

Exposure to Inhalable Dust and Endotoxin Among Danish Pig Farmers Affected by Work Tasks and Stable Characteristics

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Objective: To identify working tasks and stable characteristics that determine intensity and variability of personal exposure to dust and endotoxin among pig farmers.

Methods: Three hundred fifty-four personal full-shift measurements were performed in 231 farmers employed in 53 Danish pig farms. Filters were gravimetrically analysed for inhalable dust and for endotoxin by the *Limulus* amoebocyte lysate assay. Information on working tasks and stable characteristics were collected using self-reported activity diaries and walk-through surveys performed in conjunction with the measurements. Associations between log-transformed dust and endotoxin exposure and working tasks and stable characteristics were examined using linear mixed-effects analysis. In these models, worker and farm identity were treated as random effects and working tasks and stable characteristics as fixed effects. Both separate and combined models for tasks and stable characteristics were elaborated.

Results: Inhalable dust concentrations ranged between 0.1 and 48 mg m⁻³ and endotoxin concentrations varied between 9.2 and 370 000 EU m⁻³. Field work activities played a dominant role on the exposure variability. Indoor working tasks with intense animal activity or handling of feed materials increased exposure concentrations, whereas engagement in field work was associated with lower exposure concentrations. High-pressure water cleaning increased endotoxin exposure but did not affect exposure to inhalable dust. Stable characteristics related to feeding practices and type of ventilation were determinants of exposure to inhalable dust. For endotoxin, the most important determinants were use of dry feed and slatted floor coverage. Feeding practices solely explained all between-farms variability in exposure to inhalable dust and endotoxin.

Conclusions: These findings suggest feeding systems, flooring and ventilation to be potential areas where improved methods can reduce exposure to dust and endotoxin among pig farmers. Further, they highlight particular tasks involving feeding and intense animal handling as sources of very high levels of exposure. The pig farming industry is encouraged to focus on exposure reduction. Use of respirators during performance of working tasks where levels of exposure are particularly high ought to be considered until adequate hygienic solutions have been established.

Keywords: determinants of exposure; dust; endotoxin; pig farmers; variability

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INTRODUCTION

The respiratory tract of animal farmers is exposed to various gases and aerosols of chemical, mineral, plant, animal, and microbial origin (Schenker *et al.*, 1998). Of those, the aerosols of organic origin are widely accepted as the main and most influential on the farmers' respiratory health. One of the most active and well investigated constituents of organic dusts is endotoxin (Douwes *et al.*, 2003; Sigsgaard *et al.*, 2010). Endotoxins are lipopolysaccharides primarily of gram-negative bacteria origin commonly present in workplaces involved in plant and animal material processing or other workplaces with strong presence of human or animal faeces (Douwes *et al.*, 2003; Liebers *et al.*, 2006). Endotoxins have strong proinflammatory capabilities and can induce several respiratory and systemic disorders including chronic bronchitis, non-atopic asthma, and wheeze, fever and chills, malaise, and chronic obstructive pulmonary disease (Douwes *et al.*, 2003; Rylander, 2006; Liebers *et al.*, 2008). However, protective effects of occupational endotoxin exposure in relation to allergic asthma and sensitization have also been described (Eduard *et al.*, 2004; Smit *et al.*, 2008; Basinas *et al.*, 2012a). More recently, supportive findings towards a protective effect of endotoxin exposure against lung cancer have been published (Lenters *et al.*, 2010; McElvenny *et al.*, 2011), but relationships remain argued with no associations between endotoxin and lung cancer displayed in a recent well-established multinational population-based study (Peters *et al.*, 2012).

Livestock farmers are well documented as highly exposed to both organic dust and endotoxin (Kullman *et al.*, 1998; Nieuwenhuijsen *et al.*, 1999; Simpson *et al.*, 1999; Radon *et al.*, 2002; Spaan *et al.*, 2006). We recently assessed high levels of personal exposure to dust and endotoxin among Danish livestock farmers (Basinas *et al.*, 2012b). Accurate estimation of dust and endotoxin exposure for farmers is, however, complicated because of large temporal variability in personal levels of exposure (Kromhout and Heederik, 2005). For example, Dutch pig farmers were estimated to have their average daily dust and endotoxin concentrations within a factor 9–21, respectively, while the range between pig farmers did not exceed a factor 4 (Preller *et al.*, 1995b; Kromhout and Heederik, 2005). Similarly, in an analysis with >6000 personal dermal exposure measurements, the average daily exposure of agricultural workers to pesticides were estimated to lie

within 10- to 40-folds (Kromhout and Vermeulen, 2001). Even larger day-to-day variations in daily average concentrations have been reported among US livestock and arable farmers (Nieuwenhuijsen *et al.*, 1999; Kromhout and Heederik, 2005).

In our recent exposure study, we also reported on the variability in personal dust and endotoxin concentrations among livestock farmers (Basinas *et al.*, 2012b). Using personal repeated measurements on 231 pig and 77 dairy cattle farmers, we found average daily dust and endotoxin concentrations to vary between 25- and 250-folds, respectively. We observed up to a factor 30 increase in endotoxin day-to-day exposure variability for field workers compared with stable workers. These findings suggest a considerable potential for errors in estimation of long-term exposures, emphasizing the need for improvement in sampling and estimation strategies within prospective exposure studies.

