Classification of occupational genotoxic carcinogens on the basis of their carcinogenic potency

Onderzoek
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SUMMARY

At the request of the Directorate General of Labour of the Dutch Ministry of Social Affairs and Employment we worked out a system for classifying genotoxic carcinogens according to their estimated risk for humans in the occupational situation. Three classes were defined qualifying carcinogens of low, intermediate and high potency, respectively. After drawing up an inventory of existing and proposed systems for classification of carcinogens, we decided to develop a system based on the TD_{50}-value combined with weighing factors, finally resulting in a potency index expressed as estimated 'tumour incidence' (in fact tumour incidence corrected by means of weighing factors) in humans at 1 mg/m^3 under occupational conditions of exposure (8h/day, 5 days/week for 40 years). The rationale for this approach is discussed in chapters 2 and 3, particularly in the concluding remarks of chapter 3. Chapter 4 gives the working-out as well as further reasons for the different choices that had to be made with regard to 1) the calculation of the TD_{50} (mg/m^3, lifespan conditions) and the conversion to incidence at the workplace (mg/m^3 under occupational conditions), 2) selection of the weighing factors and the assignation of scores to these, and 3) the limits for the three potency-classes. Finally, we have scored and ranked 23 genotoxic carcinogens according to the proposed system (see Chapter 5 and Tables 5.1 and 5.2). Application of the weighing factors appeared to lead to a shift in the ranking order: 5 carcinogens (potassium bromate, 3,3'-dichlorobenzidine, 4,4'-methylene bis(2-chloroaniline), acrylonitrile and 5-nitroacenaphthene) moved from the intermediate risk class (4×10^{-5} < estimated 'tumour incidence' per mg/m^3 < 4×10^{-3}) to the high risk class (estimated 'tumour incidence' per mg/m^3 $\geq 4 \times 10^{-3}$), one carcinogen (1,2-dibromoethane) moved from the high to the intermediate risk class and one (styrene oxide) from the intermediate to the low risk class (estimated 'tumour incidence' per mg/m^3 $\leq 4 \times 10^{-5}$).

The proposed system enables the use of animal and human data for ranking of carcinogens according to their estimated risk in man in a comprehensible and consistent manner. Moreover, the proposed system uses all relevant information available. Adjustment of potency indices by means

of weighing factors remains, however, a subjective operation heavily dependent on expert judgement.

1 INTRODUCTION

The Directorate General of Labour (DGA) of the Dutch Ministry of Social Affairs and Employment proposes to categorize carcinogens on the basis of their mode of action, in non-genotoxic and genotoxic (the policy categories A and B, respectively) in order to distinguish between carcinogens with and without a threshold level, respectively (Arboraad, 1986). The genotoxic carcinogens (category B) are further differentiated according to their potency into three classes (the policy classes B1, B2, and B3). For the subdivision of the genotoxic carcinogens (category B) into three classes no directives are given. However, it has been suggested to examine the usefulness of the criteria proposed by the American Conference of Governmental Industrial Hygienists (ACGIH) for the classification of experimental animal carcinogens into three classes denoting carcinogens of low, intermediate and high potency. The main criteria used by the ACGIH are a combination of the dose causing a significant carcinogenic effect, the latency period of tumour induction, the number of animal species showing a positive response and the different routes of administration leading to a positive response.

Carcinogenic potency can be defined as the magnitude, with respect to dose, of the carcinogenic activity of a chemical in the species under consideration (ECETOC, 1982). In this context, the expression "magnitude of carcinogenic activity" refers not only to incidence but includes also the intensity of the tumour response. The parameters needed for expressing cancer incidence and intensity in animals include the proportion of tumour bearing animals, the latency of tumour formation, the malignancy of the tumours, the cell types affected and so on (confer ECETOC, 1982). Obviously, carcinogenic potency cannot be expressed in a numerical value in a simple way. Details on existing models for estimating the carcinogenic potency are given in chapter 3.

Classification of carcinogens according to their potency in animals will apply only to those carcinogens for which no or insufficient human data

are available; if adequate human data are available they will be used for classification.

The present study is aimed at examining the possibilities of classifying genotoxic carcinogens (category B) into three classes B1 through B3 according to their carcinogenic potency with the prerequisite that the system should be simple and easy to apply to both animal and human data.

2 AN OVERVIEW OF NATIONAL POLICIES FOR REGULATING CARCINOGENS

The Federal Republic of Germany

The German "MAK" (Maximale Arbeitsplatz Konzentrationen = Maximal Workplace Concentrations) committee recognizes in its overall policy (= toxic substances regulation policy) a category of carcinogenic substances that is divided into two sub-categories, A: Eindeutig als krebserzeugend ausgewiesene Arbeitsstoffe (unequivocally proven occupational carcinogens), and B: Stoffe mit begründetem Verdacht auf krebserzeugendes Potential (suspect carcinogens). Category A substances do not receive a "MAK" value, but a "TRK" value (Technische Richt Konzentration = lowest concentration technically feasible) (Deutsche Forschungsgemeinschaft, 1986).
These Category A substances are further divided into three "policy groups" (gefährdend, stark gefährdend, sehr stark gefährdend) on the basis of their physical-chemical properties, their potency, their concentration in mixtures and the extent of their use (Bundesarbeitsblatt, 1979; Tordoir, 1980). As to how far each criterion plays a role in assessing a substance to one of the three groups, and how potency is defined, has not been published. This further division has no influence on the assessment of "TRK" values.
In the future the Category B substances will possibly be split up in two groups: B1-substances that need further investigation, and B2-substances that have been extensively tested, but are still regarded as suspect carcinogens (Senatskommission der DFG, 1986).

U.K.

Apart from the "Carcinogenic Substances Regulations 1967", that forbid the use of a limited number of substances, in England's toxic-substances-policy up to now no difference is made between carcinogenic and non-carcinogenic substances (Arboraad, 1986; Lewis, 1985). A carcinogen policy is in preparation.

France

In France we recognize a policy based on proven human carcinogens: category C1 with an assigned limit value, category C2 without an assigned limit value, to which no exposure in any form is permitted, and a category of suspect human carcinogens viz. category C3 with or without an assigned limit value (Cahiers de notes documentaires, 1984).
The French carcinogen policy is part of their limit value policy dealing with dangerous substances. The distinction between proven and suspect human carcinogens is based on epidemiologic and animal studies. The reason for which some carcinogens are classified with an assigned limit value and others without a limit value is not given.

USA

In the USA several agencies are occupied with the classification of substances on the basis of their carcinogenicity. Among them are: The Occupational Safety and Health Administration (OSHA, 1980, 1982), The Environmental Protection Agency (EPA, 1986), and The American Conference of Governmental Industrial Hygienists (ACGIH, 1985 - 1986).

OSHA: In 1980 the OSHA presented its carcinogen-policy consisting of criteria and procedures for the identification, classification, and regulation of potential occupational carcinogens (OSHA, 1980). Potentially carcinogenic substances are devided into category I and category II: potential carcinogens based on either good (category I) or suggestive (category II) human or animal evidence. Regulation

is based upon this categorisation without any further classification being made on the basis of either potency or working mechanism.
In 1982 the OSHA gave notice that it was reevaluating its "Carcinogen Policy", to determine the need for modification, based on U.S. Supreme Court decisions, public requests for review, and the Agency's own experience (OSHA, 1982).

EPA: The EPA's "Guidelines for Carcinogen Risk Assessment" were published in September 1986 (EPA, 1986). The "Guidelines" present a classification system consisting of five groups: A carcinogenic to humans, B probably carcinogenic to humans, C possibly carcinogenic to humans, D not classifiable as to human carcinogenicity, and E evidence of non-carcinogenicity for humans. This classification system is based on evidence from human and animal studies. Groups A and B are considered for further regulation based on quantitative risk assessment consisting of dose-response relationships and exposure assessment. The possibility of ranking carcinogens according to mechanism of action has been suggested (Marshall, 1982), but this suggestion has not led to any effect in the present "Guidelines".

ACGIH: The ACGIH is an organisation concerned with providing guidelines for occupational exposures which, based upon current scientific judgment, should protect nearly all workers from harm.
The ACGIH, on the basis of results from epidemiologic and animal studies, recognizes two main categories of carcinogens namely: A1 human carcinogens, and A2 industrial substances suspect of carcinogenic potential for man. Either category may contain substances with or without a TLV (Threshold Limit Value).

Next to this, ACGIH gives guidelines for classification of experimental animal carcinogens into three groups denoting substances with high, intermediate and low carcinogenic potency. This classification is based on the latency period of tumour

induction, the dose at which and the number of species in which a substance elicits cancer. This subdivision serves the purpose of determining appropriate thresholds of neoplastic response by which TLV's can be assigned. Working mechanism is not taken into consideration by the ACGIH (1985 - 1986).

Denmark

The Danish authorities are at present not planning to use potency ranking of carcinogens for the purpose of classification and hazard labelling (Nordic Working Party, 1987).

Finland

The lowest total dose found to induce tumours in animal bioassays has been used for classification of carcinogenic substances by the National Board of Health into two categories: class I and class II poisons.
No potency evaluation is used sofar in the regulation of carcinogenic substances (Nordic Working Party, 1987).

Norway

The State Pollution Control Authority and The Directorate for Labour Inspection have appointed a scientific group to advise in identifying and classifying carcinogenic substances.
Substances are divided into three categories: category I: chemical substances showing sufficient evidence of carcinogenicity, category II: chemical substances showing possible carcinogenic effect, and category III: substances that are not shown to be carcinogenic, or for which the data are insufficient.
No distinction is made between substances with high and low potency (Nordic Working Party, 1987).
The Norwegian Directorate for Labour Inspection is preparing regulations for occupational toxic and carcinogenic agents and products.

Sweden

The National Swedish Board of Occupational Safety and Health has classified carcinogenic substances into three policy groups: Forbidden carcinogenic substances (substances with a proven strong carcinogenic effect.); substances that may only be produced or used after official approval (substances suspect of carcinogenic effect.); substances with a limit value (National Swedish Board, 1981).
According to the new regulations for classification and hazard labelling effective from January 1, 1986, hazardous products will be divided into four categories. Products which may cause cancer will be assigned to the category "Very dangerous products" unless the probability of the product to cause cancer is regarded as very low. In this case, the product will be assigned to the category "Hazardous products".
An official discussion has just started among the Nordic countries on the use of potency evaluation of carcinogenic substances (Nordic Working Party, 1987).

3 EXISTING METHODS FOR POTENCY AND RISK ESTIMATIONS OF CHEMICAL CARCINOGENS

Squire (1981, 1983, 1984) proposed an approach for ranking animal carcinogens based on evidence derived from animal and genotoxicity studies. Six factors are involved in his ranking system (Table 3.1), and each factor is divided in sub-factors with a scoring system, which results in total scores varying from 13 to 100. The total score is used to rank the animal carcinogens into five classes (Table 3.2). As an example Squire (1981) ranked ten animal carcinogens (Table 3.3).
Crump (1983) has made two criticisms of this scoring system. One criticism is that this system does not give adequate weight to the dose at which carcinogenic effects occur (the lack of weight given to carcinogenic potency), and a second one is that the actual numbers of malignant and benign neoplasms would be more important than the ratio of malignant to benign neoplasms as is suggested by Squire.

We feel that another deficiency in Squire's system is, that it does not take into account the presence of negative results in one or more species, the relevance of the route of administration and data on metabolism, toxicokinetics and mechanism of action. Furthermore, Squire does not give a foundation for his scoring system.

The US Environmental Protection Agency (1984) has proposed the use of a Unit Risk estimation (WHO, 1987). The Unit Risk is defined as the life-time cancer risk in a hypothetical population where the individuals are exposed daily to the agent at a concentration of 1 $\mu g/m^3$ in air they breathe, or 1 $\mu g/l$ in drinking water via ingestion from birth throughout life. Unit risk calculations have two purposes: 1) to compare the carcinogenic potency of different substances, and 2) to indicate the risk represented by the substance, if the degree of exposure is known.
For those substances where appropriate studies were available, the average relative risk model was used. Before any attempt is made to assess the risk in the general population, three assumptions and considerable mathematical treatment of the raw data are needed at each phase of the risk assessment process to fill in various gaps in the underlining data base. The assumptions are: 1) There is no threshold dose for carcinogens, 2) there is a constancy of the relative risk in the specific study situation, and 3) the response (measured as relative risk) is some function of cumulative dose or exposure.

