline silica are needed.

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CC-16; SP-A; SP-D; rock drilling; crystalline silica

Introduction

Many job tasks in the heavy construction industry may generate airborne dust containing crystalline silica. Rock drillers operate different types of rock drilling equipment. Granite, which is a common rock in Norway, may contain up to 24–40% quartz (NGU (Geological Survey of Norway) 2015). Inhalation of crystalline silica is a significant contributor to occupational mortality and morbidity globally, and silicosis and chronic obstructive pulmonary disease (COPD) have been associated with such exposure (Hnizdo and Vallyathan 2003; Bergdahl et al. 2004; Leung et al. 2012; Möhner et al. 2013; Brüske et al. 2014; Tavakol et al. 2017).

Pneumoproteins are regarded as markers of lung injury and inflammation. Surfactant protein (SP) A (SP-A) and SP-D are mainly synthesized in alveolar type II cells (AT II) and catabolized by alveolar macrophages (AMs) and AT II (Ikegami 2006; Han and Mallampalli 2015). SP-A and SP-D have a multitude of functions, e.g. the ability to recognize and bind pathogens, attach the opsonized pathogens to phagocytic cells, and to reduce inflammation through a number of cellular receptors (Sorensen 2018; Watson et al. 2019). SP-D has also important roles in maintaining pulmonary surfactant, as shown by increased alveolar and tissue phosphatidylcholine and increased number of foamy macrophages in SP-D deficient mice (Botas et al. 1998; Korfhagen et al. 1998). One important role of SP-A in the homeostasis of surfactant is to promote surfactant uptake by pneumocytes (Nathan et al. 2016; Olmeda et al. 2017). Club cell protein 16 (CC-16) is synthesized in CCs present throughout the respiratory tract with the highest density in the respiratory bronchioles. Anti-inflammatory properties of CC-16 have been shown in mice (Laucho-Contreras et al. 2016).

Human surfactant is composed of around 80% phospholipids, 10% neutral lipids, and 10% SPs (Perez-Gil and Weaver 2010). Intratracheal exposure to crystalline silica in rats resulted in higher intracellular and alveolar space levels of phospholipids (Grünspan et al. 1973; Dethloff et al. 1986). Similar observations were reported in sheep (Lesur et al. 1993). Increased lipid content in AMs collected from bronchoalveolar fluid of silicotic humans has also been observed, the increase being larger in severe than in slight silicosis (Hou et al. 2019). Rat macrophage toxicity induced by crystalline silica *in vitro* was significantly reduced by adding SP-A (Spech et al. 2000). Increased SP-D levels in alveolar cells were also observed in rats after instillation with crystalline silica intratracheally (Barbaro et al. 2002). Higher serum SP-A levels by lower SP-A levels in the lung

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tissue were observed in rats (Liu et al. 2016). The above studies indicate increased turnover and alterations of surfactant and SP levels caused by silica exposure.

Few studies have analyzed pneumoproteins in serum of crystalline silica exposed workers. Workers diagnosed with silicosis and workers currently exposed to crystalline silica had higher SP-D and lower CC-16 concentrations in serum compared to referents (Bernard et al. 1994; Wang et al. 2007), while Xue et al. (2017) reported similar concentrations of SP-D in serum among silicosis patients and referents.

This study is part of an investigation of pulmonary health among crystalline silica exposed rock drillers. We have previously reported a decline in pulmonary function (Ulvestad et al. 2020). The primary aim of the present work was to assess the impact of exposure on serum concentrations of the pulmonary biomarkers SP-A, SP-D, and CC-16. A further aim was to assess whether pulmonary function may be associated with pneumoprotein concentrations.

Materials and methods

Study design

Three major contractors carrying out rock drilling outdoors in Norway were asked to participate in this cross-sectional study. The companies, employing altogether 140 male rock drillers (defined as working in the immediate vicinity of a drill rig), accepted the invitation. All 140 workers occupationally exposed to crystalline silica (occupationally exposed) were invited to participate in the study. Four subjects declined. It also turned out that 13 of the subjects were no longer exposed at the time of the examinations and were therefore excluded. Hence, the study comprised 123 currently crystalline silica exposed rock drillers. A group of workers non-occupationally exposed to crystalline silica (non-exposed) ($N = 48$) was also recruited. They had to our knowledge never been occupationally exposed to particulate matter (PM) or crystalline silica. They were administrative personnel, typically foremen and other non-exposed heavy construction workers, working at the same construction sites as the rock drillers. All the non-occupationally exposed workers agreed to participate.

