

Department of Occupational Biohazards, Institute of Rural Medicine, Lublin
Pulmonary Department, Medical University of Lublin

MARCIN GOLEC, CZESŁAWA SKÓRSKA, BARBARA MACKIEWICZ,
JACEK DUTKIEWICZ

*Health effects of exposure to thyme dust
in a group of thyme growing farmers*

Exposure to the inhalation of organic dust while performing of professional activities may cause respiratory disorders and other health problems in agricultural workers. It has been proved that workers employed in herb processing facilities, grain mills, stores, malhouses or tobacco industry, often showed work-related symptoms and toxic or allergic reactions to organic dust and its components (2, 4, 6). Also individual farmers, who have diverse and long-termed exposure to organic dust are at risk of work-related health disorders. For example, farmers exposed to grain dust, flax dust, and dust of animal origin also suffer from health disorders caused by these dusts (17, 18). The list of health disorders caused by organic dusts of various origin is long. They include disorders of upper and lower respiratory tract (occupational asthma, organic dust toxic syndrome (ODTS), mucous membrane irritation (MMI), rhinitis), skin disorders (including contact dermatitis and other dermal allergies) and ophthalmic problems (e.g. conjunctivitis). The diseases can have both allergic and/or immunotoxic background (4).

Exposure to organic dust of herbal origin may also be a cause of many health disorders. Herbs may exert adverse respiratory, dermal and general effects due to allergic and/or toxic properties. Until recently, there has been a little information about occupational diseases among herb processing workers and farmers. It was found that people growing and processing herbs could be exposed to large concentrations of airborne microorganisms, dust and endotoxin posing a risk of work-related respiratory diseases (1,8). Medical examinations of herb processing workers showed a high frequency of work-related symptoms and positive allergic reactions to herb extracts and microbial allergens associated with herb dust in skin and precipitin tests (2,19).

Not only workers of herb processing facilities are at risk of harmful effects of organic dust from herbs but also herb growing farmers. Mackiewicz et al. (12) described the case of allergic alveolitis in a thyme growing farmer, following exposure to thyme dust. Thyme can also cause dermal problems, including airborne contact dermatitis due to occupational exposure to thyme dust (8,12,19).

The aim of the present study was to examine effects of exposure to organic dust from thyme (*Thymus vulgaris* L.) among individual farmers cultivating this herb.

Thyme is a semi-woody, aromatic, shrubby herb with grey-green leaves. It belongs to the mint family (*Labiatae* or *Lamiatae*). Thyme is native to the western Mediterranean region and southern Italy. However, nowadays it is cultivated all over the world and has naturalized in some areas including central and eastern Europe and north-eastern United States. Thyme is

widely used as a culinary spice and a component of drugs and cosmetics. Thyme has a very important place among plant medicines because of its antiseptic, antispasmodic, expectorant, stimulant and astringent properties. The herb is used against digestive, respiratory and dermal disorders (16). Thyme is cultivated by many farmers in Poland. Consumption of herbs increases by 12%–15% per year, and so it could be expected that the number of people having professional contact with thyme will be each year greater.

Farmers thresh the dried plant and then supply the herb to the facilities. Threshing of thyme takes place indoors and is associated with a great exposure to thyme dust. So far only little is known about the effects of occupational exposure to thyme dust and the purpose of this study was to increase the knowledge of this problem.

MATERIAL AND METHODS

Examined population. A group of 47 thyme growing farmers (24 females + 23 males) aged 41.46 ± 12.25 years of age (mean \pm S.D.) were examined. They worked on their individual farms located in the Lublin region (eastern Poland). The farmers were examined when reloading, cleaning and threshing thyme. As a reference group, 63 students and office workers not exposed to organic dust were examined.

Medical examinations. Farmers were interviewed with the American Thoracic Society (ATS) standard questionnaire, and with the questionnaire developed in the Institute of Agricultural Medicine in Lublin for the study of work-related symptoms caused by organic dusts (3). Next, they were subjected to routine physical examination and to lung function examinations with the LUNGTEST 500 spirometer produced by MES (Kraków, Poland). The spirometric values of the forced expiratory volume in one second (FEV_1), FEV_1/VC , vital capacity (VC) and peak expiratory flow (PEF) were determined.