Knowledge on determinants of exposure—factors which explain systematic differences in exposure over time or between individuals—is an essential component of the development process of exposure prevention and controlling strategies (Burstyn and Teschke, 1999; Burdorf, 2005). Moreover, in-depth knowledge of factors affecting workplace exposure can be used to effectively reduce the measurement error and thereby attenuation in the risk estimates for chronic diseases in epidemiological studies in populations with large temporal variability in exposure (Burdorf, 2005), such as agricultural workers and farmers (Kromhout and Heederik, 2005). Observational evaluations of determinants of personal exposures in workplaces are vital because they allow the assessment of multiple factors in real working conditions with a great degree of generalizability (Burstyn and Teschke, 1999). For pig farmers, previous observational exposure determinant studies have largely been simplistic in design and tended to oversimplify the description of the working environment by including only a few potential determinants in their assessments; usually being the type or stage of production or the measurements season (Louhelainen *et al.*, 1987; Vinzents and Nielsen, 1992; Chang *et al.*, 2001a; Kim *et al.*, 2008; Mc Donnell *et al.*, 2008; Bonlokke *et al.*, 2009). Comprehensive investigations of multiple determinants of personal exposure to dust and endotoxin have been sparse (Preller *et al.*, 1995a; O'Shaughnessy *et al.*, 2010), and so far the combined effect of both multiple farm characteristics and working tasks of pig farmers have been

examined only in one study performed during the early 1990s (Preller *et al.*, 1995a).

The objective of the present analysis was to explore factors that determine the intensity and variability of personal exposure to dust and endotoxin among pig farmers. To identify activity patterns and stable characteristics of importance for personal exposure to dust and endotoxin among pig farmers, we used data collected from self-reported activity diaries and walk-through surveys along with >300 personal exposure measurements in linear mixed-effect regression analysis.

METHODS

Study design

The design and the sampling and analytical methods applied in the exposure assessment of the Sund Stald, in English Healthy Stables (SUS) study have been described in detail in a previous publication (Basinas *et al.*, 2012b). Briefly, the SUS study was initiated in 1992 with the aims (i) to describe the prevalence and incidence of respiratory symptoms in a farming environment and (ii) to investigate the effect of farming on the development of allergy, asthma, and respiratory disease (Elholm *et al.*, 2010). The study population included all 2458 second year students at all farming schools of Denmark, and a control group of 967 conscripts in the Danish army. Overall, 1964 farming school students (80%) and 592 conscripts (61%) gave consent to participate in the study. The final population sample consisted of the 1964 students and 407 randomly selected conscripts.

The present work is a part of the follow-up of the SUS cohort (Elholm *et al.*, 2010). In the 15-year follow-up period, changes in the occupational status of the participants were to be expected. Therefore, *a priori* identification of the remaining active farming population of the initial SUS cohort was performed. Information on current and previous employment in farming, type and related farm characteristics (location, size, and number of animals) for 1239 participants (66% participation rate) was obtained from a preliminary selection questionnaire and from an exposure scheme filled out during the clinical investigations. Of the participants, 423 (34%) emerged as still active and full-time employed in farming (defined as 37 h of farm work per week and including owners that fulfilled this criterion) and 76 pig farmers out of 159 located in the area of Jutland, Denmark were selected randomly

after stratification by farm size. Of those, 22 were either excluded due to migration, change of occupation, part-time employment in farming, poor health, or lack of contact ($n = 11$), or refused to participate ($n = 11$) in the personal measurements. The remaining 54 pig farmers were interviewed.

The interview was performed in person with standardized developed schemes assessing production characteristics (i.e. number of employees, collaborations, number and type of animals, size, unit structure and locations, and building infrastructure) and farm practices (i.e. agriculture form, cleaning and disinfection schedules and frequency, manure handling) at farm level.

Farm visits, measurements, and data collection

All of the 54 pig farmers who agreed to be interviewed also agreed to allow samples to be taken on their farms. When farmers and farm owners were two different persons, consent for farm participation was acquired by the latter. Within the 54 farms, there were a total of 233 owners and workers who were monitored representing >90% of the total workforce in these farms and resulting in 358 personal full working-shift measurements. For the present analysis, four measurements from two farmers, representing one farm in our population, were excluded due to their involvement in mixed pig and cattle production activities. For every farm, two measurement visits were scheduled on randomly chosen working days (Monday to Friday) of the week during summer and winter 2008 and 2009. Summer visits were performed between May 1 and October 1 and winter visits between November 17 and April 3. Climatic data from the Danish Meteorological Institute suggested major changes in average monthly temperatures ($\pm 10^\circ\text{C}$) to occur during the months of April for summer and October to November for winter (DMI, 2008).

The performed tasks were registered by the farmers in structured activity diaries with a 30-minu interval checklists, which covered the measurement day. Twenty-four pre-selected tasks related to animal handling and movement, feeding and cleaning practices, handling of bedding materials, performance of high-pressure washing, disinfection, repairs, and outdoor activities as well as open entries for other non-specified tasks were included. The completed diary was collected at the end of the work-shift.

The structure and the production characteristics of the participating farms have been described in details elsewhere (Basinas *et al.*, 2012b). All

major types of Danish pig farms were represented in our sample. The number of housed pigs per farm ranged from 1400 to 20 700 with a median of 5650 pigs. On average, each farm comprised 8 departments (range 3–28) sized between 33 and 2110 m², and frequently belonging to more than one production site. Farm characteristics, engineering parameters, and the hygienic conditions present in each department of the visited farm were registered through walk-through surveys performed during the visiting days. Notations were kept in pre-fixed inspection sheets designed to allow assessment for >120 well-defined characteristics including basic department parameters (e.g. type of stable, dimensions, type and number of animals), applied ventilation, heating, flooring, bedding, and feeding practices, use of water and/or oil sprinkling, methods and frequency of manure handling, general hygienic parameters (e.g. floor condition, accumulation of manure) as well as parameters related to the disinfection and cleaning of the stable.

The outdoor temperature was measured locally at noon using a portable weather station (OBH Nordica A/S, Taastrup, Denmark) with a measurement accuracy of $\pm 1^\circ\text{C}$.

