American Journal of Industrial Medicine 21:177–191 (1992)

Immunological and Respiratory Changes in Animal Food Processing Workers

Eugenija Zuskin, MD, DSc, Bozica Kanceljak, MD, DSc, E. Neil Schachter, MD, Theodore J. Witek, Ph.D, Saul Maayani, PhD, Satindra Goswami, PhD, Zvi Marom, MD, and Nicholas Rienzi, MS

A group of 35 men employed in the processing of animal food was studied to assess the relation between respiratory findings and immunological status. The most frequent positive skin prick reactions to occupational allergens were to fish flour (82.9%), followed by carotene (77.1%), corn (65.7%), four-leaf clover (62.9%), sunflower (54.3%), chicken meat (31.4%), soy (28.6%), and yeast (22.7%). Increased total IgE serum levels were found in 14/35 (40.0%) animal food workers compared to 1/39 (2.6%) in a healthy population ($p < 0.01$). A significantly higher prevalence of chronic respiratory symptoms was found among the exposed workers compared to control workers. There was however, no significant difference in the prevalence of chronic respiratory symptoms between animal food workers with positive and negative skin tests to house dust or to fish flour or among those with increased or normal IgE (except for dyspnea). The frequency of acute symptoms associated with the work shift was high among the animal food workers but similar by immunological status. There were significant mean across-shift reductions for all ventilatory capacity tests, being particularly pronounced for FEF_{25}. Workers with positive skin tests to fish flour antigen had significantly larger across-shift reductions in FEF_{25} than workers with negative skin reactions. An aqueous extract of animal food dust caused a dose-related contractile response of isolated guinea pig tracheal muscle in vitro. Our data suggest that, in addition to any immunological response animal food dust may produce in vivo, it probably also causes direct irritant or pharmacological reactions on the airways as suggested by our in vitro data.

Key words: animal food dust, immunology, respiratory symptoms, ventilatory capacity, occupational allergy, skin prick tests, animal food processing workers

INTRODUCTION

Few studies have examined different food components as potential agents associated with respiratory difficulties [Brooks, 1985]. Those food-related products which have been suspected of causing respiratory difficulties include coriander [Suhonen et al., 1979], cinnamon [Uragoda, 1984], red paprika [Uragoda, 1966], spices

Andrija Stampar School of Public Health, Zagreb, Yugoslavia (E.Z.).
Institute for Medical Research and Occupational Medicine, Zagreb, Yugoslavia (B.K.).
Mount Sinai School of Medicine, New York, NY (E.N.S., T.J.W., S.M., S.G., Z.M., N.R.).
Address reprint requests to Dr. E. Neil Schachter, Mount Sinai School of Medicine, One Gustave L. Levy Place, P.O. Box 1232, New York, NY 10029-6574.
Accepted for publication July 9, 1991.

[Toorenenbergen and Dieges, 1985], celery and milk products [Nava et al., 1983], and watermelon (Enberg et al., 1988). Recently, Birnbaum et al. [1989] reported allergy to sunflower honey associated with allergy to celery. Work-related symptoms and positive skin (or radioallergosorbent test [RAST]) tests against buckwheat were reported by Gohte et al. [1983]. Knusel and Wutrich [1983] described an allergic respiratory disease induced by contact with pet-fish food. Dietschi and Wutrich [1987] further described a case of allergic bronchial asthma caused by polyvalent sensitization to various components in fish food. Raszeja-Kotelba et al. [1976] described a large number of workers employed in a fish flour factory with occupational findings including inflammatory reactions of the skin and occupational allergy. The health risks in feed-mixing plants have been described by Vyskocil et al. [1972]. The authors found a high prevalence of chronic bronchitis (43%) in men and 18% in women. Recently, Vadivieso et al. [1988] suggested the importance of cereal flour in animal formula as a cause of occupational asthma in farm animal breeders. A previous epidemiological study of workers occupationally exposed to dust in the processing of animal food by Zuskin et al. [1989] demonstrated acute and chronic changes in ventilatory capacity and the development of chronic as well as acute respiratory symptoms.

No epidemiological study of the immunological status of animal food workers has been reported. The current study represents an extension of our previous epidemiological study of respiratory function in workers employed in the animal food processing industry [Zuskin et al., 1989]. In the present study we investigated the immunological reactions to different food components and the relationship between respiratory dysfunction and immunological status in workers occupationally exposed to animal food dust.

SUBJECTS AND METHODS

Immunological Study

Occupational exposure. Animal feed for pigs and chickens was prepared with different components including soy, sunflower seeds, fish flour, corn, different wheats, four-leaf clover, carotene, vitamins (A, D, K, B12, C), iodized salt, and minerals (Fe, Cu, Se, Ca). Each separate component was individually prepared for incorporation into the food mix. Our workers were exposed to food aerosols during the grinding, weighing, mixing, and packaging of these different food components. Workers worked in two shifts and rotated through all work areas. Our study only examined workers during the first shift.

