

EXPOSURE TO AIRBORNE MICROORGANISMS, DUST AND ENDOTOXIN DURING PROCESSING OF VALERIAN ROOTS ON FARMS

Czesława Skórska, Jolanta Sitkowska, Ewa Krysińska-Traczyk, Grażyna Cholewa,
Jacek Dutkiewicz

Department of Occupational Biohazards, Institute of Agricultural Medicine, Lublin, Poland

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Abstract: The aim of this study was to determine the levels of microorganisms, dust and endotoxin in the air during various stages of valerian (*Valeriana officinalis*) roots processing by herb farmers and to examine the species composition of airborne microflora. Air samples were collected on glass fibre filters by use of personal samplers on 15 farms owned by valerian cultivating farmers, located in Lublin province (eastern Poland). The concentrations of total viable microorganisms (bacteria + fungi) in the air showed a marked variability and were within a range of $0.95\text{--}7,966.6 \times 10^3$ cfu/m³. Though median was relatively low (10.75×10^3 cfu/m³), on 4 farms the concentrations exceeded the level of 10^5 cfu/m³ and on 1 farm the level of 10^6 cfu/m³. During the processing of valerian roots, distinct changes could be observed in the composition of airborne microflora. In the first stages of processing, the freshly dug and washed roots until shaking in the drying room, the most numerous were Gram-negative bacteria of the family Pseudomonadaceae (mostly *Stenotrophomonas maltophilia*, *Pseudomonas chlororaphis* and *Pseudomonas fluorescens*). After drying, the dominant organisms were thermo-resistant endospore-forming bacilli (*Bacillus* spp.) and fungi, among which prevailed *Aspergillus fumigatus*. Altogether, 29 species or genera of bacteria and 19 species or genera of fungi were identified in the farm air during valerian processing, of these, 10 and 12 species or genera respectively were reported as having allergenic and/or immunotoxic properties. The concentrations of airborne dust and endotoxin on the examined farms were very large and ranged from 10.0–776.7 mg/m³, and from 0.15–24,448.2 µg/m³, respectively (medians 198.3 mg/m³ and 40.48 µg/m³). In conclusion, farmers cultivating valerian could be exposed during processing of valerian roots to large concentrations of airborne microorganisms, dust and endotoxin posing a risk of work-related respiratory disease.

Address for correspondence: Dr Czesława Skórska, Department of Occupational Biohazards, Institute of Agricultural Medicine, Jaczewskiego 2, 20-090 Lublin, Poland. E-mail: skorska@op.pl

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INTRODUCTION

Various authors have demonstrated that handling of grain, hay, flax and other plant materials may be associated with exposure to large quantities of organic

dust and bioaerosols causing allergic and/or immunotoxic reactions and respiratory disease in the workers [10, 14, 15, 16, 25, 33, 35, 36, 47, 48, 56, 59]. So far, relatively little is known about the risk associated with handling of various herbs, in spite of growing interest in cultivating

and processing herbs for medical, alimentary and cosmetic purposes [5, 26]. Dutkiewicz *et al.* [21] found that the workers of big herb processing plants were exposed to large concentrations of microorganisms ($40.6-627.4 \times 10^3$ cfu/m³) and endotoxin (0.2-2,681.0 µg/m³). Similarly, Krysińska-Traczyk *et al.* [32] recorded large concentrations of microorganisms ($90.4-594.8 \times 10^3$ cfu/m³) and endotoxin (37.4-2,448.8 µg/m³) during cleaning of thyme by herb farmers.

People occupationally exposed to dust from herbs showed a high proportion of allergic reactions to the extracts of herbs and microorganisms associated with herb dust and often reported work-related respiratory symptoms [22, 26]. Accordingly, our group has extended the research on exposure to herb dust and its effects onto further species of medicinal herbs cultivated in Poland, including valerian.

Valerian (*Valeriana officinalis* L.) is a perennial plant of the family Valerianaceae which has been known since ancient times as a medicinal herb. The extract of its roots has a remarkable influence on the cerebro-spinal system, and is used as a sedative and nerve relaxant. The root has also antispasmodic, carminative, diuretic and hypotensive properties. The rhizome-type roots of 2-year-old plants are harvested in the autumn and are used fresh or dried [4, 6].