Dust measurements and endotoxin analysis

All farmers were equipped with a waist belt carrying two portable AirChek XR5000 pumps, each connected through a flexible tube to a conical inhalable sampler (CIS; JS Holdings, Stevenage, UK) mounted with a 37-mm glass-fibre (GFA) filter (Whatman International Ltd, Maidstone, UK). The samplers were pinned in the farmers' pectoral area, and sampling was performed at airflow of 3.5 l min⁻¹ according to the manufacturer's instructions. Filters were gravimetrically measured in a room with controlled climatic conditions (22°C, 45% relative humidity; desiccation ≥ 24 h) using an analytical balance with 0.1 μg readability (Mettler-Toledo Ltd, Greifensee, Switzerland), and then extracted in pyrogen-free water (PFW) with 0.05% (v/v) Tween-20. Analysis for the endotoxin content in the extracts was performed in PFW (1:200 dilution) using a quantitative kinetic chromogenic Limulus amoebocyte lysate test (Kinetic-QCL 50-650U kit, Lonza, Walkersville, MD, USA; Spaan *et al.*, 2008). For quality control, 210 blank filters not subjective to sampling were included throughout the measurement series, half of which were included in the analysis for endotoxin. These were used to control for contamination of the sampled filters during

handling, transport, and storage and to adjust the collected dust mass during the gravimetric analysis. The limit of detection (LOD) for dust was 0.074 mg filter⁻¹ and for endotoxin 13.69 EU filter⁻¹; results were expressed in mg m⁻³ and EU m⁻³, respectively. For three samples with measured dust or endotoxin concentration below the LOD a 2/3 value of the corresponding LOD was used.

Stable characteristics

The daily work performed by most farmers involved presence in several stables of different types with different housing characteristics. The time a farmer spent in a stable, or with a farm or environmental characteristic was therefore expressed as a portion of his overall working time on the measurement day (Preller *et al.*, 1995a). Estimations were made for all work allocated in areas where animals were present and stable characteristics (e.g. ventilation) were functional. When time was spent on insemination and early handling of piglets (i.e. castration, tail clipping, teeth cutting), as stated in the workers activity diary, these activities were pre-allocated on the insemination and farrowing departments, respectively. The remaining animal-related working time was allocated to the compartments involved based on the number of animals present. To account for differences in animal tending requirements across the different stages of production, weighting factors of 10:2:1:1 per animal housed in (a) farrowing, (b) serve or gestation, (c) weaning, and (d) finishing stables were used, respectively. These weighting factors were estimated based on the average time needed for daily nursing of an animal in a specific stage of the production as published by the Danish Expertise Centre for Agriculture (Dansk landbrugsrådgivning, 2003). This was applied on farm characteristics earlier identified in the literature as affecting exposure to dust and endotoxin, i.e. type of accommodation, feeding, ventilation, flooring, heating, and the basic hygienic conditions present.

Data analysis

All data were analysed using the SAS statistical software version 9.2 (SAS Institute Inc, Cary, NC, USA) with exposure on the natural log scale. Log transformation was preceded by formal tests of the exposure distributions using Kolmogorov–Smirnov tests. Exposure distribution characteristics are therefore summarized as geometric means (GM) with geometric standard

deviations (GSD) provided along with arithmetic mean (AM) values.

Relationships between exposure, stable characteristics, and working tasks were assessed using mixed-effect linear models (PROC MIXED) (Rappaport *et al.*, 1999; Peretz *et al.*, 2002) with farm and worker included as random effects and tasks and farm characteristics as fixed effects. Initially, two separate models for work tasks (Model 1) and stable characteristics (Model 2) were elaborated for both dust and endotoxin concentrations. Only variables with a minimum of 10 observations were included in these models. Model entry criteria were applied to restrict the model building process to the most relevant tasks and farm characteristics and limit thereby the possibility of coincidental results. Single covariates showing a *P* value of 0.3 or smaller were included in further backward stepwise regression, where only covariates with a significance <0.1 were retained. For modelling farm characteristics, the population was restricted to only workers working indoors where stable characteristics were of interest. Therefore, 49 measurements from 38 workers with outdoor work were excluded as well as measurements from further 26 workers who either worked whole days outdoors (*n* = 23) or indoors in an environment irrelevant (e.g. high-pressure wet cleaning on completely empty stables for a whole shift) to the determinants investigated (*n* = 14). The combined effect of tasks performed under certain stable characteristics was then examined on the restricted measurement sample (*n* = 268), for dust and endotoxin exposure, respectively (Model 3), by combining the stable-related tasks identified in Model 1 with the stable characteristics identified in Model 2. These models were not optimized further to allow comparisons across model results.

In all models, tasks were included as continuous variables using the actual time spent in minutes by the farmers on the tasks, whereas stable characteristics were used either as continuous (portion of overall time spent on the presence of a characteristic) or dummy variables (Table 1; Preller *et al.*, 1995a). A compound symmetric covariance structure was assumed, and all estimations of variance components were based on the restricted maximum likelihood method. Model adequacy was assessed through influence diagnostic and residual plots. Pearson and when appropriate Spearman correlation coefficients were used to describe associations between endotoxin, dust and working seasons, and between working tasks and stable characteristics.

RESULTS

The measured levels of dust and endotoxin exposure along with the numbers of participating farms and workers are presented in Table 2. On average, sampling was performed for 368 min (SD = 89.4) during summer and 366 min (SD = 84.3) during winter. The mean (GM) exposure level for all measurements performed was 3.4 mg m⁻³ for inhalable dust and 1500 EU m⁻³ for inhalable endotoxin. Dust and endotoxin concentration during winter were significantly higher than those during summer and exposure concentrations were somewhat higher in the subsample of indoor measurements compared with the overall sample. Correlations between dust and endotoxin were moderate (overall *r* = 0.62) and between seasons low (*r* = 0.30 for dust and 0.15 for endotoxin).

Working tasks

The results from the linear mixed-effects models with working tasks for dust and endotoxin exposure among the overall population are shown in Table 3. The basic characteristics in respect to task occurrence, the average working time needed for each task, and the use of personal protective equipment (PPE) are also presented.