Informed written consent for the voluntary participation in the study was obtained from all participants. The study was approved by the South East Norwegian Regional Ethical Committee for Medical Research (REK) (2015/2116).

Job descriptions

The typical working day of a rock driller lasts 10–12 h. Rock drillers operate drilling rigs, while blasting leaders and guardrail installers work close to the drilling operation. Depending on how the jobs were performed, six different job categories were defined. Individual exposure was found to be closely connected to job category. Further details on job descriptions have been published (Ulvestad et al. 2020).

Clinical examinations

The work sites of the participants were scattered throughout the country. Hence, medical examinations were carried out at the companies' regional offices immediately after the end of a 4 d working week. Background data were recorded. Height and weight were measured for the calculation of body mass index (BMI). The participants were interviewed about earlier and current jobs, including occupational dust exposure, and type of drilling equipment they had used. Smoking habits, current and previous diseases were recorded. Finally, health examinations, including high-resolution computed tomography (HRCT) and spirometry (without the use of bronchodilators), were carried out. Blood samples were collected for the purpose of assessing biomarkers. HRCT was carried out according to Ulvestad et al. (2020).

A Spirare SPS version 330 spirometer was used according to the ERS/ATS recommendations (Miller et al. 2005). Recorded variables were forced vital capacity (FVC), forced expiratory volume in one sec (FEV_1), and maximal mid-expiratory flow (MMEF). We have previously reported significantly lower FEV_1/FVC ratio and MMEF% predicted among the occupationally exposed rock drillers (Ulvestad et al. 2020). In order to study pneumoprotein concentrations in relation to pulmonary function, a FEV_1/FVC ratio <0.70 was regarded as indicative of COPD (McNulty and Usmani 2014). We are not familiar with any similar cutoff for MMEF% predicted, and therefore set MMEF% predicted <70 as a cutoff indicative of small airways dysfunction.

Blood sampling

Ten mL of whole blood was collected from the cubital vein in vacuum tubes without additives (Becton Dickinson and Company, Franklin Lakes, NJ, USA) and coagulated for 45 min before centrifugation at 2000 *g* for 15 min. Serum was pipetted into two 4.0 mL NUNC® polypropylene cryotubes (Thermo Fisher Scientific, Waltham, MA, USA) and frozen immediately before long-term storage at −80 °C. The time of blood sampling (in minutes after midnight) was registered.

Pneumoprotein analysis in serum

Protein levels of CC-16 and SP-D were measured using DuoSet ELISA kits obtained from R&D systems (Stillwater, MN, USA). SP-A was measured using antibodies from Abnova (Abnova, Taipei, Taiwan). Samples were missing for five subjects. All analyses were performed according to the manufactures' instructions. Intra-assay coefficients of variance were <11% for all assays.

Occupational exposure measurements

Exposure assessment has been previously described (Ulvestad et al. 2020). In brief, during the years 2015–2018, 23 construction sites of the three companies were surveyed

in order to assess exposure for each job task by personal air sampling. Ninety-eight and 93 samples were collected for the determination of PM of the respirable and the thoracic aerosol fraction, respectively. The crystalline silica content was determined in 67 respirable samples. PM in the respirable and the thoracic aerosol fraction was collected as previously described (Ulvestad et al. 2020). Crystalline silica was not measured among the occupationally non-exposed workers. The PM mass was determined by weighing the filters before and after sampling, using a Sartorius MC 5 microbalance (Sartorius AG, Göttingen, Germany). Respirable α-quartz was measured by X-ray diffraction spectrometry according to the silver filter method NIOSH Method 7500, with a Malvern Panalytical X'Pert^{3}Powder diffractometer, equipped with a PIXcel^{1D} detector and an Empyrean X-ray tube (Malvern Panalytical B.V., Eindhoven, Netherlands).