Agar-gel precipitation tests. These tests were performed with antigens of microorganisms which occur commonly in the agricultural environment in the Lublin region and entire eastern Poland. All allergens (lyophilised extracts of bacterial and fungal mass) were prepared for the tests in the Department of Occupational Biohazards in our Institute according to a unified procedure as described earlier (2). The following twelve allergens were used: *Acinetobacter calcoaceticus*, *Alcaligenes faecalis*, *Arthrobacter globiformis*, *Bacillus subtilis*, *Pantoea agglomerans* (syn. *Erwinia herbicola*, *Enterobacter agglomerans*), *Saccharopolyspora rectivirgula* (syn. *Micropolyspora faeni*, *Faenia rectivirgula*), *Streptomyces albus*, *Thermoactinomyces vulgaris*, *Alternaria alternata*, *Aspergillus candidus*, *Aspergillus fumigatus*, *Penicillium citrinum*. The agar-gel precipitation test was performed by Ouchterlony double diffusion method in purified 1.5 % Difco agar with farmers, sera and the antigens of above mentioned bacteria and fungi dissolved in saline at a concentration of 30 mg/ml.

Skin tests. The tests were carried out by prick technique with four allergens: *Pantoea agglomerans*, *Saccharopolyspora rectivirgula*, *Streptomyces albus* and *Aspergillus fumigatus*. The antigens were dissolved in phosphate buffered saline (PBS, Biomed, Kraków), at a concentration of 5 mg/ml, sterilized by filtering and checked for sterility and lack of toxicity. The test was performed on the forearm with the antigenic extracts and PBS as a control. The tested sites were observed after 20 min. The wheal and/or erythema reactions of 3 mm or more in diameter were regarded as positive.

Statistical analysis. The obtained results were analyzed by the Student's t-test and by Pearson's test for correlation. All tests were done with the use of the Statistica™ ver. 4.5 package (Statsoft, USA).

RESULTS

OCCURRENCE OF WORK-RELATED SYMPTOMS IN THYME HANDLING FARMERS

Thirty out of 47 interviewed farmers (63.8%) reported work-related symptoms while handling thyme (Tab. 1). The most common complaint was blocking of a nose due to swelling of nose's mucous membrane reported by 18 farmers (60%), followed by dry cough reported by 14 farmers (46%), general weakness by 13 farmers (43.3%), chest tightness, dyspnoea, headache, eyes itching—each symptom reported by 12 farmers (40%), fatigue by 11 persons (36.8%), hoarseness by 10 persons (33.3%), shivering by nine farmers (30.0%), fever and body itching each by seven persons (23.3%), rash by six farmers (20.0%). Two farmers (6.7%) reported nausea and chest rattling each.

Among 30 farmers having work-related symptoms, three persons reported only one symptom, one person – two symptoms, five farmers – three symptoms, three – four symptoms, six – five symptoms, four – six, five – seven symptoms, one person – eight and two – twelve symptoms. Seventeen interviewed persons did not report any work-related symptoms.

Table 1. Prevalence of work-related symptoms in farmers engaged in reloading, cleaning and threshing thyme (N=47)

Work-related symptoms	Farmers reporting symptoms	
	number	percent
Dry cough	14	29.8
Productive cough	4	8.51
Dyspnoea	12	25.5
Chest tightness	12	25.5
Blocking of the nose	18	38.3
Chest rattling	2	4.25
Hoarseness	10	21.3
Fever	7	14.9
Shivering	9	19.2
Nausea	2	4.25
Vomiting	0	0
Headache	12	25.5
General weakness	13	27.7
Sweating	0	0
Joint and muscle aching	0	0
Body aching	0	0
Fatigue	11	23.4
Body aching	7	14.9
Rash	6	12.8
Eyes itching	12	25.5
Total symptomatic farmers	30	63.8

Table 2. Mean spirometric values in farmers engaged in threshing thyme and in control subjects
Examined group of farmers (N = 47)

Spirometric values	Occurrence of work-related symptoms	Mean differences of spirometric values before and after work \pm SD	Difference between subgroups
VC	Yes (N = 30)	7.3 \pm 25.3	p>0.1 (not significant)
	No (N = 17)	11.4 \pm 21.2	p>0.1 (not significant)
FEV ₁	Yes (N = 30)	18.2 \pm 31.8	p>0.1 (not significant)
	No (N = 17)	24.1 \pm 46.6	p>0.1 (not significant)
FEV ₁ %VC	Yes (N = 30)	8.1 \pm 15.2	p>0.1 (not significant)
	No (N = 17)	14.7 \pm 28.4	p>0.1 (not significant)
PEF	Yes (N = 30)	10.9 \pm 24.8	p>0.1 (not significant)
	No (N = 17)	13.6 \pm 15.4	p>0.1 (not significant)