Overall, the models consisted of nine tasks for dust and seven tasks for endotoxin that together explained >26% of the within-workers variability in exposure. The explained overall variability in dust exposure was 27% and in endotoxin exposure 30%. The most influential task was field work, which was associated with a reduced exposure to dust and endotoxin. A model with field work as the only fixed effect explained 24% of the within-workers variability for both exposure to dust and to endotoxin.

Handling of feeding materials was one of the strongest predictors for both dust and endotoxin exposure. Handling feeding materials for 30 min (the average time performed in our population) increased the level of dust exposure by 17% and of endotoxin exposure by 13%. When high-pressure washing is being performed for 60 min, an increase equal to exp(0.005 * 60 min) or of 35% is to be expected. Other tasks that significantly increased exposure, included manual feeding, preparation and spread of bedding, moving of weaners and finishers, handling and nursing of piglets, and injection and handling of sick animals. Sensitivity analyses were performed by fitting the same multivariate models on the sample of indoor

Table 1. Outline of the developed database and basic information for working tasks performed by 231 pig farmers employed in 53 Danish pig farms, and stable characteristics for a sub-group of 181 indoor workers including direct animal exposure.

Working tasks ^a	<i>n</i>	Department characteristics (cut-off time level) ^b	<i>n</i>	Coding (median) ^c
Tasks related to stables and affiliated areas		Outdoor temperature	268	Continuous (12°C)
Controlling/inspecting	240	Housing		
Weighing	43	Animals in a loose housing system	58	Continuous (15%)
Moving breeding animals	112	Animals housed in batch pens	205	Continuous (39%)
Moving and loading weaners and finishers	143	Animal housed in crates (including farrowing)	211	Continuous (63%)
Handling and nursing piglets (ear tagging, castrating, cutting tails, teeth cutting)	118	Ventilation		
Inseminating	112	Mechanical with neutral pressure (>60%)	15	Present (1) or absent (0)
Ultrasound scanning of pregnant/inseminated sows	14	Mixed type(including natural)	19	Present (1) or absent (0)
		Mechanical with negative pressure (>60%)	234	Ref.
		Mechanical with pit exhaust	48	Continuous (17.3%)
Injection or handling sick animals	172	Heating		
Handling dead animals	93	Floor heating (>50%)	153	Present (1) or absent (0)
Preparation and handling of feed and seeds	104	Radiator heating (>50%)	57	Present (1) or absent (0)
Cleaning feed troughs	8	Floor type		
Manual feeding	159	Full slatted floor (>50%)	22	Present (1) or absent (0)
Automatic feeding (including adjusting/inspecting)	137	Mostly slatted (>50%)	101	Present (1) or absent (0)
Preparation and spread of bedding	108	Mostly concrete	145	Ref
Removing manure (in pens and stalls)	84	Deep litter	21	Continuous (17.4%)
Sweeping or scraping corridors	58	Water showering system	83	Continuous (42.9%)
Washing stables with high pressure	71	Feeding characteristics		
Disinfecting pens/stalls/stables	17	Dry feed (>80%)	147	Present (1) or absent (0)
Emptying slurry pits	9	Dry and wet feed	50	Present (1) or absent (0)
Repair and maintenance of animal buildings/feed rooms and stable installations	83	Wet feed (>80%)	71	Ref
Administrative/office work	46	Ad libitum feeding method	157	Continuous (33.8%)
Tasks related to outdoor work		Hygienic conditions		
		Floor condition		
Repairing/maintaining machinery and equipment (e.g. tractor, track, harvester)	48	Wet floor (>80%)	48	Present (1) or absent (0)
Handling manure tanks and dunghills	5	Mixed floor condition	75	Present (1) or absent (0)
Logging, splitting or cutting wood	7	Dry floor (>80%)	145	Ref
Work in the fields (working the soil, sowing, harvesting, applying fertilizers, draining, etc.)	25	Very dusty feeding path	73	Continuous (11.1%)
		Very high dung accumulation	104	Continuous (11.3%)
		Disinfected with bacterial agents (only endotoxin)	202	Continuous (63.6%)
Diverse ^d	66			

n = number of observations.

^aFor all pig farmers included.

^bOnly for workers with a full indoor working shift and time spend dealing with the characteristics, cut-off level indicates the level of stable working time used to consider the characteristic present.

^cMedian value of portion of time spend in presence of a stable characteristic, estimated for positive values.

^dNot belonging to any of the above categories and including sparsely performed (*n* < 5) tasks.

Table 2. Basic measurement characteristics and personal levels of dust (mg m⁻³) and endotoxin (EU m⁻³) exposure of Danish pig farmers.

Period	n	f	k	Dust			Endotoxin			r
				AM	GM (GSD)	Range	AM	GM (GSD)	Range	
<i>All measurements</i>										
Overall	354	53	231	4.9	3.4 (2.6)	<LOD–48	6200	1500 (4.4)	<LOD–370 000	0.62*
Summer	181	52	181	4.3	2.8 (2.6) ^a	0.1–48	6000	1100 (4.2) ^a	14–370 000	0.66*
Winter	173	53	173	5.5	4.1 (2.5)	<LOD–20	6600	2100 (4.2)	<LOD–290 000	0.54*
<i>Only indoor measurements</i>										
Overall	268	51	181	5.3	4.0 (2.1)	0.47–48	5300	1800 (3.2)	<LOD–370 000	0.51*
Summer	135	47	135	4.7	3.4 (2.2) ^a	0.50–48	5200	1400 (3.2) ^a	160–370 000	0.66*
Winter	133	49	133	5.9	4.8 (1.9)	0.47–20	5300	2400 (3.1)	<LOD–110 000	0.25*

n = number of measurements.

f = number of farms.

k = number of workers.

AM = arithmetic mean.

GM = geometrical mean.

GSD = geometrical standard deviation.

r = Pearson correlations between measured dust and endotoxin concentrations.