ECETOC (1982) discusses various factors, which ECETOC believes are involved in making a distinction between carcinogens of widely-differing potency. Besides the dose, also human data, experimental animal data, metabolism studies, the mode of action, are needed to determine the carcinogenic potency of a chemical. They concluded, that because the process of categorisation is neither primarily numerical nor invariable (it will differ from chemical to chemical) an accurate potency value can not be determined. Only judging a group of factors from animal and human data would lead experts to the categorisation for individual chemicals.

Theiss (1983) presented a classification system, which utilizes in a tier system, short-term test results, data from animal experimentation, and epidemiologic evidence to differentiate between weak and strong carcinogens (Table 3.4). If a chemical produces positive results in several short-term tests (= tier 1) it would be classified as a strong carcinogen. When sufficient experimental animal data (=tier 2) are available to make a judgment regarding carcinogenic potency, this judgment would supersede that made after considering only short-term tests. Epidemiologic evidence (=tier 3) would be considered in conjunction with the experimental animal evidence in making a determination regarding carcinogenic potency. Evidence from animal experimentation supersedes epidemiologic evidence only when this epidemiologic evidence is of questionable nature. An example: if the experimental animal evidence suggests that the chemical is a strong carcinogen, but well-conducted epidemiologic studies do not detect carcinogenic effects, then the chemical would be classified as a weak carcinogen. In the proposed tier system it is not clear what the definitions are of "several" and "sufficient".

Peto et al. (1984) proposed the TD_{50}, defined as that chronic dose rate (in mg/kg body weight/day) which would give 50% of the animals tumours within some standard experiment time -the "standard lifespan"- for species, as a measure of carcinogenic potency. In the accompanying paper Gold et al. (1984) present the Carcinogenic Potency Database, as the TD_{50}, which includes results of about 3000 long-term, chronic experiments of 770 test compounds, while in the paper of Sawyer et al. (1984) nonparametric procedures are developed and described for estimating the TD_{50} and confidence intervals. According to Barr (1985) Peto et al. (1984) confirmed the 10^7 range of potencies, but also found up to 10^3 range of potencies within the data for single substances (e.g. vinyl chloride). This same range of estimates is seen (according Barr, 1985) when full risk assessments are performed with the results of vinyl chloride bioassays, and this variation has been used by OSHA as a reason for not considering the risk assessment methodology valid or useful.

Calculation of the TD_{50} value implicit the knowledge of the data of the standard lifespan of the test animal, the mean (lifelong) mg/kg body weight/day dose, a correction factor for experiments which terminate prior to the standard lifespan, a method of selecting tumour sites on which to base the TD_{50}, statistical methods for estimating the TD_{50} when time-to-tumour data are available, and an analysing system when actuarial correction is impossible.

Ames et al. (1987) have introduced the HERP (Human Exposure dose/Rodent Potency dose) as a measure of possible hazard for humans. The HERP is derived from the TD_{50} and is calculated by expressing each human exposure (daily lifetime dose in milligrams per kilogram) as a percentage of the rodent TD_{50} dose (in milligrams per kilogram). In the paper it is said, that it would be a mistake to use the HERP index as a directe estimate of human hazard, because 1) at low dose rates human susceptibility may differ systematically from rodent susceptibility, 2) the general shape of the dose-response relationship is not known, and 3) it may be necessary to deal with carcinogens that differ in their mechanisms of action and thus in their dose-response relationship. This HERP-system can be seen as an extension c.q. application of the TD_{50} system of Peto.

Gaylor and Chen (1986) compared cancer rates across animal species for a wide variety of compounds (190) administered by various routes of exposure, by using the TD_{50} data of Gold et al. (1984). It was found that the geometric means of the ratios of minimum TD_{50}'s for rats: mice are 1/2.2 and 1/1.3 for diet and gavage, respectively. A mean ratio for rats:mice of 1/1.48 was obtained for compounds administered in the diet when the tumour site was the liver for both species. The geometric mean of the minimum TD_{50}'s for rats:hamsters was 1/13.7 for diet. In general, comparisons of minimum TD_{50}'s across the three rodent species were generally within a factor of 100 for a wide variety of compounds.

The Nordic Working Party (1987) suggested the use of an index of carcinogenic potency, represented by TDx, the lowest daily dose (in mg test substance per kg body weight) which has induced a statistically

significant increase in the number of tumours, or has induced a statistically significant increase in tumour frequency as shown by trend analysis (x denotes the per cent of tumour bearing animals). In an attempt to define practical administrative limits between the groups of carcinogens with high, intermediate, and low potency, experimental data concerning very potent carcinogens were examined by the working group in detail. It was found that the doses of benzo(a)pyrene and N-nitrosodiethylamine which induced tumours in 50% of the test animals, differed between experiments by a factor of 100-150. Based on experimental data available, it was concluded that the variation in TDx doses for substances in well performed experiments probably do not differ by a factor of more than 600. To define an administrative limit between the groups of carcinogens of intermediate and low potency, the following reasoning was applied: if the most potent of the substances of intermediate potency have TDx values of about 1 mg/kg b.w./day, and if the variation of the TDx values of these substances corresponds to that of the very potent substances, a distinction between the two groups could be set in the dose interval between 150 and 600 mg/kg b.w./day. It is concluded by the Working Party, that TDx values in most cases will be the most important factor, but the TDx values should always be regarded in conjunction to other relevant data such as: dose-response relationships, information on mechanisms, information on toxicokinetics, epidemiologic data etc.

Lutz (1986) suggested that the determination of liver DNA binding of mutagenic compounds could be used as a semiquantitative short-term test for carcinogenic potential to predict that mutagens exhibiting a Covalent Binding Index, (CBI=umol chemical bound per mol DNA nucleotide)/ (mmol chemical administered per kg body weight) of below 10 are most probably weak carcinogens, and that a CBI in the order of 100 stands for potent carcinogens.

Concluding remarks

A few systems or proposals for systems for an administrative ranking of carcinogens according to their potency were described. In the proposed systems potency indices are derived from epidemiologic data., from animal data (TD_{50}, TDx etc.), from the results of tests to which a predictive value is attributed e.g. "Covalent DNA Binding Index", or from a combination of these factors such as for example in the system proposed by Squire (1981, 1984).

It is obvious that ranking of carcinogens according to their estimated potency in humans, should preferentially be based on human data, and in the absence of relevant human data, on animal data. Different indices are described in the literature for expressing carcinogenic potency, on the basis of animal experiments, in a numerical value such as TD_{50} and TDx. These potency indices have in common that they are based on tumour incidence in relation to the dose. For practical reasons we prefer the TD_{50} calculated according to the method of Peto et al., 1984. This method is widely applied, while an extensive data base as well as the TD_{50} of a number of carcinogens have meanwhile been published.

However, since a system based on TD_{50} values derived from animal data only, is too simple as a system for ranking carcinogens according to their estimated potency in humans, a number of other factors can be added to the system in the form of weighing factors. A system that in this sense is more appropriate, has been proposed by Squire (1981; 1984). Disadvantages of the latter system are:

1. The less research has been performed on a chemical, the less dangerous the chemical turns out to be in the Squire system;
2. Not enough weight is attached to negative observations;
3. The system is not adapted to the occupational situation, for example, with respect to number of years of exposure.

We decided to develop a system based on combining the TD_{50} value with weighing factors, finally resulting in a potency index expressed as estimated "tumour incidence" (in fact tumour incidence corrected by means

of weighing factors) in humans at 1 mg/m^3 under occupational conditions of exposure time (8h/day, 5 days/week for 40 years).

4 SET UP OF THE CLASSIFICATION SYSTEM, WITH STARTING POINTS AND RATIONALE

4.1 Introduction

As pointed out in the first chapter, this report is primarily aimed at examining the possibilities of categorizing genotoxic carcinogens according to their potency into three classes: viz. genotoxic carcinogens that may be of low, intermediate or high risk for humans in the occupational situation. If available epidemiological data in humans should be used for such a categorization. However, epidemiological data are scarce and mostly not available. The next best is potency ranking on the basis of data from long-term animal experiments, that mimick as closely as possible the occupational situation. Categorization in the absence of epidemiological data is complicated by the different steps that have to be taken to 'translate' the results obtained in animal experiments into a reasonable estimate of the carcinogenic potency in humans at the workplace. These steps can be described as:

(1) Quantification of the carcinogenic response in animals. For this purpose dose-response extrapolation models are used to estimate cancer risks associated with low-dose exposure.
(2) Extrapolation from animals to humans. Examples of important factors are: differences in toxicokinetics between animals and man, and the relevance of the set up of animal experiments with respect to the occupational situation.

It is self-evident that an estimation of the health risk associated with the production and use of a genotoxic carcinogen also depends on the exposure assessment including both the number of workers that actually will be exposed and the degree of exposure at the work place. However, this aspect of cancer risk estimation is not included in the potency classification given in the present report.

In this chapter we tried to work out a procedure enabling categorization of category B carcinogens according to their estimated potency in humans in the occupational situation by combining carcinogenic potency as determined in animals and expressed as tumour incidence/mg carcinogen/m^3 air under occupational conditions, supplemented with a number of weighing factors thought to be of importance with regard to the (possible) tumour response in humans. This procedure regards steps 1 and 2 enumerated above, exposure assessment is not included in this procedure and has to be worked out separately.

4.2 Quantification of carcinogenic potency, extrapolation from high to low doses.

Animal cancer tests can be analyzed quantitatively to give an estimate of the relative carcinogenic potencies of chemical carcinogens. In the foregoing years various mathematical models have been proposed to fit the dose-response relationships in long term animal experiments. Once the curve has been established, the potency can be expressed in various ways and the confidence limits can be calculated. Frequently used potency indices are TD_{50}, TDx, and incidence per unit of exposure etc. For ranking of carcinogens according to potency it is not essential which potency index is used, as long as these indices are calculated using the same mathematical model they can be directly converted into each other. In contrast, the mathematical model used for fitting the dose response curve and the conventions applied for calculating the potency index such as correction factors, standard values, number of dose levels included etc. are very important.

Well-known are the models worked out by Meselson and Russell, 1977; Crouch and Wilson, 1979; the National Academy of Science, 1980, and the group of Gold, Peto, Sawyer and coworkers. For a review see chapter 3 and G. Charnley and T.W. Thorslund, Biologically-based models to predict cancer risk, in Carcinogen Risk Assessment, 1988, pp 105-113, ed. C.C. Travis, Plenum Press, New York and London).

In the Netherlands an estimation of the potency of chemical carcinogens for humans is made by calculating the incidence per mg/m^3 or per mg/kg body weight according to the so called 'Simple Dutch Method' (SDM). A description of this method together with the potency indices calculated according to this method is given in Appendix 4-3.

4.2.1 Advantages of the Peto TD_{50}

The methodical approach described by the group of Gold et al. (1984) and Peto et al. (1984) seems most appropriate to us. It is an attractive, rather flexible approach, that is widely accepted. Their ongoing "Potency Database" comprises at the moment already TD_{50} values of more than 1000 chemicals (Gold et al., 1984, 1986b, 1987). These investigators have worked out in detail two methods of statistical analysis of bioassay results: one using life table data and the other using summary incidence data. From either type of analysis, the carcinogenic potency is estimated as TD_{50}: the dose rate in mg per kg body weight per day which would halve the probability of an animal remaining tumour free by the end of the standard life span for the species (Peto et al., 1984).
Comparison of these two methods has shown that there is substantial agreement between the summary incidence and the life table methods of analysis in terms of potency estimation, but there appeared some notable differences in the estimated shape of the dose-response curve (Gold et al., 1986a). The analysis using life table data is far more complex than the summary incidence method, that does not take into account correction for mortality.
From these considerations it is clear that for our purpose, potency estimation in a simple way, the summary incidence method is most appropriate. Moreover, in most long term animal experiments results are presented as summary incidence data and mostly no life table data are given.

In order to provide the most accurate estimates of the TD_{50} and to provide a large resource with comparable data to facilitate the analysis of the results of animal cancer tests, Gold et al. calculated the TD_{50} in a standardized way using a number of strictly defined starting points

regarding 1. selection of tissue and tumour types for calculation of carcinogenic potency, 2. the estimation of the average daily dose level, 3. the extrapolation of TD_{50} to the standard lifespan (Gold et al., 1984).