Reference group (N = 63)

Spirometric values	Mean differences \pm SD	Difference compared to normal values
VC	6.21 \pm 19.60	p>0.1 Not significant
FEV ₁	-1.02 \pm 34.01	p>0.1 Not significant
FEV ₁ %VC	-11.16 \pm 20.66	p>0.1 Not significant
PEF	8.55 \pm 31.44	p>0.1 Not significant

None of the members of the reference group reported the occurrence of work-related symptoms.

Lung function changes. The mean baseline spirometric values in the farmers' group did not show significant differences compared to the normal values (Tab. 2). The same situation was noted in the reference group. Considerable differences between the measured and normal spirometric values were found only in four farmers – which is not a statistically significant difference. No significant differences were found between the spirometric values in farmers reporting work-related symptoms and those who did not report such symptoms (Tab. 2). A slight before and after work decrease of the mean values of FEV₁/VC, FEV₁ and PEF were noted in a group of farmers – however, the above differences were not significant and remained within a normal range. No differences between the spirometric values before and after work were found in the reference group.

Oxymetric changes. All oximetric changes were within a normal range – blood saturation with oxygen in both groups remained constant before and after the work.

ALLERGIC REACTIONS

Skin reactions. The frequency of positive skin response to four microbial allergens associated with organic dust was within a range of 4.3–14.9% (Tab. 3). Seven farmers (14.9%) showed immediate positive skin reactions to *Pantoea agglomerans*. Four persons had positive skin reactions to *Saccharopolyspora rectivirgula*. Two out of 47 examined farmers (4.3%) each showed positive skin reactions to *Aspergillus fumigatus* and to *Streptomyces albus*. The frequency of positive skin response to the same allergens was within a of 0–1.6% in the reference group and was significantly lower in cases of *Pantoea agglomerans* and *Saccharopolyspora rectivirgula* compared to the group of thyme growing farmers.

Table 3. Frequency of positive skin reactions in thyme farmers and reference group

Allergens	<i>Saccharopolyspora Rectivirgula</i>	<i>Pantoea agglomerans</i>	<i>Aspergillus fumigatus</i>	<i>Streptomyces albus</i>	Control (PBS)
Farmers group (N = 47)	8.5%*	14.9%**	4.3%	4.3%	0%
Reference group (N = 63)	0%	1.6%	1.6%	1.6%	0%

Significantly greater compared to reference group; * $p < 0.05$, ** $p < 0.01$

Table 4. Frequency of positive precipitin reactions in thyme farmers and referents (%)

Antigens	Thyme farmers (N = 46)	Reference group (N = 63)
<i>Saccharopolyspora rectivirgula</i>	2.2	0
<i>Thermoactinomyces vulgaris</i>	2.2	0
<i>Streptomyces albus</i>	4.3	1.6
<i>Arthrobacter globiformis</i>	2.1	0
<i>Pantoea agglomerans</i>	56.5**	20.6
<i>Acinetobacter calcoaceticus</i>	8,7	3.2
<i>Aspergillus fumigatus</i>	28.3*	9.5
<i>Penicillium citrinum</i>	4.3	0
<i>Aspergillus candidus</i>	0	0
<i>Alternaria alternata</i>	2.2	0
<i>Bacillus subtilis</i>	2.2	0
<i>Alcaligenes faecalis</i>	28.3*	9.5

Significantly greater compared to reference group; * $p < 0.05$; ** $p < 0.01$

Precipitin reactions. The frequency of positive precipitin reactions to twelve microbial allergens associated with thyme dust in eastern Poland was within a range of 2.2–56.5% in the farmers group and 0–11.1% in the reference group (Tab. 4). The frequency of positive reactions to the above mentioned twelve allergens was in almost all cases higher in the farmers' group, except for *Aspergillus candidus*, which did not show any positive reaction either in the farmers

or reference groups. The highest percentage of positive reactions among thyme farmers was noted in the case of the extract *Pantoea agglomerans* – 56.5%. It was much greater compared to the reference group and the difference was highly significant ($p < 0.001$). The incidence of positive precipitin reactions with the antigens of *Aspergillus fumigatus* and *Alcaligenes faecalis* was also very high among farmers (28.3%) and occurred significantly more frequently than in the reference group ($p < 0.05$). Positive precipitin reactions to *Saccharopolyspora rectivirgula*, *Thermoactinomyces vulgaris*, *Streptomyces albus*, *Arthrobacter globiformis*, *Acinetobacter calcoaceticus*, *Penicillium citrinum*, *Alternaria alternata* and *Bacillus subtilis* were less frequent in the farmers group compared to three above mentioned allergens, being within a range of 2.2–8.7%, but were also higher than in the reference group.