^aSignificantly different from winter.

*P < 0.05.

measurements used for modelling farm characteristics. This did not show systematic differences in direction or size of the associations. Personal protection equipment was not used frequently with a prevalence >10% only for high s washing. The overall prevalence of PPEs use was 12%.

Stable characteristics

Factors eligible to enter the multivariate model for endotoxin included type of animal housing, type of feed, outdoor temperature, water showering and ad libitum feeding system, floor exhaust ventilation, feed path dustiness, application of disinfection, dung accumulation, and slatted floor coverage. For dust, the eligible factors were outdoor temperature, type of feed and ventilation, heating parameters, the ad libitum feeding system, the slatted floor coverage, the feed path dustiness, and the level of floor dampness.

The final model for dust consisted of five factors (i.e. outdoor temperature, type of feed and ventilation, ad libitum feeding system, and floor dampness) explaining 80% of the variability between farms, but only 22% of the total variability (Table 4). For endotoxin, the outdoor temperature, the type of feed, the floor coverage, and the use of water showering remained in the final model, which explained all the between-farms variability and 13% of total variability. The use of dry feed was a strong determinant of both exposure to dust and to endotoxin. Farmers with >80% of their stable working time spent on stables with

dry feeding had a factor 1.5–1.8 higher exposure compared with those exposed for the same time in an environment with wet feeding. Time spent on a department with an ad libitum feeding installation elevated personal dust exposure, and a water showering system for pigs elevated endotoxin exposure. Dust exposure was decreased in workers spending most of their time working under mechanical ventilation with negative pressure and in presence of a wet floor. Increased slatted floor area coverage was related to increased exposure to endotoxin.

Work tasks and stable characteristics combined

In Table 5, the results of the model assessing the combined effects of tasks and stable characteristics are presented. The inclusion of work tasks in the model for stable characteristics increased the proportion of explained variability to 35% (from 22%) for exposure to dust and to 27% (from 13%) for exposure to endotoxin. Feeding practices and the outdoor temperature remained strong determinants of exposure as did also the type of ventilation for dust, and the showering system and type of floor for endotoxin exposure. There were no substantial changes in estimates for most of the stable-related tasks included, except for injection and handling of sick animals and performance of repair work which no longer appeared to affect exposure to dust. The effect of manual feeding on exposure to dust and of preparation and distribution of bedding on exposure to endotoxin were also weakened.

Table 3. Effect of working activities modelled as continuous variables on the log-transformed personal level of exposure to dust (mg m^{-3}) and endotoxin (EU m^{-3}) among Danish pig farmers. Results estimated on the basis of 354 measurements performed in 231 farmers employed in 53 farms.

	<i>N</i>	PPE (<i>n</i>)	MDN (min)	Dust				Endotoxin			
				β^a	<i>e</i>	Change factor ^b	<i>P</i>	β^a	<i>e</i>	Change factor ^b	<i>P</i>
Model with tasks											
Intercept				0.90	0.097		<0.001	7.2	0.11		<0.001
Moving breeding animals	112	3	57.5	0.0020	0.0010	1.13	0.04				
Moving and loading weaners and finishers	143	4	85	0.0022	0.0010	1.14	0.002	0.0029	0.0010	1.19	0.006
Handling and nursing piglets (ear tagging, castrating, cutting tails, teeth cutting)	118	5	90	0.0019	0.0007	1.12	0.005				
Injection or handling sick animals	172	5	45	0.0023	0.0010	1.15	0.03				
Preparation and handling of feed and seeds	104	7	30	0.0051	0.0011	1.36	<0.001	0.0040	0.0016	1.27	<0.001
Manual feeding	159	8	30	0.0027	0.0016	1.18	0.09				
Preparation and spread of bedding	108	6	30					0.0045	0.0027	1.31	0.01
Washing with high pressure	71	8	60					0.0050	0.0012	1.35	<0.001
Disinfection	17	3	30					-0.019	0.0051	0.31	<0.001
Repair and maintenance of animal buildings/ feed room and stable installations	83	1	75	0.0013	0.0007	1.08	0.05				
Work in the fields (working the soil, sowing, harvesting, applying fertilizers)	25	0	210	-0.0041	0.0006	0.78	<0.001	-0.0090	0.0009	0.58	<0.001
Administrative/ office work	46	0	60	-0.0032	0.0011	0.83	0.004	-0.0051	0.0016	0.74	0.002
$\text{bf}\sigma^2$ (naive estimate)				0.028 (0.042)	0.024		0.1	0.068 (0.0021)	0.05		0.09
$\text{bw}\sigma^2$ (naive estimate)				0.14 (0.19)	0.071		0.02	0.074 (0.19)	0.14		0.3
$\text{ww}\sigma^2$ (naive estimate)				0.48 (0.65)	0.066		<0.001	1.4 (2.0)	0.17		<0.001
Explained variability											
Between farm				33%				0%			
Between worker				26%				61%			
Within worker				26%				30%			
Total				27%				30%			

Naive estimates are derived from a model without fixed effects.

n = number of observations; PPE = number of cases reported for use of personal protection equipment; MDN = median time spent on an activity estimated only for positive responses on the day of the measurements; β = regression coefficient for log-transformed exposure data; *e* = standard error; *P* = *P* value; $\text{bf}\sigma^2$ = between-farm variance; $\text{bw}\sigma^2$ = between-worker (within-farms) variance; $\text{ww}\sigma^2$ = within-worker (day-to-day) variance.

^aPer 1 min of actual task performance.

^bFactor for change in exposure for 60 min of task performance.

Table 4. Mixed-effect models results for stable characteristics affecting the log-transformed personal dust (mg m^{-3}) and endotoxin (EU m^{-3}) exposure of pig farmers. All characteristics are estimated on the worker level on the basis of 268 indoor measurements.