It will be clear that with regard to the occupational situation these starting points need some adaptation.

4.2.2 Conventions used for calculation of the Peto TD_{50} with adaptations for the occupational situation

The discussion about the various conventions and starting points of Gold for the calculation of the TD_{50} under lifespan conditions and our adaptations with regard to the occupational situation follows, as closely as possible, the conventions as described in the paper of Gold et al. (Gold et al., 1984, pp 11-14).

A. Selection of species tissue and tumour types for calculation of carcinogenic potency.

Gold et al., 1984 do'nt make a choice, but calculate the TD_{50} for different types of treatment-related tumours and for the number of animals with treatment-related tumours if possible. For our purpose, i.e. risk estimation in the occupational situation, we have decided to calculate the TD_{50} preferably from studies in which the test chemical was administered in a way relevant for the occupational situation. Further, if possible we prefer a TD_{50} based on the number of animals with treatment-related tumours albeit a mixture rather than a TD_{50} based on the number of animals with one selected tumour type. For chemicals with more than one adequately performed positive test, we select the lowest significant TD_{50} value from those experiments thought to be the most relevant with respect to the occupational situation.

B. Estimation of the average daily dose level,

For estimation of the dose level averaged over the duration of the experiment we propose to use exactly the same procedure as that used by Gold applying the same assumptions and standard values for dose calculation (see appendix 4-1).

C. Extrapolation of TD_{50} to the standard lifespan (f^2d).

Here also we prefer the use of the procedure of Gold applying the same conventions and standard lifespan values (Gold et al. 1984, pp 13 and 14). A procedure for calculating the TD_{50}- value for tumour bearing animals according to the summary incidence method is given in appendix 4-1. In case there are not enough data for calculating a 'Peto TD_{50}', we propose to calculate a 'stripped TD_{50}' from the best experimental data available, using the same rules and conventions as for the 'Peto TD_{50}'.
To convert the TD_{50} from mg/kg b.w. into mg/m^3 the standard values as published by Gold et al., 1984 are used (Appendix 4-1, p.91 this report).

D. Conversion of the TD_{50} (lifespan conditions) to incidence at the workplace (occupational conditions)

Potency estimates for the occupational situation are based on 8-hour shifts, 5 working days per week, and a total working life of 40 years. To convert TD_{50} values expressed in mg/m^3 and calculated for the standard life span of the laboratory animal in question to incidence values at an exposure concentration of 1 mg/m^3 under occupational conditions it is assumed that the average man works 8 hours/day, 5 days/week for 40 years.

4.3 Weighing factors for carcinogenic potency estimation in humans

This paragraph lists the weighing factors, that are expected to be of relevance in estimating the potency of carcinogens for humans in the occupational situation. Application of these weighing factors may improve the estimation of carcinogenic potency in humans on the basis of available animal data.

4.3.1 Selection of weighing factors

We have selected the following 6 weighing factors:

(1) Relevance of the way the chemical has been administered with regard to occupational exposure.
(2) Number of different species affected/number examined.
(3) Malignancy of induced neoplasms.
(4) Dose-response relationship.

(5) Toxicokinetics, metabolism, mechanism of action.
(6) Epidemiologic data that can not be used for quantitative extrapolation

Two kinds of reflections have played a role in awarding scores to the weighing factors:

(1) Scores that are based on a lack of information; information that we feel should have been available. For example:
Weighing factor 1: We consider it normal that information is available from studies in which the chemical has been administered in a way relevant for the occupational situation.
Weighing factor 2: We consider it normal that a chemical that is classified as a genotoxic carcinogen has been examined for carcinogenicity and found positive in at least two different animal species.
Weighing factor 3: malignancy is the normal situation.
Weighing factor 4: it is considered normal that dose-response data are available
Weighing factor 5: Normally, data on toxicokinetics, metabolism or mode of action are not available.
Weighing factor 6: Normally, no data on epidemiology are available.

(2) Scores that originate from indications for increased/decreased risk. For example:
Weighing factor 2. Test chemical has been examined in 3 or more species: ratio of positive tests/negative tests may point to increased or decreased risk to be expected for humans. Besides the availability of several adequately performed positive tests enables the selection of the most potent (lowest relevant) TD_{50}-value; this has consequences for the assignment of scores to weighing factor 2.
Weighing factor 3: low degree of malignancy leads to a lower estimated risk for humans
Weighing factor 5: available data point to clear differences with humans.

Squire selected as weighing factors: (1) number of different species affected, (2) number of different neoplasms induced, (3) spontaneous incidence of neoplasms, (4) dose-response relationship, (5) malignancy of induced neoplasms, (6) genotoxicity.
Weighing factors 2 through 5 of Squire are included in the TD_{50} value and if not or only partly they are included in the weighing factors we propose, for example the dose-response relationship, if not available, a factor for increased risk is given; weighing factor 1 of Squire is included as weighing factor 2 in our system.
We did not select genotoxicity as weighing factor (weighing factor 6 of Squire) because we are dealing with proven genotoxic carcinogens only. The scores for the weighing factor can vary between 1 and 100, a score of 1 for a factor means that the contribution of the weighing factor regarding to risk estimation is neutral. The scores of the weighing factors can be positive, negative or neutral.

Of course, adjusting potency indices with the aid of weighing factors is a subjective, but has the advantage that extrapolation from animal to man can be done comprehensibly and as consistently as possible, and in such a way that the potency indices of the chemical substances can be compared among each other. Besides, it contributes to make use of all relevant information available.

4.3.2 Assignation of scores to the weighing factors

(1) Relevance of the way the chemical has been administered with regard to occupational exposure.
- Carcinogenicity data derived from studies in which the test chemical was administered in a way corresponding to the occupational exposure: neutral, no adaptation of potency.
- TD_{50} derived from a study in which the test chemical was administered in a way irrelevant for the occupational situation, while an adequate negative study using a relevant way of administration is available: factor up to 100 for decreased potency.
See also note 1, page 25.

- Way of administering the test substance irrelevant and no relevant other study available: factor up to 10 for increased potency. The potency should not be adapted, when data on metabolism, toxicokinetics, etc. indicate that the carcinogenic effect is most probably independent of the way the chemical is administered.

(2) Number of different species affected/number examined.

- The test chemical appears carcinogenic in the two species examined: neutral, no adaptation of potency.
- The test chemical is carcinogenic in one species and not carcinogenic in the second species: factor up to 10 for decreased potency. See also note 1, page 25.
- Only data from one animal species are available: factor up to 10 for increased potency.
- The test chemical has been examined in three or more different species:

-- in case of negative tests: factor up to 10 for decreased potency

-- in case of positive tests there are two possibilities,

a) Data from several adequately performed experiments allow selection of most potent (lowest significant) TD_{50}: neutral, no adaption of potency,

b) Although several experiments have been performed only data from one of the experiments are suitable for calculation of the TD_{50}: factor up to 10 for increased potency.

(3) Malignancy of induced neoplasms.

- Induced tumours are malignant: neutral, no adaptation of potency.
- The results indicate a low degree of malignancy: factor up to 5 for decreased potency.

(4) Dose-response relationship.

- Dose-response data available: neutral, no adaptation of potency.
- Only one dose examined: factor up to 20 for increased potency.

(5) Toxicokinetics, metabolism, mechanism of action.

- No data available or available data point to qualitative and quantitative similarities with humans: neutral, no adaptation of potency.

- Available data point to clear differences with humans: factor up to 100 for decreased potency.

(6) Epidemiologic data, that can not be used for quantitative extrapolation
- Inadequate data: neutral, no adaptation of potency
- Adequate, negative epidemiologic studies: factor up to 100 for decreased potency. Adequate, negative data can and should also be used to calculate the maximum tumour incidence to be expected in case the compound indeed would be a human carcinogen.
- Positive epidemiologic studies, that are not suitable for risk evaluation: factor up to 100 for the increased potency.

NOTES
1. Adequate negative data can be used to calculate the maximum tumour incidence to be expected in case the compound would be an animal carcinogen. This value can be helpful in assigning scores to the weighing factors in question.
2. If biotransformation and toxicokinetic studies or studies on mechanism of action conclusively demonstrate the irrelevance of the carcinogenicity data in animals for humans, one might decide to remove such a genotoxic animal carcinogen from the list of carcinogens to be classified, and, thus to consider the compound as a non-carcinogen in humans.

4.4 Limits for the three potency-classes as proposed by the Directorate General of Labour

The following limits are chosen for the three potency classes:
Class B1 includes carcinogens with 'estimated potency-values' equal to or higher than 4×10^{-3} at an exposure level of 1 mg/m^3 under occupational conditions; these are called strong carcinogens;
Class B2 consists of chemicals with 'estimated potency-values' between 4×10^{-3} and 4×10^{-5} at an exposure level of 1 mg/m^3 under occupational conditions; these are called moderate carcinogens;

Class B3, the weak carcinogens, include chemicals with potency estimates equal to or lower than 4×10^{-5} at an exposure level of 1 mg/m^3 under occupational conditions.
The risk limit of 4×10^{-3} is chosen because this is the widely accepted risk for serious accidents at the workplace over a period of 40 years. In no case and under no conditions a cancer risk equal to or higher than 4×10^{-3} (class B1) can be accepted. The second limit of 4×10^{-5} is chosen arbitrarily and implies that an estimated cancer risk at the workplace equal to or lower than 4×10^{-5} (class B3) is considered acceptable provided certain (moderately strong) safety measures are taken. Obviously, class B2 carcinogens are carcinogens of intermediate potency.
The exposure concentration of 1 mg/m^3 as a unit of exposure for comparison of accompanying carcinogenic potency of occupational carcinogens is a technical, empirical choice. The major justification for the choice of the unit of exposure and of the limits for the different classes is the usefulness of the system in terms of being sufficiently discriminating with respect to (potency) classes (or potency ranking) of occupational carcinogens. If it would appear that the suggested limits are not discriminating enough (too many carcinogens show up in one class), the limits should be adapted. The limits chosen are in the range of the levels given by ACGIH (factor 10 difference), and the Nordic Working Party (factor 2 difference).

5 CLASSIFICATION IN PRACTICE

5.1 Classification procedure

This paragraph lists the steps that are to be taken to classify a genotoxic carcinogen.

I. Are there sufficient epidemiologic data for quantitative risk assessment and thus, for classification in humans?
   - Yes, the chemical is classified using the epidemiologic data. If necessary, these data have to be adapted to the occupational situation.

- No, —> II. Classification to be based on animal data.

II. Are there enough animal data for calculating the 'Peto TD_{50}' according to the 'Summary Incidence Method'?
- Yes, —> IV (for the actual calculation see appendix 4-1).
- No, —> III

III. Caculate a 'stripped TD_{50}' from the best experimental data available. Use for this calculation the same rules and conventions as for the 'Peto TD_{50}' with our adaptations for the occupational situation, see appendix 4-1. The only difference between the 'Peto TD_{50}' and the 'stripped TD_{50}' is the absence of data on dose response relationships in animal experiments. (See Paragrapgh 4.3.1 Weighing factor 4) —> IV.

IV. Calculate the cancer incidence per mg/m^3 under occupational conditions (see paragraph 4.2.2 D).

V. Consider and score the weighing factors applicable as exemplified in paragraph 4.3.2.

VI. Classify the carcinogen as a B1, B2 or B3 carcinogen (see paragraph 4.4).

5.2 Classification of 23 genotoxic carcinogens

Twenty three genotoxic carcinogens have been scored according to the proposed system and ranked with and without weighing factors.
Table 5.1 gives a survey of the genotoxic carcinogens included in this exercise together with the estimated cancer incidences in the occupational situation without and with weighing factors.
Table 5.2 gives a ranking of the carcinogens according to their estimated potency indices with and without weighing factors.
This latter table shows that the application of the weighing factors causes a shift in the ranking order of the carcinogens with the result

that 5 carcinogens (indicated with ↑) moves from class B2 into class B1 and 1 carcinogen (indicated with ↓) moves from class B1 into class B2.

The data on the individual compounds are given below.

Vinyl chloride monomer (VCM)
Consulted literature Swaen et al. (1987).

I There are sufficient epidemiologic data for quantitative risk assessment in humans. A lifetime exposure to 0.001 mg/m^3 VCM results in an additional risk to the general population of 1×10^{-6} (Swaen et al., 1987).
Translated to the occupational situation this means an expected incidence for humans of 1.4×10^{-4} at an exposure of 1 mg VCM per m^3.

