

# Effects of wood dust exposure and diethylnitrosamine in an animal experimental system

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An experimental study was conducted in Syrian golden hamsters (Wilhelmsson et al., 1984). Four exposure chambers made of steel were built. A special suction unit for controlled evacuation and particle generators with rotating grinders for generation of fresh wood dust under controlled conditions with respect to particle size and distribution were constructed. Special auxiliary motors for the particle generators were built to achieve the desired efficiency. The particle size and the concentration of wood dust were checked each week during the experiment both under the microscope and by weighting millipore filter papers.

Male Syrian golden hamsters were used. At the start of the experiment the animals were approximately 10 weeks old and weighed 90-120 g. Every week they were re-weighed. Tapwater was administered ad libitum and standard laboratory animal pellets were provided. A reversed 24 h cycle was employed in an attempt to ensure that the animals would be as active as possible while exposed. The air was maintained at a temperature of  $20^{\circ} \pm 1^{\circ}\text{C}$  and the relative humidity was 45-60 per cent. Two series of animal exposure experiments were performed, study I with 15 mg wood dust/m<sup>3</sup> and study II with 30 mg wood dust/m<sup>3</sup>. In each of the studies the animals were divided into four groups; two of them in each study were exposed to fine particles of fresh beech wood dust, for 6 h a day, 5 days a week, for 36 and 40 weeks respectively. One of these groups was also given 1.5 mg of DEN subcutaneously in study I and 3.0 mg of DEN subcutaneously in study II, in both cases once a week for the first 12 consecutive weeks. Another group in each study was given a corresponding dose of DEN but without exposure to wood dust. The fourth group served as a control, and was exposed neither to wood dust nor to DEN, but was placed in an exposure chamber at the same time as the other animals. At the end of the experiments the animals were killed by an intraperitoneal injection of Mebumal. While the animals were in a narcotic state the larynx, trachea, lungs, liver and kidneys were removed. The animals heads

were deepfrozen 10 min after death. When frozen, the nose were cut into 4-5 slices, each 3-5 mm thick. In study I the slices were placed in Carnoy's fluid and later stored in ethylbenzoate. In study II animals which were alive at the end of the study were fixated by perfusion with 4% neutral formaldehyde, through the left cardiac ventricle. Animals which died spontaneously during the experiments were deep-frozen as soon as possible. This was done not later than within 15 hours.

After dehydration the samples were embedded in paraffin, and sections approximately 7 $\mu$ m thick were cut and stained with hematoxylin and eosin. The pathologist was not informed from which animal group the material derived.

It was found in study I that low doses of wood dust and diethylnitrosamine, either alone or combined, did not give rise to any metaplasia, dysplasia or tumours in the nasal mucosa of the Syrian golden hamsters. There was one spontaneous tracheal squamous cell papilloma in the control group. The same frequency of tracheal papillomas was found among animals, exposed to DEN regardless of whether or not they were exposed to wood dust. Low doses of wood dust alone did not give rise to any detectable tracheal changes (Table 1).

Table 1.

tumour	control	exposure to		
		wood dust	DEN	wood dust + DEN
nose	0/12	0/12	0/12	0/12
larynx-trachea	1/7 (14%)	0/8	3/8 (38%)	4/8 (50%)
lung	0/7	0/8	0/8	0/8

In the animal study with higher doses (study II) there was a spectrum of multiple lesions in the DEN exposed animals, ranging from hyperplasia and dysplasia of respiratory epithelium, the olfactory mucous membrane, in the submucous glands at various locations to papillomas and carcinomas. The papillomas were mainly of the columnar cell type, with the presence of more or less cuboidal metaplasia. All of the malignant neoplasias were adenocarcinomas with various grades and patterns of differentiation as could be seen.

In this material no significant difference was found between the group exposed to DEN alone and that exposed to DEN combined with wood dust. About half of the animals had nasal tumours (Table 2).

However, the most interesting findings in the whole study were that in the group exposed to wood dust alone one animal exhibited nasal focal metaplasia (Figure 1) of cuboidal type with mild dysplasia and another animal had a malignant nasal tumour (Figure 2). Unfortunately, these two animals, as so many of the others, were among those which died spontaneously, and the tumour was there-