The aim of the present work was to determine the levels of microorganisms, dust and endotoxin in the farm air during various stages of valerian roots processing, and to examine the species composition of airborne microflora.

MATERIALS AND METHODS

Examined farms. Air sampling was performed on 15 farms owned by valerian cultivating farmers, located in Lublin province (eastern Poland) on the territory of 1 village, at the distance of circa 30 km to the west of the city of Lublin. Samples were collected in the autumn 2004 (23 September - 14 October) during processing of harvested valerian roots (rhizomes) by farmers.

Before harvesting of roots, leaves were cut down by a cutting machine. Then, valerian roots were dug out by another machine, washed in a river and loaded for a week in drying rooms at 40-50°C, either threaded on wire loops in old-type drying rooms or loosely in new-type drying rooms. The dried roots were sacked for disposal to herb processing facilities.

Air samples were collected during performing by herb farmers following activities at processing valerian roots: 1) Manual cleaning of dug valerian roots, trimming leaves remnants and outgrowths (on 2 farms: 1A and 1B); 2) Shaking of moist (washed) roots before threading on wire (on 1 farm: 2); 3) Threading of moist roots on wire loops (on 1 farm: 3); 4) Shaking of loose roots in a modern drying room (on 2 farms: 4A and 4B); 5) Unloading of loose roots from a modern drying room (on 1 farm: 5); 6) Taking out loops with threaded roots from an old-type drying room (on 1 farm: 6); 7) Stripping off dried roots

from wire loops (on 3 farms: 7A, 7B and 7C); 8) Sacking of dried valerian roots (on 4 farms: 8A, 8B, 8C and 8D).

Microbiological examination of the air. Air samples were taken by use of an AP-2A personal sampler (TWOMET, Zgierz, Poland), at the flow rate of 2 l/min. Glass fibre filters, with 1 µm pore size and 37 mm diameter, were used. On each farm, 3 samples were collected in parallel using 3 samplers during 30 minutes: 1 for determination of the concentration and species composition of microorganisms, and the other 2 for determination of the concentration of dust and endotoxin. The concentration of dust in the air was determined gravimetrically from the difference between weight of the filter measured before and after sampling. The concentration of airborne dust estimated for each farm was a mean of 3 single determinations.

In addition, on each farm 3 air samples were collected in parallel on the polyvinyl chloride filters by use of 3 stationary AS-50 samplers (TWOMET, Zgierz, Poland), at the flow rate of 50 l/min, for determination of the dust and endotoxin concentrations. The concentration of airborne dust, estimated gravimetrically for each farm, was a mean of 3 single determinations.

The concentration and species composition of microorganisms in collected air samples were determined by dilution plating. The filters were extracted in 3 ml of sterile saline (0.85% NaCl) with 0.05% Tween 80, and after shaking, serial 10-fold dilutions were made. The 0.1 ml aliquots of each dilution were spread on duplicate sets of 4 agar media: blood agar for estimation of total mesophilic Gram-negative and Gram-positive bacteria, eosin methylene blue (EMB) agar (Merck, Darmstadt, Germany) for estimation of Gram-negative bacteria, half-strength tryptic soya agar (Sigma, St. Louis, MO, USA) for estimation of thermophilic actinomycetes, and malt agar (Difco, Detroit, MI, USA) for estimation of fungi. The blood agar plates and EMB agar plates were subsequently incubated for 1 day at 37°C, then 3 days at 22°C and finally 3 days at 4°C. The malt agar plates were subsequently incubated for 4 days at 30°C and 4 days at 22°C [14]. The prolonged incubation at lower temperatures aimed to isolate as wide a spectrum of bacteria and fungi as possible. The tryptic soya agar plates were incubated for 5 days at 55°C. The grown colonies were counted and differentiated and the data reported as cfu per 1 cubic metre of air (cfu/m³). The total concentration of viable microorganisms in the air was obtained by the addition of the concentrations of total mesophilic bacteria (grown on blood agar medium), thermophilic actinomycetes and fungi. The percent composition of the total microflora of the air was then determined.