Benzene

Consulted literature Swaen et al. (1988).

I There are sufficient epidemiologic data for quantitative risk assessment in humans. A lifetime exposure to 0.65 $\mu g/m^3$ benzene results in an additional risk to the general population of 1×10^{-6} (Swaen et al., 1988).
Translated to the occupational situation this means an expected incidence for humans of 2.1×10^{-4} at an exposure level of 1 mg benzene per m^3.

Ethylene oxide

Consulted literature: WGD 87-127-17; Vermeire et al. (1985a); Garman et al. (1986); Snellings et al. (1984); IARC (1987).

I Not enough epidemiologic data for quantitative risk assessment in humans. A causal relationship between exposure to ethylene oxide and leukaemia is thought possible, but the five small epidemiological studies so far available suffer from various disadvantages, especially confounding exposures, which make their interpretation difficult (IARC, 1987).

II Ethylene oxide was tested by intragastric intubation in rats and produced local tumours, mainly squamous-cell carcinomas of the forestomach. When rats were fed diets with ethylene oxide, no increased incidence of tumours was observed. When rats were exposed by inhalation, ethylene oxide increased the incidences of mononuclear-cell leukaemia, brain tumours and proliferative lesions of the adrenal cortex in animals of each sex and of peritoneal mesotheliomas in males. In mice inhalation of ethylene oxide resulted in increased incidences of alveolar/bronchiolar lung tumours and tumours of the Harderian gland in animals of each sex and of uterine adenocarcinomas, mammary carcinomas and malignant lymphomas in females. Subcutaneous injection of ethylene oxide in mice produced local tumours, which were mainly fibrosarcomas.

Adequate data are available for calculating the TD_{50} according to the "Summary Incidence Method". Data expressed as adjusted ratios (= number of animals with primary brain tumours/number alive at the time the first tumour was observed in any group) were taken from a paper by Garman et al. (1986): inhalation study in Fischer-344 rats exposed by inhalation to 0, 10, 33 and 100 ppm ethylene oxide, 6 hr/day, 5 days/week, for up to 24 months. Incidences amounting to 2/369 in controls, 2/186 in the 10 ppm group, 8/177 in the 33 ppm group and 11/167 in the 100 ppm group.

The TD_{50} based on these data amounts to 1115 ppm, which corresponds to 2040 mg/m^3. A correction is not necessary.

III Not applicable

IV Based on the TD_{50} of 2040 mg/m^3 the estimated incidence for humans in the occupational situation amounts to $0.5/[2040 \times 7/4 \times 6/8] = 1.9 \times 10^{-4}$ per mg/m^3.

V (1) Neutral: TD_{50} based on inhalation study.

(2) Neutral: The test chemical appears carcinogenic in the two species examined.

(3) Neutral: tumours malignant

(4) Neutral: dose response data available.

(5) Neutral: available data point to similarities with humans.

(6) Factor 5 for increased potency: Epidemiologic data suggest that ethylene oxide might be carcinogenic in humans.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $1.9 \times 10^{-4} \times 5 = 9.5 \times 10^{-4}$.

Propylene oxide

Consulted literature: NTP technical report 267 (1985); Vermeire et al. (1985b); Vermeire et al. (1987); IARC (1987)

I Not enough epidemiologic data for quantitative risk assessment in humans.

II Propylene oxide was tested by oral gavage in rats and produced local tumours, mainly squamous-cell carcinomas and papillomas of the forestomach. When tested by inhalation in mice and in rats, it produced haemangiomas and haemangiosarcomas of the nasal submucosa in mice and an increased incidence of papillary adenomas of the nasal turbinates in rats. Subcutaneous administration in mice, induced local sarcomas, mainly fibrosarcomas.
Adequate data are available for calculating the TD_{50} according to the "Summary Incidence Method". For the calculation of the TD_{50} the adjusted rates for the incidences of hemangioma and hemangiosarcoma's in mice were used. These data originate from NTP technical report 267 (1985), inhalation study in mice exposed by inhalation to 0, 200, or 400 ppm propylene oxide, 6 hr/day, 5 days/week, for 103 weeks. The adjusted rates (males + females) amounted to 0 % in the control group, 0 % in the 200 ppm group and 32.2% in the 400 ppm group. The TD_{50} based on these data amounts to 713 ppm, which corresponds to 1691 mg/m^3.

III Not applicable

IV Based on the TD_{50} of 1671 mg/m^3 the estimated incidence for humans in the occupational situation amounts to $0.5/[1671 \times 7/4 \times 6/8] = 2.3 \times 10^{-4}$ per mg/m^3.

V (1) Neutral: TD_{50} based on inhalation study.
(2) Neutral: the test chemical has been examined and was found positive in rats and mice.
(3) Factor 5 for decreased potency: The results indicate a low degree of malignancy.
(4) Neutral: dose response data available.
(5) Neutral: no data available.
(6) Neutral: no adequate morbidity and mortality studies in man.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $2.3 \times 10^{-4} : 5 = 4.6 \times 10^{-5}$.

1,2-dibromoethane (EDB)

Consulted literature: Gold et al. (1984), WGD rapport RA 5/87 (1987), IARC (1977)

I Not enough epidemiologic data available for quantitative risk assessment in humans.

II 1,2-Dibromoethane is carcinogenic in mice and rats after its oral administration, it produced squamous-cell carcinomas of the forestomach. Results from inhalation studies in rats and mice are available.

TD_{50}-values calculated for various tumour types in different organs have been published (Gold et al., 1984).

We have selected a rat inhalation study performed in the frame of the National Toxicology Program. In this study groups of 100 rats (50 males, 50 females) were exposed to atmospheres containing 0, 10 or 40 ppm EDB for 78-103 weeks. To calculate the TD_{50} we have used the number of animals showing a mixture of different tumours per number of animals being at risk as given by Gold et al., (1984) and assumed that the exposure was 8 hours/day for 5 days/week. The incidences were 7/100, 87/100, 93/100 for the 0, 10 and 40 ppm groups, respectively, males and females taken together.

The TD_{50} was based on the results obtained with the control and the lowest dose animals, as inclusion of the data of the 40 ppm group caused a significant downward departure from linearity of the dose response curve. Based on the results obtained at 0 and 10 ppm the TD_{50} amounts to 27 mg/m^3.

III Not applicable

IV Based on the TD_{50} of 27 mg/m^3 the estimated cancer incidence in humans in the occupational situation amounts to $0.5/[27 \times 7/4] = 1 \times 10^{-2}$ per mg/m^3

V (1) Neutral: TD_{50} based on inhalation study.

(2) Neutral: the test chemical has been examined in rats and mice.

(3) Neutral: induced tumours are of varying degrees of malignancy.

(4) Neutral: dose response data available.

(5) Factor 5 for decreased potency:: available data of toxicokinetic studies point to differences with humans.

(6) Factor 2 for decreased potency: epidemiologic data do not point to increased risks for humans in the occupational situation.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $1 \times 10^{-2} : 10 = 1.0 \times 10^{-3}$.

Acrylonitrile

Consulted literature: Gold et al. (1984), Quast et al. (1980), VROM (1984a), IARC (1987).

I Not enough epidemiologic data for quantitative risk assessment in humans.
According to the IARC (1987) there is limited evidence of carcinogenicity to humans.

II After oral administration as well as inhalation, tumours were induced in central nervous system, Zymbal's gland, small intestine, tongue, mammary glands and stomach of rats.
Results from inhalation studies in rats are available. TD_{50}-values calculated for various tumour types in different organs have been published (Gold et al., 1984).
We have selected a rat inhalation study performed by Quast et al., (1980). The data used for the TD_{50} calculation were taken from a Criteria Document on acrylonitrile, which gives a summary of the study of Quast et al. (VROM, 1984a). In this study groups of 100 male and 100 female rats were exposed to atmospheres containing 0, 44 and 176 mg/m^3 acrylonitrile 6 hours/day, 5 days per week for 2 years. The TD_{50} was calculated for the number of animals (males + females) with primary brain tumours, being 0/200 in the control group, 8/199 in the mid-dose group and 31/199 in the high-dose group (Table 7.2 of the VROM, 1984a publication). The TD_{50} based on these data amounts to 712 mg/m^3.

III Not applicable

IV Based on the TD_{50} of 712 mg/m^3 the estimated incidence for humans in the occupational situation amounts to $0.5/[712 \times 7/4 \times 6/8] = 5.4 \times 10^{-4}$ per mg/m^3.

V (1) Neutral: TD_{50} based on inhalation study.
(2) Factor 10 for increased potency: Only one species examined.
(3) Neutral: induced tumours are of varying degrees of malignancy.
(4) Neutral: dose response data available.
(5) Neutral: available data of toxicokinetic studies do not point to dissimilarities with humans.

(6) Factor 5 for increased potency: Epidemiologic data suggest that acrylonitrile might be carcinogenic to humans.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $5.4 \times 10^{-4} \times 50 = 2.7 \times 10^{-2}$.

Epichlorohydrin

Consulted literature: Laskin et al. (1980), Van Duuren et al. (1974), VROM, (1984b), IARC (1987)

I Not enough epidemiologic data for quantitative risk assessment in humans.

II Epichlorohydrine was tested in rats by oral administration, inducing papillomas and carcinomas of the forestomach and by inhalation, inducing papillomas and carcinomas of the nasal cavity. It was also tested in mice by skin application and by subcutaneous (producing local sarcomas) and intraperitoneal (producing mouse-lung tumours) injection; it gave negative results after continuous skin painting but was active as an initiator on skin.

TD_{50}-values calculated for various tumour types in different organs (malignant lymphoma's, liver tumours) have been published for the inhalation study (Gold et al., 1984).

We have calculated the TD_{50} for the same inhalation study using the data of the lifetime study published by Laskin et al. (1980). In this study male rats were exposed by inhalation to atmospheres containing epichlorohydrin at levels of 0, 10 or 30 ppm for their lifetime (16-136 weeks), 6 hours per day and 5 days per week.

Unadjusted TD_{50} based on the incidence of squamous cell carcinomas in the 0 (0/100), 10 (0/100) and 30 (1/100) ppm group amounts to 1931 ppm, which corresponds to 7588 mg/m^3. Not sufficient data for adjustment of the TD_{50} according to the criteria agreed on.

III Not applicable.

IV Based on the TD_{50} of 7588 mg/m^3, the estimated incidence for humans in the occupational situation amounts to $0.5/[7588 \times 7/4 \times 6/8] = 5 \times 10^{-5}$ per mg/m^3.

V (1) Neutral: TD_{50} based on inhalation study.

(2) Neutral: The test chemical appears carcinogenic in the two species examined.

(3) Neutral: induced tumours are of varying degrees of malignancy.

(4) Neutral: dose-response data available.

(5) Neutral: no data available.

(6) Factor 5 for increased potency: Epidemiologic data give a suggestion that epichlorohydrin might be able to induce lung cancer in humans (VROM, 1984b).

VI The estimated incidence per mg/m^3 for humans in the occupational situation amounts to $5 \times 10^{-5} \times 5 = 2.5 \times 10^{-4}$.

Potassium bromate

Consulted literature: IARC (1986)

I No epidemiologic data for quantitative risk assessment in humans are available.

II Only one carcinogenicity with rats has been found in the literature (IARC, 1986)

Adequate data are available for calculating the TD_{50} according to the "Summary Incidence Method". Data used were taken from a paper by Kurokawa et al., 1983 cited in IARC 40, 1986, 211: Groups of 52-53 male and 52-53 female Fischer-344 rats were administered 0, 250 or 500 mg/l potassium bromate in the drinking water for 110 weeks. Treatment-related increases in the incidence of renal adenocarcinomas were males: 3/53 (controls), 24/53 (low-dose), 24/53 (high-dose); females: 0/47 (controls), 21/50 (low-dose) and 36/49 (high-dose). The TD_{50} based on the incidence of renal adenocarcinomas in control and low-dose group (males and females combined) amounts to 16.89 mg/kg body weight/day. After application of the correction factor f^2 ($f = \frac{110}{104}$), the TD_{50} amounts to 18.9 mg/kg b.w./day or to 54 mg/m^3 in case of exposure by inhalation.

III Not applicable.

