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VARIETY IN DUSTINESS AND HYGIENE QUALITY OF PEAT BEDDING

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Abstract: Respiratory exposure to organic dust induces chronic pulmonary diseases both in farmers and horses. The aim of this study was to examine the variation of dustiness and hygiene quality of peat moss bedding. Materials studied were weakly decomposed sphagnum peat (A), weakly decomposed sphagnum peat warmed up in storage (> 30ºC) (B) and two more decomposed few-flowered sedge peats (C and D). The geometric mean of mesophilic fungi, thermotolerant fungi and thermophilic actinomycetes were determined from the material. Samples of inhalable dust and endotoxins were collected with IOM samplers and respirable dust with 10M foam samplers when the peat was rotated in a cylinder. The number of particles was detected with an optical particle counter. An LAL assay was used for analysing endotoxins from the filter samples. There were differences in the hygiene quality and dustiness between peat materials ($p < 0.01$). The geometric mean of fungi was smallest in material A. Warming-up increased the number of fungi in sphagnum peat, but on the other hand, it decreased the content of endotoxin ($p < 0.01$). Few-flowered peat materials contained thermophilic actinomycetes and material D also contained *Aspergillus fumigatus*. The concentrations of inhalable dust, respirable dust and the number of particles were smaller in the few-flowered peats (C-D) than in the sphagnum peats (A-B). It is concluded that there are differences in the dustiness and hygiene quality of peat bedding.

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INTRODUCTION

Respiratory exposure to organic dust induces chronic pulmonary diseases both in farmers and horses. Horses suffering from chronic bronchitis are more often sensitised to *Aspergillus fumigatus* and *Alternaria alternata* allergens than horses in the control group, and they are partly sensitised to the same fungal proteins as mould-allergic human patients [4]. Especially the feeding of horses and handling of beddings increases the content of dust and fungal spores in the stable air [2, 22]. The amount of air impurities is according to the bedding material used in stable [21]. The horse is long-lived compared to other agricultural animals and its athletic ability is closely linked to its respiratory well-being. According to Holcombe *et al.* [8], stabling is associated with inflammation of both the upper and lower airways of young horses.

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Table 1. Median and range of volume weight, pH-value and humidity content of peat materials analysed in the study (n = 5).

Material	Volume weight (g/l) Median (Range)	pH Median (Range)	Humidity (%) Median (Range)
A. Light, weakly decomposed sphagnum peat, von Post H 1-2	82 (82–88)	3.9 (3.8–4.0)	26 (25–27)
B. Light, weakly decomposed sphagnum peat, von Post H 1-2, warmed-up in storage (>30ºC)	138 (136–140)	3.5 (3.4–3.6)	28 (28–32)
C. More decomposed few-flowered sedge peat, von Post H 3-4	188 (176–196)	4.3 (4.2–4.4)	43 (41–44)
D. More decomposed few-flowered sedge peat, von Post H 4-5	258 (256–270)	4.6 (4.6–4.7)	44 (41–44)

The usage of peat moss as bedding is quite common in Finland and Sweden. The influence of peat bedding on the air quality in piggeries [16] and horse stables [14] has been evaluated in few studies. The quality of peat materials used as bedding varies even when assessed with visual perception. However, comparative studies about the dustiness and hygiene quality of peat have not been published. Laboratory conditions allow comparison of the different materials without external influences such as ventilation, external temperature and animal activity affecting the results. The dustiness of straw and wood chip beddings has studied also been in some extent in the laboratory [10, 20].

Peat moss bedding absorbs liquid and ammonia very well [1]. The fertilization use of horse manure with peat bedding is easy [9] when compared to manure with wood chip bedding. This may be seen as an advantage to horse owners. Nevertheless, problems due to the dustiness of peat, that is, high contents of inhalable dust and microbes (fungi, bacteria), in the workplace have been reported [16]. The geometric mean of mesophilic fungi in clean peat bedding has varied from 10^2–10^8 cfu/g [1, 14, 16] and thermotolerant fungi from 10^3–10^5 cfu/g [1, 15]. In the study by Mäittälä *et al.* [16], clean peat bedding contained 10^2-10^3 cfu/g of thermophilic actinomycetes. Mesophilic fungi, such as *Oidiodendron* and *Penicillium* and yeasts have been found to be mainly species of clean peat [15]. Rautiala *et al.* [17] reported that mesophilic *Penicillium* and yeast have been common also in air samples in composting swine confinement buildings using peat as composting bedding. Thermotolerant *Paecilomyces* and *Penicillium* species [6, 16] have also been common in peat. Clarke and Madelin [3] found only *Aspergillus candidus* and *Streptomyces* species from clean peat bedding.