DISCUSSION

The obtained results indicate that farmers exposed to large concentrations of thyme dust and associated microorganisms during threshing thyme are under increased risk of work-related pulmonary and skin disorders such as allergic alveolitis, occupational asthma, chronic bronchitis, Organic Dust Toxic Syndrome and skin allergies like contact dermatitis. This presumption is supported by high frequency of work-related symptoms and results of skin and precipitin tests.

The frequency of work-related symptoms among farmers exposed to thyme dust was high (63.8%), distinctly higher compared to farmers exposed to grain dust examined in an earlier study who reported 44.7% of work-related symptoms (18). The frequency of work-related symptoms while thyme threshing was even slightly higher than among people professionally exposed to flax dust (62.7%) (17). The frequency of ODTS – like respiratory symptoms (dry cough – 46.7%, dyspnoea – 40.0%) was similar to general symptoms (general weakness – 36.7%, headache – 40.0).

The structure of work-related symptoms in thyme growing farmers, showing similar incidence of respiratory and general symptoms, was different compared to other professionals exposed to organic dust. Among flax handlers, the most common were general symptoms, and among grain handlers typical ODTS - like respiratory symptoms prevailed. The difference could be caused by a high frequency of work-related symptoms in the thyme farmers group.

To summarize, the results shows that work-related symptoms observed in this study among a population of thyme farmers correspond to the Organic Dust Toxic Syndrome (ODTS), which probably is the initial stage of allergic alveolitis. It should also be mentioned that the examined population was rather young (mean age was 41.46 years), while the severe occupational disease due to exposure to organic dust may appear yet after a long job duration (about 30 years). The observed occurrence of work-related symptoms should be considered as a significant health problem in the examined occupational group.

The high frequency of work-related symptoms among thyme farmers was not accompanied by changes in lung function. The spirometric values were within a normal range and did not show a significant post work decline. Similar results were obtained in the earlier studies in the populations of flax farmers (17) and grain farmers (20). Oxymetric values also did not show any decline after work. These parameters decreased after work among flax handling farmers (although even in this case the oximetric values were within a normal range) 17.

The occupational risk in the examined group of thyme farmers was confirmed by the common occurrence of positive skin prick tests results. Frequency of positive response to all microbial allergens tested (*Saccharopolyspora rectivirgula*, *Pantoea agglomerans*, *Aspergillus fumigatus*, *Streptomyces albus*) was significantly higher than in the reference group. The greatest frequency of positive response was noted to the extract of *Pantoea agglomerans*

(14.9%) and *Saccharopolyspora rectivirgula* (8.5%). It is noteworthy that *Streptomyces albus* and *Pantoea agglomerans* were reported as causative agents of allergic alveolitis (7,13).

Frequency of positive skin reactions to microbial allergens in the examined population of thyme farmers was lower compared to positive skin prick tests response to allergens associated with herb dust among workers of herb processing facilities (2).

In contrast, the frequency of positive precipitin reactions to twelve microbial allergens was much greater in thyme farmers group than among workers of pharmaceutical industry exposed to dust from herbs (2). In the examined group of thyme farmers, reactions to almost all microbial antigens occurred significantly more frequently than in the reference group. The only exception was response to *Aspergillus candidus* characterized by lack of positive reactions in both groups.

It appears that bacterium *Pantoea agglomerans* is the most important hazard to persons exposed to thyme dust. Frequencies of positive skin tests reactions (14.9%) and precipitin reactions (56.5%) were very high and significantly greater compared to reference group (skin tests – $p < 0.01$, precipitin tests – $p < 0.001$). Allergic reactions to the extract of this Gram-negative bacterium were more common than to other examined microbial antigens. *Pantoea agglomerans* is the source of strong allergens and endotoxin and may be a cause of allergic alveolitis among persons exposed to organic dust in Poland (5, 6, 10, 13–15).