Subjects. A group of 35 men employed in the animal food processing industry was studied. They were volunteers from the 82 workers in an initial clinical study [Zuskin et al., 1989]. There was no difference in baseline lung function between those workers who participated in this study and those who did not. The mean age of the workers was 40 years (range: 26–58 years), the mean height was 173 cm (range: 160–189 cm) with a mean duration of employment in the industry of 14 years (range: 3–30 years). Most of the workers (89%) were regular smokers, smoking on the average 20 cigarettes daily (range: 15–40). A group of 30 men employed as clerical office workers in the industry were studied as a control group for respiratory symptoms. These two cohorts were similar in age, smoking habits, and duration of employment. The control workers were not exposed to dusts or fumes.

Immunological studies. Food processing workers were skin tested with aqueous allergens of different food components and standard allergens using skin prick testing. We did not perform control skin testing on non-exposed workers. Occupational allergens were prepared from the material collected in the workrooms and included dust of carotene, sunflower, corn, chicken meat, soya, four-leaf clover, fish flour, and yeast. Allergens were prepared by the method of Sheldon et al. [1967]. Skin prick tests with these allergens were performed using a dilution of 1:10 w/v. Workers were also tested with standard extracts of house dust, molds, *Dermatophagoides pteronyssinus,* histamine base (0.1 mg/ml), and buffer as a control solution. Commercial mold antigens were prepared as a mixture of *Alternaria, Penicillium mucor, Cladosporium, Aspergillus niger,* and *Aspergillus fumigatus* as 0.2% w/v solution [Sheldon et al; 1967]. The skin reactions were read after 20 minutes. A skin prick test was considered positive if the diameter of the observed wheal was larger than 3 mm (corrected for the control reaction of buffer). We illustrate our results with findings for skin tests to fish flour (the most commonly positive food allergen); similar findings were seen with other animal food components. Similarly we illustrate our results for common allergens using data for extracts of house dust.

Serum levels of total IgE antibody were measured by Paper radioimmunosorbent test (PRIST), using a direct radioimmunological sandwich technique based on paper discs as a solid phase [Wide and Porath, 1966]. Levels of IgE below 125 kU/L were considered normal [Johansson, 1968]. A group of 39 healthy male and female volunteers (not occupationally exposed to food dusts) were simultaneously studied for IgE levels.

Respiratory symptoms. Chronic respiratory symptoms were recorded by using a modification of the Medical Research Council Committee questionnaire [1960] with additional questions on occupational asthma [WHO, 1986]. The following definitions were used:

1. chronic cough/phlegm: cough or phlegm production or both on most days for at least 3 months in the year;
2. chronic bronchitis: cough and phlegm for a minimum of 3 months in the year and for not less than 2 successive years;
3. dyspnea grade: 3, shortness of breath when walking with other people on level ground; grade 4, shortness of breath when walking at one's own pace on level ground;
4. occupational asthma: chest tightness, cough, wheezing, shortness of breath, and acute decrease in ventilatory capacity at or following work.

The workers were asked additional questions about acute symptoms which developed while at work, such as cough, dyspnea, irritation or dryness of the throat, eye irritation, bleeding, secretion or dryness of the nose, and headache.

Ventilatory capacity studies. Ventilatory capacity was measured by recording maximum expiratory flow-volume (MEFV) curves on which forced vital capacity (FVC), one-second forced expiratory volume (FEV_1), and flow rates at 50% and 25% of the vital capacity (FEF_{50}, FEF_{25}) were determined. The MEFV curves were recorded using a Pneumoscreen spirometer (Jaeger, Germany). The acute effect of exposure to animal food dust on ventilatory capacity was studied by recording MEFV curves on Monday, before and after a work shift (day shift). The measured pre-shift values were compared with the expected normal values of Quanjer [1983]. The spirometer was calibrated for volume on a daily basis. Two physicians familiar with environmental and epidemiological surveys administered the questionnaire and lung function testing throughout the survey.

Environmental dust measurements. Airborne dust was sampled during an 8 hour work shift in the work place of the examined workers. Personal sampler (Casella-London Millipore Field Monitors) with Millipore AA membrane filters were used to estimate total dust exposure. Only selected workers wore personal samplers so that it was not possible to correlate physiological measurements with dust levels. In addition, two stage stationary samplers, consisting of a membrane filter preceded by a horizontal elutriator, were used to collect the respirable fraction in the work areas.

Statistical analysis. The results of ventilatory capacity measurements were analyzed using the Student's t-test for paired differences (acute effect) and unpaired (chronic effect) differences. The chi-square test or, where appropriate, Fisher's exact test was used to test differences in the prevalence of respiratory symptoms. The value of $p < 0.05$ was considered as significant.

Animal Food Extract Assay

In order to investigate the potential of animal food dust to cause airway smooth muscle contraction, we tested the bronchoconstricting potential of animal food extract on isolated guinea pig tracheal rings in vitro. Extract was made by placing one gram of animal food dust in 10 ml of sterile water for 24 hours at a temperature of +4°C. The solution was filtered through a sterile gauze sponge and centrifuged at 16,000 rpm for 20 minutes. The supernatant was decanted and used as the extract for the experiment.