Bacterial isolates were identified with microscopic and biochemical methods, as recommended by Bergery's Manual [30, 58, 62] and Cowan & Steel [8]. Additionally, the selected isolates were identified with microtests: API Systems 20E and NE (bioMérieux, Marcy l'Etoile, France) and BIOLOG System (Biolog, Inc., Hayward, CA, USA).

Fungi were classified by microscopic methods, according to Barron [1], Larone [38], Litvinov [40], Ramirez [49], and Raper & Fennell [50].

The concentration of bacterial endotoxin in the airborne dust was determined by the *Limulus* amoebocyte lysate gel tube test (LAL) [39]. The filters were extracted for 1 hour in 10 ml of pyrogen-free water at room temperature, heated to 100°C in a Koch apparatus for 15 min (for better dissolving of endotoxin and inactivation of interfering substances), and after cooling, serial dilutions were prepared. The 0.1 ml dilutions were mixed equally with the "Pyrotell" *Limulus* reagent (Associates of Cape Cod, Inc., Falmouth, MA, USA). The test was incubated for 1 hour in a water bath at 37°C, using pyrogen-free water as a negative control and the standard lipopolysaccharide (endotoxin) of *Escherichia coli* 0113:H10 (Difco) as positive control. The formation of a stable clot was regarded as a positive result. The estimated concentration of endotoxin in dust (ng/mg) was multiplied per estimated concentration of dust in the air (mg/m³) and the results were reported as micrograms of the equivalents of the *E. coli* 0113:H10 endotoxin per 1 m³ of air. To convert to Endotoxin Units (EU), the value in nanograms was multiplied by 10.

Statistical analysis. The data were analysed by Shapiro-Wilk test for distribution, Spearman correlation

test and Wilcoxon matched-pairs test, using STATISTICA for Windows v. 5.0 package (Statsoft©, Inc., Tulsa, Oklahoma, USA).

RESULTS

The concentrations of total viable microorganisms in the farm air during processing valerian roots varied within a wide range from 0.95-7,966.6 × 10³ cfu/m³ (Tab. 1). Though median was relatively low (10.75 × 10³ cfu/m³), on 4 farms, the concentrations exceeded the level of 10⁵ cfu/m³ and on 1 farm at the level of 10⁶ cfu/m³. The median amounts of mesophilic bacteria and fungi were similar, while thermophilic actinomycetes formed only small portion of total microflora. Mesophilic bacteria prevailed on 8 farms, fungi on 6 farms, and on 1 farm their amounts were equal (Tab. 1).

During the processing of valerian roots, distinct changes could be observed in the composition of airborne microflora. In the first stages of processing the freshly dug and washed roots, until shaking in the drying room, the most numerous were Gram-negative bacteria, except the sampling site at threading of moist roots on wire loops, where fungi prevailed (Fig. 1). After drying, the dominant organisms were thermo-resistant endospore-forming bacilli and fungi. Bacilli prevailed at unloading valerian roots from drying rooms, while fungi were most

Table 1. Concentrations of microorganisms (cfu/m³ × 10³) in farm air during processing valerian roots.

No.	Activity	Sampling site	Total mesophilic bacteria (Blood agar)	Gram-negative bacteria (EMB agar)	Thermophilic actinomycetes (Tryptic soya agar)	Fungi (Malt agar)	Total microbes*
1	Cleaning and trimming of dug valerian roots	Farm 1A	120.0	69.0	1.2	0.05	121.25
		Farm 1B	0.6	0.5	0	0.6	1.2
2	Shaking of moist roots before threading on wire	Farm 2	7,965.0	1,005.0	0	1.6	7,966.6
3	Threading of moist roots on wire loops	Farm 3	0.2	0	0.1	75.0	75.3
4	Shaking of loose roots in a drying room	Farm 4A [#]	0.8	0	0	0.15	0.95
		Farm 4B [#]	795.0	399.0	0.9	8.3	804.2
5	Unloading of loose roots from drying room	Farm 5 [#]	11.4	0	0.6	4.0	16.0
6	Taking out loops with threaded roots from drying room	Farm 6	2.4	0.7	0.08	0.7	3.18
7	Stripping off dried roots from wire loops	Farm 7A	1.7	0	0.04	2.5	4.24
		Farm 7B	2.6	0.8	0.7	4.1	7.4
		Farm 7C	4.0	0.5	0.15	6.6	10.75
8	Sacking of dried valerian roots	Farm 8A	1.3	0	0.15	25.0	26.45
		Farm 8B	3.5	7.5	1.2	2.5	7.2
		Farm 8C	5.4	14.7	0.7	3.4	9.5
		Farm 8D	4.5	5.3	0.5	135.0	140.0
	Median		3.5	0.7	0.15	3.4	10.75

