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Influence of Building Maintenance, Environmental Factors, and Seasons on Airborne Contaminants of Swine Confinement Buildings

Eight swine confinement buildings, selected to cover the widest possible range of cleanliness, were visited twice during winter and once during summer to verify the range, seasonal variations, and correlations between biological and chemical contaminants. Physical aspects were graded for dirtiness (1=clean, 10=dirty), ventilation, air temperature, number of animals, building, and room size. Air samples were taken to measure relative humidity, CO₂, ammonia, total dust, and microbiological counts and/or identification (bacteria and molds); endotoxin levels also were measured. During winter, average measurements and ranges were: CO₂ = 0.304% (0.254 to 0.349%); ammonia = 19.6 ppm (1.9 to 25.9 ppm); dust = 3.54 mg/m³ (2.15 to 5.60 mg/m³). There were 883 cfu/m³ (547 to 2862 cfu/m³) of molds, 4.25 × 10⁵ cfu/m³ (1.67 × 10⁵ to 9.30 × 10⁵ cfu/m³) of total bacteria, 29 cfu/m³ (3 to 94 cfu/m³) of thermophilic actinomycetes). A significant decrease in bacterial levels (p=0.04), dust (p=0.0008), ammonia (p=0.005), and CO₂ (p<0.0001) was observed during summer sampling when compared with winter levels. Mold counts were positively correlated (p=0.03) with dirtiness scores, while bacterial counts were negatively correlated with this parameter (p<0.002), whereas bacteria and endotoxins were correlated with the number of animals (p<0.05). Ambient gases (CO₂ and ammonia) correlated with each other (p=0.006). Bacteria were the most important contaminant in swine confinement buildings, and endotoxin levels found were also very high (mean=4.9 × 10³ EU/m³). We conclude that a wide range of air contamination exists in swine confinement buildings of different maintenance. There is a decrease in some of these contaminants during summer. Observed dirtiness of the swine confinement buildings has a poor predictive value concerning air quality.

Keywords: aerobiology, agricultural health, air sampling, biological contaminants

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Airborne pollutants found in swine confinement buildings are harmful for the human respiratory tract. Swine confinement workers can develop chronic airway inflammation caused, in all likelihood, by the inhalation of organic contaminants and other airborne pollutants.⁽¹⁾ Pig farmers have a high prevalence of chronic bronchitis, asthma, and

organic dust toxic syndrome.⁽²⁾ In 1989, Donham et al. proposed values of environmental contaminants that could induce a decrease in pulmonary functions.⁽³⁾

The airborne dust found in swine confinement buildings contains large numbers of bacteria (gram positive in majority) and mesophilic molds.⁽⁴⁾ This environment also contains high

levels of endotoxins and ammonia.⁽³⁾ Endotoxins, organic dust, microorganisms, and gases including ammonia could all be responsible for the respiratory symptoms associated with the exposure to this environment.^(2,3,6)

Modern pig farmers have increased animal density and confinement to decrease feeding time, optimize space use, and minimize heat requirement during cold winter months. In most countries, as in Canada, where swine production is on an industrial scale, there is a large variability of swine confinement buildings in term of size, types of ventilation and heating, cleanliness, and dung collection and disposal systems. Some older buildings are poorly maintained, whereas newer and modern facilities are sometimes kept spotless. These physical aspects of swine confinement buildings could have a significant impact on airborne contamination. For example, dust deposition and humidity on the walls and ceilings of poorly maintained buildings could facilitate microbial growth and proliferation, thus increasing airborne contamination if these sources become aerosolized. The type of indoor dung collection and the frequency at which it is emptied could also influence airborne contaminant concentration. The impact of these parameters and other variables such as the number of pigs and their density could also have an effect. The number of animals and their activity influence the CO₂, water vapor, and organic dust levels.

In many northern countries like Canada, there is a wide range of outside temperatures between summer and winter: average daytime high temperature for July in Eastern Canada is 26°C, whereas the average minimal temperature in January is -20°C. Because of these extreme climatic variations between seasons, the ventilation of agricultural buildings is kept at a minimum during winter and at its maximum in summer. In summer, high temperature could enhance bacterial and fungal growth, therefore increasing their levels in swine confinement buildings. However, any increase may be compensated for by more ventilation. In winter, low ventilation would tend to increase the airborne concentration of these contaminants, but this could be compensated for by a potentially slower growth. The net effect of these divergent variables remains to be clarified.

Two important studies have been published on the effect of some physical and environmental parameters on airborne contaminants in swine buildings.^(7,8) In these two studies, the predictive value of dirtiness and the day-to-day variation of bacterial contaminants were not evaluated. Kiekhaefer et al.⁽⁸⁾ (American study) reported higher levels of bacterial contaminants during summer/fall than during winter/spring. In their study the difference of outdoor temperature between the two seasons could be very small: according to their definition, winter/spring outdoor temperature had to be below 4°C and summer/fall outside temperature above the same limit. The present authors wanted to study periods in which temperatures differences were the largest possible in Canada (January/February versus July/August) to verify whether the same observations would remain true. In Atwood et al. (European study),⁽⁷⁾ only low correlations were found between different physical parameters and contaminants and no cross-seasonal analysis was performed.