We used the trachea of young Albino Hartley male guinea pigs (300–390 g) purchased from Charles River Labs, Wilmington, MA. Animals were sacrificed by CO_2 asphyxiation for 2 minutes and the trachea was removed within 3 minutes. The animal tissues were manually trimmed of excess fat and connective tissues. Four segments (''rings'' each 4–6 mm wide) were cut from a single trachea, and each was suspended between two L-shaped stainless steel hooks mounted in a 20 ml organ chamber containing Krebs-Hanseliet buffer of the following composition (μM): NaCl, 110.0; KCl, 4.80; $CaCL_2$, 2.35; $MgSO_4$, 1.20; $KHPO_4$ 1.20; $NaHCO_3$, 25.0; dextrose, 110; and Na2EDTA, 0.03, in glass distilled water. Organ chambers were maintained at 36.5 ± 0.5°C, and were continuously aerated with 95% O_2 and 5% CO_2 to maintain pH = 7.5 ± 0.1. The tissue segments were initially set to 2 g of tension, and were allowed to relax for approximately 1.5 hours before the experiment began. During that period the tissue was washed at 15 minute intervals. After the relaxation period, the tension in each tissue segment was adjusted to 2 g for all subsequent assays. Isometric contractions were recorded using a Grass FT03C force displacement transducer attached to a Grass polygraph recorder. Before the contraction-response assay with food dust extract was performed, a challenge with carbachol 10^{-5} M was run. A dose-response curve with food dust extract was obtained by adding progressively increasing volumes of extract or Krebs (used as a control) into the tissue bath in progressive aliquots of 10, 30, 100, 300, and 1,000 μl. The potency of the extract was determined by comparing the biological activity with the maximal contraction induced by carbachol (10^{-5} M) on the same tissue. The data were expressed as a percentage of the initial carbachol contraction (10^{-5} M).

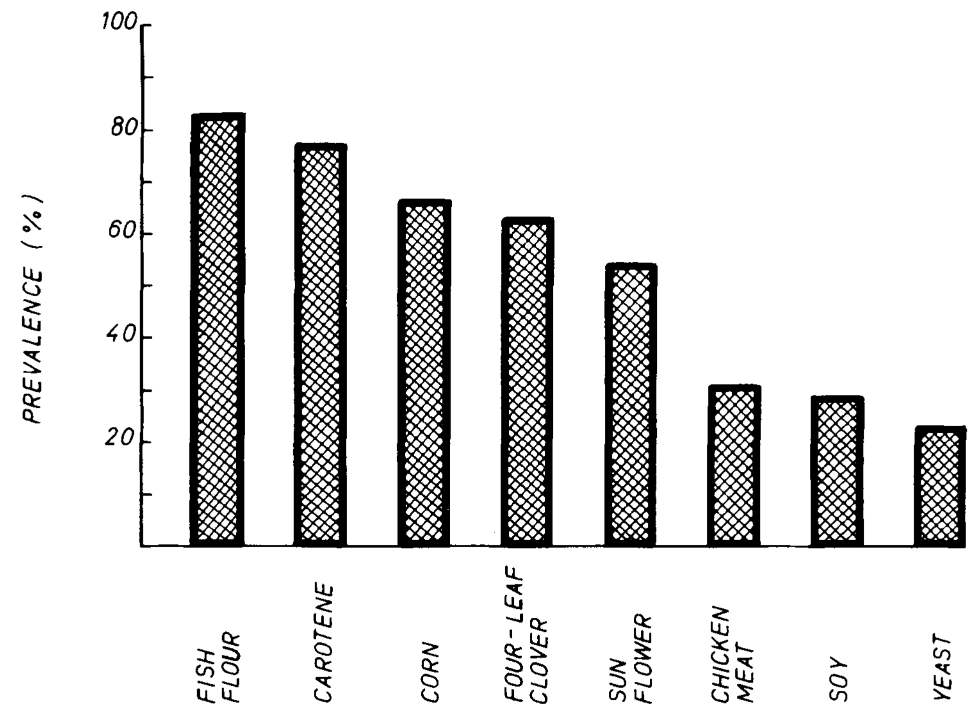

Fig. 1. Prevalence of positive skin prick tests to occupational allergens in animal food workers.

The protein content in animal food dust extract was determined by the Lowry method [Lowry et al., 1951]. The endotoxin content was assayed in animal food extract by Sigma, E-TOXATE concentrate (Limulus Amebocyte Lysate) kit No. 210-A for detection and semiquantitation of endotoxin.

RESULTS

Immunological Study

All tested workers had typical skin reactions to histamine and none reacted to the buffer control solution. Thirty-one percent of the subjects reacted to extracts of *D. pteronyssinus*, 28.6% to house dust, and 5.7% to molds. Immediate skin reactions to different animal food components are illustrated in Figure 1. The highest prevalences of positive immediate skin reaction were found for fish flour (82.9%), followed by carotene (77.1%), corn dust (65.7%), four-leaf clover (62.9%), sunflower (54.3%), chicken meat (31.4%), soy (28.6%), and yeast (22.7%).