IV Based on the TD_{50} of 54 mg/m^3 the estimated cancer incidence in humans in the occupational situation amounts to $0.5/[54 \times 7/5 \times 7/4 \times 24/8 = 1.3 \times 10^{-3}$ per mg/m^3.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study.

(2) Factor 10 for increased potency: Only data from one animal species are available.

(3) Neutral: Induced tumours were of varying degrees of malignancy.

(4) Neutral: dose response data available.

(5) Neutral: no data available.

(6) Neutral: no data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $1.3 \times 10^{-3} \times 50 = 6.5 \times 10^{-2}$.

1-Methyl-3-nitro-1-nitrosoguanidine (MNNG)

Consulted literature: IARC (1974, 1987), Gold et al. (1984), Gold et al. (1986b), Arffmann et al. (1981).

I No epidemiologic data available.

II MNNG is carcinogenic in all species tested: mouse, rat, hamster, rabbit and dog. Following its oral administration, papillomas and squamous-cell carcinomas of the oesophagus and forestomach, adenocarcinomas of the stomach, small intestine and large bowel, and sarcomas of the gastrointestinal tract were reported. After subcutaneous injection of mice, it produced lung and liver tumours and haemangioendotheliomas. After intrarectal instillation in rats and guinea pigs and after intrauterine and intravaginal application to rats, it produced local tumours. Data after oral (drinking water) administration are available for calculating the TD_{50} according to the "Summary Incidence Method". Data were taken from a study of Arffmann et al. (1981). Groups of 30 male Wistar rats, 6-7 weeks old, were given continuously MNNG in drinking water for 32 weeks, followed by 55 weeks without MNNG. Dose levels 0 mg/L, 20 mg/L and 83 mg/L. Unadjusted TD_{50} based on the incidence of gastrointestinal tumours in the 0 (0/30), 20 (10/30) and 83 (20/30) mg/L group amounts to 1.00 mg/kg bw/day. With the correction of $(87/104)^2 = 0.699$, the TD_{50} amounts to 0.699 mg/kg bw/day or to 2.43 mg/m^3 in case of exposure by inhalation.

III Not applicable

IV Based on the TD_{50} of 2.43 mg/m^3 b.w. the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[2.43 \times 7/5 \times 7/4 \times 24/8] = 2.8 \times 10^{-2}$ per mg/m^3.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity.

(2) Neutral: the availability of several adequately conducted positive carcinogenicity studies allowed the selection of a relevant TD_{50}-value.

(3) Neutral: tumours malignant.

(4) Neutral: dose-response data available

(5) Neutral: not enough data available.

(6) Neutral: no data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $2.8 \times 10^{-2} \times 5 = 1.4 \times 10^{-1}$.

1,1-dimethylhydrazine

Consulted literature: Gold et al. (1984), IARC (1974, 1987), WGD report (1987), Toth (1973).

I Not enough epidemiological data for risk assessment in humans available. One negative cohort study is known in the literature.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III Data after oral administration by drinking water are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Toth (1973). Administration of 0.01% 1,1-dimethylhydrazine in the drinking water of Swiss albino mice [50 males and 50 females, (5 weeks old)] during their whole lifespan (males: 67 weeks, females: 77 weeks) resulted in angiosarcomas of the blood vessels. Incidence amounting to 6/220 in controls, and 79/100 in the dose group (males and females combined). Other tumours observed inluded tumours of the lungs, kidneys and liver.
The TD_{50} based on these data amounts to 8.32 mg/kg bw/day. With the correction of $(67/104)^2 = 0.42$, the TD_{50} amounts to 3.45 mg/kg bw/day or to 2.18 mg/m^3 in case of exposure by inhalation.

IV Based on the TD_{50} of 2.18 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[2.18 \times 7/5 \times 7/4 \times 24/8] = 3.1 \times 10^{-2}$.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study. 1,1-Dimethylhydrazine is absorbed rapidly through the skin of dogs and appears in the blood within 30 seconds (IARC, 1974).

(2) Neutral: the test chemical has been examined in two animal species (mouse and rat). However, it is noticed that according to IARC (1974) the observation of only a few liver tumours in rats after oral administration does not allow a proper evaluation of the carcinogenicity in rats.

(3) Neutral: tumours malignant.

(4) Factor 10 for increased potency: only one dose examined.

(5) Neutral: not enough data available.

(6) Neutral: not enough data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $3.1 \times 10^{-2} \times 50 = 1.6$.

1,2-Dichloroethane

Consulted literature IARC (1979c, 1987), Gold et al. (1984), WGD-report 87-27-23 (1987).

I No epidemiological data in humans available.

II Data after oral administration by gavage are available for calculating the TD_{50} according to the "Summary Incidence Method". Data were taken from a NCI study as published in IARC Monograph (1979, page 437-439). Groups of 50 male and 50 female B63CF1 mice, 5 weeks old, were administered 1,2-dichloroethane in corn oil by gavage on 5 days/week for 78 weeks. A group of 20 mice served as matched controls. High-dose males received 150 mg/kg/day for 8 weeks and then 200 mg/kg bw/day for 70 weeks, followed by 13 weeks without treatment. High-dose females received 250 mg/kg/day for 8 weeks, 400 mg/kg bw/day for 3 weeks, and 300 mg/kg bw for 67 weeks, followed by 13 weeks without treatment. Low-dose males received 75 mg/kg/day for 8 weeks and then 100 mg/kg bw/day for 70 weeks, followed by 12 weeks without treatment. Low-dose females received 125 mg/kg/day for 8 weeks, 200 mg/kg bw/day for 3 weeks, and 150 mg/kg bw for 67 weeks, followed by 13 weeks without treatment. Increased incidences of the following neoplasms were observed: mammary adenocarcinomas, uterine adenocarcinomas, endometrial stromal neoplasms of the uterus, and squamous-cell carcinomas of the forestomach in females; lung adenomas and malignant histiocytic lymphomas in males and females; and hepatocellular carcinomas in male mice. Number of animals with tumours and the numbers of animals examined histopathologically were 4/19 in control males, 15/46 in the low-dose males, 28/47 in the high-dose males, 6/20 in control females, 33/50 in low-dose females and 29/48 in the high-dose females.

The not corrected TD_{50} based on the male data amounts to 123 mg/kg bw/day. With the correction of $(90/104)^2 = 0.75$, the estimated TD_{50} amounts to 92 mg/kg bw/day or to 320 mg/m^3 in case of exposure by inhalation.

III Not applicable

IV Based on the TD_{50} of 320 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[320 \times 7/5 \times 7/4 \times 24/8] = 2.1 \times 10^{-4}$.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study. Inhalation studies were performed (Gold et al. 1984), but data are not available.

(2) Neutral: The test chemical appears carcinogenic in the two species examined.

(3) Neutral: Tumours malignant.

(4) Neutral: Dose-response data available

(5) Neutral: No enough data available.

(6) Neutral: not data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $2.1 \times 10^{-4} \times 5 = 1.1 \times 10^{-3}$.

1,3,-propanesultone

Consulted literature IARC (1974, 1987), Ulland et al. (1971). No data from Gold et al. (1984, 1986) available.

I No epidemiologic data for risk assessment in humans available.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III Data are available on oral administration (rat), subcutaneous and/or intramuscular administration (mouse and rat). Data after oral (by gavage) administration are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Ulland et al. (1971). 1,3-Propane sultone was given twice weekly at doses of 56 mg/kg bw for 32 weeks and 28 mg/kg bw for 60 weeks. The incidence of gliomas in animals treated with 28 mg/kg bw for 60 weeks amounted to 27/52 (males and females combined). The incidence in controls was 0/6 and 1/6 in males and females, respectively. In addition, several rats had leukaemia, ear duct tumours and adenocarcinomas of the small intestine.
The not corrected TD_{50} based on the data of the 28 mg/kg bw exposure amounts to 8.59 mg/kg bw/day. With the correction of $(60/104)^2$ = 0.33, the estimated TD_{50} amounts to 2.86 mg/kg bw/day or to 8.17 mg/m^3 in case of exposure by inhalation.

IV Based on the TD_{50} of 8.17 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[8.17 \times 7/5 \times 7/4 \times 24/8] = 8.3 \times 10^{-3}$.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study.

(2) Neutral: The test chemical appears carcinogenic in the two species examined (rat and mouse).

(3) Neutral: tumours malignant.

(4) Factor 10 for increased potency: Only one dose examined.

(5) Neutral: not enough data available.

(6) Neutral: no data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $8.3 \times 10^{-3} \times 50 = 4.2 \times 10^{-1}$.

2-methylaziridine (propylene imine)

Consulted literature IARC (1975, 1987), Ulland et al. (1971). No data from Gold et al. (1984, 1986) available.

I No epidemiologic data for risk assessment in humans available.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III Only data after oral (by gavage) administration are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Ulland et al. (1971). 2-Methylaziridine given twice weekly to rats at doses of 20 mg/kg bw for 28 weeks or 10 mg/kg bw for 60 weeks produced gliomas, ear-duct squamous-cell carcinomas, intestinal adenocarcinomas, leukaemias, breast tumours and miscellaneous tumours (not specified).
The incidence of tumour bearing animals (males plus females) amounted to 22/52, 37/52 and 2/12 for the high dose (2 x 20 mg, 28 weeks), the low dose (2 x 10 mg, 60 weeks) and the control groups respectively. The TD_{50} based on these data amounts to 2.56 mg/kg bw/day. With the correction of $(60/104)^2 = 0.33$, the TD_{50} amounts to 0.85 mg/kg bw/day or to 2.43 mg/m^3 in case of exposure by inhalation.

IV Based on the TD_{50} of 2.43 mg/mg^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[2.43 \times 7/5 \times 7/4 \times 24/8] = 2.8 \times 10^{-2}$ per mg/m^3.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study.
(2) Factor 10 for increased potency: Only data from one animal study are available.
(3) Neutral: tumours malignant.
(4) Factor 5 for increased potency: No real dose response data available.
(5) Neutral: No data available.
(6) Neutral: No data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $2.8 \times 10^{-2} \times 250 = 7$.

2-nitropropane

Consulted literature: Gold et al. (1984), IARC (1982c), Griffin et al. (1980, 1981), WGD-report (1985), Lewis et al. (1979).

I Not enough epidemiologic data for quantitative risk assessment in humans.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III An inhalation study in rabbits (negative result, Lewis et al., 1979) was considered to be inadequate for evaluation (IARC, 1982). Evidence for the carcinogenicty of 2-nitropropane in rats (inhalation studies) by Griffin et al. (1980, 1981) and Lewis et al. (1979). The study of Lewis et al. (1979) was not used, because a calculation of the TD_{50} was not possible (Two dose levels). At the highest dose level all rats had liver tumours, and at the lowest dose level none of the rats had a tumour).

Data after inhalatoir administration are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Griffin et al. (1980). Group of 125 male and 125 female rats were exposed by inhalation to 2-nitropropane at a concentration of 0 or 25 ppm for 7 hours per day, 5 days per week over a period of 22 months (95 weeks). Focal areas of hepatocellular nodules were noted in 3/250 control animals and 13/249 exposed animals. The TD_{50} based on these data amounts to 417.05 ppm. With the correction of $(95/104)^2 = 0.83$, the estimated TD_{50} amounts to 346 ppm, which corresponds to 1263 mg/m^3.

IV Based on the TD_{50} of 1263 mg/m^3 The estimated incidence for humans in the occupational situation expressed in mg per m^3 amounts to $0.5/(1263 \times 7/4 \times 7/8) = 0.5/1933.97 = 2.5 \times 10^{-4}$ per mg/m^3.

V (1) neutral: TD_{50} based on inhalation study.

(2) Factor 10 for increased potency: The test chemical has been examined in rat (positive result) and rabbit [negative result, exposure was short (6 months) and small number of animals].

(3) Neutral: Tumours not malignant, but in the study of Lewis et al. (1979) all 10 rats exposed to 754 mg/m^3 (207 ppm) for 6 months exhibited multiple hepatocellular carcinomas. No lesions were noted after exposure to 98 mg/m^3 (27 ppm).

(4) Neutral: Dose response data available.

(5) Neutral: not enough data available.

(6) Neutral: not enough data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $2.5 \times 10^{-4} \times 10 = 2.5 \times 10^{-3}$.

3,3'-dichlorobenzidine

Consulted literature Gold et al. (1984), Stula et al. (1975), IARC (1982c, 1987).