* Sum of the concentrations of mesophilic bacteria, thermophilic actinomycetes and fungi.

[#] Farms with modern drying rooms, roots not threaded on wire loops.

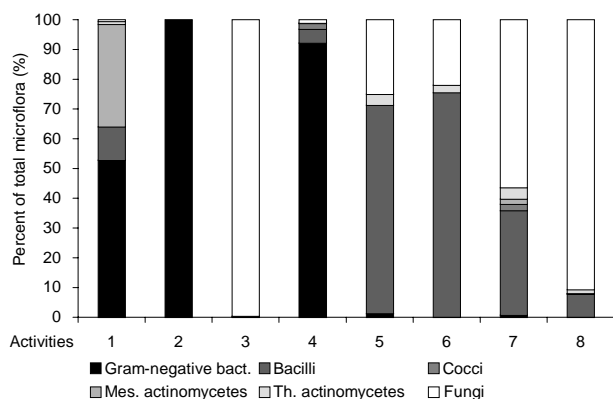


Figure 1. Composition of airborne microflora on farms during particular activities at processing valerian roots: total count, including mesophilic bacteria, thermophilic actinomycetes and fungi.

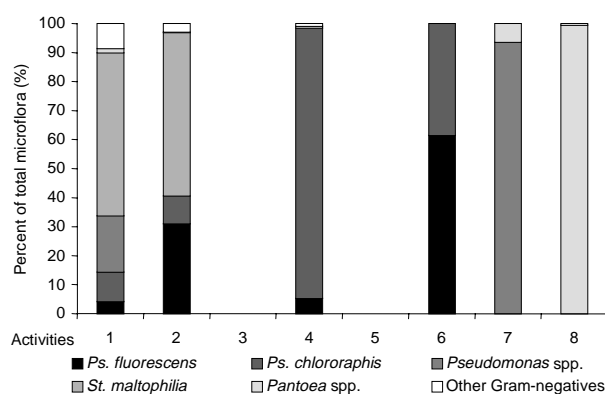


Figure 2. Composition of airborne Gram-negative bacteria on farms during particular activities at processing valerian roots.

numerous at later activities - during stripping off dried roots from wire loops and packing them into sacks (Fig. 1).

The concentrations of airborne Gram-negative bacteria recovered on EMB agar were large at the initial stages of valerian roots processing, up to the level 10^6 cfu/m³ (Tab. 1). Among them, there distinctly prevailed rods belonging to the family Pseudomonadaceae (Fig. 2), which formed 97.4% of the total Gram-negative strains isolated from the farm air during processing of valerian roots. The most common were isolates belonging to 3 species: *Stenotrophomonas maltophilia*, *Pseudomonas chlororaphis* and *Pseudomonas fluorescens*. Bacteria of the family Pseudomonadaceae were abundant in the farm air during the initial stages of the valerian roots processing, and their number sharply dropped after drying the roots. In the final stage of sacking dried roots, the concentration of Gram-negatives was relatively low (mostly 10^3 cfu/m³) and characterized by the prevalence of *Pantoea* spp., which constituted 99.4% of the total Gram-negative count (Fig. 2).

The concentrations of thermophilic actinomycetes were low throughout the whole cycle of valerian roots processing, ranging from $0-1.2 \times 10^3$ cfu/m³. *Thermo-*

actinomyces strains (*Th. vulgaris*, *Th. thalpoophilus*) distinctly prevailed at the majority of processing stages (Fig. 3).

