

Ocular irritation from product of pesticide degradation among workers in a seed warehouse

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Abstract: Four workers at a seed supply warehouse in Chiba Prefecture, Japan, complained of ocular irritation on the job. Pesticide-coated seeds were stored in the warehouse but no significant amount of pesticide was detected in the air inside the warehouse. To identify the cause of the ocular irritation and to determine an appropriate solution to the problem, the authors used thermal desorption gas chromatography-mass spectrometry to analyze the profiles of volatile organic compounds (VOCs) in the air of the two warehouses at the site—warehouse A, where the four workers experienced ocular irritation, and warehouse B, where no workers experienced ocular irritation. Comparing the profiles of VOCs in these warehouses indicated that *n*-butyl isocyanate, a hydrolyzed product of the fungicide benomyl, was the cause of the workers' ocular irritation. *n*-Butyl isocyanate is known to be a contact irritant and if the benomyl-coated seeds were not properly dried before storage in the warehouse *n*-butyl isocyanate would have been produced. The results of the study suggest that more attention should be paid both to the pesticide itself and to the products of pesticide degradation. In this study, *n*-butyl isocyanate was identified as a product of pesticide degradation and a causative chemical affecting occupational health.

Key words : *n*-Butyl isocyanate, Benomyl, Occupational exposure, Ocular irritation, Pesticide exposure

Introduction

In 2010, four men employed at a seed supply company in Chiba Prefecture, Japan, came to an occupational health physician of the company complaining of ocular irritation while working in a seed warehouse (warehouse A). To identify the cause of the problem and to take appropriate measures to resolve it, the company asked the authors to assess the air inside the warehouse, giving particular attention to the environmental chemicals stored there.

In the summer of 2009, the seed supplier built a new plant with two warehouses (A and B) for seed storage.

Soon after the warehouses became operational in the spring of 2010, workers at warehouse A began to experience ocular irritation without any other symptoms such as respiratory irritation. Interviews with the workers revealed that ocular irritation was experienced not only by the four men who went to see the occupational health physician but also by other workers at warehouse A. In contrast, none of the workers at warehouse B reported experiencing ocular irritation. The capacities of the two warehouses were different: the volume of warehouse A was 250 m³ while the volume of warehouse B was 550 m³. Neither warehouse had a ventilation system but, to protect the stored seeds, both warehouses had air conditioning that maintained an air temperature of 20 °C and a humidity level of 40%.

Initially, it was thought that the workers' symptoms were caused by formaldehyde, a chemical commonly released by new building materials. However, formaldehyde

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levels in the warehouses were low (less than 0.1 ppm), indicating that some other chemical substance must be causing the ocular irritation. Pesticides were then suspected as the cause of the ocular irritation because research has shown that workers involved in treating seeds with pesticides are at high risk for pesticide exposure¹. The seed supplier reported using a mixture of polymers and five pesticides as seed coating, to improve the stability of the seeds during storage. The five pesticides were benomyl, captan, iprodione, mepronil, and thiuram. Only thiuram was a known ocular irritant. None of these five pesticides was detected either in the air or the suspended particles, which were collected by high efficiency particulate air filters, inside the warehouses. Therefore, we focused our attention on the products of pesticide degradation in the workplace. In some cases, the products of hydrolysis, oxidation, biodegradation, and photolysis of pesticides can be as toxic or more toxic than the pesticides themselves².

Materials and Methods

The profiles of airborne volatile organic compounds (VOCs) were analyzed in warehouses A and B on 16 March, 2011. Pumped sampling was performed using thermal desorption cartridges [Air Toxics (200 mg, 60–80 mesh); Spelco, Inc., Bellefonte, PA, USA]. Each thermal desorption cartridge collected VOCs in 6.0 l of air. All pumped sampling was carried out with diaphragm pump equipped with an integrating flow meter (GSP-250FT; Gastec Corp., Kanagawa, Japan) at a flow rate of 0.2 l/min. The collected VOCs were determined by thermal desorption gas chromatography-mass spectrometry (TD-GC-MS). TD was performed with an automatic thermal-desorption unit (ATD 400, Perkin-Elmer, Norwalk, CT, USA) connected by a transfer line (maintained at 290 °C) to a gas chromatograph (Auto System XL, Perkin-Elmer, Norwalk, CT, USA) with a quadrupole mass spectrometer (Turbo Mass Gold, Perkin-Elmer, Norwalk, CT, USA). The TD conditions were as follows: desorb flow of 30 ml/min during 10 min at 320 °C, trap temperature maintained at 5 °C by Peltier effect and rapid heating to 320 °C at 40 °C/s, outlet split of 10 ml/min and temperature of 225 °C for the valve. The cooling was achieved using a stream of helium. All samples were injected with a 5:1 split ratio, with respect to the injection from the trap to the GC column. A capillary column (60 m × 0.32 mm ID) with a film thickness of 1.8 μm (Inertcap-624, GL Science, Tokyo, Japan) was connected directly to the ion source and the carrier gas was helium at a flow rate of 1 ml/min.