I Not enough epidemiologic data for quantitative risk assessment in humans.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III Following oral administration 3,3'-dichlorobenzidine liver-cell tumours in mice, hepatocellular carcinomas in dogs, mammary and Zymbal-gland tumours in rats and carcinomas of the urinary bladder in hamsters and dogs. Increased incidences of leukaemias were observed in rats following oral administration and in mice following transplacental exposure. Also data on subcutaneous and/or intramuscular administration are available, but according to IARC (1982) these data are inadequate.

Data after oral (diet) administration are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Stula et al. (1975). A group of 44 male and 44 female 5-week-old ChR-CD rats were given a diet containing 1000 ppm 3,3'-dichlorobenzidine. The control group consisted of 44 males and 44 females. As the group size of the 1000 ppm group was spontaneously reduced to six rats when 69 weeks on test, a terminal sacrifice was conducted on this group. Incidence of tumours observed in male treated rats: 9/44 granulocytic leukaemias, 7/44 mammary adenocarcinomas and 8/44 Zymbal gland carcinomas; corresponding incidences in the control group were 2/44, 0/44 and 0/44. Of female rats, 26/44 treated animals developed mammary adenocarcinomas versus 3/44 in control rats.

The TD_{50} based on the mammary adenocarcinomas in females amounts to 42.1 mg/kg bw/day. With the correction of $(69/104)^2 = 0.44$, the TD_{50} amounts to 18.5 mg/kg bw/day, which corresponds to 45 mg/m^3 in case of exposure by inhalation.

IV Based on the TD_{50} of 45 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[45 \times 7/5 \times 7/4 \times 24/8 = 1.5 \times 10^{-3}$ per mg/m^3.

V (1) Factor 5 for increased potency: No inhalation data available.

(2) Factor 10 for increased potency: The test chemical was positive in more than 3 species, but only data from one of these experiments were suitable for calculation of the TD_{50}.

(3) Neutral: malignant tumours.

(4) Factor 5 for increased potency: More than 1 dose examined, but no dose-response data available.

(5) Neutral: not enough data available.

(6) Neutral: not enough data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $1.5 \times 10^{-3} \times 5 \times 10 \times 5 = 3.8 \times 10^{-1}$.

4-aminobiphenyl

Consulted literature: Gold et al. (1984), IARC (1972, 1979b, 1982d, 1987), Clayson et al. (1967).

I Not enough epidemiologic data for quantitative risk assessment in humans.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III 4-Aminobiphenyl is carcinogenic in mice, rats, rabbits and dogs after oral administration, producing principally cancer of the urinary bladder. After subcutaneous administration (rat) the yield of mammary gland and intestinal tumours was significantly raised. Data after oral (by gavage) administration are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Glayson et al. (1967). Administration of 0.5 mg of 4-aminobiphenyl given by stomach tube to C57xIF F_1 hybrid mice 3 times weekly for 50 weeks, followed by 20 weeks without treatment resulted in malignant hepatomas. Incidence amounting to 0/50 in controls, 17/49 in the dose group (males and females combined).
The TD_{50} based on these data amounts to 8.87 mg/kg bw/day. With the correction of $(70/104)^2 = 0.45$, the estimated TD_{50} amounts to 3.99 mg/kg bw/day, which corresponds to 2.54 mg/m^3 in case of exposure by inhalation.

IV Based on the TD_{50} of 2.54 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[2.54 \times 7/5 \times 7/4 \; 24/8] = 2.7 \times 10^{-2}$ per mg/m^3.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study.
(2) Factor 10 for increased potency: The test chemical has been examined and was positive in more than three different species, but only data from one of these experiments were suitable for calculation of the TD_{50}.
(3) Neutral: Tumours malignant.
(4) Factor 5 for increased potency: More doses were examined in other studies, but no dose-response data available.

(5) Neutral: not enough data available.

(6) Factor 10 for increased potency: positive epidemiology, data not adequate for risk evaluation in humans.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $2.7 \times 10^{-2} \times 5 \times 10 \times 5 \times 10 = 67.5$.

4,4'-methylene bis (2-chloroaniline)

Consulted literature IARC (1974, 1987) Stula et al. (1975), Gold et al. (1984).

I Not enough epidemiologic data for risk assessment in humans.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III After oral administration of 4,4'-methylene bis (2-chloroaniline), mice developed haemangiosarcomas and hepatomas; rats developed lung, liver, mammary gland and Zymbal gland tumours and haemangiosarcomas; and dogs developed urinary bladder tumours. Tumours of the lung and liver were produced after subcutaneous injection of rats. Data are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Stula et al. (1975). Administration of 1000 ppm of 4,4'-methylene bis (2-chloroaniline) in the diet of ChR-CD rats (44 males and 44 females) during their whole lifespan (up to 2 years) resulted in lung adenocarcinomas. Incidence amounting to 0/88 in controls, 48/88 in the dose group.
The TD_{50} based on these data amounts to 39.56 mg/kg bw/day, which corresponds to 113 mg/m^3 in case of exposure by inhalation.

IV Based on the TD_{50} of 113 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[113 \times 7/5 \times 7/4 \times 24/8] = 6.0 \times 10^{-4}$.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study.

(2) Factor 10 for increased potency: The test chemical was positive in 3 species but only data from one of these experiments were suitable for calculation of the TD_{50}.

(3) Neutral: tumours malignant.

(4) Factor 5 for increased potency: No real dose-response data available.

(5) Neutral: not enough data available.

(6) Neutral: not enough data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $6.0 \times 10^{-4} \times 250 = 1.5 \times 10^{-1}$.

5-nitroacenaphthene

Consulted literature IARC (1978), Gold et al. (1984), Takemura et al. (1974).

I No epidemiologic data for risk assessment in humans available.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III 5-Nitroacenaphthene is carcinogenic in female rats and female hamsters following its oral administration; it produced mainly adenocarcinomas of the small intestine and mammary carcinomas in female but not in male rats and cholangiomas in female but not in male hamsters. It produced leukaemia and reticulum-cell sarcomas in mice following its intraperitoneal injection. Data after oral (diet) administration are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Takemura et al. (1974) as published in IARC vol. 16 (1978). Of 24 weanling female and 10 male Syrian hamsters that received 1% 5-nitroacenaphthene in their diet continuously for 6 months 7/24 females developed cholangiomas. No tumors were observed in the 20 control females or in the treated male hamsters. The experiment was stopped after 500 days.
The TD_{50} based on the data of the females amounts to 359.94 mg/kg bw/day after application of the correction factor (0.47), which corresponds to 458 mg/m^3 in case of exposure by inhalation.

IV Based on the TD_{50} of 458 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[458 \times 7/5 \times 7/4 \times 24/8] = 1.5 \times 10^{-4}$ pr mg/m^3.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study.
(2) Neutral: the test chemical appears carcinogenic in the two species examined.
(3) Neutral: tumours malignant.
(4) Factor 10 for increased potency: Only one dose examined.
(5) Neutral: not enough data available.
(6) Neutral: no data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $1.5 \times 10^{-4} \times 50 = 7.5 \times 10^{-3}$.

Benzidine

Consulted literature Gold et al. (1984, 1986), IARC (1972, 1982a, 1982b, 1987).

I There is sufficient evidence for carcinogenicty to humans. However, there are no data for quantitative risk assessment in humans.

II No animal data are available for calculating the "Peto TD_{50}" according to the "Summary Incidence Method".

III Benzidine is carcinogenic to mice, rats, hamsters and dogs after oral-, subcutaneous and/or intramuscular-, intraperitoneal- and inhalatoir administration. Following oral administration of benzidine and its hydrochloride, increases in the incidence of benign and malignant liver neoplasms were observed in mice and hamsters and of mammary cancer in rats; benzidine induced bladder carcinomas in dogs. Following subcutaneous administration of benzidine and its sulphate to rats, Zymbal-gland tumours were observed. After intraperitoneal administration of benzidine to rats, an increase in the incidence of mammary-gland and Zymbal-gland neoplasms was observed. Data after inhalatoir administration are available for calculating the TD_{50} according to the "stripped TD_{50}". Data were taken from a study of Zabezhinsky (1970) as published in the IARC vol. 29 (1982b). A group of 48 white outbred rats (Rappolovo stock) of both sexes, weighing 100-120 g. was exposed to benzidine in inhalation chambers for 4 hours/day, 5 days/week over 20 months. Total dose 27 mg/rat. The first myelogenous leukaemia was found 13 months after the start of the experiment, at which time 28 rats were still alive. By the end of the study (28 months) 5 myeloid leukaemias, 2 breast fibroadenomas, 1 squamous-cell cancer of the Zymbal gland, 1 hepatoma and 1 breast adenocarcinoma were found in 8 animals. Mammary adenomas were found in 2/21 control animals.

The TD_{50} based on these data amounts to 1.19 and 0.198 mg/kg bw/day for 4 h and 24 h respectively. After application of the correction factor f^2 ($f=\frac{28}{24}$), the estimated TD_{50} (24 h) amounts to 0.27 mg/kg bw/day, which corresponds to 0.77 mg/m^3 in case of exposure by inhalation.

IV Based on the TD_{50} of 0.77 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[0.77 \times 7/5 \times 7/4 \times 24/8] = 8.8 \times 10^{-2}$ per mg/m^3.

V (1) Neutral: Inhalation studies were performed.

(2) Neutral: The availability of several adequately conducted positive carcinogenicity studies allowed the selection of a relevant TD_{50}-value.

(3) Neutral: Tumours malignant.

(4) Neutral: Dose response data are available.

(5) Neutral: no data available.

(6) Factor 10 for increased potency: There is sufficient evidence for carcinogenicty for humans.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $8.8 \times 10^{-2} \times 10 = 8.8 \times 10^{-1}$.

Cadmium chloride.

Consulted literature: IARC (1973, 1976, 1987), Gold et al. (1984), Taneka et al. (1983).

I Not enough epidemiologic data for risk assessment in humans.

II Cadmium chloride produced local sarcomas and pancreatic islet-cell tumours in rats after subcutaneous injection, testicular tumours in mice and rats after subcutaneous administration. Administration up to 50 mg/kg cadmium chloride in the diet to rats did not increase the incidence of tumours. Cadmium chloride produced a dose-dependent increase in the incidence of lung carcinomas in rats after exposure by inhalation. Data after exposure by inhalation are available for calculating the TD_{50} according to the "Summary Incidence Method". Data were taken from a study of Taneka et al. (1983). Groups of male inbred W rats were exposed by inhalation to atmospheres containing nominated Cd concentrations of 12.5, 25 or 50 $\mu g/m^3$. The exposure time amounted to 18 months, 23 hours/day, 7 days/week. The survivors were killed 13 months after the end of the exposure time. The incidence of lung carcinomas was 25/38 in the group exposed to 50 μg Cd/m^3, 20/38 in the 25 μg Cd/m^3 group, 6/39 in the 12.5 μg Cd/m^3 group, and 0/38 in the control group.
The TD_{50} based on these male data amounts to 50.68 $\mu g/m^3$ = 0.051 mg/m^3 and to 0.085 mg/m^3 after application of the correction factor f^2 ($f = \frac{31}{24}$).

III Not applicable.

IV Based on the TD_{50} of 0.085 mg/m^3 the estimated TD_{50} for humans in the occupational situation amounts to 0.085 x 70/40 x 7/5 mg/m^3 = 0.207 mg/m^3 applying the assumptions enumerated in section 4.2.2.
The estimated incidence for humans in the occupational situation expressed in mg per m^3 amounts to 0.5/0.207 = 2.4 per mg/m^3.

V (1) Neutral: TD_{50} based on an inhalation study.
(2) Neutral: the test chemical appears carcinogenic in the two species examined.
(3) Neutral: tumours malignant.

(4) Neutral: Dose-response data available

(5) Neutral: not enough data available.

(6) Factor 2 for increased potency: According to IARC limited evidence.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of 2.4 x 2 = 4.8.

Styrene oxide

Consulted literature: IARC (1976, 1979a, 1982d, 1985, 1987). Gold et al. (1987).

I Not enough epidemiologic data for risk assessment in humans.