The purpose of this study was to examine the variation of dustiness and hygiene quality of peat moss which may be used as bedding. The objective of this trial was also to look into the influence of the stage of decomposition, humidity content and warming-up on the quality of peat and its suitability for bedding.

MATERIALS AND METHODS

Materials analysed. Peat moss materials studied were light, weakly decomposed sphagnum peat, von Post H 1-2 (material A), light, weakly decomposed sphagnum peat, von Post H 1-2, warmed-up in storage (> 30ºC) (material B), more decomposed few-flowered sedge peat, von Post H 3-4 (material C) and more decomposed few-flowered sedge peat, von Post H 4-5 (material D). The volume weight (g/l), pH-value and humidity content (%) of peat materials are presented in Table 1. Analyses were done with a well-mixed compiled sample (volume of 30 litre). The compiled sample included about 60 partial samples taken from the peat material. The pH-value of peat material was measured (n = 5) from the peat-water extract (ratio of volume was 2.5:1) after the peat was soaked for 20 hours in deionised water at room temperature (Knick-pH-Einstabmebketten, Typ SE 101, Elektronische Meβgeräte GmbH, Germany). The volume weight and humidity content of materials were determined gravimetrically (n = 5). Peat samples were dehydrated for 20 hours at 105ºC before weighing for humidity content determination (Sartorius analytic, Type A 120 S, Sartorius GmbH Göttingen, Germany).

Microbial analyses. The content of viable microbes (mesophilic fungi, thermotolerant fungi and thermophilic actinomycetes) was determined from peat (n = 4). The number of colony forming units (cfu/g) was counted after incubation and identified microscopically. The culture media, incubation temperature and incubation time used were: Hagem agar, 25ºC, 7 days (mesophilic fungi), Hagem agar, 40ºC, 5 days (thermotolerant fungi) and half strength nutrient agar, 55ºC, 3 days (thermophilic actinomycetes). The detection limit was 1,000 cfu/g for materials A and B and 100 cfu/g for materials C and D.

Dust and endotoxin analyses. The aim of this procedure was to simulate the dust exposure for workers in an actual working situation. Spreading of the dust from the peat material in the cylinder was attempted so as to make it similar to that caused by changing bedding in a horse stable. This procedure was chosen to of allow the comparison of the different peat materials without external influences, such as ventilation and horse activity affecting the results.

A one-litre sample was taken and analysed for dustiness using a rotating drum with a cylinder 70 cm long and 30 cm in diameter, containing eight mixing plates 5 cm in height. Three air samples were taken from the cylinder on three different filters at the same time, and the number of particles released during rotation was

Table 2. Geometric average (cfu/g) and range of viable micro-organisms in a gram of peat material. Detection limit was 1,000 cfu/g for materials A and B and 100 cfu/g for materials C and D (n = 4). (The other legends are as in Tab. 1.)

Material	Geometric mean and range of microorganism, cfu/g		
	mesophilic fungi	thermotolerant fungi	thermophilic actinomycetes
A	2.4×10^5 (2.0×10^5-3.1×10^5)*	5×10^2 (<10^2-10^2)*	<2.5×10^2
B	7.2×10^7 (5.7×10^7-1.1×10^9)*	6.6×10^6 (5.1×10^6-7.6×10^6)*	<2.5×10^2
C	1.8×10^7 (1.5×10^7-2.0×10^7)*	3.3×10^5 (2.8×10^5-3.9×10^5)*	<2.5×10^1
D	1.4×10^6 (9.2×10^5-2.6×10^6)*	4.2×10^4 (3.0×10^4-5.2×10^4)*	1.2×10^3 (3.0×10^2-2.4×10^3)*

*p < 0.05 Asymp. Sig (2-tailed). Mann-Whitney U –test.