Fungi recovered from the farm air during the valerian roots processing showed a marked variability, both in concentration and in species composition. The concentration of airborne fungi varied between $0.15-135.0 \times 10^3$ cfu/m³ (Tab. 1) and was the largest at threading of moist roots on wire loops (75.0 cfu/m³) and at sacking of dried roots (up to 135.0×10^3 cfu/m³). During threading of moist roots the *Penicillium* strains predominated, while during sacking of dried roots the *Aspergillus* strains prevailed (Fig. 4), among which the most common was the species *Aspergillus fumigatus* which constituted 57.5% of the total count.

Statistical analysis revealed that mesophilic bacteria showed a significant correlation with Gram-negative bacteria ($p < 0.01$) and endospore-forming bacilli ($p < 0.05$), while thermophilic actinomycetes showed a significant correlation with aspergilli ($p < 0.05$) and highly significant correlation with endospore-forming bacilli ($p < 0.001$).

In the air samples taken on the examined farms, 29 species or genera of bacteria and 19 species or genera of

Table 2. List of microbial species and genera identified in samples of farm air during processing of valerian roots.

Gram-negative bacteria: *Aeromonas* sp. (4B), *Pantoea agglomerans**+ (synonyms: *Erwinia herbicola*, *Enterobacter agglomerans*) (1A, 2, 4B, 7C, 8B, 8D), *Pantoea* spp. (2, 4B, 8D), *Pseudomonas chlororaphis* (1A, 2, 4B, 6), *Pseudomonas fluorescens**+ (1A, 2, 4B, 6), *Pseudomonas mesophilica* (1A), *Pseudomonas multivorans* (1B), *Pseudomonas vesicularis* (7B, 7C), *Pseudomonas* spp. (1A), *Serratia plymuthica* (2), *Sphingomonas paucimobilis* (1A, 2, 8C), *Stenotrophomonas maltophilia* (1A, 2).

Bacilli: *Bacillus badius* (1A-8D), *Bacillus cereus* (5, 7A), *Bacillus licheniformis**+ (8A), *Bacillus megaterium* (1A-8D), *Bacillus pumilus* (4A, 5, 7C, 8B, 8C), *Bacillus* spp. (1A-8D).

Gram-positive cocci: *Micrococcus* spp. (4A, 4B, 7A, 7B, 7C), *Staphylococcus* spp. (3, 8D).

Mesophilic actinomycetes: *Streptomyces albus** (1A, 7A, 7C), *Streptomyces* spp. (1A, 4A, 7A, 7B, 8B).

Thermophilic actinomycetes: *Saccharomonospora viridis** (5, 7B, 8D), *Saccharopolyspora rectivirgula** (7C, 8D), *Thermoactinomyces thalpoophilus** (1A, 4B, 5, 6, 8A, 8B, 8C, 8D), *Thermoactinomyces vulgaris** (1A, 4B, 5, 6, 7A, 7B, 8A, 8B, 8C, 8D), *Thermomonospora fusca** (3, 4B, 8B, 8C), *Thermomonospora chromogena** (8A, 8C, 8D), *Streptomyces* spp. (3, 7B).

Fungi: *Alternaria alternata**+ (1A, 4A, 6, 8B, 8C), *Aspergillus candidus**+ (8C), *Aspergillus fumigatus**+ (8A, 8B, 8C, 8D), *Aspergillus glaucus* (5, 7B, 7C, 8A, 8B, 8C, 8D), *Aspergillus niger**+ (4B, 8B), *Aspergillus terreus**+ (8C), *Candida* spp.* (1B, 7A, 7B, 8B, 8C, 8D), *Cylindrocarpon* spp. (2), *Fusarium* spp.+ (4B, 8D), *Humicola* spp. (7A), *Monilia* spp. (3, 4B, 8B), *Mucor mucedo** (2, 4B, 8D), *Mucor* spp.* (5, 7B, 8A, 8B, 8C), *Oidiodendron flavum* (4A), *Penicillium* spp. (1B, 3, 5, 6, 7B, 7C, 8B, 8C, 8D), *Rhizopus nigricans**+ (4B, 8B), *Rhodotorula rubra* (7C), *Trichoderma album* (7B), *Trichothecium roseum* (8C).