To assure a proper evaluation of the above variables, one must question the type of sampling used in the analysis. During a 4- or 5-hour working day, animal and human activity is constantly changing. Check point air analyses are sometimes performed, whereas some authors prefer a long period of sampling for adequate evaluation of contaminants and exposure. Measurement of diurnal variability in airborne contaminants is required to determine the usefulness of a checkpoint analysis and verify whether

there is a constancy in the temporal level of these contaminants for a given building.

This study was performed to verify (1) the variability in airborne contaminants between swine operations with buildings of different visual and physical aspects and farming practices, (2) the influence of season with extreme difference in outside temperature on these contaminants, and (3) the importance of multiple samplings in the proper evaluation of airborne contamination of swine confinement buildings.

MATERIAL AND METHODS

Choice of Swine Buildings

Eighteen buildings were visited by one of the authors (YC) to select 8 buildings that covered the widest possible range of building designs, cleanliness, and technologies of production. Only swine fattening operations were chosen. The 8 selected swine buildings were visited 3 times: twice during the winter of 1997 (Visits W1 and W2, between January 13 and March 5) and once during the following summer time (Visit S, in July or August).

Physical Aspects of Buildings

Nine nonfarmer volunteers were sent to each of these buildings: five volunteers evaluated the building at Visit W1 and four at Visit W2. Each person was asked to fill out a questionnaire on the building characteristics. Each evaluator completed the questionnaire without comparing answers with other volunteers. Based on the presence of dust and other visual aspects, a 1 to 10 score was given for dirtiness (1 being the cleanest, and 10 the dirtiest). A similar scale was used for odors. Information on the frequency at which the indoor dung collection system was emptied, the number of ventilators in use, the kind of feeding material, the number of animals, and the building dimensions was taken. The indoor temperature and humidity and outdoor temperature were measured with a thermohygrometer (VWR, Quebec, Canada) three times during the sampling procedure.

Air Sampling and Analysis

All the samples and measures were taken three times: at the beginning, the middle, and the end of the 4-hour period, except for ammonia and dust, which were sampled continuously for 4 hours. Sampling sites were selected to be the most representative of the building's environment. All air samples were taken on a table, 1 m above the floor. The sampling sites were always positioned at one extremity of the animal enclosure. This position was consistent throughout the different buildings. The samplers (Andersen, AGI, filters; Grasby Andersen, Atlanta, Ga.) were always about 50 cm from the enclosure. No specific study was done to evaluate whether this site was the most representative, but visually it was the closest to the sources of contamination and as far away as possible from the doors, windows, or other ventilation sources.

Ammonia

The 4-hour ammonia samplings were done in triplicate. Sulfuric acid pretreated silica gel columns (Dur-Pro, Brossard, Canada) were used with a low flow-rate pump (0.15 L/min) calibrated with an SKC UltraFlo Electronic Calibrator (Dur-Pro). Ammonia was eluted from the columns and analyzed in a reference laboratory using the Aquatic 5400 methodology (Tecator A.B., Höganåf, Sweden). Control columns were brought to the sampling site

TABLE 1. Number of Swine Building, Visual Evaluation of Dirtiness (Mean of Nine Evaluations), Number of Pigs per Building and Per Room, and Building Size

# Swine Building	Visual Evaluation ^a 1 to 10 (range)	Number of Pigs/Building Visit			Number of Pigs/Room Visit			Building Size (m ²)
		W1 ^b	W2 ^c	S ^d	W1	W2	S	
1	1.375 (1-2)	834	854	784	2	72	70	2100
2	6.75 (5-9)	300	200	260	40	40	49	816
3	8.25 (7-9)	350	350	250	96	96	96	523
4	4.5 (4-6)	800	800	700	180	180	180	1920
5	1.5 (1-3)	363	388	360	363	388	360	1226
6	5.88 (5-7)	108	108	588	108	108	108	1223
7	5.38 (3-6)	364	360	352	364	360	352	799
8	2.37 (2-3)	665	650	615	111	111	111	1579

^a1 = best, 10 = worst.

^bFirst visit, winter.

^cSecond visit, winter.

^dThird visit, summer.

and exposed to the ambient environment, but without pump sampling, and were analyzed by the same procedure.

CO₂

Carbon dioxide levels were measured with an ADC direct reader (IRSST, Montreal, Canada) calibrated with a 0.105% CO₂ standard. These measures were performed in triplicate three times during the 4-hour period.