Among the tested workers, 14 (40.0%) had increased serum levels of IgE. Of the 39 healthy volunteers 1 (2.6%) had a level of IgE greater than 125 kU/L ($p < 0.01$). All of the animal food workers with elevated IgE had positive skin prick reaction to fish flour, carotene, and four-leaf clover. Two workers with asthma had positive skin reactions to fish flour and four-leaf clover dust. One worker also reacted to soy, and one worker to sunflower. Both workers with asthma had increased IgE serum levels (278 kU/L and 288 kU/L).

Respiratory symptoms. As previously described [Zuskin et al., 1989], the

TABLE I. Prevalence of Chronic Respiratory Symptoms in Animal Food Workers and in Control Workers

Group	Mean age (years)	Mean exposure (years)	Chronic cough (%)	Chronic phlegm (%)	Chronic bronchitis (%)	Dyspnea (%)	Asthma (%)	Chest tightness (%)	Rhinitis (%)
Animal food workers (N = 35)	40	14	19 (54.3)	18 (51.4)	15 (42.9)	11 (31.4)	2 (5.7)	17 (48.6)	9 (25.7)
			< 0.05	<0.04	NS[a]	<0.03	NS	<0.001	NS
Control workers (N = 30)	36	11	8 (26.7)	7 (23.3)	7 (23.3)	2 (6.7)	0 (0)	2 (6.7)	2 (6.7)

[a]NS, difference statistically not significant ($p > 0.05$).

prevalence of chronic respiratory symptoms was higher in animal food workers than in control subjects, being significantly different for chronic cough ($p < 0.05$), chronic phlegm ($p < 0.04$), dyspnea ($p < 0.03$), and chest tightness ($p < 0.001$) (Table I).

There were no significant differences in the prevalence of chronic respiratory symptoms between workers with and without positive skin reactions to fish flour ($p < 0.05$). Table II presents the prevalence of chronic respiratory symptoms separately in animal food workers by IgE and skin test (to house dust) status. No consistent differences were seen between workers with normal and elevated IgE or between workers with positive and negative skin tests to house dust. As previously noted the prevalence of symptoms for this group (and the above subgroup) was significantly higher than that of the control population. Data on symptoms (Tables I, II) are not adjusted for smoking since most of these workers were smokers.

There was a high prevalence of acute symptoms during the work shift ranging from 48–55% for cough, dyspnea, eye irritation, irritation and dryness of the throat and dryness of the nose, and 32% for headache. The prevalence for secretion or bleeding of the nose was 3/35 (9%) and 4/35 (11%), respectively. Workers with normal and increased IgE serum level had a similar prevalence of acute symptoms. There was no significant difference in symptom prevalence between workers with positive and negative skin tests.

Ventilatory capacity studies. As shown in our previous study of animal food workers [Zuskin et al., 1989] significant differences exist in baseline lung function and in across-shift changes when compared to controls. There was no significant difference in the magnitude of acute across-shift reductions between animal food workers with positive and negative skin tests to specific animal food (fish flour) antigen except for FEF_{25}, which was significantly higher in workers with positive (−13.4%) than in those with negative (−9.1%) skin tests to fish flour ($p < 0.001$) (Table III).

Comparison of baseline ventilatory capacity by positive and negative skin tests to non-specific (house dust) antigen (Table IV) demonstrated no consistent pattern in across-shift changes in lung function.

Table V displays the ventilatory capacity data by worker serum IgE levels. There was no difference in across-shift reductions between those workers with normal and increased IgE.

TABLE II. Prevalence of Chronic Respiratory Symptoms in Animal Food Workers With Increased and Normal IgE and Positive and Negative Skin Prick Test to House Dust

Group	N	Chronic cough	Chronic phlegm (%)	Chronic bronchitis (%)	Asthma (%)	Dyspnea grade 3 or 4 (%)	Chest tightness (%)	Rhinitis (%)
Increased IgE	14	7 (50.0) NS[a]	7 (50.0) NS	5 (35.7) NS	2 (14.3) NS	5 (35.7) NS	6 (42.9) NS	4 (28.6) NS
Normal IgE	21	12 (57.1)	11 (52.3)	10 (47.6)	0 (0)	6 (28.6)	11 (52.4)	5 (23.8)
Positive skin test	10	5 (50.0) NS	5 (50.0) NS	5 (50.0) NS	2 (20.0) NS	2 (20.0) NS	5 (50.0) NS	3 (30.0) NS
Negative skin test	25	14 (56.0)	13 (52.0)	10 (40.0)	0 (0)	9 (36.0)	12 (48.0)	6 (24.0)

[a]NS, difference statistically not significant ($p > 0.05$).

Finally we examined lung function effects by duration of employment (Table VI). No consistent differences in across-shift changes in lung function were noted.

Environmental dust measurement. Total dust concentration ranged from 0.77 mg/m^3–10.62 mg/m^3, with the respirable fraction ranging from 0.34 mg/m^3 to 2.94 mg/m^3. This total dust concentration was not in general higher than that allowed by the current Yugoslav standard (10 mg/m^3).