Calculation of cumulative exposure

Rock drillers may have performed several types of drilling-related jobs over the years. The mean exposure of each of six defined job categories was calculated based on concentrations measured in the collected air samples. Based on a thorough occupational history, the cumulative exposure to respirable crystalline silica was calculated for each worker by adding up the arithmetic mean (AM) exposure for each year in a particular drilling job. The individual cumulative exposures within each job category were finally added to give an individual cumulative exposure. Further details have been published (Ulvestad et al. 2020).

Statistics

The distributions of the variables were examined visually and data were log-transformed when the skewness exceeded 2.0. For those variables, geometric means (GMs) are presented while AM is used otherwise. Group differences were assessed with the Students t-test. Multiple linear regression analysis (backwards procedure) was used to assess associations between pneumoprotein concentrations and several independent variables simultaneously. Independent variables were ever being exposed to crystalline silica (1/0), current smoker (1/0), age (years), BMI (kg/m^2), and the time of blood sampling (in minutes after midnight). If ever being exposed to crystalline silica was associated with pneumoprotein concentrations, the variables cumulative exposure and current exposure were added separately (to avoid collinearity) to the model instead of ever being exposed, and the multiple linear regression analysis performed among the exposed rock drillers only. As a final approach, the exposed subjects were stratified into three equally large groups; low, medium, and high cumulative exposure, for the purpose of visualizing dose-response associations adjusted for relevant confounders. General linear models were used for the adjustments. Two-tailed p values <0.05 were considered to be of statistical significance. The statistical data package SPSS version 25.0 (IBM Corporation, Somers, NY, USA) was used for the analyses.

Results

The exposed rock drillers were comparable to the non-occupationally exposed workers with respect to age, smoking habits and BMI (Table 1). They had been exposed for 10.7 years on average, and their GM cumulative exposure to crystalline silica was 239 μg/m^3.

The concentrations of SP-D were significantly higher ($p < 0.001$) and SP-A was nearly significantly higher ($p = 0.051$) among the rock drillers, as compared to the non-occupationally exposed workers (Table 2). The CC-16 concentrations were similar in the two groups. The results changed only slightly when adjusting for variables that were associated with the pneumoprotein concentrations in the regression analysis (Table 3, Panel A).

The results from the multiple linear regression analysis showed that the concentrations of SP-A and SP-D were

Table 2. The concentrations of serum pneumoproteins in rock drillers occupationally exposed to crystalline silica and occupationally non-exposed workers.

	Exposed ($N = 123$) AM† (min–max)	Non-exposed ($N = 48$) AM (min–max)	p Value
SP-A (ng/L)	1847(43–3934)[c]	1378 (1–3912)[e]	0.051
SP-A $_{Adj}$[a]	1839	1402	0.06
SP-D (μg/L)‡	12.7 (4.0–100)[c]	8.8 (3.6–17.2)[d]	<0.001
SP-D$_{Adj}$[a]	12.8	8.6	<0.001
CC-16 (μg/L)	12.4 (2.4–31.3)[c]	12.3 (3.3–34.4)[d]	0.89
CC-16$_{Adj}$[b]	12.4	12.5	0.97

†Arithmetic mean; ‡geometric mean; $_{Adj}$[a] adjusted for current smoking; $_{Adj}$[b] adjusted for BMI, current smoking, and time of blood sampling; c) two subjects missing; d) three subjects missing; e) four subjects missing.

Table 1. Background characteristics of the study participants.

	Exposed ($N = 123$) AM† (min–max)	Non-exposed ($N = 48$) AM (min–max)	p Value
Age (years)	38.9 (18–72)	37.3 (22–65)	0.38
BMI (kg/m^2)	27.0 (20.2–41.7)	26.2 (21.0–41.4)	0.23
Current smokers (%)#	20.3	25.0	0.51
Former smokers (%)#	20.3	14.6	0.39
Sampling of blood after midnight (min)	693 (482–1004)	695 (523–909)	0.92
FEV$_1$/FVC	0.79 (0.59–0.91)[a]	0.81 (0.67–0.88)[b]	0.01
MMEF%	84 (43–153)[a]	94 (46–148)[c]	0.02
Years exposed	10.7 (1–42)	–	–
Cumulative quartz (years × μg/m^3)‡	239 (5 – 5885)[a]	–	–
Current respirable particulate matter (μg/m^3)‡	227 (91–1360)	–	–
Current quartz (μg/m^3)‡	36 (12–440)	–	–

†Arithmetic mean; ‡geometric mean; #prevalence; a) means one subject missing, b) means two subjects missing, and c) means three subjects missing.