Dust

Dust sampling also was for 4 hours and in triplicate. Preweighed 37-mm PVC filter (0.8 µm) housed in closed-face cassettes were used with SKC 224-44XR personal sample pumps (Dur-Pro) calibrated at 1.5 L/min (Kurz flowmeter; Instruments Inc., Carmel, Calif.). The sampling was carried out with the port of entry pointing upward. Filters were stored in the freezer until the end of the study. Filters were stored in a drying chamber until constant weight and weighed under controlled atmosphere to avoid rehydration. Control filters were brought to the sampling site and exposed, but not subjected to sampling, and weighed by the same procedure.

Bacteria

Airborne bacteria were sampled with all-glass impingers 30 (AGI-30) (Ace Glass Inc., Vineland, N.J.) connected to Gilian Aircon II pumps (Levitt Security, Montreal, Canada) at a flow rate of 12.5 L/min for 16 min, three times during the 4-hour period. Pump flow rate was set with a Kurz flowmeter. Sterile AGIs contained 20 mL of sterile saline water (0.08% NaCl) and were kept on ice after the sampling. Back at the laboratory (maximum 1 hour after the sampling procedure), sample volume was measured (to evaluate evaporation) and completed at 30 mL with sterile saline water containing 0.15% Tween 80 (final concentration of ~0.05%) and diluted to 10⁻². Undiluted and diluted samples were plated in triplicate on tryptic soy agar (TSA) (Difco, Detroit, Mich.) containing cycloheximide (500 mg/L) to avoid mold growth and incubated at 30°C for 60 hours. Total bacteria were counted at the dilution where the plates showed between 30 and 300 colonies. Control samples were taken outside, about 1 km upwind from the swine building, when the outside temperature was above -4°C. The levels were compared with indoor values. If outside colony numbers or visual population seemed obviously similar, these values were subtracted from the inside.

Molds

Airborne molds were sampled with a six-stage Andersen impactor (Grasby Andersen) connected to Gilian Aircon II pumps (Levitt security, Montreal, Canada) at a flow rate of 28.3 L/min for 2 min, three times during the 4-hour period. Pump flow rate was set with a Kurz flowmeter. Andersen samplers were loaded with rose-bengal agar (Difco) containing chloramphenicol (50 mg/L) to avoid bacterial growth. Dishes were incubated at 30°C for 5 days. Molds were identified with microscopic and macroscopic observations.⁽⁹⁾ Control samplings were performed as for the bacteria. The control samples were used in comparison with the indoor samplings. If similar mold colonies were found outside and inside, the outside level was subtracted. If the population was different, the controls were not used.

Thermophilic Actinomycetes and *Saccharopolyspora Rectivirgula*

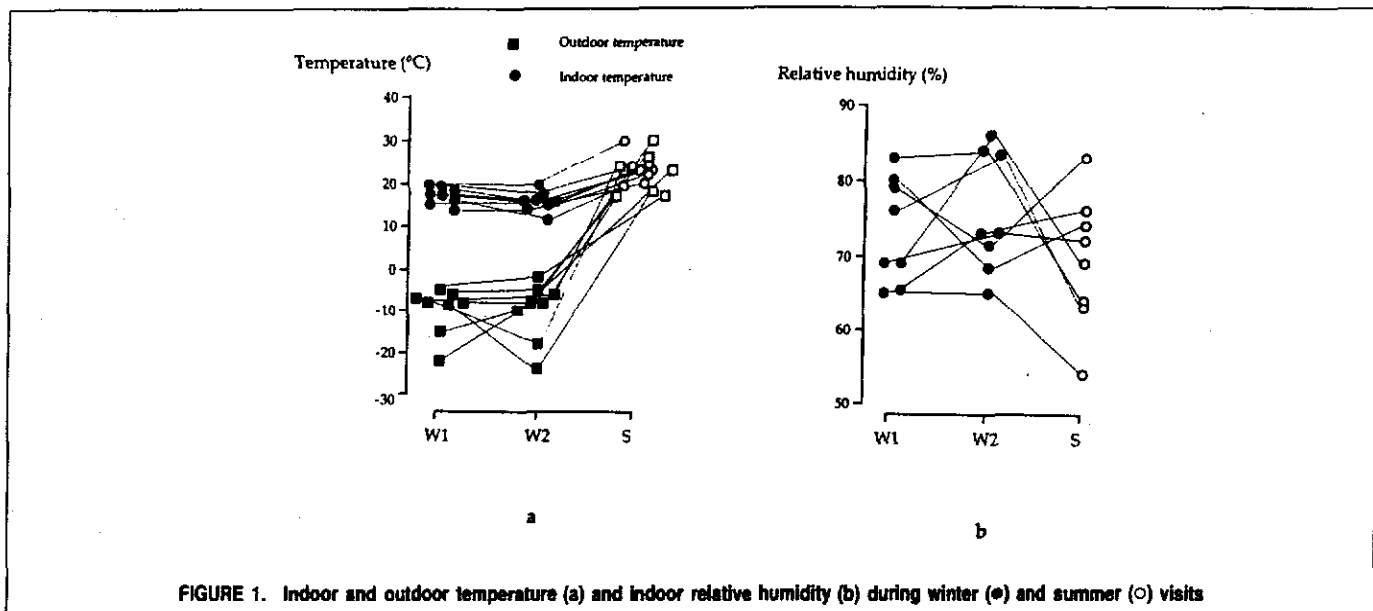
Thermophilic actinomycetes also were sampled with the Andersen impactor at the same flow rate but for 20 min, three times during the 4-hour period. Andersen samplers were loaded with TSA containing cycloheximide (500 mg/L) to avoid mold growth. Dishes were incubated at 52°C for 5 days. Thermophilic actinomycetes were counted and the presence of *Saccharopolyspora rectivirgula*, the bacteria most frequently responsible for farmer's lung disease, was evaluated using common growth characteristics.⁽¹⁰⁾