Animal Food Extract Assay

The mean values for the contractile effect of animal food extract on guinea pig tracheal smooth muscle are shown in Figure 2. A total of 24 guinea pigs were studied and the contractile response to 10, 30, 100, 300, 1,000 μl of standardized (by protein content) animal food dust extract was tested. This figure demonstrates a dose-related contractile effect of the water soluble animal food extract. The results are presented as a percentage of an initial maximal carbachol contraction produced by stimulation with 10^{-5} M carbachol. Dose-response parameters were calculated using a curve fitting from the software of PROPHET (Boston, MA).

Aqueous extracts increased the tone of tracheal rings inducing a dose-related smooth muscle contraction. The mean smallest contraction (10 μl) was 27% and the mean greatest contraction (1,000 μl) was 69% of maximal carbachol contraction. A control Krebs solution added in the same volume into the organ bath did not cause changes in the tone of the guinea pig tracheal rings. Extrapolated response parameters for dose-response curves of animal food extract gave the following data: Emax (% maximum carbachol) = 126.6%; EC50 (μl) = 594.2 μl.

Determination of protein content in animal food dust extract demonstrated that the extract used for the in vitro study on isolated guinea pig tracheal rings contained 5.15 mg/ml of protein. The endotoxin assay did not demonstrate the presence of endotoxin in the animal food extract used for the in vitro study on isolated guinea pig tracheal rings.

DISCUSSION

The preparation of animal food creates a respirable aerosol with multiple food components which can result in immunological changes in many factory workers.

TABLE III. Ventilatory Capacity in 35 Animal Food Workers in Relation to Skin Prick Test to Fish Flour Allergen*

	FVC			FEV_1			FEF_{50}			FEF_{25}		
	Before shift	Difference before-after shift		Before shift	Difference before-after shift		Before shift	Difference before-after shift		Before shift	Difference before-after shift	
Group	L	%	p	L	%	p	L/s	%	p	L/s	%	p
Positive skin test (N = 29)	4.33 ± 0.83 <0.001 5.35 ± 0.71[a]	−4.9	<0.001	3.42 ± 0.63 <0.001 3.99 ± 0.65[a]	−6.1	<0.001	4.76 ± 1.18 <0.001 5.66 ± 0.41[a]	−9.5	<0.001	2.39 ± 0.69 <0.026 2.74 ± 0.5[a]	−13.4	<0.001
Negative skin test (N = 6)	4.71 ± 0.86 NS[b] 5.81 ± 1.16[a]	−4.9	<0.06	3.41 ± 0.52 <0.043 4.44 ± 0.94[a]	−5.9	NS	4.30 ± 1.59 <0.053 5.85 ± 0.45[a]	−9.3	<0.007	1.98 ± 0.73 <0.028 2.88 ± 0.44[a]	−9.1	<0.05

*The data are presented as mean ± SD.
[a]Predicted normal values.
[b]NS, difference statistically not significant ($p > 0.05$).

TABLE IV. Ventilatory Capacity in 35 Animal Food Workers in Relation to Skin Prick Test to House Dust*

	FVC			FEV_1			FEF_{50}			FEF_{25}		
	Before shift	Difference before-after shift		Before shift	Difference before-after shift		Before shift	Difference before-after shift		Before shift	Difference before-after shift	
Group	L	%	p	L	%	p	L/s	%	p	L/s	%	p
Positive skin test (N = 10)	4.02 ± 0.69 <0.001 5.10 ± 0.55[b]	−3.9	<0.022	3.23 ± 0.67 NS[a] 3.69 ± 0.61[b]	−8.1	<0.042	4.54 ± 1.27 <0.02 5.63 ± 0.42[b]	−13.0	<0.016	2.26 ± 0.69 NS 2.74 ± 0.47[b]	−9.3	<0.028
Negative skin test (N = 25)	4.55 ± 0.85 <0.001 5.56 ± 0.86[b]	−5.1	<0.001	3.49 ± 0.57 <0.001 4.22 ± 0.71[b]	−5.4	<0.001	4.74 ± 1.25 <0.001 5.72 ± 0.43[b]	−8.0	<0.001	2.35 ± 0.72 0.014 2.78 ± 0.43[b]	−14.0	<0.001

*The data are presented as mean ± SD.
[a]NS, difference statistically not significant ($p > 0.05$).
[b]Predicted normal values.

TABLE V. Ventilatory Capacity in 35 Animal Food Workers in Relation to Serum IgE Level*

	FVC			FEV_1			FEF_{50}			FEF_{25}		
	Before shift	Difference before-after shift		Before shift	Difference before-after shift		Before shift	Difference before-after shift		Before shift	Difference before-after shift	
Group	L	%	p	L	%	p	L/s	%	p	L/s	%	p
Increased IgE (N = 14)	4.41 ± 0.84 <0.006 5.31 ± 0.78[a]	−5.4	<0.006	3.31 ± 0.58 <0.034 3.86 ± 0.71[a]	−6.9	<0.013	4.24 ± 1.24 <0.001 5.60 ± 0.37[a]	−8.3	<0.004	2.01 ± 0.67 <0.003 2.68 ± 0.38[a]	−10.9	<0.010
Normal IgE level (N = 21)	4.39 ± 0.85 <0.001 5.51 ± 0.83[a]	−4.6	<0.003	3.49 ± 0.62 <0.001 4.21 ± 0.70[a]	−5.4	<0.001	4.98 ± 1.18 <0.008 5.75 ± 0.45[a]	−10.0	<0.001	2.53 ± 0.65 NS[b] 2.82 ± 0.47[a]	−13.4	<0.001

*The data are presented as mean ± SD.
[a]Predicted normal values.
[b]NS, difference statistically not significant ($p > .05$).