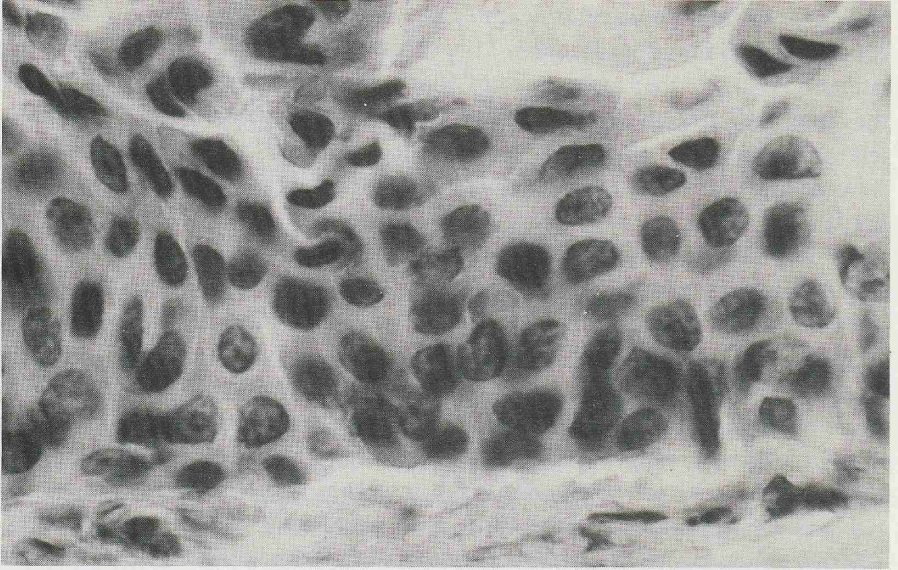


Figure 1. Metaplastic epithelium with mild dysplasia in the nasal mucosa of a Syrian golden hamster ( $\times 1800$ ). (Wood dust only).

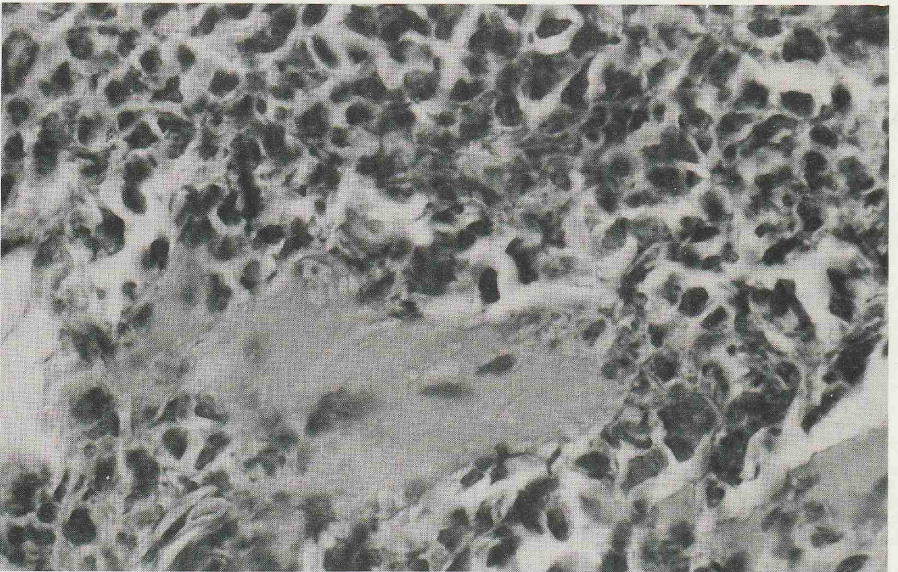


Figure 2. Undifferentiated malignant nasal tumour without detectable epithelial differentiation in an animal exposed to wood dust only ( $\times 1800$ ).

Table 2.

tumour	control	exposure to		
		wood dust	DEN	wood dust + DEN
nose-dysplasia	0/19	1/22 (5%)	8/18 (44%)	4/17 (24%)
nose-tumour	0/21	1/22 (5%)	10/22 (46%)	11/21 (52%)
larynx-trachea	0/22	0/23	10/19 (53%)	11/18 (61%)
lung	0/22	0/23	0/19	1/18 (6%)

fore necrotic and thus unclassifiable, but the pattern of growth was malignant, destroying the adjacent bone tissue and infiltrating the olfactory bulb. Furthermore, this group exposed to wood dust alone, showed slight inflammatory reactions in the respiratory epithelium and the submucosal stroma which were not observed in the control group.

In the larynx and the trachea all classified tumours were papillomas of the cuboidal or squamous epithelial type. One Syrian golden hamster, exposed to both DEN and wood dust, was found to have a small adenocarcinoma of the lung. The death rate was very high in all groups and significantly higher in the two DEN-treated groups. The explanation is presumably the effect of frequent, often multiple obstructive papillomas of the larynx and the trachea in animals exposed to DEN, but cannibalism was also a problem in this investigation, as often is the case in studies of this type.

## REFERENCES

1. Wilhelmsson B, Lundh B, Drettner B, Stenkvist B. Effects of wood dust and diethylnitrosamine - a pilot study in Syrian golden hamsters. *Acta Otolaryngol* (Stockh) 1985; 99:160-71.

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