Endotoxins

To determine airborne endotoxin concentrations, AGI-30 samples were used. Samples were kept frozen (-20°C) in plastic (winter) or glass tubes (summer) before measurement. Endotoxin was measured with limulus amoebocyte assay (LAL) endpoint chromogenic test⁽¹¹⁾ (Associates of Cape Cod, Woods Hole, Mass.). Controls were obtained with sterile saline water containing 0.05% Tween 80 with which sterile AGI-30 samplers were washed for a few minutes. This procedure allowed measurement of the initial endotoxin contamination of the samples and material.

Statistical Analysis

According to the type of data, comparisons were performed with Student's paired t-test or Wilcoxon signed test. Spearman correlation coefficient was used to measure the relationships between different parameters. A p-value <0.05 was considered significant.



Comparisons were also performed between the two winter values and between winter visit and summer visit values. The results show the winter-summer comparison performed between the average winter values and the summer values.

RESULTS

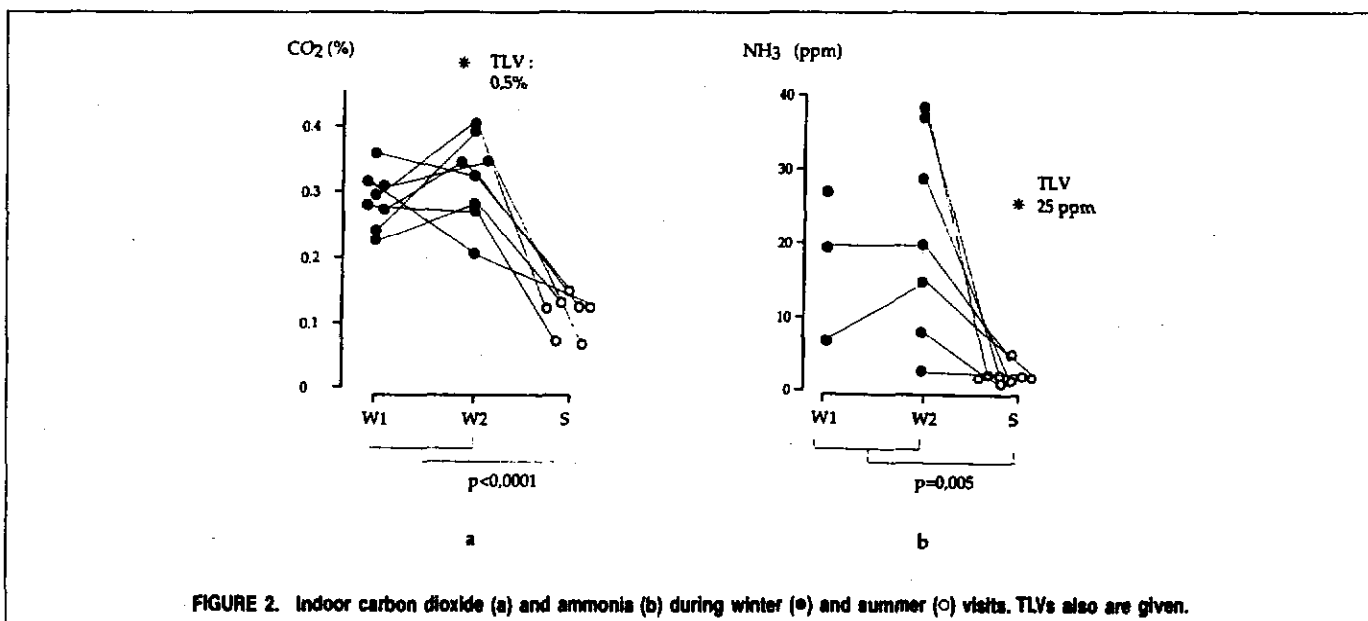
Visual Aspect and Physical Parameters

Table I shows the variability of the visual aspects: scores given are the average for the nine evaluations and the range of scores for each building. The individual evaluations were therefore very consistent. Total number of pigs and the size of the buildings where samplings were performed also are given. Indoor and outdoor temperatures were similar for the two winter visits (W1 and W2)

and, as expected, both of these temperatures were higher in summer (Figure 1a). However, although higher in summer, the difference in inside temperature was rather modest. Indoor air relative humidity remained constant (Figure 1b).