	<i>n</i>	Dust				Endotoxin			
		β	<i>e</i>	Change factor ^a	<i>P</i>	β	<i>e</i>	Change factor ^a	<i>P</i>
Model with farm characteristics									
Intercept		1.3	0.11		<0.001	7.5	0.20		<0.001
Outdoor temperature, °C	268	-0.025	0.0047	0.78	<0.001	-0.044	0.0087	0.64	<0.001
Ventilation (1/0)									
Neutral pressure	15	0.36	0.18	1.43	0.06				
Mixed type (including natural)	19	0.26	0.16	1.30	0.1				
Negative pressure	234	Ref.							
Feed type (1/0)									
Dry	147	0.43	0.11	1.53	<0.001	0.59	0.26	1.80	0.03
Dry and wet	50	0.35	0.13	1.42	0.008	0.22	0.15	1.25	0.2
Wet	71	Ref.				Ref.			
Ad libitum feeding system (%)	157	0.0046	0.0016	1.05	0.005				
Water showering system (%)	83					0.0051	0.0029	1.05	0.08
Floor type (1/0)									
Full slatted	22					0.51	0.17	1.66	0.004
Mostly slatted	101					0.65	0.21	1.92	0.002
Mostly concrete	145					Ref.			
Floor condition (1/0)									
Wet floor	48	-0.25	0.11	0.78	0.03				
Mixed floor condition	75	-0.055	0.093	0.95	0.6				
Dry floor	145	Ref.							
$\text{bf}\sigma^2$ (naive estimate)		0.011(0.052)	0.024		0.3	0(0.056)			
$\text{bw}\sigma^2$ (naive estimate)		0.12(0.091)	0.055		0.02	0.086(0)	0.15		0.3
$\text{ww}\sigma^2$ (naive estimate)		0.30(0.41)	0.049		<0.001	1.1(1.3)	0.17		<0.001
Explained variability									
Between farm		79%				100%			
Between worker		0%				0%			
Within worker		27%				15%			
Total		22%				13%			

Naive estimates are derived from a model without fixed effects.

β = Regression coefficient; *e* = standard error; *P* = *P* value; $\text{bf}\sigma^2$ = between-farm variance; $\text{bw}\sigma^2$ = between-worker (within-farms) variance; $\text{ww}\sigma^2$ = within-worker (day-to-day) variance.

^aFactor for change in exposure on presence versus absence of a characteristic or for an increase of 10 U in °C or in portion (%) of working time.

DISCUSSION

Study approach

In this study, we identified tasks and stable characteristics that affected personal exposure to dust and to endotoxin among Danish pig farmers using an observational study approach based on collected repeated exposure measurements.

We applied a 'real-life' scenario using a random sampling design by performing measurements on common working days for the farmers. This approach enabled a comparative assessment of working tasks performed indoors and outdoors as well as in stables bearing certain engineering parameters and farm characteristics. Our statistical model results suggested specific work tasks

Table 5. Mixed-effect models results on determinants (farm characteristics and working tasks) of log-transformed personal dust (mg m^{-3}) and endotoxin (EU m^{-3}) exposure among Danish pig farmers. Estimations are based on 268 indoor measurements.

	<i>n</i>	Dust				Endotoxin			
		β	<i>e</i>	Change factor ^a	<i>P</i>	β	<i>e</i>	Change factor ^a	<i>P</i>
Model with tasks and farm characteristics									
Intercept		0.97	0.14		<0.001	7.2	0.20		<0.001
Tasks (min)									
Moving breeding animals	98	0.0014	0.0008	1.09 ^b	0.08				
Moving weaners and finishing pigs	124	0.0015	0.0006	1.09 ^b	0.02	0.0023	0.0010	1.15 ^b	0.02
Handling and nursing piglets (ear tagging, castrating, cutting tails, etc.)	111	0.0017	0.0006	1.11 ^b	0.01				
Injection or handling sick animals	146	0.0008	0.0008	1.05 ^b	0.3				
Preparation and handling of feed and seeds	81	0.0054	0.0010	1.38 ^b	<0.001	0.0052	0.0017	1.37 ^b	0.003
Manual feeding	146	0.0014	0.0013	1.09 ^b	0.3				
Preparation and spread of bedding	89					0.0038	0.0028	1.26 ^b	0.2
Washing with high pressure	60					0.0038	0.0011	1.26 ^b	<0.001
Disinfection	16					-0.022	0.0042	0.27 ^b	<0.001
Repair and maintenance of animal buildings/ feed room and stable installations	64	0.0001	0.0007	1.00 ^b	0.9				
Administrative/ office work	36	-0.0032	0.0012	0.83 ^b	0.01	-0.0035	0.0020	0.81 ^b	0.08
Farm characteristics									
Outdoor temperature, °C	268	-0.024	0.0047	0.79 ^c	<0.001	-0.048	0.0081	0.62 ^c	<0.001
Ventilation (1/0)									
Neutral pressure	15	0.41	0.17	1.51	0.02				
Mixed type (incl. Natural)	19	0.23	0.15	1.26	0.1				
Negative pressure	234	Ref.							
Feed type (1/0)									
Dry	147	0.42	0.099	1.52	<0.001	0.58	0.16	1.79	<0.001
Dry and wet	50	0.41	0.12	1.51	0.001	0.87	0.19	2.39	<0.001
Wet	71	Ref.				Ref.			
Ad libitum feeding system, %	157	0.0052	0.0015	1.05 ^c	0.001				
Water showering system, %	83					0.0067	0.0026	1.07 ^c	0.01
Floor type (1/0)									
Full slatted	22					0.42	0.24	1.52	0.09
Mostly slatted	101					0.20	0.14	1.22	0.2
Mostly concrete	145					Ref.			
Floor condition (1/0)									
Wet floor	48	-0.13	0.11	0.88	0.3				

Table 5. *Continued*

	<i>n</i>	Dust				Endotoxin			
		β	<i>e</i>	Change factor ^a	<i>P</i>	β	<i>e</i>	Change factor ^a	<i>P</i>
Mixed floor condition	75	0.012	0.094	1.01	0.9				
Dry floor	145	Ref.							
$b_f\sigma^2$ (naive estimate)		0 (0.052)				0 (0.056)			
$b_w\sigma^2$ (naive estimate)		0.042 (0.091)	0.045		0.2	0.015 (0)	0.11		0.5
$w_w\sigma^2$ (naive estimate)		0.32 (0.41)	0.050		<0.001	0.98 (1.3)	0.14		<0.001
Explained variability									
Between farm		100%				100%			
Between worker		54%				0%			
Within worker		22%				25%			
Total		35%				27%			

Naive estimates are derived from a model without fixed effects.