The GC temperature program was as follows: 40 °C (held for 1 min) then increased at 5 °C/min to 140 °C and finally increased at 10 °C/min to 240 °C (held for 5 min). The mass spectrometer was operated in the electron ionization mode with voltage of 70 eV and current of 200 μA. Full scan mass spectra, over a mass-to-charge (m/z) range $35 \leq m/z \leq 400$, were acquired at a rate of 2 scan/s. The compound identification was based on the National Institute of Standards and Technology (NIST) mass spectra database, according to the match of m/z between the fragmentation pattern of the “peak” of interest and that of the standard database. *n*-butyl isocyanate was purchased from Wako Pure Chemical Industries Ltd (Osaka, Japan).

Results

Figure 1 presents the total ion chromatograms of GC-MS for air samples collected in warehouse A and warehouse B. For comparison, both of these chromatograms have been placed in the same figure with the chromatogram from warehouse A being vertically offset from that of chromatogram B. The scale of the vertical axis in both of these chromatograms is the same. The chromatograms show 19 major peaks and numerous minor ones. Comparing the chromatogram profiles of the two warehouses, it can be seen that six peaks that occurred in the warehouse B chromatogram did not occur in the warehouse A chromatogram. However, one major peak in the warehouse A chromatogram was barely visible in the warehouse B chromatogram. The retention time of this peak was 16.8 min and the height was 13.7 times that of the same peak in the warehouse B chromatogram. The signal-to-noise ratio of the peak from warehouse B was greater than 10, which made this peak large enough to be distinguished from the baseline noise. The compound was identified as *n*-butyl isocyanate by comparing the mass spectral fragmentation pattern of the compound with the NIST mass spectra database (Fig. 2). The retention time of the compound also matched that of *n*-butyl isocyanate standard.

Identification of *n*-butyl isocyanate as the cause of the workers' ocular irritation pointed to the need to reduce the amount of *n*-butyl isocyanate in the warehouse air. The source of the *n*-butyl isocyanate was thought to be the pesticide benomyl [methyl 1-(butylcarbamoil)-2-benzimidazolecarbamate] which, coated on seeds, was used as a fungicide by the seed supplier. Benomyl is known to hydrolyze rapidly to *n*-butyl isocyanate and carbendazim in aqueous solutions (Fig. 3)³. The seed supplier used an aqueous solution of pesticides and polymers to prepare

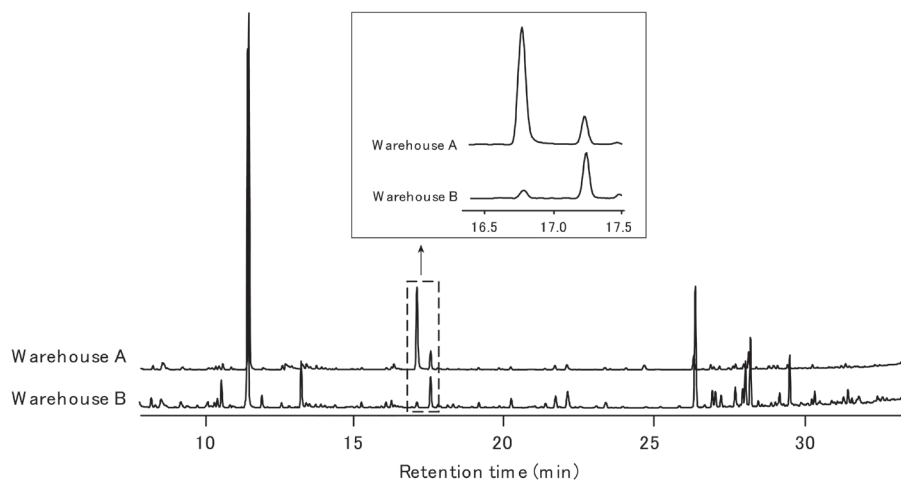


Fig. 1. Comparison of total ion chromatograms of VOCs by TD-GC-MS for air samples collected inside warehouse A and warehouse B. The vertical axis is proportional to the total ion signal intensity.