Air Sampling Results

Ammonia, CO₂ levels, and their threshold limit values (TLVs[®]) according to American Conference of Governmental Industrial Hygienists guidelines are shown in Figures 2a and 2b (missing data points in Figure 2a are due to technical problems). Ammonia and CO₂ levels were lower in summer (S) than in winter ($p=0.005$ and $p<0.0001$, respectively) and this was observed for all buildings. No difference was observed between the two winter samplings (W1 and W2) and, at W1, levels of ammonia were higher than the proposed TLV for one building; this was also observed in W2 for three buildings. Dust levels and TLV for total dust and



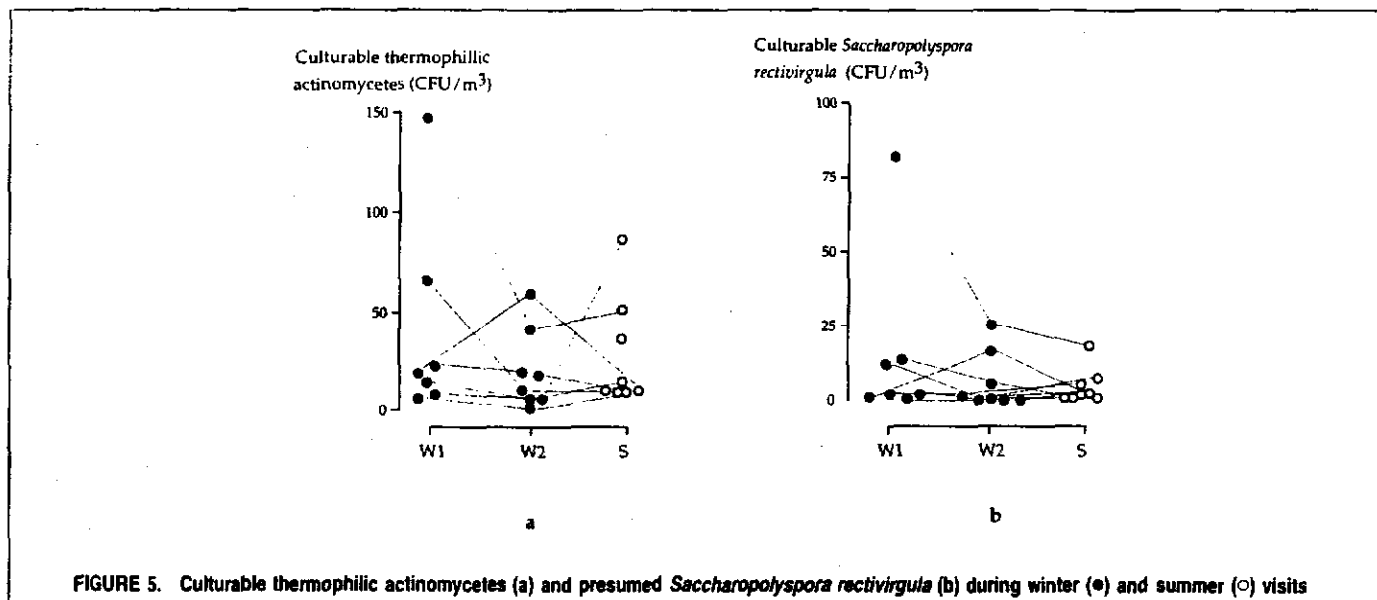


FIGURE 5. Culturable thermophilic actinomycetes (a) and presumed *Saccharopolyspora rectivirgula* (b) during winter (●) and summer (○) visits

modern facilities where cleanliness was strictly maintained contained as many, sometimes more, bacteria than that of the filthiest swine buildings. This was true despite the fact that a wide range of swine confinement buildings had been selected, as can be appreciated by the range in mean scores given by the nine volunteer evaluators. This apparent paradox can perhaps be explained by the importance of the number of pigs: the greater the number of pigs, the larger the bacterial counts. The negative relationship observed between dirtiness and number of pigs confirms that recently built buildings are larger and therefore shelter a greater number of pigs and are better maintained: new and clean facilities housed, on average, more pigs than the old and dirty ones. Another hypothesis to explain the decrease in bacterial levels with dirtiness would be that dust and dirt accumulation on the ceiling and walls could adsorb airborne bacteria produced by the pigs and the manure and thus help decrease the number of airborne bacteria. To prove this hypothesis, it would be interesting to perform a study on microbial contamination on wall and ceiling dust and dirt. It is also important to mention that even if correlations were observed, they may not express a causal relationship. The correlation may result from variables that are correlated with a causal factor.