Table 3. Multiple linear regression analysis including all subjects (Panel A) and occupationally exposed rock drillers only (Panel B).

	SP-A (ng/L) β-coef	p value	SP-D (lg) (μg/L) β-coef	p value	CC-16 (μg/L) β-coef	p value
A) All subjects (exposed and non-exposed)[a] ($N = 171$)						
Exposed	240	<0.05	0.09	<0.001	–	
Current smoking	−724	<0.01	0.16	<0.001	−2.6	<0.05
Age	–		–		–	
BMI	–		–		–	
Time of day collecting blood	–		–		−0.01	<0.01
Multiple R	0.28	<0.01	0.39	<0.001	0.29	<0.01
B) Only occupationally exposed rock drillers[b] ($N = 123$)						
Cumulative exposure	–		0.07	<0.05	–	
Current smoking	−819	<0.01	0.16	<0.01	−3.2	<0.05
Age	–		–		–	
BMI	–		–		–	
Time of day collecting blood	–		–		−0.01	<0.01
Multiple R	0.24	<0.01	0.32	<0.01	0.38	<0.01

SP-A, SP-D (lg), and CC-16 are used as dependent value.
(–) Not included in the final model; [a]6, 5, and 5 subjects missing for SP-A, SP-D, and CC-16, respectively; [b]Two subjects missing.

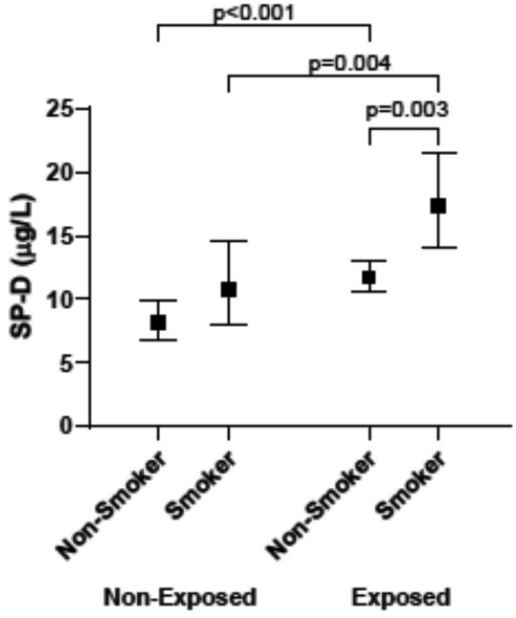

Figure 1. SP-D concentrations in smoking ($N = 25$) and nonsmoking ($N = 96$) rock drillers occupationally exposed to crystalline silica and in smoking ($N = 12$) and nonsmoking ($N = 33$) occupationally non-exposed workers according to current smoking habits. Geometric mean (95% CI).

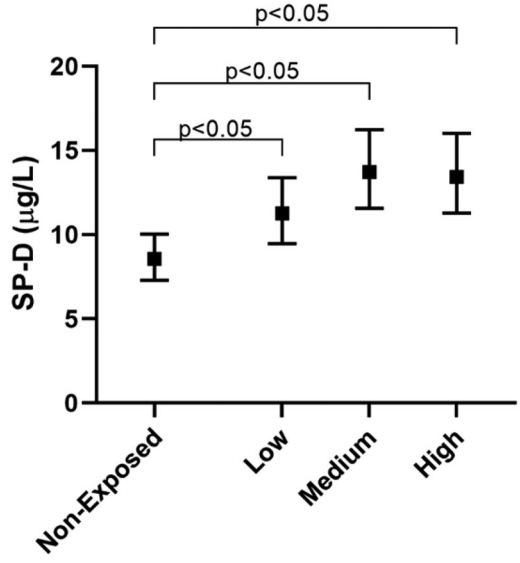

Figure 2. SP-D in non-exposed workers ($N = 45$) and in rock drillers occupationally exposed to crystalline silica stratified into low ($N = 38$, GM 44 μg/m^3, min–max 5–114), medium ($N = 41$, GM 217 μg/m^3, min–max 118–354), and high ($N = 41$, GM 1269 μg/m^3, min–max 365–5885) cumulative quartz exposure adjusted for age and smoking. Geometric mean (95% CI).

significantly associated with being occupationally exposed as a rock driller (Table 3, Panel A). Being a current smoker was associated with all serum pneumoprotein concentrations. The CC-16 concentrations, but not SP-A and SP-D, were significantly associated with the time of day when the blood samples were collected.