Sites of isolation are given in parentheses. The names of the species reported as having allergenic and/or immunotoxic properties (see text) are in bold and marked as follows: * allergenic species; + immunotoxic species.

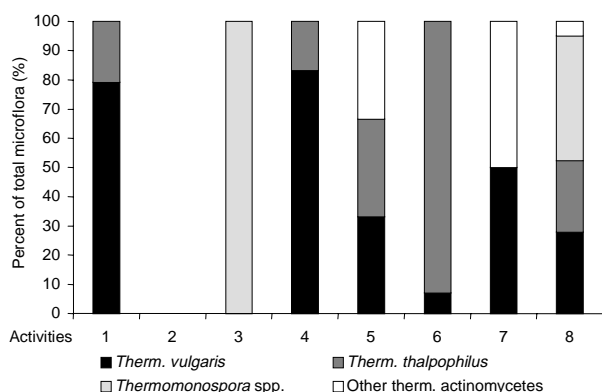


Figure 3. Composition of airborne thermophilic actinomycetes on farms during particular activities at processing valerian roots.

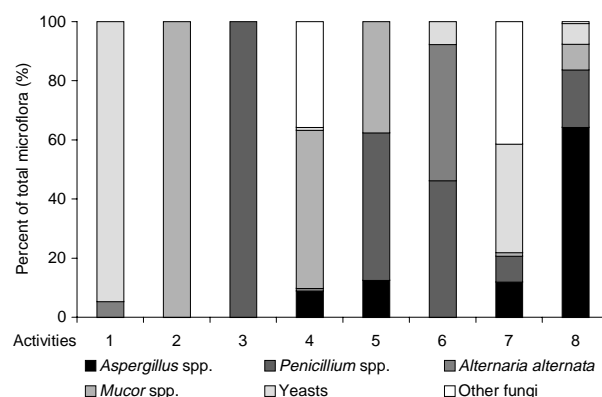


Figure 4. Composition of airborne fungi on farms during particular activities at processing valerian roots.

fungi were identified; of these, 10 and 12 species or genera respectively were reported as having allergenic and/or immunotoxic properties [2, 16, 24, 29, 35, 36, 56] (Tab. 2). These figures are certainly an underestimation, as a part of bacterial and fungal strains could be identified only to generic level.

The concentrations of airborne dust and endotoxin determined on the examined farms during valerian processing with the use of personal sampling were very large and ranged from 10.0-776.7 mg/m³, and from 0.15-24,448.2 µg/m³, respectively (medians 198.3 mg/m³ and 40.48 µg/m³) (Tab. 3). The concentrations of these factors determined in parallel with the use of stationary sampling were significantly lower ($p < 0.05$) and ranged from 1.5-158.9 mg/m³, and from 0.0045-981.0 µg/m³, respectively

(medians 33.4 mg/m³ and 33.43 µg/m³) (Tab. 3). The values of airborne dust and endotoxin showed a significant correlation when determined with the use of personal sampling ($p < 0.01$), but not when determined with the use of stationary sampling ($p > 0.05$).

DISCUSSION

The concentrations of bioaerosols recorded during processing of valerian roots showed a great variability, by 5-6 orders of magnitude. The median concentration of total airborne microorganisms (10.75×10^3 cfu/m³) approximated values found in sawmills [20] and at processing of hops [27] and potatoes [23] but was lower compared to those reported for working environments

Table 3. Concentrations of dust and bacterial endotoxin in farm air during processing valerian roots.