Table 3. Median and range of the content of inhalable dust, respirable dust and endotoxin released into the air during cylinder rotation of peat material 100 g (n = 5). (The other legends are as in Tab. 1.)

Material	Inhalable dust, mg/m^3 Median (Range)	Respirable dust, mg/m^3 Median (Range)	Endotoxin, EU/m^3 Median (Range)
A	47.3 (32.0–87.6)	5.3 (3.4–6.7)**	73,000 (35,000–85,000)**
B	3.3 (1.6–6.9)	2.3 (1.0–3.0)**	280 (180–790)
C	1.7 (1.4–2.5)	0.4 (0.4–0.6)	360 (220–440)
D	2.1 (0.9–2.6)	0.3 (0.3–0.7)	600 (430–630)

**p < 0.01 Asymp. Sig (2-tailed). Mann-Whitney U –test.

counted. The cylinder was rotated six times at a speed of 34 rpm, for three minutes at a time, at 10-minute intervals.

The sampling time for each filter was 60 minutes, during which the stationary samples for inhalable dust, respirable dust and endotoxin were collected with IOM samplers for 18 minutes. Endotoxin and inhalable dust were sampled with IOM samplers and respirable dust with an IOM sampler provided with polyurethane foam. Dust samples were taken with the use of calibrated pumps (Model 224, SKC, USA) at the airflow of 2.0 litre/minute. The dust was analysed gravimetrically.

The number of dust particles released into the air was counted using an optical particle counter (Hiac/Royco, Model 5000, Spacific Scientific®, USA). The particle counter measured the number of particles with different sizes in five categories: 0.3-0.5 µm, 0.5-1 µm, 1-3 µm, 3-5 µm and 5-10 µm. The endotoxin was analysed with the kinetic Bio Whittaker-QCL method, based on a LAL (Limulus amebozyte lysate) enzyme. The sampling procedure was performed five times consecutively for each peat material. The cylinder was vacuumed thoroughly after every rotated material.

Statistical analyses. Statistical analyses were carried out with the statistical software package SPSS 10.0 for Windows. The non-parametric Kruskal-Wallis and Mann-Whitney U tests were used to detect differences in measured qualities of peat materials. Correlation between measured qualities was determined by calculating values of Spearman's correlation coefficient (*r*). The value of *r* was considered statistically significantly different from zero when the p-value for a correlation coefficient test was less than 0.05.

RESULTS

Hygiene quality after logarithm transformation and dustiness of studied peat materials were significantly different from each other ($p < 0.01$). Light, weakly decomposed sphagnum peat (material A) contained the least fungi (Tab. 2). The content of mesophilic and thermotolerant fungi was highest in light, weakly decomposed sphagnum peat which warmed-up (> 30ºC) during storage (material B), and the second highest in the more decomposed few-flowered sedge peat, von Post H 3-4 (material C). Thermophilic actinomycetes were found only in the most decomposed few-flowered sedge peat (material D).

Penicillium was the main fungus in all peat materials studied (Tab. 4). Yeasts were present only in the more decomposed few-flowered sedge peats (materials C and D), which contained also more mesophilic fungi species than light, weakly decomposed sphagnum peat. Materials C and D contained also *Aspergillus fumigatus* fungus and cellulose-decomposing *Trichoderma* fungus. Sterile fungi in materials B, C and D were fungi which did not sporulate in the culture media used. Thus, those non-sporing fungi could not be identified microscopically.

The concentrations of inhalable dust, respirable dust and endotoxin released into the air during cylinder rotation of peat are shown in Table 3. Light, weakly decomposed sphagnum peat contained significantly more inhalable dust and endotoxin than other peat materials ($p < 0.01$). The concentrations of respirable dust in material A was about ten-fold when compared to materials C and D.