TABLE VI. Ventilatory Capacity in 35 Animal Food Workers in Relation to Duration of Exposure*

Exposure	FVC			FEV_1			FEF_{50}			FEF_{25}		
	Before shift	Difference before-after shift		Before shift	Difference before-after shift		Before shift	Difference before-after shift		Before shift	Difference before-after shift	
(years)	L	%	p	L	%	p	L/s	%	p	L/s	%	p
< 10	4.86 ± 0.62	−6.8	<0.008	3.72 ± 0.43	−7.8	<0.001	4.77 ± 1.13	−11.7	<0.006	2.31 ± 0.61	−9.5	<0.03
(N = 11)	<0.029			<0.015			<0.003			<0.001		
	5.53 ± 0.71[a]			4.31 ± 0.60[a]			5.99 ± 0.28[a]			3.14 ± 0.22[a]		
> 10	4.18 ± 0.84	−3.8	<0.002	3.28 ± 0.63	−5.2	<0.003	4.64 ± 1.31	−8.4	<0.001	2.33 ± 0.75	−14.2	<0.001
(N = 24)	<0.001			<0.001			<0.002			NS[b]		
	5.39 ± 0.86[a]			3.96 ± 0.74[a]			5.55 ± 0.40[a]			2.60 ± 0.41[a]		

*The data are presented as mean ± SD.
[a]Predicted normal values.
[b]NS, difference statistically not significant ($p > 0.05$).

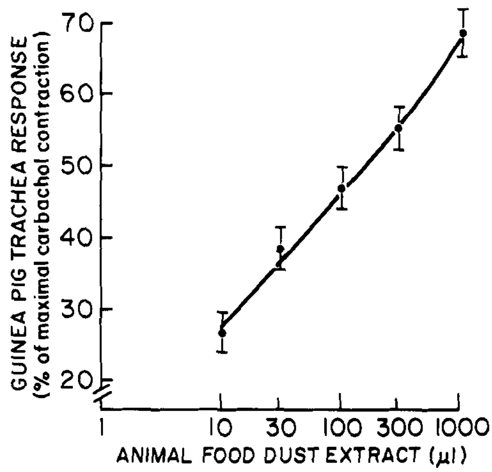

Fig. 2. Contractile activity of aqueous animal food extract on isolated guinea pig tracheal smooth muscle as percentage of initial carbachol 10^{-5} M contraction (mean ± SE).

Our study of animal food workers demonstrates that a majority reacted to some components of the preparation. The largest number of workers, 29 (82.9%) reacted to fish flour, followed by carotene, 27 (77.1%), corn, 23 (65.7%), four-leaf clover, 21 (62.9%), sun-flower, 19 (54.3%), chicken meat, 11 (31.4%), soy , 10 (28.6%), and yeast, 8 (22.7%). In a previous study of immunological findings in spice factory workers, we found positive intradermal skin tests with mixed spice dust allergen in a high percentage of exposed workers (73.3%) and in 33.3% of control workers. Increased total IgE serum levels were found in 36.8% of exposed and in 9.7% of the control workers [Zuskin et al., 1988b]. By comparison, coffee and tea workers demonstrated a lower prevalence of positive skin reactions to green coffee dust allergen (40%) and to some tea dust allergens (range: 10–45%) [Zuskin et al., 1981; 1985]. The large number of workers with positive skin tests in the present study may indicate, at least in part, a non-specific reaction. Beck and Nissen [1983] described that 65% of non-exposed individuals had a positive patch test to one or more species of fish. In our previous study of spice factory workers, 73.3% demonstrated positive skin reaction to mixed spice dust [Zuskin et al., 1988b]. In that study there was no consistent correlation between skin reactivity and chronic respiratory symptoms. However, acute symptoms in spice factory workers during the work shift were more prevalent in workers with positive than in those with negative skin tests to mixed spices. In the current study, because we did not have control (unexposed) workers tested with animal food allergens, it is not possible to definitively characterize these reactions as non-specific or allergic. However, the very high prevalence of positive skin tests suggests a non-specific reaction.

Fish protein has previously been described as a cause of disease "Fishmeal worker's lung" [Hogg, 1988]. Orford and Wilson [1985] studied 10 workers with respiratory illness associated with the processing of frozen king crab. They found FEV_1, FVC, and mid-expiratory flow rate (MEFR) decreased during the day in four crab processors, and precipitins were evident in the sera of nine workers. The same authors suggest that the inhalation of aerosolized crab antigen(s) may cause respiratory symptoms and can cause immunological effects similar to those caused by other occupational respiratory allergens. In our study we did not find a significant difference in the prevalence of chronic respiratory symptoms between workers with positive and negative skin prick tests to fish flour antigen. However, workers complained of more intense acute symptoms when they were mixing fish flour into the food, particularly cough, dyspnea, and irritation of the throat. Since our workers were mostly exposed to an aerosol composed of a mixture of foods, it was not possible to study the effect of fish flour alone.

