

Incidence of bladder cancer in a cohort of workers exposed to 4-chloro-o-toluidine while synthesising chlordimeform

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Abstract

Between 1982 and 1990 seven cases of bladder cancer were detected in a group of 49 workers who were synthesising chlordimeform from 4-chloro-o-toluidine. Latency periods ranged from 15 to 23 years. The incidence of bladder tumours in this group was significantly higher than that of the cancer registers of the former GDR, Saarland, and Denmark by factors of 89.7, 53.8, and 35.0 respectively. This provides further evidence that monocyclic aromatic amines such as 4-chloro-o-toluidine may be carcinogenic in humans.

In 1987 the Commission for the Investigation of Health Hazards of Chemical Compounds in the Work Area of the Deutsche Forschungsgemeinschaft (DFG) classified 4-chloro-o-toluidine as a substance capable of inducing malignant tumours.¹⁻³ This classification was based on a communication from a chemical company reporting an increased incidence of bladder cancer in workers engaged in the synthesis of 4-chloro-o-toluidine.⁴ Recently, an increased incidence of bladder cancer has been found in another chemical plant in Germany where 4-chloro-o-toluidine was used to synthesise the insecticide chlordimeform.⁵ In 1985 chlordimeform was classified by the DFG Commission as a III A2 compound ("compounds which in the Commission's opinion have proven so far to be unmistakably carcinogenic in animal experimentation only; namely under conditions which are comparable to those for possible exposure of a human being at the workplace, or from which such comparability can be deduced"), but was not officially listed at the time.^{2,3} The present publica-

tion is to describe the known details of exposure and incidences of bladder cancer in this German chemical plant.

Subjects and methods

The insecticide chlordimeform was manufactured from 4-chloro-o-toluidine from December 1965. Production was not carried out continuously but in response to orders so that workers were subjected to different periods of exposure. Exact data for exposure were not available between 1965 and 1976 because measurements of concentration in the air or monitoring of urine excretion was not carried during that time. According to the works medical department haematurias only occurred during the synthesis of chlordimeform and then only when breakdown or accidents led to conditions resulting in high exposure. A haematuria was remembered in a mechanic who had come into contact with 4-chloro-o-toluidine. Some cases of methaemoglobinaemia were also seen. The plant was able to provide data about the total number of days of exposure for individual workers.

The situation changed after 1976 when chlordimeform production was halted to improve the working conditions and minimise human exposure. Chlordimeform production recommenced in 1980 with substantially improved containment. Exposure to 4-chloro-o-toluidine and chlordimeform was then analytically checked by monitoring of urine and shown to be minimal. In 1986 production of chlordimeform was finally stopped at the plant.

According to information from the Medical Health Department of the company 170 workers had come into contact with chlordimeform by the end of production in 1986; many of these employees only had minimal exposure—for instance, in laboratories. Only 49 (all male) workers were involved in the synthesis of chlordimeform. Until 1990, bladder cancers were found in this group only.

Those 49 workers who had been involved in the synthesis of chlordimeform from 4-chloro-o-toluidine were chosen to make up the study group. The period under investigation began with the entry

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of the subject into the company's employment (1950–9, $n = 17$; 1960–9; $n = 4$; 1970–9; $n = 28$) and finished with the occurrence of bladder cancer ($n = 7$), death from other causes ($n = 2$), premature termination of employment ($n = 8$), or the end of the year 1990. The expected incidence of bladder cancer was calculated by extracting the sex and age specific incidence of bladder cancer from the cancer registers of Saarland (1988)⁶ and of Denmark and the former GDR (for 1978–82 in each case),⁷ multiplying them with the person-years under investigation at five year intervals, and then adding them. These cancer registers were chosen because no central cancer register for the Federal Republic of Germany exists until now, and because the closeness of the chemical plant to the regions of the registers made it likely that they really reflected the incidence of cancer at the region of the chemical plant. The standard incidence rate (SIR) was the ratio of the number of cases observed (O) to the expected number (E). The 95% confidence interval (95% CI) and the significance level (p) were calculated under the assumption that the square root of O is roughly normally distributed at a standard deviation (SD) of 0.5 when E is small.⁸

The phenotype of N-acetylation was determined according to the method of Grant *et al*⁹ and Kalow.¹⁰

Results

Subjects started work at the plant at an average age of 30 (range 18–51). The exposure ranged from three to 956 (mean 271) days. By the end of 1990 ($n = 39$) an average of 18 (10–25) years had passed since the start of exposure; 28 persons were smokers, 15 were non-smokers, and in six persons smoking habits could not be evaluated. Bladder cancer was detected in seven of the 49 subjects by the end of 1990. Table 1 lists the data on the cancer patients. Table 2 lists the SIRs for bladder cancer with respect to the cancer registers used. The acetylator phenotype could be determined in five patients. Table 1 includes the results. One patient with bladder cancer died (myocardial infarction) before the investigation was completed and a further member of the group had a brain tumour.