No. Activity	Sampling site	Personal sampling		Stationary sampling	
		Dust mg/m ³ *	Endotoxin µg/m ³	Dust mg/m ³ *	Endotoxin µg/m ³
1 Cleaning and trimming of dug valerian roots	Farm 1A	776.7 ± 621.1	6,387.09	30.6 ± 13.2	52.87
	Farm 1B	397.8 ± 214.1	92.07	40.5 ± 18.4	3.45
2 Shaking of moist roots before threading on wire	Farm 2	488.3 ± 38.6	876.67	17.3 ± 2.5	33.43
3 Threading of moist roots on wire loops	Farm 3	16.7 ± 2.7	1.04	1.5 ± 0.3	0.03
4 Shaking of loose roots in a drying room	Farm 4A [#]	10.0 ± 0	31.25	30.0 ± 30.8	504.03
	Farm 4B [#]	666.7 ± 276.7	16,404.08	112.1 ± 29.9	981.00
5 Unloading of loose roots from drying room	Farm 5 [#]	571.1 ± 110.1	24,448.20	118.4 ± 51.7	180.97
6 Taking out loops with threaded roots from drying room	Farm 6	342.2 ± 186.9	6.48	76.3 ± 44.9	34.12
7 Stripping off dried roots from wire loops	Farm 7A	19.2 ± 3.1	0.15	3.9 ± 0.8	0.0045
	Farm 7B	570.0 ± 169.9	40.48	81.3 ± 44.9	12.57
	Farm 7C	131.7 ± 19.3	137.16	104.0 ± 42.4	6.77
8 Sacking of dried valerian roots	Farm 8A	55.6 ± 18.1	23.15	17.2 ± 1.3	77.8
	Farm 8B	138.3 ± 23.2	37.59	33.4 ± 5.5	16.18
	Farm 8C	198.3 ± 40.9	19.37	32.4 ± 4.7	5.78
	Farm 8D	130.0 ± 29.4	1,015.63	158.9 ± 27.6	33.43
Median		198.3	40.48	33.40	33.43

*Mean ± S.D.; [#]Farms with modern drying rooms, roots not threaded on wire loops.

with the highest bioaerosol pollution, such as: grain stores, seed stores, animal feed factories, malt houses, herb processing plants, pig farms, poultry farms, flax farms and waste composting facilities [10, 14, 17, 18, 19, 21, 25, 33, 35, 41, 47, 59].

As, so far, there are no internationally recognised Occupational Exposure Limit (OEL) values for bio-aerosols, the results obtained in the present work could be compared only to the proposals raised by particular authors. As regards total viable airborne microorganisms, the OEL values proposed by Malmros *et al.* (10×10^3 cfu/m³) [43], and by Dutkiewicz & Jabłoński (100×10^3 cfu/m³) [16] were exceeded respectively on 8 and 4 of the 15 farms examined. The OEL values for airborne Gram-negative bacteria proposed by Clark [7] and Malmros *et al.* [43] (1×10^3 cfu/m³), and by Dutkiewicz & Jabłoński [16] and Górny & Dutkiewicz [28] (20×10^3 cfu/m³) were exceeded respectively on 6 and 3 of the 15 farms examined. The OEL value proposed by Dutkiewicz & Jabłoński [16] and Górny & Dutkiewicz [28] for airborne fungi (50×10^3 cfu/m³) was exceeded on 2 of the 15 examined farms, while nowhere was the OEL value proposed by these authors for airborne thermophilic actinomycetes (20×10^3 cfu/m³) exceeded.

The species composition of viable airborne microflora found on examined farms showed a distinct variability depending on the stage of valerian roots processing. From initial handling of freshly dug and washed roots until shaking of roots in a drying room, the airborne microflora was distinctly dominated by Gram-negative bacteria of the family Pseudomonadaceae. During a one-week drying they were killed by high temperature and replaced by thermo-resistant endospore-forming bacilli which became the prevalent constituent of the airborne microflora in the later stages of valerian roots processing. The most common organisms in the last stages of valerian roots processing were fungi, mainly *Aspergillus fumigatus* and other species of the genus *Aspergillus*. Gram-negative flora at these stages was less common and consisted mainly of *Pantoea* species. The most striking feature of the airborne microflora recovered at valerian processing was a total absence of corynebacteria that occur commonly in the air of working environments polluted with organic dusts, such as animal farms [14, 17], sawmills [20], herb processing plants [21], potato processing plant [23], and during handling of grain and hops [19, 27].