The SP-D concentrations were associated with cumulative exposure to crystalline silica when only the rock drillers were considered in the multiple linear regression analysis, while no such association was observed for SP-A (Table 3, Panel B). Current smoking was associated with the concentrations of all three pneumoproteins.

Figure 1 shows that the highest SP-D concentrations were measured among exposed rock drillers that were current smokers. The concentrations were significantly ($p = 0.003$) higher among smoking rock drillers compared to nonsmoking rock drillers. Occupationally exposed nonsmokers had significantly ($p < 0.001$) higher concentrations than nonsmoking non-occupationally exposed workers. The difference among smokers was also of statistical significance ($p = 0.004$).

Figure 2 shows the SP-D concentrations (adjusted for age and current smoking) among non-occupationally exposed workers and exposed rock drillers stratified into three equally large groups according to cumulative exposure. Stratifying the exposed rock drillers into three groups according to years of exposure did not reveal any significant group differences. Neither current exposure measures to crystalline silica nor being a former smoker were associated with any of the pneumoprotein concentrations (data not shown).

We have previously shown that the pulmonary measures MMEF% predicted and FEV_1/FVC ratio were significantly lower in the exposed rock drillers than in the non-occupationally exposed workers, while all other pulmonary

Table 4. The concentrations of pneumoproteins in rock drillers occupationally exposed to crystalline silica according to having MMEF% predicted below or above 70% or FEV_1/FVC ratio below or above 0.70.

	MMEF% predicted		
	<70 (N = 29) AM† (min–max)	≥70 (N = 91) AM (min–max)	p Value
SP-A (ng/L)	1545 (171–3917)	1957 (43–3934)	0.13
$SP\text{-}A_{Adj}$	1693	1910	0.48
SP-D (μg/L)‡	17.3 (5.5–100)	11.4 (4.0–30.9)	0.001
$SP\text{-}D_{Adj}$	16.6	11.5	0.004
CC-16 (μg/L)	11.5 (2.4–29.2)	12.8 (2.8–31.3)	0.32
$CC\text{-}16_{Adj}$	11.7	12.7	0.46
	FEV_1/FVC		
	<0.70 (N = 11) AM (min–max)	≥0.70 (N = 109) AM (min–max)	p Value
SP-A (ng/L)	1370 (372–3914)	1907 (43–3934)	0.12
$SP\text{-}A_{Adj}$	1578	1886	0.49
SP-D (μg/L)‡	16.5 (8.7–80.6)	12.2 (4.0–100)	0.11
$SP\text{-}D_{Adj}$	15.0	12.4	0.30
CC-16 (μg/L)	8.2 (2.4–15.5)	12.9 (2.8–31.3)	0.02
$CC\text{-}16_{Adj}$	8.3	12.9	0.02

†Arithmetic mean; ‡geometric mean; Adj: adjusted for age, current smoking, body mass index, and time of blood sampling.

Table 5. The concentrations of serum pneumoproteins in rock drillers occupationally exposed to crystalline silica and non-exposed workers with both FEV_1/FVC ratio ≥0.70 and MMEF% ≥70 predicted.