Table 2 Standard incidence rates (SIRs) of bladder carcinoma in the group of workers ($n = 49$) engaged in chlordimeform synthesis

O	E (country)	SIR	95% CI	p Value
7	0.078 (GDR)	89.7	35.6–168.6	0.000002
7	0.200 (Denmark)	35.0	13.9–65.7	0.00001
7	0.130 (Saarland)	53.8	21.3–101.1	0.000005

Discussion

The 49 persons involved in the synthesis of chlordimeform from 4-chloro-*o*-toluidine included seven persons suffering from bladder cancer by the end of 1990. A further member of the group developed a brain tumour. The SIRs for bladder cancer in this group depending on the cancer registers in which they are referred (table 2) were between 35 and 89. The increase in the rates was statistically significant. The real risk may be somewhat greater, as expected incidence has only been taken from cancer registers of the later 1980s whereas the entry date for the cohort was earlier and the incidence of bladder cancer has tended to increase over the last decades (Frentzel-Beyme R, personal communication).

All the seven workers suffering from bladder tumours were exposed to 4-chloro-*o*-toluidine while synthesising chlordimeform before 1976. None of those workers who were merely handling the final product chlordimeform at the formulation or packing plants rather than synthesising it from 4-chloro-*o*-toluidine had developed bladder cancer by the end of 1990. The cohort investigated was engaged in chlordimeform production for a limited period (8–12 weeks) of each year. During the rest of the time they handled other chemicals at the plant. None of the resulting potential exposures were quantified by chemical analysis at the time. Some of the workers were handling another aromatic amine, 4-chloro-aniline. This was used, however, for appreciably shorter periods and in smaller quantities than 4-chloro-*o*-toluidine. We consider it probable that any such exposure will be less important in the increased incidence of bladder cancer found.

Table 1 Smoking habits, times of exposure and latency, age at diagnosis, and acetylator phenotype in seven workers with bladder cancer

Subject	Diagnosis	Smoking habit	Exposure	Latency (y)	Age at diagnosis (y)	Acetylator phenotype
1	Transitional cell carcinoma	Non-smoker	Sporadic before 1976	21	57	Deceased
2	Transitional cell carcinoma	10† a day	291 days 1966–71	23	47	
3	Papillary carcinoma	Non-smoker	555 days 1968–75	16	48	Slow
4	Transitional cell carcinoma	10 a day	617 days 1968–76	17	62	Slow
5	Transitional cell carcinoma	Non-smoker	766 days 1968–76	21	62	Slow
6	Transitional cell carcinoma	20 a day	644 days 1968–76	15	43	Slow
7	Transitional cell carcinoma	20 a day	Sporadic 1966–74	17	56	Fast
Average			575 days*	19	54	

*Without subjects 1 and 7.

†Number of cigarettes.

On the other hand the exposure to 4-chloro-o-toluidine, which is not only the raw material to synthesise chlordimeform but is also a metabolite of chlordimeform in mammals,² and possibly chlordimeform, seem to play a considerable part in the genesis of these cancers. Both 4-chloro-o-toluidine and chlordimeform have been shown to be carcinogenic in mice.² For 4-chloro-o-toluidine, indications also exist for its carcinogenic potential in humans: Uebelin and Pletscher¹¹ did not find any cancers in subjects working in production of 4-chloro-o-toluidine, but Müller¹² described a worker developing a lung tumour 12 years after chlorotoluidine intoxication. Another publication reported a (non-significant) increase in gastrointestinal and respiratory tract cancers in workers who had been exposed to 4-chloro-o-toluidine and other aromatic amines.¹³ Currie¹⁴ mentioned the occurrence of one case of bladder tumour among nine cases of 4-chloro-o-toluidine intoxication; the tumour was diagnosed only three years after the haematuria and there was some doubt about the degree of exposure. Finally Stasik⁴¹⁵ found eight cases of bladder tumours in a group of 116 persons exposed to 4-chloro-o-toluidine and calculated an SIR of 72.7; the order of magnitude of the excess incidence of bladder tumours is in good agreement with our results.

Increased incidence of bladder tumours has been reported among a Chinese population living in counties where the insecticide chlordimeform has been used in agriculture for several years.¹⁶ The evidence to support this study is limited, however, and needs further investigation.

It remains an open question whether chlordimeform is a bladder carcinogen in its own right. There is sufficient evidence, however, that its metabolite 4-chloro-o-toluidine exhibits such a potential. Therefore the same precautions used for 4-chloro-o-toluidine should be applied to chlordimeform.

The exposure and latency times (table 1) in our study are in good agreement with those found for other aromatic amines and with the results of Stasik.⁴¹⁵ We could determine the N-acetylator phenotype in five of the seven cancer patients; four of them were slow acetylators. This agrees with other studies reporting an increased risk of bladder tumours caused by arylamines in slow acetylators.¹⁷¹⁸ Four of the carcinoma patients in our study were smokers and three were non-smokers; 76% of the workers in the Federal Republic of Germany are smokers or former smokers,¹⁵ so smoking seems not to be an important confounding factor in interpreting the results of the study.

Conclusions

In our view the results confirm that exposure to 4-chloro-o-toluidine is associated with an increased

risk of developing bladder cancer. They suggest, moreover, that exposure to chlordimeform may also be a contributing factor. On the basis of published work and the fact that 4-chloro-o-toluidine is a metabolite of chlordimeform, we think that 4-chloro-o-toluidine plays an essential part in the initiation of the increased incidence of bladder cancer. This conclusion agrees with recent findings that show an increased risk for bladder carcinogenesis after exposure to other monocyclic aromatic amines.¹⁹

In our study some workers could not be included as they had left the production plant before 1985. We will report about the incidence of bladder cancer among these workers and about any new cases of bladder cancer in our study group in a subsequent publication.

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