This study showed that, in some cases during winter (and on average), viable bacterial levels were higher than the 4.3×10^5 cfu/m³ level associated with respiratory symptoms in humans.⁽¹³⁾ During summer the bacterial counts were lower than those obtained during the winter period, but still higher than 10^4 and 5×10^3 , the Danish and Swedish proposed values for work environment exposure, respectively.⁽¹²⁾ Surprisingly, endotoxin levels were higher during summer than winter, even if the total bacterial count was lowered by the increase in summer ventilation. The values reached are very high during summer (up to 10,000 EU/

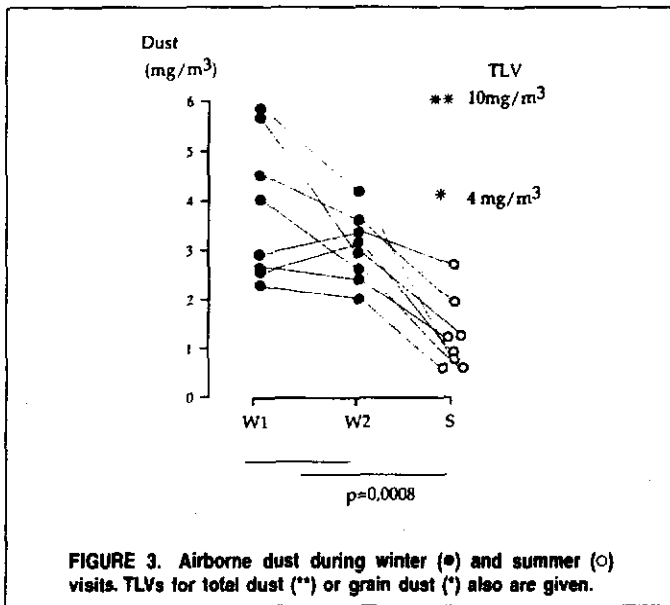
m³). This value is much higher than the recent proposed occupational exposure limit⁽¹³⁾ of 50 EU/m³. Because of technical differences in the analysis procedure, winter levels of endotoxins cannot be compared with those obtained in summer. Winter samples were frozen in plastic tubes whereas summer samples were frozen in glass tubes. Since plastic material has the ability to adsorb endotoxin, some endotoxin could have been lost before measurement. It is possible, therefore, that the levels found in winter were undervalued.

In the two studies discussed in the introduction, correlations were found between some airborne contaminants and physical parameters. Atwood et al.⁽⁷⁾ verified these relationships during winter only, whereas Kiekhaefer et al.⁽⁸⁾ performed all the analyses on a cross-seasonal basis. The correlations found in the present study are somewhat different from those found in those two studies. Most important, Kiekhaefer et al. demonstrated, in finishing buildings, a lower viable bacterial level during winter/spring when compared with summer/fall levels. The results shown in the present study demonstrate a significantly lower bacterial count in summer. This discrepancy may be due to the large difference between the two studied seasons in terms of ranges of outside temperature. In 1990 swine confinement building contamination was studied between January and April and no significant variability in bacterial contents was found within a relatively narrow range of outdoor temperature.⁽¹⁴⁾ Because of the greater temperature contrasts in the current study, ventilation rates were likely very different between winter and summer; carbon dioxide and ammonia levels support this suggestion.

Outdoor temperature had, on average, no effect on mold counts. The species recovered, and their relative proportions were also similar in winter and summer, with *Scopulariopsis*, *Aspergillus*,

TABLE II. Correlation Coefficients (r_s) and p-Values (in Brackets) for Different Combinations of Parameters

	Dirtiness	Bacteria	# Pigs	NH ₃	Dust
Molds	0.8 (0.002)				
Bacteria	-0.6 (0.03)				0.5 (0.02)
# Pigs	-0.6 (0.01)	0.4 (0.04)			
Endotoxins		0.6 (0.02)	0.7 (0.01)		
CO ₂				0.68 (0.006)	0.86 (<0.0001)



grain dust are shown in Figure 3. Dust levels were significantly higher in winter ($p=0.0008$). This higher level was observed in all buildings. At the Visit W1, three buildings had dust levels higher than the TLV for grain dust. This was observed in one building at Visit W2.

Bacterial counts (Figure 4a) were lower in summer than in winter ($p=0.04$). However, most of the summer bacterial counts were still higher than the Danish proposed threshold limit value for bacteria (10,000 cfu/m³).⁽¹²⁾ Mold counts (Figure 4c) and types were not influenced by seasons. The most important species found are also given on this figure. The control samples showed no molds (or very few) in winter samples and, in a few cases in summer samples outside isolates identical to inside ones were subtracted from the inside values (data not shown).