The number of particles of all sizes were highest in material A ($p < 0.02$). The difference in the number of

Table 4. Geometric average (cfu/g) and species of mesophilic and thermotolerant fungi in the peat materials studied (n = 4). (A. = Aspergillus; the other legends are as in Tab. 1.)

Geometric mean and species of mesophilic and thermotolerant fungi in peat							
A		B		C		D	
mesophilic fungi							
Penicillium	2.3×10^5	*Penicillium*	7.1×10^7	*Oidiodendron*	1.4×10^7	Yeast $_{white}$	7.0×10^5
Monocillium	6.8×10^3	*Blastobotrys*	1.0×10^6	Yeast $_{white}$	2.3×10^6	*Penicillium*	5.5×10^5
Absidia	2.3×10^2			*Geomyces*	8.9×10^5	Yeast $_{red}$	6.6×10^4
				Yeast $_{red}$	2.5×10^5	*Oidiodendron*	5.4×10^4
				Penicillium	1.8×10^5	*Acremonium*	1.1×10^4
				Paecilomyces	6.8×10^4	*Rhinocladiella*	1.1×10^4
				Mucor	6.8×10^4	*Monicillium*	9.0×10^3
				A. fumigatus	4.5×10^4	Sterile fungi	6.8×10^3
				Trichoderma	2.3×10^4	*Blastobotrys*	6.8×10^3
				Aspergillus	2.3×10^4	*Trichoderma*	6.8×10^3
						Engyodontium	2.3×10^3
						Lecotyphora	2.3×10^3
						A. fumigatus	2.3×10^3
thermotolerant fungi							
Paecilomyces	5.0×10^2	*Penicillium*	4.9×10^6	*Penicillium*	1.6×10^5	*A. fumigatus*	2.0×10^4
		Sterile fungi	1.3×10^6	*A. fumigatus*	8.6×10^4	*Penicillium*	9.7×10^3
		Paecilomyces	3.6×10^5	Sterile fungi	6.1×10^4	Sterile fungi	7.5×10^3
				Paecilomyces	2.0×10^4	*Paecilomyces*	4.5×10^3
				Mucor	2.3×10^3		

particles in size 3-10 µm was over 350-fold between materials A and D. There was a variation in the amount of 3-5 µm sized particles released from the peat material onto the cylinder during rotation periods (Fig. 1). The number of particles was higher in the first rotation period than in the second period in materials B and C. The rotation of the cylinder increased slightly the amount of particles in size 3-5 µm in all peat materials studied.

The humidity content of peat was noticed to correlate significantly with the concentration of inhalable dust ($r = -0.72$, $p = 0.001$), respirable dust ($r = -0.88$, $p < 0.001$), number of particles ($r < -0.80$, $p < 0.001$) and the content of endotoxin ($r = -0.54$, $p < 0.02$) released from peat during handling. Any correlation between the fungi and humidity content could not be shown.

DISCUSSION

Light, weakly decomposed sphagnum peat (material A) released most inhalable and respirable dust during handling, but on the other hand, it contained the least mesophilic and thermotolerant fungi. The dustiness of material A may be partly explained by the lower humidity content than in the others. Larsson *et al.* [15] have suggested that a humidity content of 50% would keep the dust content of air low enough during peat bedding handling. Studied peat materials represent the conventional humidity content of peat bedding conveyed to the horse stables in Finland where their humidity content was distinctly under 50%.

In agreement with Mäittälä *et al.* [16], *Penicillium* was the main fungus in all the peat materials studied. Mesophilic yeasts which have been mentioned to be one of the main species in clean peat [15] were not found from weakly decomposed sphagnum peat. More decomposed few-flowered sedge peat contained *Aspergillus fumigatus* and material D also contained thermophilic actinomycetes. Those microbes have been associated with pulmonary diseases in humans [11]. According to Eder *et al.* [4], also horses suffering from chronic bronchitis are more often sensitised to some *Aspergillus fumigatus* allergens than horses in the control group. The occurrence of *Trichoderma* in materials C and D may be seen as a sign of far-advanced decomposition of those peat materials.