Forest et al. [1983] found a high prevalence of occupational asthma (14.7%) in crab processing workers and concluded that atopy was not a predisposing factor. A high prevalence of occupational asthma (15.6%), rhino-conjuctivitis (76.1%), and urticaria (34.8%) was described in snow crab processing workers by Cartier et al. [1984]. This is a higher prevalence of occupational asthma than we found in our animal food workers (5.7%). Droszcz et al. [1981] found 23% of the workers studied in a fish meal factory had possible fish allergy. However, only two of them had elevated serum IgE levels. In their study, chronic bronchitis was diagnosed in 54.7% of the studied workers and slight obstruction was found in 33% of workers. A high prevalence (40%) of wheezing and other respiratory symptoms was reported in the prawn processing factory by Gaddie et al. [1980]. Skin prick tests to prawn extract gave positive reaction in 39% of affected workers and 39% had increased prawn-specific IgE.

Our results confirm the data of our previous study of animal food workers, indicating that occupational exposure to animal food aerosols can be responsible for the development of acute and chronic respiratory symptoms and lung function reductions [Zuskin et al., 1989]. Because of the presence of many varieties of organic material in this work place we suspected a possible allergic component in this disease. However, in our current study, there was no association between chronic respiratory symptoms and increased serum IgE levels and positive skin tests to fish flour or to house dust.

A large number of workers in the animal food processing industry complained of acute symptoms associated with the work shift. These prevalences were similar to those obtained in coffee and tea workers [Zuskin et al., 1979; Zuskin and Skuric, 1984], and in workers processing soy bean and spices [Zuskin et al., 1988a,c]. Such acute symptoms unrelated to immunological indices may suggest a non-specific irritant nature of the animal food dust.

Mean across-shift reductions of ventilatory capacity in animal food workers were statistically significant and similar to those seen in previous study of animal food workers [Zuskin et al., 1989]. The largest across-shift reductions were observed for FEF_{25} followed by FEF_{50}, FEV_1, and FVC. In general, no consistent relationship was found between the severity of across-shift changes in lung function and immunological markers (skin tests, serum IgE). Finally, there was no relationship between decreased baseline pulmonary function and immunological indices. This suggests that any chronic impairment is not related to these markers of allergy.

Our study on isolated guinea pig trachea suggests that clinical respiratory findings obtained in humans can be simulated in guinea pig tracheal smooth muscle in vitro. Previous experience suggests that guinea pig airways are pharmacologically similar to human airway muscle [Brink et al., 1980]. The guinea pigs used in our experiments were not pre-sensitized to animal food. We suggest therefore that extract of animal food can induce airway constriction in vitro by a direct action on guinea pig airway smooth muscle. This contractile activity is concentration dependent. The response may represent a direct irritant effect or possibly some pharmacologically related phenomenon. These data are similar to the results obtained with green coffee dust and spices on isolated guinea pig tracheal rings [Zuskin et al., 1983; 1988b].

The production of animal food is a complex process including many nutritional components as well as the addition of antibiotics and hormones. In addition, these foods are frequently contaminated with microorganisms [Vyskocil et al., 1972]. Our findings suggest that immunological reactions are frequent in animal food workers, but are not generaly correlated with respiratory findings. While the mechanisms by which the organic aerosols in this industry cause airway damage remain to be better defined, our studies suggest that effects not mediated by classic immunological reactions may be involved.

ACKNOWLEDGMENTS

Supported in part by grant YF 733 from the National Institutes of Health, Bethesda, MD, grant RO1-OHO 2593-O1A1 from the National Institute of Occupational Safety and Health, Centers for Disease Control, Atlanta, GA, and by the Henry and Catherine Gaisman Fund, New York, NY.

REFERENCES

Beck HI, Nissen BK (1983): Type-I reactions to commercial fish in non-exposed individuals. Contact Dermatitis 9:219–223.

Birnbaum J, Tafforeau M, Vervloet D, Charpin J, Charpin D (1989): Allergy to sunflower honey associated with allergy to celery. Clin Exp Allergy 19:229–230.

Brink C, Dunkan PG, Midzenski M, Douglas JS (1980): Responses and sensitivity of female guinea pig respiratory tissues to agonists during ontogenesis. J Pharmacol Exp Ther 215:426–433.

Brooks SM (1985): Occupational Asthma. In: Weiss EB, Segal MS, Stein M (eds): "Bronchial Asthma." 2nd ed. Boston: Little Brown and Co., pp 461–493.

Cartier A, Malo JL, Forest F, Lafrance M, Pineau L, St-Aubin JJ, DuBois JY (1984): Occupational asthma in snow crab-processing workers. J Allergy Clin Immunol 74:261–269.

Dietschi R, Wutrich B (1987): "Aquarium" allergy bronchial asthma caused by polivalent sensitization to various components in fish food. Hautarzt 38:160–161.

Droszcz W, Kowalski J, Piotrowska B, Pawlowicz A, Pietruszcwska E (1981): Allergy to fish mean factory workers. Int Arch Occup Environ Health 49:13–19.