Some thermophilic actinomycetes (Figure 5a) and *Saccharopolyspora rectivirgula* (Figure 5b) were found, but their counts were very low and not season-dependent.

The endotoxin values are shown in Figure 4b. The dotted line means that no comparison should be performed between the summer and winter levels, because the method used was slightly different (see Discussion).

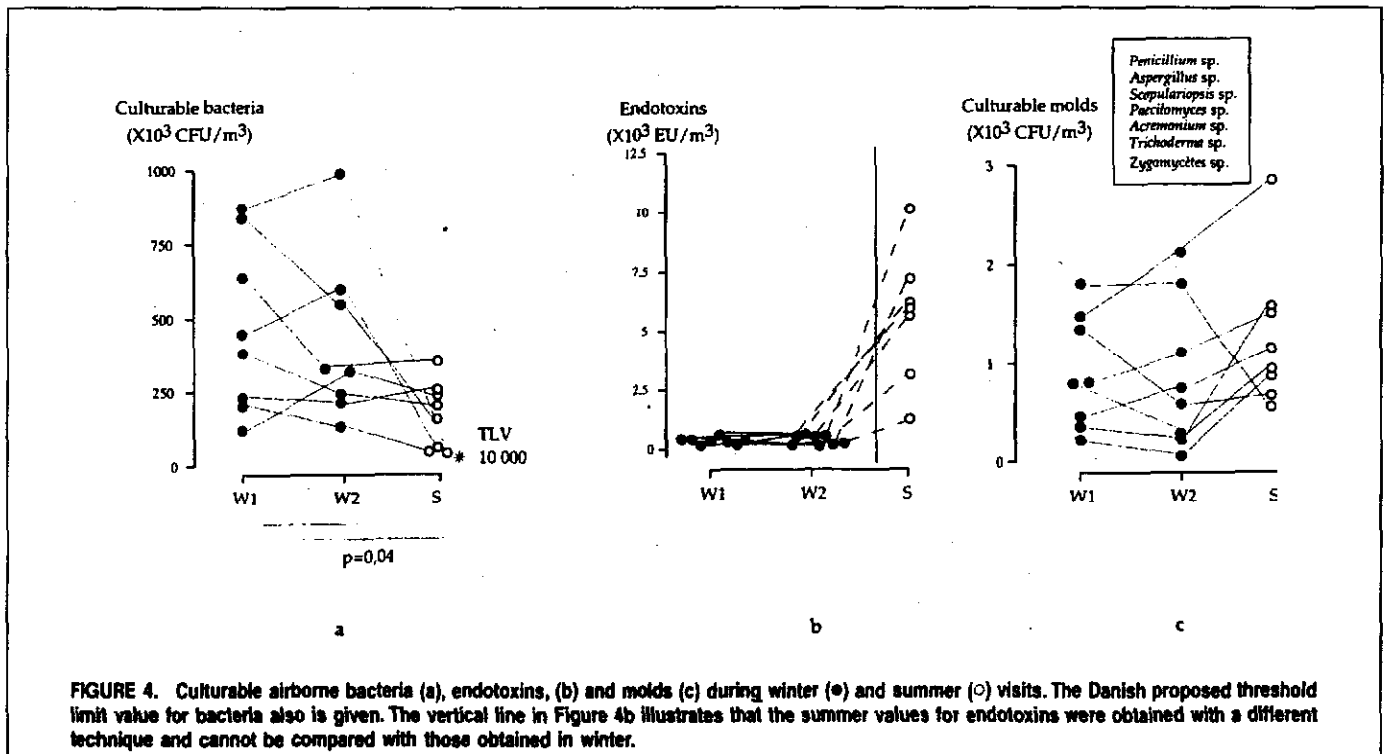
Spearman correlations between different air contaminants and physical parameters are shown in Table II. There were good positive correlations between visual aspects and airborne mold counts, ammonia, and carbon dioxide levels. Carbon dioxide also positively correlates ammonia and dust, and bacteria correlates number of pigs and endotoxins. Negative relationships were demonstrated between dirtiness and the number of pigs, and between dirtiness and bacterial levels. Paired Student t-test showed that indoor and outdoor temperatures (in other words, seasons) significantly affect carbon dioxide, bacterial counts, and dust.

Variation within the 4-hour samplings for bacterial counts is shown on Figure 6. This figure demonstrates that there is little constancy in bacterial contamination in the morning, at midday, or in the afternoon.

Results on the frequency at which the indoor dung collection system was emptied, other maintenance practices, and odor scores showed no constancy or correlation within themselves or with the other parameters (data not shown).