In this study, the content of endotoxin released into air during cylinder rotation of peat materials was 280-73,000 EU/m^3. According to Rieger *et al.* [18], the endotoxin content in horse stable air varied - 2,000 EU/m^3 in stables with sawdust bedding, 14,000 EU/m^3 in stables with straw bedding, and 22,000 EU/m^3 in stables with hemp bedding. Light, weakly decomposed sphagnum peat (material A) contained significantly more endotoxin than the other peat materials ($p < 0.01$). One reason for this may be the low stage of decomposition in sphagnum peat. According to Su *et al.* [19], the early stage of cotton textile processing seems to generate high endotoxin and bacteria contamination. The concentration of biologically-active endotoxin has been reported to be associated quite well with workers' reported symptoms [13]. According to the results

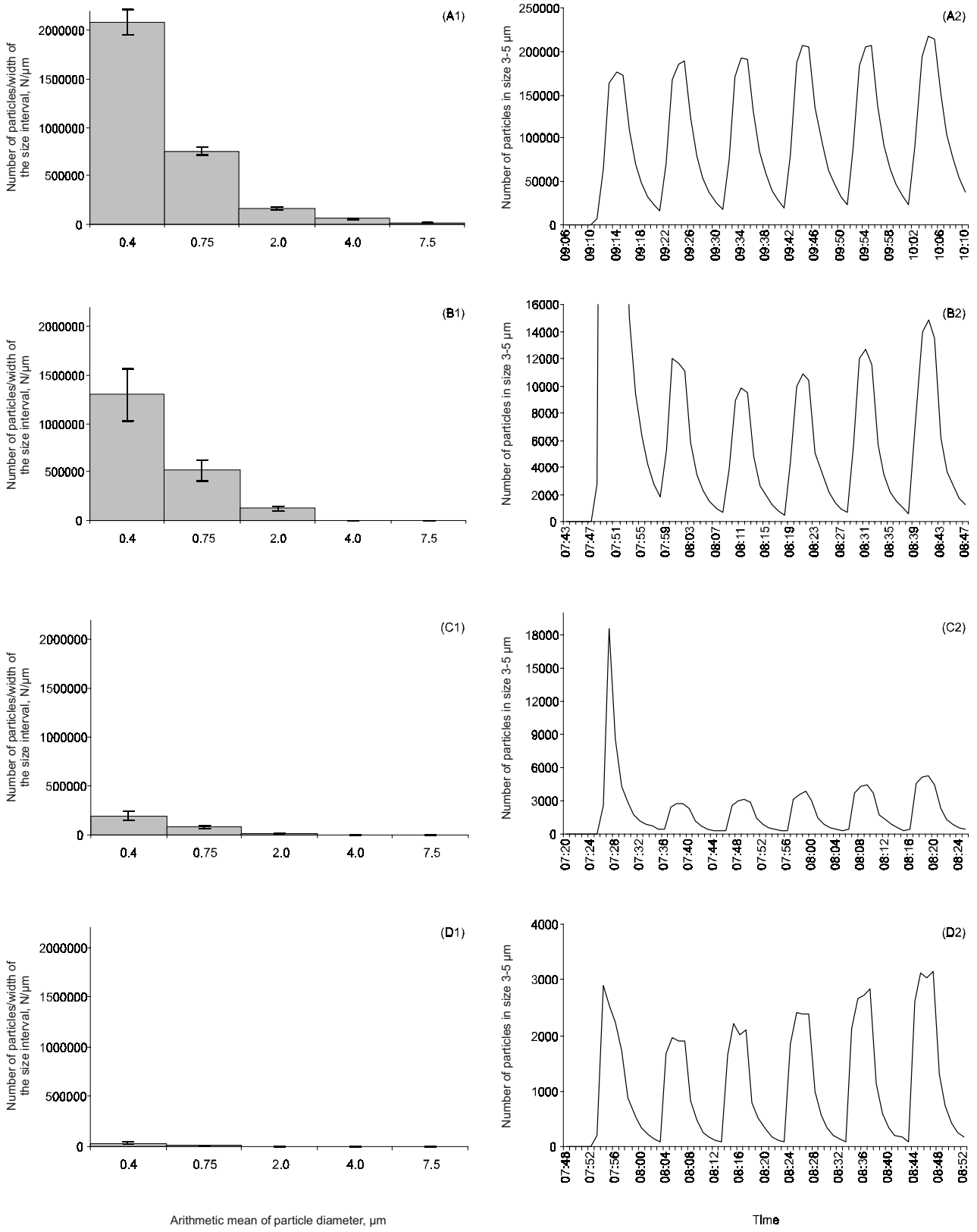

Figure 1. Number of dust particles in relation to the width of size interval vs mean particle diameter (A1-D2), and number of 3-5 µm sized particles in the cylinder during the rotation management (A2-D2) of weakly decomposed sphagnum peat, von Post H 1-2 (A1 and A2), weakly decomposed sphagnum peat, von Post H 1-2, and warmed-up (B1 and B2), more decomposed few-flowered sedge peat, von Post H 3-4 (C1 and C2), and more decomposed few-flowered sedge peat, von Post H 4-5 (D1 and D2).

of this study, it may be assumed that using material A as bedding might cause higher bacteria exposure than the bedding used from other studied materials. Workers' respiratory symptoms have been reported to increase when the air content of endotoxin is > 250 EU/m^3 [12]. The Dutch Expert Committee on Occupational Standards has recommended a health-based occupational exposure limit of 50 EU/m^3 over an eight-hour period of endotoxin exposure [7].

Warming over 30ºC increased the number of fungi in peat material but, on the other hand, it decreased the content of endotoxin in light, weakly decomposed sphagnum peat. No correlation between warming and the content of peat's dust was noticed.

The concentrations of inhalable dust (2-47 mg/m^3) measured from the studied peat materials cannot be compared directly to the occupational exposure limits for organic inhalable dust (5 mg/m^3 $_{8\ h}$ and 10 mg/m^3 $_{15\ min}$ [5]), nor to the air content of inhalable dust measured from a piggery (3.5-41.0 mg/m^3 [16]) using peat bedding. According to Kaliste *et al.