β = regression coefficient; *e* = standard error; *P* = *P* value; $b_f\sigma^2$ = between-farm variance; $b_w\sigma^2$ = between-worker (within-farms) variance; $w_w\sigma^2$ = within-worker (day-to-day) variance.

^aEstimated factor of change in exposure on presence, unless otherwise stated.

^bFactor for change in exposure for 60 min of task performance.

^cFactor for change in exposure for an increase of 10 U.

and layout of the working environment to influence exposure variability, and they highlighted feed type as the most important determinant of differences of exposure to dust and to endotoxin between farms.

When interpreting the above findings, there are certain issues that must be considered. Our statistical models have been developed on the basis of information for only a certain number of farm characteristics earlier identified as influential on the level of dust and endotoxin exposure of pig farmers. In addition, model building was restricted by the presence of high correlations between certain variables (e.g. type of stable and use of ad libitum feeding) or the absence of variation in others (e.g. use of natural ventilation or infrequently performed working tasks). Infrequency can severely affect the reliability of the derived estimates (Boleij *et al.*, 1995), and therefore to increase precision all tasks with <10 observations were excluded from the modelling process. Moreover, distortion of estimates due to the presence of correlations with unmeasured and thereby also unmodelled factors cannot be excluded. Potential large differences in farm practices between time periods and countries (e.g. use of non-industrial farming methods, process changes through time) limit the generalization of our model results to the period of exposure monitoring and to countries with similar farming practices as to the ones

applied in Denmark although comparable findings have been reported in a study among Dutch pig farmers performed in the early 1990s (Preller *et al.*, 1995a).

Considering the validity of our reported inhalable dust and endotoxin exposure levels, as discussed earlier (Basinas *et al.*, 2012b), these are in good agreement with the results of previous studies that used comparable sampling (Radon *et al.*, 2002; Spaan *et al.*, 2006) and analytical methodologies (Spaan *et al.*, 2006). The partly systematic selection of farms (i.e. random after stratification by farm size) in our study is unlikely to have biased the representativeness of Danish pig farms in our farm sample. The distribution of farms in Denmark in our initial sampling was similar to the one reported by the Danish authorities, with >85% of the farms located in the areas of Jutland and Funen (StatBank Denmark, 2010). A formal analysis showed that selected farms did not differ in size from farms in the initial sample, and there were no differences in average personal dust and endotoxin concentration across farm size strata (data not shown).

Influence of feed, floor, and ventilation

Our analysis showed the type of feed along with the ventilation as the most important determinants for exposure to dust, and the type of feed along with the flooring for exposure to endotoxin.

Feed was the most influential parameter, explaining in univariate analysis all between-farms variability. Most (74–96%) of the farmers variability in exposure to dust and endotoxin though was allocated to the temporal within-worker component, whereas the between-farms variability was generally small with farms contributing <10% of the total variability in exposure (Tables 3 and 4). The observed increased levels of exposure for workers mostly working in stables with dry feed (increase between 52 and 79%) or for workers mostly working in stables with full slatted floor coverage (increase of 52%) are supported by Preller *et al.* (1995a). The authors used a data collection and analysis strategy similar to ours, and reported a >20% decrease in dust exposure when wet feeding was used and an increase of 16% when full slatted floor was present. Feed is recognized as a source of dust and endotoxin exposure within pig buildings (Donham *et al.*, 1986; Pearson and Sharples, 1995), and in their exposure assessment study of 171 Dutch pig stables. Attwood *et al.* (1987) reported considerably lower dust concentrations in stables using wet feed compared with stables using dry feed. The strong association between the slatted floor coverage and endotoxin probably reflects the increased exposure to faeces, which is another known source for exposure to endotoxin (Spaan *et al.*, 2006).

The positive association between the ad libitum feeding and exposure we demonstrated contradicts previously reported results by Crook *et al.* (1991). Considering the strong correlation ($r = 0.78$, $P < 0.0001$) that we found between ad libitum feeding and batch pen housing (a system applied primarily in weaning and finishing houses), the effect of ad libitum feeding could reflect the expected higher animal movement and animal intensity. These are both strong exposure determinants (Attwood *et al.*, 1987; Gustafsson, 1999; Duchaine *et al.*, 2000; Thorne *et al.*, 2009), in weaning and finishing houses compared with departments housing sows where restricted feeding is most commonly used.

An increase in outdoor temperature by 10°C was associated with a decrease in exposure as high as 30% in both our study and the study by Preller *et al.* (1995a). This can probably be attributed to the higher rate of ventilation used at higher temperatures. The outdoor temperature can be an indirect indicator of the ventilation rate (Gustafsson, 1999) and to optimize production pigs require temperatures within specific ranges (Wathes and Whitemore, 2007).

The observed increase of 51% (Model 3; Table 5) in levels of dust exposure for workers mostly exposed to a neutral ventilation system compared with those mostly exposed to a negative pressure ventilation system is difficult to explain, but it could relate to air movement and air distribution within the animal house. Further investigation will be needed to validate this finding.