DISCUSSION

Although dirtiness was positively correlated with the number of molds, this characteristic was negatively associated with bacterial level. A possible explanation for the positive correlation between dust and molds is that dust and dirt accumulation on walls and ceilings could promote mold growth (molds are more xerophilic than bacteria). To the authors' surprise, the air inside



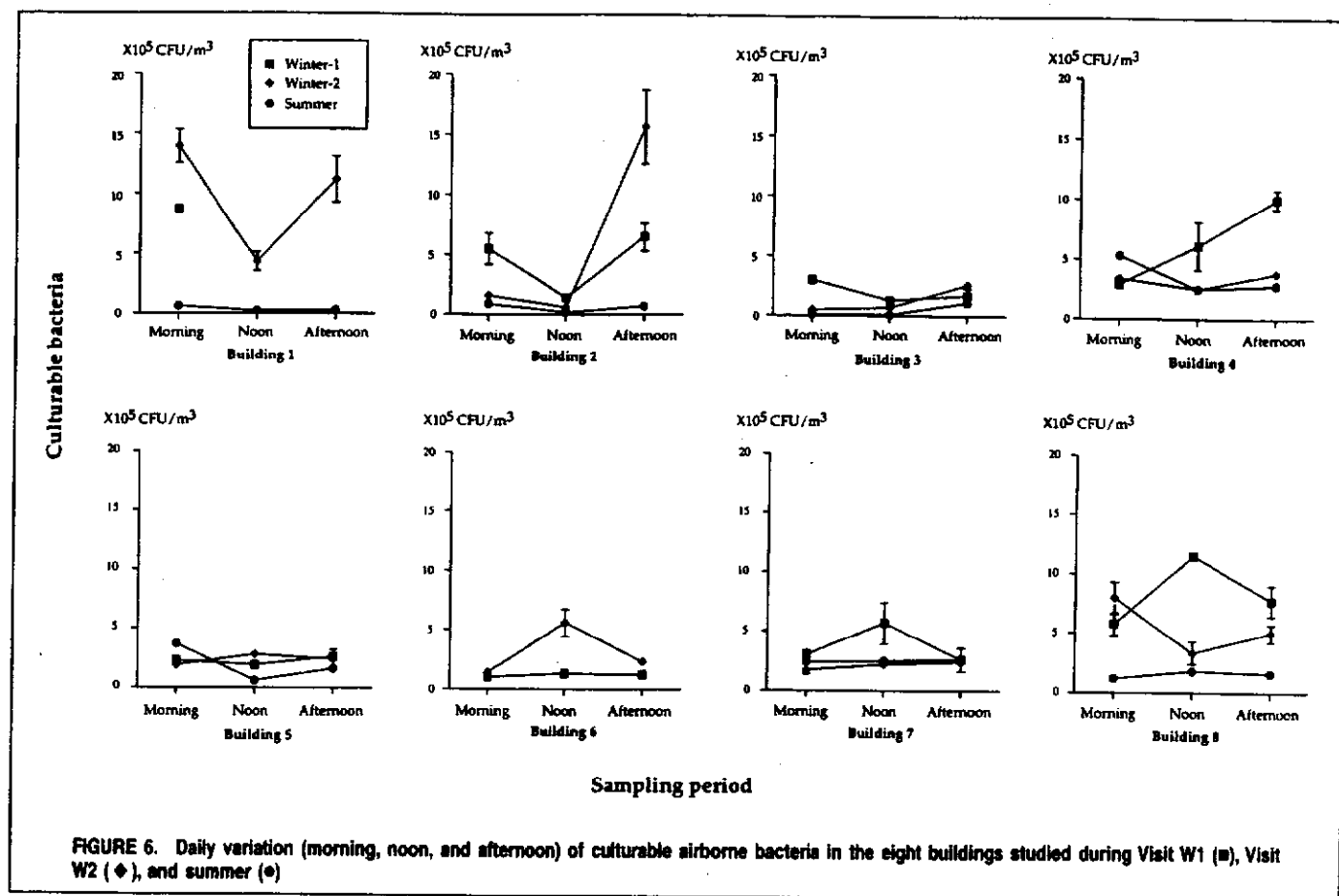


FIGURE 6. Daily variation (morning, noon, and afternoon) of culturable airborne bacteria in the eight buildings studied during Visit W1 (■), Visit W2 (◆), and summer (●)

and *Penicillium* being the most frequently recovered. Mold counts were not very high (from 2.82×10^2 to 3.82×10^3) when compared with levels found in some other highly contaminated environments such as dairy barns⁽²⁵⁾ but were comparable with levels usually found in swine buildings.⁽⁸⁾ Culturable mold levels were, in all cases, lower than the levels associated with respiratory symptoms in humans (1.3×10^4).⁽³⁾

The lack of constancy between highest and lowest levels of bacteria over a 4-hour period for a given swine building confirms the necessity of performing long-term sampling to evaluate worker exposure levels and their variability better.

CONCLUSIONS

A wide range of air contamination exists among swine confinement buildings of different maintenance, and there is a decrease in some of these contaminants during summer. Observed dirtiness of the swine confinement buildings has a poor predictive value of air quality, even if it is a good predictor for culturable fungi.

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