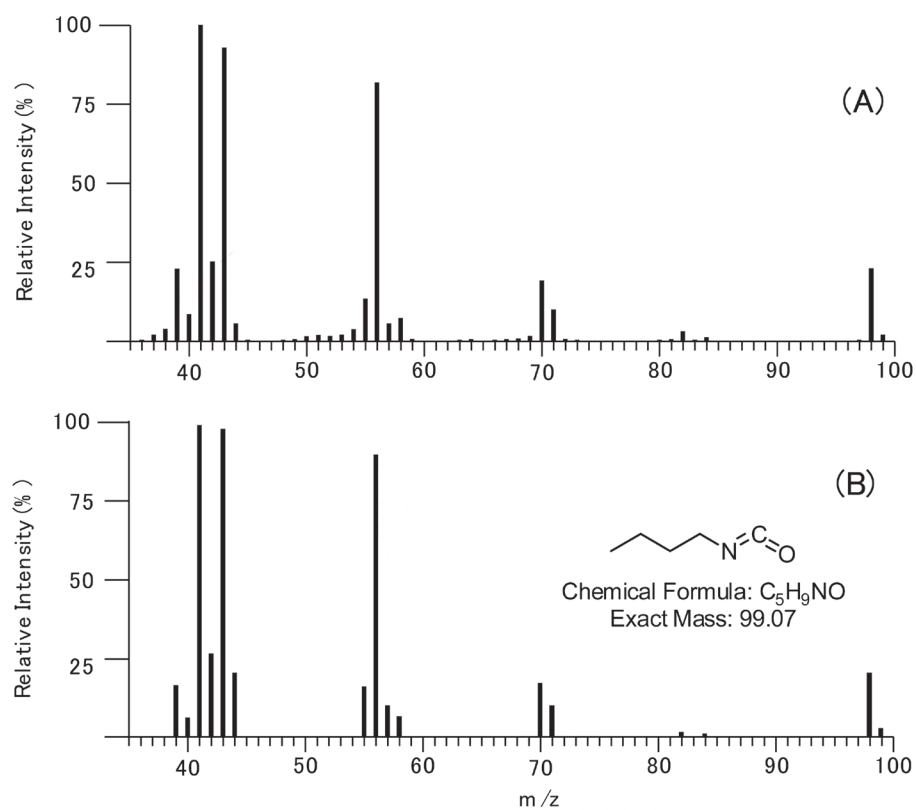


Fig. 2. Mass spectra of the peak at the retention time of 16.8 min (A) and that of the standard of *n*-butyl isocyanate (B).

the coating for the seeds by coating processing apparatus in the same plant, so *n*-butyl isocyanate could have been released from the benomyl-coated seeds during the drying process. Additionally, if the seeds were not thoroughly dry

when they were stored in the warehouse, *n*-butyl isocyanate could have been released into the air. To avoid the problem of airborne *n*-butyl isocyanate in the warehouse, we suggested to the workers that they extend the drying

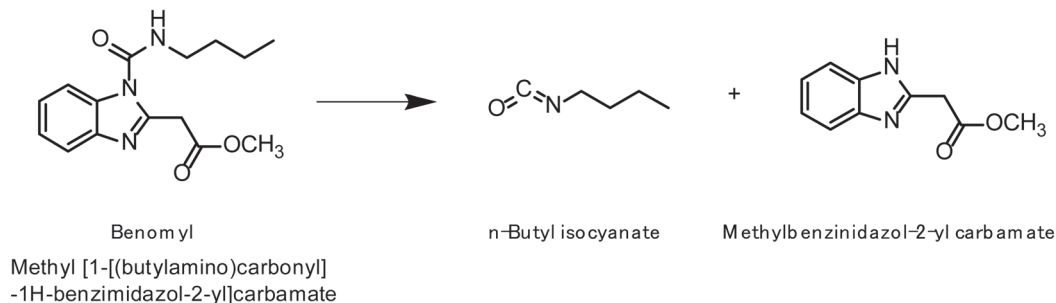


Fig. 3. Hydrolysis of benomyl to *n*-butyl isocyanate and carbendazim.

Table 1. Profile of warehouse A and B showing summary information

Variable	Warehouse A	Warehouse B
Number of workers with ocular irritation	4	0
Total amount of seeds (tons)	113	148
Amount of benomyl-coated seeds (tons)	28.9	4.2
Warehouse volume (m ³)	250	550
Amount of benomyl-coated seeds per unit volume (kg/m ³)	115.6	7.6

time for the coated seeds, before storage in the warehouse. After implementing this change in the production process, the workers in warehouse A reported decreased ocular irritation.