* [10], the content of inhalable dust released from clean wood chip bedding was < 1-25 mg/m^3 when measured with the rotation cylinder. The sample volume (0.5 litre) and rotation time for sampling (6 minutes) were, however, slightly different in Kaliste's work to those used in this study (1 litre and 18 minutes). The rotation of peat material in the cylinder may considered more aggressive handling when compared to the bedding used in horse stables.

The number of 3-5 µm sized particles stayed quite steady in the light, weakly decomposed sphagnum peat during rotation management. This might be due to the higher number of fine particles in material A because the number of particles did not decrease between the first and second rotation periods, as happened with the other peat materials. One reason for the occurred particle reduction might be the coincident dust sampling. However, the rotation of the cylinder increased slightly the amount of 3-5 µm sized particles in all the peat materials studied. This may be seen as a sign of peat materials grinding during the rotation. The same grinding of peat might happen also in bedding use, especially when dry peat material is used. The number of 0.5-5 µm sized particles in the air released during handling of the more decomposed few-flowered sedge peat was the same as the content measured released from good quality straw and wood shavings [20].

CONCLUSIONS

The variation of dustiness and hygiene quality in peat moss bedding was studied. There were significant differences in the quality of different materials which should be observed when bedding is chosen. Conclusions arrived at in this study are as follows:

a) Light, weakly decomposed sphagnum peat (von Post H 1-2) included less fungi but a significantly higher content of endotoxin than more decomposed peat materials (von Post H 3-5).

b) The warming-up of peat increased the number of fungi in the material but, on the other hand, it decreased the endotoxin content of weakly decomposed sphagnum peat.

c) The more decomposed few-flowered sedge peats (von Post H 3-5) included *Aspergillus fumigatus* and material D also contained thermophilic actinomycetes.

d) The content of inhalable dust, respirable dust and the number of particles released during handling of the materials were smaller in more decomposed few-flowered sedge peat (von Post H 3-5) than in less decomposed sphagnum peat (von Post H 1-2).

e) The humidity content of peat correlated significantly with the content of dust released during peat handling.

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