Enberg R, McCullough J, Ownby DR (1988): Antibody responses in watermelon sensitivity. J Allergy Clin Immunol 82:795–800.

Forest F, Cartier A, Malo JL, Lafrance M, Pineau L, DuBois JY, St-Aubin JJ (1983): Epidemiology of occupational asthma in crab-processing workers of Iles de la Madeleine, Canada. Am Rev Respir Dis 127:155.

Gaddie J, Legge J, Friend JAR, Reid TMS (1980): Pulmonary hypersensitivity in prawn workers. Lancet 2:1350–1353.

Gohte CJ, Wieslander G, Ancker K, Forsbeck M (1983): Buckwheat allergy: health food, and inhalation risk. Allergy 38:155–159.

Hogg JC (1988): Extrinsic allergic alveolitis and occupational asthma in Churg A, Green FH (eds): "Pathology of occupational lung disease." New York: IGAKU-SHOIN, pp. 331–349.

Johansson SGO (1968): Serum IgND levels in healthy children and adults. Int Arch Allergy 34:1–8.

Knusel EL, Wutrich B (1983): "Aquarium allergy": fish food, another domestic allergen. Schweiz Med Wochenschr 113:658–662.

Lowry OH, Rosenbrough NJ, Farr AL, Randall RL (1951): Protein measurements with the folin phenol reagent. J Biol Chem 193:265–275.
Medical Research Council Committee on the Aetiology of Chronic Bronchitis (1960): Standardised questionnaire on respiratory symptoms. Br Med J 2:1665.
Nava C, Brambilla G, Briatico-Vangosa M, Marchisio M, Talamo F (1983): Occupational asthma in food workers: study of three cases. Med Lav 74:302–307.
Orford RR, Wilson JT (1985): Epidemiologic studies in processing of the king crab. Am J Ind Med 7:155–169.
Quanjer PH (1983): Standardized lung function testing. Report of Working Party on "Standardization of Lung Function Tests" of the European Community for Coal and Steel. Bull Eur Physiopathol Respir 19 (suppl 5):1–95.
Raszeja-Kotelba B, Buschke M, Jablonska H, Skuza M, Slomko M, Wanic M, Hryniewiecka I (1976): Skin lesions in workers employed in the manufacture of fish flour. Przegl Der 63:435–439.
Sheldon JM, Lowel RG, Mathews KP (1967): "A Manual of Clinical Allergy." Philadelphia: Saunders, pp 507–531.
Suhonen R, Keskinen H, Bjorksten F, Vaheri E, Zitting A (1979): Allergy to coriander: a case report. Allergy 34:327–330.
Toorenenbergen AW, Dieges PH (1985): Immunoglobulin E antibodies against coriander and other species. J Allergy Clin Immunol 76:477–481.
Uragoda C (1966): Symptoms among chilli grinders. Br J Ind Med 24:162–164.
Uragoda C (1984): Asthma and other symptoms in cinnamon workers. Br J Ind Med 41:224–227.
Vadivieso R, Pola J, Zapata C, Puyana J, Cuesta J, Martin C, Losada E (1988): Farm animal feeders: another group affected by cereal flour asthma. Allergy 43:406–410.
Vyskocil J, Chroma M, Berka I, Lejhanoova G, Pospisil I (1972): The health risks in feed-mixing plants. Int Arch Arbeitsmed 29:291–311.
Wide L, Porath J (1966): Radioimmunoassay of proteins with the use of Sephadex-coupled antibodies. Biochim Biophys Acta 130:257–260.
World Health Organization (1986): "Detection of Occupational Disease." Geneva, pp 311–318.
Zuskin E, Duncan PG, Douglas JS (1983): Pharmacological characterisation of extracts of coffee dust. Br J Ind Med 40:193–198.
Zuskin E, Kanceljak B, Pokrajac D, Schachter EN, Witek TJ (1988a): Respiratory symptoms and ventilatory capacity in soy bean workers. Am J Ind Med 14:157–165.
Zuskin E, Kanceljak B, Skuric Z, Ivankovic D (1985): Immunological and respiratory changes in tea workers. Int Arch Occup Environ Health 56:57–65.
Zuskin E, Kanceljak B, Skuric Z, Pokrajac D, Schachter EN, Witek TJ, Maayani S (1988b): Immunological and respiratory findings in spice-factory workers. Environ Res 47:95–108.
Zuskin E, Mataija M, Pokrajac D, Schachter EN, Witek TJ (1989): Respiratory function in animal food processing workers. Am J Ind Med 16:179–187.
Zuskin E, Skuric Z (1984): Respiratory function in tea workers. Br J Ind Med 41:88–93.
Zuskin E, Skuric Z, Kanceljak B, Pokrajac D, Schachter EN, Witek TJ (1988c): Respiratory findings in spice factory workers. Arch Environ Health 43:335–339.
Zuskin E, Valic F, Kanceljak B (1981): Immunological and respiratory changes in coffee workers. Thorax 36:9–13.
Zuskin E, Valic F, Skuric Z (1979): Respiratory function in coffee workers. Br J Ind Med 36:117–122.