The exposure of farmers to hazardous microorganisms changed in the course of valerian root processing. In the initial stages the farmers could be exposed to large amounts of Gram-negative bacteria of the family Pseudomonadaceae. Though so far little is known about allergenic and immunotoxic properties of these bacteria, they have been identified as common constituents of oil mist in metallurgic industry facilities [9, 61] and implicated as causative agents of allergic alveolitis in exposed workers [2]. They should be also considered as a source of environmental endotoxin. In the initial stages of

valerian root processing, the farmers could be also exposed to large amounts of *Penicillium*, known as a common cause of allergic alveolitis [35, 36].

After drying of valerian roots, the main hazard to the farmers is posed by allergenic fungi of the genus *Aspergillus*, in particular *Aspergillus fumigatus* which may be a cause of asthma, allergic alveolitis and pulmonary aspergillosis [11, 31]. The potentially pathogenic role of the endospore-forming bacilli, commonly occurring at this stage, is poorly known. It has been documented that amylolytic and proteolytic enzymes produced by *Bacillus licheniformis* and *Bacillus subtilis* may evoke asthma in exposed workers of detergent producing facilities [3, 29]. The risk of allergic alveolitis at this stage is posed by thermophilic actinomycetes which occurred in relatively small numbers but showed a diverse species composition. The epiphytic species *Pantoea agglomerans* (synonyms: *Erwinia herbicola*, *Enterobacter agglomerans*), prevailing among Gram-negative bacteria at the final stage of sacking dried valerian roots was proved to possess strong endotoxic and allergenic properties [13, 15, 34, 44, 45, 46, 53, 57, 60]. The results obtained by Mackiewicz *et al.* [42] and Golec *et al.* [26] suggest the important role of this bacterium as an occupational allergen in herb dust.

The concentrations of dust and bacterial endotoxin in the farm air recorded during valerian processing by personal sampling were very large. The concentrations of dust were of the order 10^1 - 10^2 mg/m³, exceeding on all farms the Polish OEL value of 4 mg/m³ [52] by 2.5-194 times.

The concentrations of airborne endotoxin recorded during valerian roots processing showed a great variability in the range of 10^1 - 10^4 µg/m³. On all but one farms they exceeded the OEL values proposed by various authors [7, 12, 16, 28, 37, 43, 54] and the values supposed to cause decrease of lung function during work shift and ODS symptoms [55]. The median concentration of endotoxin in the air exceeded over 200 times the OEL values proposed by Clark [7] (0.1 µg/m³), Rylander [54] (0.1-0.2 µg/m³), Malmros *et al.* [43] (0.1 µg/m³), and by Górny & Dutkiewicz [28] (0.2 µg/m³), and over 1,500 times the OEL values proposed by Laitinen *et al.* [37] (0.025 µg/m³), and by Dutch Expert Committee on Occupational Standards (DECOS) [12] (0.005 µg/m³).

A drastic rise in the concentration of airborne endotoxin up to the extraordinary high levels of 16,404.08-24,448.20 µg/m³ have been observed after drying of valerian roots in modern drying rooms. This could be the result of elevated temperature, as it is known that heating might enhance the biological activity of the endotoxin by changing of its physical structure [51, 54]. A similar rise in the concentration of airborne endotoxin has been observed by our group at a study in a potato processing plant, during steaming of potatoes at the blanching process [23]. These data suggest the possibility of a particular respiratory risk that might arise when endotoxin containing organic materials are dried,

steamed, roasted or burnt in the course of various production or heating processes.

Nevertheless, possibility of unspecific, false-positive *Limulus* reactions must be also considered for the explanation of the extraordinary high levels of airborne endotoxin recorded during processing of valerian roots. It cannot be excluded that these results might be affected, at least in part, by an unspecific *Limulus* reaction with unknown constituents of valerian roots that had been changed by drying.

The concentrations of dust and endotoxin measured with the use of stationary sampling were also large but distinctly and significantly lower compared to those measured with the use of personal sampling. This suggests that personal sampling is a better method for assessment of occupational risk from exposure to organic dust than stationary sampling.

CONCLUSION

Farmers cultivating valerian could be exposed during processing of valerian roots to large concentrations of airborne microorganisms, dust and endotoxin posing a risk of work-related respiratory disease.

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