We reported increases in exposure to endotoxin when showering pigs with water. This finding is supported by the results from a previous study among Australian piggeries (Banhazi *et al.*, 2008). They used stationary sampling and reported strong positive correlations between inhalable endotoxin exposure and relative humidity. Both microbial growth (Chang *et al.*, 2001b) and bacterial survival time (Zucker *et al.*, 2000) have been suggested to enhance in the presence of high moisture, thus potentially increasing contamination after air suspension (Banhazi *et al.*, 2008).

Influence of field work

Using the farmers self-reported survey information on performed tasks, we established exposure models that explained >25% of the given between-farms, between-workers, and within-workers variability for exposure to dust and 61 and 30% of the between- and within-workers variability for exposure to endotoxin (Model 1; Table 3). Performance of outdoors (field) work was a strong protective factor for both exposure to dust and to endotoxin. Our measurements were distributed over a long-time period, and the field working tasks performed consisted mostly of common tasks related to soil preparation (e.g. ploughing, land rolling, and tilling), sowing and post sowing handling (e.g. manure and fertilizer spreading), and less to crop harvesting. All farms in our study were equipped with cabined tractors. The protective effect of field work is generally supported by a previous study among Norwegian farmers that reported lower levels of dust and endotoxin exposure during hay and grain harvesting compared with pig animal tending (Melbostad and Eduard, 2001). Similarly, in an earlier study among Californian farmers (Nieuwenhuijsen *et al.*, 1999), task-based measured endotoxin concentrations were in general lower for field crop-related tasks compared with those related to livestock tending. However, in a large Dutch exposure assessment study on different branches of the primary agricultural production, workers involved in potato cultivation and grain harvesting were exposed to considerable dust and endotoxin concentrations (Spaan

et al., 2006). These measurements were worst-case scenarios; given the cyclic nature of field-related activities, lower concentrations will occur more frequently. Among Norwegian farmers, yearly average exposure levels of dust endotoxin were reported to be substantially lower for crop farmers compared with livestock farmers (Eduard *et al.*, 2009).

Influence of tasks requiring near contact with animals

Considering animal-related working tasks, movement of pigs and tasks that included intense animal handling like castration, teeth cutting as well as the injection and handling of sick animals (a very common task for workers in weaning and finishing herds) were significantly associated to an increased exposure to organic dust. A recent study among US pig breeders (O'Shaughnessy *et al.*, 2010), also reported greater dust concentrations in tasks related to animal movement during the weaning process, and pig load-out has been described as a task with very high exposure to dust and endotoxin (O'Shaughnessy *et al.*, 2012). Likewise, in the study of Preller *et al.* (1995a), activities related to intense animal handling (castration, teeth cutting, ear tagging) and movement (re-penning) were reported to increase both exposure to dust and endotoxin.

In our study, tasks related to feed handling were strong determinants of both exposure to dust and endotoxin and performance of high-pressure washing was found to significantly increase exposure to endotoxin. These findings are not unexpected. High levels of dust and endotoxin exposure have been measured among animal feed and seed processing workers (Smid *et al.*, 1992; Spaan *et al.*, 2006), and Preller *et al.* (1995a) found a strong association between tasks related to feeding and cleaning of food storage areas and exposure. Similarly, in a very recent study that used task-based sampling approaches, personal endotoxin exposure levels during high-pressure washing activities were reported to average at 40 000 EU m⁻³ with a range between 5401 and 180 864 EU m⁻³ (O'Shaughnessy *et al.*, 2012).

Use of personal protection equipment

Twelve percent of the workers included in our study used respirators during certain working activities. This is in agreement with the low prevalence of PPE use previously reported among US (Carpenter *et al.*, 2002) and Brazilian (Costa *et al.*, 2007) farmers. In a more recent study among Canadian pig farmers prevalence of self-reported

PPE use was 25% (Bonlokke *et al.*, 2009). Farmers have been suggested to face difficulties and discomfort when using PPEs (Mpfu *et al.*, 2002). Amid the current absence of effective exposure-control strategies and the difficulty and large costs in implementing engineering methods to reduce exposure inside pig stables, a panel of US experts recently advocated the development of a respiratory protection program based on the use of PPEs during performance of specific tasks (Von Essen *et al.*, 2010). This recommendation was based on results from experimental studies among subjects previously unexposed to pig farming, which demonstrated a significant decrease in inflammatory reactions among those wearing a respirator compared with those unprotected (Dosman *et al.*, 2000; Palmberg *et al.*, 2004; Sundblad *et al.*, 2006). Our study, apart from showing the need for a similar educational program among Danish farmers, provides suggestions on potential tasks to be subject of PPE usage.

Yet, predictions based on our combined task and stable characteristic models suggest that a reduction of up to 63% in endotoxin exposure and 56% in dust exposure is possible through best management practices in feeding, ventilation and flooring, i.e. use of wet feed, negative pressure ventilation and a mostly concrete floor. Further reductions towards a maximum 80% can be expected through additional optimization of the ventilation rate and reduction of water showering to a minimum. However, the day-to-day variability in personal exposure also depends on the tasks performed, and thereby further control measures like process changes may be required, e.g. use of mechanical or robotic straw distributors. A preventive strategy based on use of PPEs should only be considered as an intermediate stage until adequate hygienic solutions can be established.

CONCLUSIONS

Overall, this study suggests activities related to nursing and movement of animals, work related to feed handling as well as high-pressure water cleaning to increase the level of personal exposure to dust or endotoxin. Farm characteristics that appeared to be associated with exposure to dust and endotoxin included type of feed, use of an ad libitum feeding system, type of ventilation, and slatted floor coverage. Use of dry feeding had the strongest effect, explaining all variability in exposure between farms. These findings provide information to support a special attention on specific high exposure working

tasks and furthermore suggest feeding, flooring and ventilation parameters as potential areas for developing risk management methods to reduce exposure to dust and endotoxin among pig farmers.

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