II Styrene oxide was tested by intragastric intubation in Sprague-Dawley rats and induced squamous-cell carcinomas and papillomas of the forestomach. Prenatal exposure followed by postnatal oral administration of styrene oxide to BDIV rats also produced squamous-cell carcinomas and increased the incidence of papillomas of the forestomach. No increase in the incidence of skin tumours was observed in mice of two strains following topical application of styrene oxide. Data after intragastric intubation administration are available for calculating the TD_{50} according to the "Summary Incidence Method". Data were taken from Maltoni et al. (1979). Groups of 40 male and 40 female Sprague-Dawley rats, 13 weeks old, received 0, 50 or 250 mg/kg bw styrene oxide by intragastric intubation once daily, on four to five days per week for 52 weeks. This period was followed by 83 weeks without treatment. The incidences (referring to corrected numbers of animals alive at 51 weeks) of squamous-cell carcinomas (invasive plus in situ) were 0/65 in control, 12/62 in low-dose, and 27/58 in high-dose males plus females.
The TD_{50} based on these data amounts to 175.63 mg/kg bw/day and to 296 mg/kg b.w./day after application of the correction factor f^2 ($f = \frac{135}{104}$), which corresponds to 846 mg/m^3 in case of exposure by inhalation.

III Not applicable

IV Based on the TD_{50} of 846 mg/m^3 the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[846 \times 7/5 \times 7/4 \times 24/8] = 8.0 \times 10^{-5}$ per mg/m^3.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study.

(2) Factor 10 for decreased potency: The test chemical appears carcinogenic in the rat (oral application) and negative in the mouse (skin application).

(3) Neutral: Tumours malignant.

(4) Neutral: Dose-response data available

(5) Neutral: Not enough data available.

(6) Neutral: Not enough data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $8.0 \times 10^{-5} \times 0.5 = 4.0 \times 10^{-5}$.

3,3'-dimethylbenzidine (o-tolidine)

Consulted Literature: IARC (1972), Hazleton study (October 16, 1985, pag. 21).

I Not enough epidemiologic data for risk assessment in humans.

II 3,3'-dimethylbenzidine (0-tolidine) is a systemic carcinogen in the rat when given subcutaneously. In feeding experiments, the substance failed to produce tumours in hamsters. Data after oral (drinking water) administration are available for calculating the TD_{50} according to the "Summary Incidence Method". Data were taken from a Hazleton study (1986). F344 rats (6-8 weeks old) received 3,3'-dimethylbenzidine in drinking water. Dose schedule 0% (70 males and 70 females), 0.003% (45 males and 45 females), 0.007% (75 males and 75 females) and 0.015% (70 males and 70 females). The experiment was terminated after 60 weeks. Neoplasms were seen in the intestinal tract, liver, oral cavity, skin, lungs, Zymbal's gland and preputial glands. Nonneoplastic lesions were seen in liver, bone marrow, spleen, adrenal gland, parathyroid, mandibular lymph node, skin, heart, preputial/clitoral gland, lungs, Zymbal's gland and kidneys. Number of animals with malignant neoplasm(s) and the numbers of animals examined were 4/140 in control animals, 34/90 in the 0.003% group (1.6 mg/kg bw/day), 112/150 in the 0.007% group (3.75 mg/kg bw/day), and 103/140 in the 0.015% group (8.05 mg/kg bw/day). The TD_{50} was calculated excluding the high dose group.
The not corrected TD_{50} based on these data amounts to 1.92 mg/kg bw/day. With the correction of $(60/104)^2 = 0.33$, the estimated TD_{50} amounts to 0.64 mg/kg bw/day, which corresponds to 1.83 mg/m^3 in case of exposure by inhalation.

III Not applicable

IV Based on the TD_{50} of 1.83 mg/m^3. the estimated cancer incidence for humans in the occupational situation amounts to $0.5/[1.83 \times 7/5 \times 7/4 \times 24/8] = 3.7 \times 10^{-2}$.

V (1) Factor 5 for increased potency: TD_{50} based on oral carcinogenicity study.

(2) Factor 10 for decreased potency: The test chemical is carcinogenic in one species and not carcinogenic in the second species.

(3) Neutral: Tumours malignant.

(4) Neutral: Dose-response data available

(5) Neutral: Not enough data available.

(6) Neutral: No data available.

VI Application of the above weighing factors results in an estimated incidence per mg/m^3 for humans in the occupational situation of $3.7 \times 10^{-2} \times 0.5 = 1.9 \times 10^{-2}$.

Dichloromethane (DCM) (suspect carcinogen!)
Consulted literature: IARC (1986a)

I Not enough epidemiologic data for quantitative risk assessment in humans.

II Adequate data are available for calculating the TD_{50} according to the "Summary Incidence Method". Data used were taken from an experiment performed in the frame of the National Toxicology Program, 1986 and cited in IARC 41, 1986a, 57: Groups of 50 male and 50 female $B6C3F_1$ mice were exposed to 0, 6940 or 13880 mg/m^3 DCM by inhalation for six hours per day on five days per week for 102 weeks and were killed after 104 weeks on study. The incidences of alveolar/bronchiolar adenomas amounts to: 3/50 (control), 19/50 (low dose) and 24/50 (high dose) and 2/50 (control), 23/48 (low dose) and 28/48 (high dose) in males en females respectively. The TD_{50} based on the incidence of lung tumours (alveolar/bronchiolar carcinomas) amounts to 11441 mg/m^3 x 102/104 = 11221 mg/m^3.

III Not applicable.

IV Based on the TD_{50} of 11221 mg/m^3 the estimated cancer incidence in humans in the occupational situation amounts to $0.5/[11221 \times 7/4 \times 6/8] = 3.4 \times 10^{-5}$ per mg/m^3.

V (1) Neutral: TD_{50} based on inhalation study.
(2) Neutral: DCM was carcinogenic in mice and rats.
(3) Neutral: induced tumours were of varying degrees of malignancy.
(4) Neutral: dose response data available.
(5) Neutral: not enough conclusive data available.
(6) Neutral: inadequate data.

VI Based on the above considerations the estimated incidence per mg/m^3 for humans in the occupational situation amounts to 3.4×10^{-5}.

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Table 3.1 Proposed system for ranking carcinogens

Factor	Score
A. Number of different species affected	
Two or more	15
One	5
B. Number of histogenetically different types of neoplasms in one or more species	
Three or more	15
Two	10
One	5
C. Spontaneous incidence in appropriate control groups of neoplasms induced in treated groups	
Less than 1 percent	15
1 to 10 percent	10
10 to 20 percent	5
More than 20 percent	1
D. Dose-response relationships (cumulative oral dose equivalents per kilogram of body weight per day for 2 years)*	
Less than 1 microgram	15
1 microgram to 1 milligram	10
1 milligram to 1 gram	5
More than 1 gram	1
E. Malignancy of induced neoplasms	
More than 50 percent	15
25 to 50 percent	10
Less than 25 percent	5
No malignancy	1
F. Genotoxicity, measured in an appropriate battery of tests	
Positive	25
Incompletely positive	10
Negative	0

*Based on estimated consumption of 100 grams of diet per kilogram of body weight. Scoring could also be developed for inhalation or other appropriate routes.

Squire (1981)

Table 3.2 Ranking animal carcinogens into five classes according to total factor score

Total factor score	Carcinogen class	Regulatory options
86 to 100	I	Restrict or ban
71 to 85	II	
56 to 70	III	
41 to 55	IV	
Less than 41	V	Several options (no action, limited use, labeling, public education)

Squire (1981)

Table 3.3 Approximate rank of ten animal carcinogens based on the proposed system

Carcinogen	Score	Rank
Aflatoxin	100	I
Dimethylnitrosamine	95	I
Vinyl chloride	90	I
Tris(2,3-dibromopropyl)-phosphate (Tris)	90	I
2-Naphthylamine	81	II
Chloroform	65	III
NTA	51	IV
Chlordane	40	V
Saccharin	36	V
DDT	31	V

Squire (1981)

Table 3.4 Response–dependent classification of carcinogenic potency

Type of evidence		Weak carcinogen		Strong carcinogen
Short-term tests	Biochemical assays Submammalian assays Mammalian mutagenic assays Mammalian cytogenetic assays Mammalian cell transformation assays	Limited positive tests		Many positive tests
Experimental animal tests	Histological evidence of neoplasia	+		
	Visible tumors			+
	Single target tissue	+		
	Multiple target tissues			+
	Single species	+		
	Multiple species			+
	Neoplasia appears late	+		
	Neoplasia appears early and/or leads to death			+
	Prolonged exposure	+		
	Short duration of exposure			+
Epidemiologic studies	Negative	+		
	Questionable	+	or	+
	Positive			+

Theiss (1983)

Table 5.1 Survey of the estimated cancer incidences for humans in the occupational situation without (-) and with (+) weighing factors

	Incidence per mg/m^3 (-)	weighing factors (1)	(2)	(3)	(4)	(5)	(6)	incidence per mg/m^3 (+)
vinyl chloride monomer	1.4×10^{-4}							1.4×10^{-4}
benzene	2.1×10^{-4}							2.1×10^{-4}
ethylene oxide	1.9×10^{-4}	1	1	1	1	1	5↑	9.5×10^{-4}
propylene oxide	2.3×10^{-4}	1	1	5↓	1	1	1	4.6×10^{-5}
1,2-dibromoethane	1.0×10^{-2}	1	1	1	1	5↓	2↓	1.0×10^{-3}
acrylonitrile	5.4×10^{-4}	1	10↑	1	1	1	1	2.7×10^{-2}
epichlorohydrin	5×10^{-5}	1	1	1	1	1	5↑	2.5×10^{-4}
potassium bromate	1.3×10^{-3}	5↑	10↑	1	1	1	1	6.5×10^{-2}
MNNG	2.8×10^{-2}	5↑	1	1	1	1	1	1.4×10^{-1}
1,1-DMH	3.1×10^{-4}	5↑	1	1	10↑	1	1	1.6
1,2-dichloroethane	2.1×10^{-3}	5↑	1	1	1	1	1	1.1×10^{-3}
1,3-propanesultone	8.3×10^{-2}	5↑	1	1	10↑	1	1	4.2×10^{-1}
2-methylaziridine	2.8×10^{-4}	5↑	10↑	1	5↑	1	1	7
2-nitropropane	2.5×10^{-3}	1	10↑	1	1	1	1	2.5×10^{-3}
3,3'-dichlorobenzidine	1.5×10^{-2}	5↑	10↑	1	5↑	1	1	3.8×10^{-1}
4-aminobiphenyl	2.7×10^{-4}	5↑	10↑	1	5↑	1	10↑	67.5
4,4'-methylene bis (2-chloroaniline)	6.0×10^{-4}	5↑	10↑	1	5↑	1	1	1.5×10^{-1}
5-nitroacenaphthene	1.5×10^{-4}	5↑	1	1	10↑	1	1	7.5×10^{-3}
benzidine	8.8×10^{-2}	1	10↑	1	1	1	10↑	8.8×10^{-1}
cadmiumchloride	2.4	1	1	1	1	1	2↑	4.8
styrene oxide	8.0×10^{-5}	5↑	10↓	1	1	1	1	4.0×10^{-5}
3,3'-dimethylbenzidine	3.7×10^{-2}	5↑	10↓	1	1	1	1	1.9×10^{-2}
dichloromethane	3.4×10^{-5}	1	1	1	1	1	1	3.4×10^{-5}

Table 5.2 Ranking of genotoxic carcinogens according to estimated cancer incidence per mg/m^3 for humans in the occupational situation without (-) and with (+) weighing factors*

(-)		(+)	
Class B1		Class B1	
cadmiumchloride	2.4	4-aminobiphenyl	67.5
benzidine	8.8×10^{-2}	benzidine	8.8×10^{-1}
3,3'-dimethylbenzidine	3.7×10^{-2}	2-methylaziridine	7
1,1-DMH	3.1×10^{-2}	cadmiumchloride	4.8
MNNG	2.8×10^{-2}	1,1-DMH	1.6
2-methylaziridine	2.8×10^{-2}	1,3-propanesultone	4.2×10^{-1}
4-aminobiphenyl	2.7×10^{-2}	3,3'-dichlorobenzidine↑	3.8×10^{-1}
1,2-dibromoethane↓	1.0×10^{-2}	4,4'-methylene bis (2-chloroaniline)↑	1.5×10^{-1}
1,3-propanesultone	8.3×10^{-3}	MNNG	1.4×10^{-1}
Class B2		potassium bromate↑	6.5×10^{-2}
3,3'-dichlorobenzidine↑	1.5×10^{-3}	acrylonitrile↑	3.7×10^{-2}
potassium bromate↑	1.3×10^{-3}	3,3'-dimethylbenzidine	1.9×10^{-2}
acrylonitrile↑	7.3×10^{-4}	5-nitroacenaphthene↑	7.5×10^{-3}
4,4'-methylene bis (2-chloroaniline)↑	6.0×10^{-4}	Class B2	
2-nitropropane	2.5×10^{-4}	2-nitropropane	2.5×10^{-3}
propylene oxide	2.3×10^{-4}	1,2-dichloroethane	1.1×10^{-3}
benzene	2.1×10^{-4}	1,2-dibromoethane↓	1.0×10^{-3}
1,2-dichloroethane	2.1×10^{-4}	ethylene oxide	9.5×10^{-4}
ethylene oxide	1.9×10^{-4}	epichlorohydrin	2.5×10^{-4}
5-nitroacenaphthene↑	1.5×10^{-4}	benzene	2.1×10^{-4}
vinyl chloride monomer	1.4×10^{-4}	vinyl chloride monomer	1.4×10^{-4}
styrene oxide↓	8.0×10^{-5}	propylene oxide	4.6×10^{-5}
epichlorohydrin	5×10^{-5}	Class B3	
Class B3		styrene oxide↓	4.0×10^{-5}
dichloromethane	3.4×10^{-5}	dichloromethane	3.4×10^{-5}

* Arrows indicate that the application of weighing factors resulted in a shift as to risk class for the chemical at issue; ↑: higher risk class ↓: lower risk class.