	Exposed (N = 91) AM† (min–max)	Non-exposed (N = 39) AM (min–max)	p Value
Age	38 (18–72)	37 (22–58)	0.47
FEV_1/FVC	0.82 (0.72–0.91)	0.82 (0.72–0.88)	0.83
MMEF%	94 (70–153)	97 (70–148)	0.36
SP-A (ng/L)	1957 (43–3934)	1287 (1–3912)	0.01
$SP\text{-}A_{Adj}$	1949	1307	0.02
SP-D (μg/L)‡	11.4 (4.0–30.9)	9.0 (3.6–17.2)	0.007
$SP\text{-}D_{Adj}$	11.5	8.7	0.003
CC-16 (μg/L)	12.8 (2.8–31.3)	12.8 (4.5–34.4)	0.98
$CC\text{-}16_{Adj}$	12.7	12.9	0.90

†Arithmetic mean; ‡geometric mean; Adj: adjusted for age, current smoking, body mass index, and time of blood sampling.

measures did not differ between the two groups (Ulvestad et al. 2020). Table 4 shows the pneumoprotein concentrations among occupationally exposed rock drillers with MMEF% predicted <70% or FEV_1/FVC <0.70 compared to rock drillers having values above these cutoff levels. Exposed rock drillers with MMEF% predicted <70% had substantially higher SP-D concentrations than rock drillers having MMEF% predicted 70%, also after adjusting for age, BMI, current smoking, and time of blood sampling ($p < 0.004$). The SP-A and CC-16 concentrations were similar in the two groups. Exposed rock drillers with FEV_1/FVC <0.70 had significantly ($p = 0.02$) lower CC-16 concentrations than rock drillers with FEV_1/FVC ≥0.70 after adjusting for relevant potential confounders. Since only one non-exposed worker had FEV/FVC <0.70 and three had MMEF% predicted <70%, no further statistical evaluation was performed.

Table 5 shows the pneumoprotein concentrations among all occupationally exposed rock drillers and non-occupationally exposed workers with both MMEF% ≥70 predicted and FEV_1/FVC ratio ≥0.70, which can be regarded as normal pulmonary functions. Although age and the pulmonary measures were similar in the two groups, the concentrations of SP-A and SP-D were significantly higher in the exposed rock drillers than in the non-occupationally exposed workers.

Occupationally exposed rock drillers who had been diagnosed with emphysema by HRCT ($N = 4$) had significantly higher SP-D concentrations compared to those who had not been diagnosed with emphysema ($N = 31$) (GM 26.3 μg/L, min–max 18–51 vs. 13.3 μg/L, min–max 4.0–100, $p = 0.04$) (results not tabulated).

Discussion

This study showed that rock drillers occupationally exposed to crystalline silica had higher concentrations of SP-A and SP-D in serum than non-occupationally exposed workers of similar age and smoking habits. Exposed workers with airways obstruction had higher mean level of SP-D and lower mean level of CC-16 in serum. However, SP-D and SP-A concentrations were also significantly higher in occupationally exposed workers when excluding workers with airway obstruction. Exposed rock drillers with emphysema ($n = 4$) diagnosed by HRCT had substantially higher SP-D concentrations than rock drillers without radiological signs of emphysema.

Occupationally exposed rock drillers had higher levels of SP-D in serum compared to occupationally non-exposed workers, although no silicosis was detected. Higher SP-D levels have been reported in workers with slight silicosis and ongoing silica exposure (Wang et al. 2007). In contrast, SP-D was not increased in silicosis patients with past exposure to crystalline silica (Xue et al. 2017). Wang et al. (2007) also observed lower CC-16 concentrations in patients with silicosis. This observation may be compatible with the results of our study, as we observed significantly lower serum CC-16 concentrations in exposed rock drillers with airways obstruction (FEV_1/FVC <0.70). Also, Bernard et al. (1994) reported lower CC-16 concentrations in silica exposed workers, although presence of silicosis or airways obstruction was not reported. Taking these considerations into account, it may be possible that having silicosis without current exposure does not necessarily result in increased concentrations of SP-D in serum. It may also be possible that airways obstruction, but not silicosis, may contribute to cause a reduction of CC-16 and increase of SP-D in serum of crystalline silica exposed workers. It is important to record time of blood sampling due to the substantial diurnal variation of CC-16 in serum (Helleday et al. 2006). Adjusting for diurnal variation was not reported by Bernard et al. (1994) and Wang et al. (2007), making the comparison of their results with our results regarding CC-16 more uncertain.