The second important biohazard for thyme farmers seems to be *Aspergillus fumigatus*. This filamentous fungus is causative factor of various respiratory diseases: allergic alveolitis, aspergillosis, bronchial asthma and ODS (11). Frequencies of allergic reactions to this fungus were very high in the examined group of thyme farmers – 28.3% positive reactions in precipitin test and 4.3% in skin prick test, being in both tests significantly higher than in the reference group.

A high frequency of positive precipitin reactions with the antigen of *Alcaligenes faecalis* confirmed results of the earlier studies of our group, in which large quantities of *Alcaligenes faecalis* were found in the air of herb processing plants (1), which was associated with high frequency of positive skin and precipitin reactions to this bacterium herb processing workers (2), and positive response in inhalation challenge to *Alcaligenes faecalis* allergen in 52.2% of the workers reporting work-related symptoms (9). The frequencies of positive skin and precipitin tests to allergen of *Saccharopolyspora rectivirgula* were significantly higher among thyme farmers compared to the reference group. This thermophilic actinomycete was noted as a causative agent of allergic alveolitis among farmers (11). *Arthrobacter globiformis*, also described as a cause of allergic alveolitis in eastern Poland (13), gave 2.1% positive reactions in the precipitin test.

CONCLUSION

Farmers engaged in cultivating and threshing of thyme (*Thymus vulgaris*) represent a group at elevated professional risk because of the high incidence of work-related symptoms and high frequency of allergic reactions to bacterial and fungal antigens associated with organic dust from thyme.

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SUMMARY

Medical examinations were performed in a group of 47 thyme farmers from eastern Poland while threshing thyme (*Thymus vulgaris*). As a reference group 63 urban dwellers were examined not exposed to organic dusts. The examinations included an interview concerning the occurrence of respiratory disorders and work-related symptoms, physical examination, lung function tests, skin prick tests with four microbial allergens and agar-gel precipitation tests with 12 antigens. 63.8% of thyme handling farmers reported occurrence of work-related symptoms while threshing thyme. The most common complaints were: blocking of the nose (reported by 18 farmers – 38.3%), dry cough (14 persons – 29.8%) and general weakness (13 persons – 27.7%). The mean spirometric values in the farmers group were within a normal range and did

not show a significant post-work decline. The farmers showed positive skin reactions to microbial antigens in the range of 4.3–14.9%, and a frequency of positive precipitin reactions 0–56.5%. The reference group responded to most allergens with a significantly lower frequency of positive results compared to the examined group. In conclusion, thyme farmers engaged in threshing this herb represent a group of elevated professional risk because of high incidence of work-related symptoms and common occurrence of positive skin and precipitin reactions to bacterial and fungal allergens associated with organic dust.

Ekspozycja na pył organiczny pochodzący z uprawy tymianku a występowanie objawów alergii u rolników uprawiających tymianek

Badaniami objęto grupę 47 rolników uprawiających tymianek (*Thymus vulgaris*) oraz, jako grupę kontrolną, 63 mieszkańców miasta niemających kontaktu z pyłami organicznymi. Przeprowadzono wywiad ze szczególnym uwzględnieniem schorzeń układu oddechowego i objawów chorobowych związanych z pracą, badanie fizykalne i czynnościowe płuc, testy skórne z czterema antygenami i testy precypitacji w żelu z 12 antygenami drobnoustrojowymi związanymi z pyłem organicznym. U 63,7% rolników stwierdzono w wywiadzie występowanie objawów związanych z pracą. Najczęściej zgłaszanymi objawami były: zaczopowanie nosa (18 osób – 38,3%), suchy kaszel (14 osób – 29,8%) oraz ogólne złe samopoczucie (13 osób – 27,7%). Średnie wartości spirometryczne pozostawały w przedziale uznawanym za normę. Odsetki dodatnich wczesnych reakcji skórnych z alergenami środowiskowymi występowały w grupie plantatorów tymianku z częstością 4,3–14,9%, dodatnie odczyny precypitacji w żelu zawierały się w przedziale 0–56,5%. Częstość występowania dodatnich reakcji alergicznych była w przypadku większości alergenów znamienne wyższa w grupie badanej niż w grupie kontrolnej. Uzyskane wyniki badań pokazują, że rolnicy uprawiający tymianek są grupą zawodową w wysokim stopniu narażoną na alergeny wziewne i choroby zawodowe układu oddechowego.