Discussion

The seed warehouse study found that *n*-butyl isocyanate was the predominant ocular irritant inside the warehouse. Like most isocyanates, *n*-butyl isocyanate can cause irritation to both the eyes and the respiratory system⁴. The contribution of exposure to *n*-butyl isocyanate in sensitization and development of ocular irritation and asthma has been demonstrated in several animal studies^{5–7}. An industrial hygiene survey⁸ conducted at a facility where *n*-butyl isocyanate was used as a chemical intermediate reported that noticeable ocular irritation (without odor or respiratory irritation) was associated with exposures of 0.005–0.010 ppm. In the present study, no workers suffered from serious respiratory irritation in warehouse A, suggesting that *n*-butyl isocyanate concentrations were less than 0.010 ppm.

Benomyl is used extensively on farms because of its relatively low toxicity⁹, except for mild irritation to skin and eyes⁴. Benomyl decomposes spontaneously, creating a reservoir for slow release of *n*-butyl isocyanate and carbendazim³. In the case of the seed supplier surveyed in this study, benomyl was used as the coating agent to pro-

tect the seeds when planted. Seeds were coated in another part of the same plant prior to storage in the warehouse. The coating agent was prepared as an aqueous solution and applied to the seeds in a rotating drum, through several spray nozzles. Hot air was used to evaporate the water, resulting in a buildup of the coating material on the surface of the seeds. The seeds were then cooled to room temperature and exposed to the air to dry. Probably because of insufficient exposure to the air, the benomyl-coated seeds were not thoroughly dry at the time of storage. *n*-Butyl isocyanate, therefore, could have been evaporating from the benomyl-coated seeds when they reached the warehouse. The assumption was made that prolonging the drying process before storage of benomyl-coated seeds would decrease the amount of benomyl hydrolysis taking place in the warehouse. This assumption was confirmed by decreases in ocular irritation among workers in warehouse A after implementation of the longer drying process for coated seeds.

The ocular irritation reported by workers in warehouse A was not reported by workers in warehouse B. This pattern is explained by differences in the amount of benomyl-coated seeds in the two warehouses and differences in the capacities of the two warehouses (Table 1). According to the seed supplier's records, warehouse A had a total of 113 tons of seed stored while warehouse B had 148 tons. However, in the spring of 2011, the amount of *benomyl-coated seeds* in warehouse A (28.9 tons) was seven times

the amount in warehouse B (4.2 tons). At the same time, the capacity of warehouse A (250 m³) was smaller than that of warehouse B (550 m³). Therefore, the amount of benomyl-coated seeds per unit volume (kg/m³) in warehouse A (115.6) was more than 15 times the amount per unit volume (kg/m³) in warehouse B (7.6). The ratio of the amount of benomyl-coated seeds was close to that of the peak intensities for *n*-butyl isocyanate on the GC-MS chromatograms for the two warehouses (warehouse A/B =13.7).

The seed warehouse workers obtained information on toxicity only from the safety data sheet (SDS) and not from other independent sources. The SDS covering benomyl did not mention ocular irritation from exposure to *n*-butyl isocyanate. As a result, the workers were unaware of the effects of hydrolysis on benomyl in a non-conventional usage of the pesticide, such as seed coating. It is suggested, therefore, that there is a need for 1) better SDS information, to assist workers in dealing with new technologies such as seed coating, 2) improved occupational training for workers starting use of new technologies, and 3) programs to improve the skills of workers who are currently employed.

Although the present study identified *n*-butyl isocyanate in the warehouse, we were unable to estimate its concentration quantitatively. We collected 6 liters (0.2 l/min × 30 min) of the workplace air on the TD cartridge; if it had been possible to obtain a suitable collection efficiency for *n*-butyl isocyanate on the cartridge, we could have calculated its airborne concentrations. However, we could not get the collection efficiency of *n*-butyl isocyanate on the cartridge. Additionally, we could not establish a sufficiently reliable calibration curve for GC-MS analysis, because of a technical error. In a future study, it would be useful to compare airborne concentrations of *n*-butyl isocyanate before and after prolonged drying of the benomyl-coated seeds.

Conclusion

The ocular irritation reported by the seed warehouse workers in this study is thought to have been caused by exposure to airborne *n*-butyl isocyanate, a product of pesticide hydrolysis degradation. Increasing the length of the

drying process after the seeds were coated with the pesticide benomyl reduced the occurrence of ocular irritation in the warehouse workers. In occupational health, more attention needs to be paid to possible toxic chemicals in the products of pesticide degradation, particularly regarding seed warehouses.

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