The occupationally exposed rock drillers included in this study had on a group basis, signs of airways obstruction, mainly related to the small airways (Ulvestad et al. 2020). The FEV_1/FVC ratio and MMEF% predicted were significantly lower in exposed rock drillers than in the non-occupationally exposed workers. Exposed rock drillers with

MMEF% <70% predicted, indicating small airways obstruction, had substantially higher SP-D concentrations than rock drillers with MMEF% ≥70% predicted. The results did not change noteworthy when four rock drillers with emphysema diagnosed with HRCT were excluded. Occupationally exposed rock drillers with FEV_1/FVC <0.70 had significantly lower CC-16 concentrations compared to those with ratio above 0.70, while SP-A and SP-D did not differ. The FEV_1/FVC ratio may be more related to the function of the larger airways and MMEF to the smaller airways. This could indicate that small airways obstruction caused by exposure to crystalline silica may result in pathological alterations with concomitant increased concentrations of SP-D in serum, while CC-16 is more active in the larger airways.

It is tempting to speculate that crystalline silica exposure may cause airways obstruction with concomitant SP and CC-16 alterations. Such information should be considered in future studies of silicosis in crystalline silica exposed workers. However, when comparing exposed rock drillers with non-occupationally exposed workers with MMEF% ≥70% predicted, SP-A and SP-D concentrations were significantly higher among the rock drillers, although age and smoking habits were similar. This indicates increased serum concentrations of SP-A and SP-D also in the absence of known pulmonary pathology, and therefore most likely the result of exposure only. We have in this study measured both current and cumulative exposure. Current exposure had no association with any of the biomarkers, while some association with cumulative exposure was observed for SP-D. This could indicate long-term exposure to be a more likely cause for the increase than current exposure. SP-A and SP-D have important roles in the homeostasis and maintenance of pulmonary surfactant (Botas et al. 1998; Korfhagen et al. 1998; Nathan et al. 2016; Olmeda et al. 2017) and several studies have pointed to an involvement of pulmonary surfactant in animals exposed to crystalline silica (Grünspan et al. 1973; Dethloff et al. 1986; Lesur et al. 1993). Thus, it is tempting to speculate that the increased levels of SP-A and SP-D that we observed in the exposed rock drillers may reflect an increased turnover of pulmonary surfactant, a possibility that should be further elucidated.

Four occupationally exposed rock drillers had emphysema detected by HRCT, while no case of silicosis was detected. They had substantially higher SP-D concentrations than rock drillers without detectable emphysema. This observation is contradictory to observations in a large study showing declining SP-D concentrations by declining lung density (Coxson et al. 2013). However, a subsequent study demonstrated the inhomogeneity of the COPD patients and suggested that high SP-D levels were associated with emphysema that was progressing, while low levels were associated with severe emphysema (Rennard et al. 2015). Significantly higher SP-D in patients with combined pulmonary fibrosis and emphysema compared to those without emphysema has also been shown (Kokuho et al. 2015; Papaioannou et al. 2016). Thus, the issue whether subjects with emphysema have higher SP-D in serum than those without is unresolved. The results with respect to emphysema and pneumoproteins in our study should be interpreted with caution since only four subjects had emphysema.

The most consistent finding in this study is the increased concentrations of SP-D in serum of the exposed rock drillers. We also observed a dose–response relationship between SP-D levels and cumulative exposure to crystalline silica, while current exposure levels were not associated with SP-D. Also having small airways obstruction was associated with higher SP-D levels among the rock drillers. Further, SP-D concentrations were significantly higher in the absence of airways obstruction, indicating that crystalline silica exposure above a certain level in apparently healthy individuals results in increased SP-D concentrations, and that the concentrations would increase further in the presence of small airways obstruction. Increased serum SP-D levels appear to be associated with COPD according to recent reviews on the subject (Nandy et al. 2019; Wang et al. 2019). Crystalline silica was not determined in the non-occupationally exposed workers. A slight exposure among them can therefore not be completely excluded. This would reduce the exposure contrast between the two groups, thereby reducing the difference in pneumoprotein concentrations between the groups.

In conclusion, rock drillers exposed to crystalline silica without known silicosis had higher serum concentrations of SP-D and higher serum SP-A concentrations. Thus, we would recommend that future studies of SPs in silicosis patients also should take into consideration the presence of emphysema and small airways obstruction as this may contribute to higher SP levels.

Disclosure statement

No potential conflict of interest was reported by the author(s).

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