APPENDIX 4-1

CALCULATION OF THE TD_{50}

DEFINITION: For any particular sex, strain, species and set of experimental conditions, the TD_{50} is the dose rate (in mg/kg b.w./day)* that, if administered chronically for a standard period - the "standard lifespan" of the species - will halve the mortality corrected estimate of the probability of remaining tumourless throughout that period.

first step: Experiments must meet the following set of standard criteria, in order to be able to compare the TD_{50} values derived from results of quite divers experimental designs as well as the author's choice of information to report.

A) animals on test were mammals.
B) administration was begun early in life. (100 days or less for rats, mice and hamsters.)
C) route of administration was diet, water, gavage, inhalation, intravenous or intraperitoneal injection.
D) test agent was administered alone.
E) exposure was chronic, with not more than 7 days between administrations.
F) duration of exposure was at least one-fourth the standard lifespan for that species.
G) duration of experiment was at least half the standard lifespan for that species.
H) research design included a control group.
I) research design included at least 5 animals per group.
J) pathology data were reported for the number of animals with tumours rather than the total number of tumours.
K) results reported were original data.

* For our purpose i.e. risk estimation in the occupational situation we prefer to express the TD_{50} in mg/m^3.

second step: The selection of tumour sites on which to base TD_{50}'s

A TD_{50} can be calculated for each category of neoplasm, benign or malignant, which an author evaluates as treatment related.
In addition, a TD_{50} can be estimated for the category "all tumour bearing animals" wherever this is reported.

third step: Estimation of Mean (lifelong) mg/kg b.w./day.

By assuming 100 % absorption, and adopting a set of standard values for each sex/species group which includes factors for daily food, water and air intake and average weight (Table 1, next page), we convert dose to mg/kg body weight/day.

$$\text{dose rate} = \frac{\text{dose x intake/day x number of doses/week}}{\text{animal weight x 7 days week}}$$

In many experiments the administration of the test compound is stopped before the terminal sacrifice or before the death of the last animal. By convention we then take the total dose administered and spread this over the entire experimental period.

Appendix 4-1

Table 1 Standard values for dose calculation: animal lifespans, weights, and intake by diet, water, and inhalation[a]

Experimental animal	Sex	Standard lifespan yr[b]	Weight kg[c]	Food/day g[c]	Food as % body weight/day	Water mL/day[d]	Inhalation volume L/min[e]
Rodents							
Mouse	Male	2	0.03	3.6	12.00	5	0.03
	Female	2	0.025	3.25	13.00	5	0.03
Rat	Male	2	0.5	20	4.00	25	0.10
	Female	2	0.35	17.5	5.00	20	0.10
Hamster	Male	2	0.125	11.5	9.20	15	0.06
	Female	2	0.110	11.5	10.45	15	0.06
Monkeys							
African green (Cercopithecus aethiops)	Both	20					
Cynomolgus (Macaca Fascicularis)	Both	20					
Rhesus (Macaca mulatta)	Both	20					
Prosimians							
Bush babies (Galago crassicaudatus)	Both	10					
Tree shrews (Tupaia glis)	Both	4.5					
Dog	Both	11	16	400	2.50	500	

[a] Although values sometimes vary depending on the source, those given here are within reasonable limits of those usually found in the published literature. No value is given when this information was not necessary for our dose calculation.

[b] Rat and mouse: data based on NCI trichloroethylene bioassay (12); hamster: data of Williams (13); nonhuman primates: data of S.M. Sieber (Laboratory of Chemical Pharmacology, NCI, National Institute of Health, Bethesda, MD), personal communication; bush babies: ages adapted from Dittmer (14); tree shrews: data of D.J. Reddy (Northwestern University, Chicago, IL.) personal communication; dog: data of M.S. Redfearn (Division of Animal Resources, University of California, Berkeley, CA.) personal communication.

[c] Rat and mouse: data based on NCI trichloroethylene bioassay (12); hamster and dog: data of D. Brooks (University of California, Davis) personal communication.

[d] Mouse, rat and dog: data from NIOSH (15); hamster: data from Hoeltge, Inc. (16).

[e] Mouse: data of Sanoskij (17); rat: data of Baker et al. (18); hamsters: data of Guyton (19).

From Gold et al. 1984.

fourth step: The calculation of the TD_{50} [1)]

I) $\frac{1}{N}\frac{dT}{dt} = a + bd$

T = number of tumour bearing animals
N = number of animals at risk
d = dose rate

thus the number of tumour bearing animals, as a fraction of the number of animals at risk, at time t, is dependent of dose rate d.

Equation I), in terms of number of animals at risk $\left(\frac{1}{N}\frac{dT}{dt} = -\frac{1}{N}\frac{dN}{dt}\right)$ leads to

II) $N(t) = N(o)^{-(a + bd)t}$

$\frac{N(t)}{No} = e^{-(a + bd)t}$ = probability of being tumourless conditional on being alive at time t and dose d [= Q(d)].

As all concentrations are assumed to be administered over a same period t*, we may write

III) $Q(d) = e^{-(a_1 t^* + b_1 dt^*)}$

$Q(d) = e^{-(a_1 + b_1 d)}$

1) A computer-program written in the Genstat Statistical Package by Drs E.D. Schoen, designated to calculate the TD_{50} with and without correction for number of tumour bearing animals in the control group, is available.

IV) For what concentration d is

$$Q(d) = 1/2\ Q(0)\ ?$$

$$\left.\begin{aligned} Q(0) &= e^{-a_1} \\ Q(d) &= e^{-(a_1 + b_1 d)} \end{aligned}\right\} \quad e^{-(a_1 + b_1 d)} = 1/2\ e^{-a_1}$$

$$\log e^{-(a_1 + b_1 d)} = \log 1/2\ e^{-a_1}$$

$$-a_1 - b_1 d = \log 1/2 - a_1$$

$$-b_1 d = \log 1/2$$

$$b_1 d = \log 2$$

$$d = \frac{\log 2}{b_1}$$

V) How do we calculate b_1?

$$Q(d) = e^{-(a_1 + b_1 d)}$$

$$\log Q(d) = -a_1 - b_1 d$$

$$-\log Q(d) = a_1 + b_1 d$$

thus b_1 is the slope of the line fitting through the different $-\log Q(d)$'s, $Q(d)$ being the probability of remaining tumourless at dose d.

VI) $\hat{b}_1 = \dfrac{\Sigma_{i=1}^{n} (d_i - \bar{d})(-\log Q_i - \bar{Q})}{\Sigma_{i=1}^{n} (d_i - \bar{d})^2}$

$\bar{a}$ = the mean of all $-\log Q_i$'s

$\bar{d}$ = the mean of all d_i's

$\hat{b}_1$ = estimate of b_1

VII) A 95 % confidence interval for b is given by

$$\hat{b} - t(N-2,0.05)\sqrt{\frac{\hat{\sigma}^2}{\Sigma_{i=1}^{n}(d_i - \bar{d})^2}} \leq b \leq \hat{b} + t(N-2,0.05)\sqrt{\frac{\hat{\sigma}^2}{\Sigma_{i=1}^{n}(d_i - \bar{d})^2}}$$

$$\hat{\sigma} = \frac{Sd}{(N-2)}$$

$$Sd = S - Sr$$

$$S = (1/N)[N\Sigma_{i=1}^{n} -\log Q_i^2 - Gq^2]$$

Gq = the sum of all values for $-\log Q$

$$Sr = \frac{[(1/N)(\Sigma_{i=1}^{n} d_i -\log Q_i - GdGq)]^2}{(1/N)(N\Sigma_{i=1}^{n} d_i^2 - GD^2)}$$

Gd = the sum of all values for d

VIII) When the 95 % confidence interval for b is $z \leq b \leq x$, then the 95 % confidence interval for the TD_{50} is given by

$$\frac{\log 2}{x} \leq \frac{\log 2}{b} \leq \frac{\log 2}{z}$$

fifth step: Correction of the TD_{50} for the possible difference in experiment time and standard lifespan.

In an experiment which is terminated before the standard lifespan, the number of tumours found will be reduced, and the dose rate d needed to halve the proportion of tumourless animals at the end of the reduced period of observation will then be greater than the true TD_{50}.
For this reason we estimate the true TD_{50} as f^2d, where f = (duration of experiment)/(standard lifespan).

APPENDIX 4-2

Appendix 4-2 Calculation of the cancer incidence per mg/m³ according to the 'Simple Dutch method' (SDM)

Extrapolation of data from animal experiments to humans and to lower exposure levels and longer exposure times rests on the assumptions of a similar metabolism in humans and rats, and a linear relationship between dose (exposure level and exposure period) and tumour response.

The following formula is applied to calculate the incidence per mg/m^3 or per mg/kg body weight/day under lifetime conditions:

The incidence in mg/m^3 under lifetime conditions =

$$[I_e - I_c] : [conc._e \times \frac{O}{Tl} \times \frac{Te}{Tl} \times \frac{\text{exposure hours/day}}{24} \times \frac{\text{exposure days/week}}{7}]$$

(I_e = tumour incidence in experiment, I_c = tumour incidence in control, O = observation time (= experimental time of Gold), Tl = lifetime animal, Te = exposure time, $conc._e$ = concentration used in experiment).
The lifetime of rats is assumed to be 1000 days.

The incidence per mg/m³ or per mg/kg body weight/day is calculated linearly using data found in the lowest dose group with a significant increase in the number of treatment-related tumour-bearing animals; the data to be used are the dose and the number of rats with a treatment-related tumour minus the number of control rats bearing the same type of tumour. Next the dose is calculated to lifespan conditions in mg/m³ or mg/kg bw/day. Some of the standard values are different from those used by Gold. For example 1000 days is taken as standard lifespan for the rat, while Gold uses a standard lifespan of 2 years for rat and mouse.

APPENDIX 4-2

Appendix 4-2 Potency indices of the 23 genotoxic carcinogens calculated according to the 'Simple Dutch Method' were as follows:

cadmiumchloride	24.7 per mg/m^3
1,2-dibromoethane	8.3×10^{-2} per mg/m^3
acrylonitrile	9.6×10^{-3} per mg/m^3
2-nitropropane	4.8×10^{-3} per mg/m^3
ethylene oxide	3.1×10^{-3} per mg/m^3
propylene oxide	1.95×10^{-3} per mg/m^3
epichlorohydrin	5.2×10^{-4} per mg/m^3
dichloromethane	3.1×10^{-4} per mg/m^3

$CdCl_2$

MNNG	2.4 per mg/kg bw	(6.9×10^{-1} per mg/m^3)*
3,3'-dimethylbenzidine	1.3 per mg/kg bw	(4.6×10^{-1} per mg/m^3)
2-methylaziridine	11×10^{-1} per mg/kg bw	(3.9×10^{-1} per mg/m^3)
benzidine	9.1×10^{-1} per mg/kg bw	(3.2×10^{-1} per mg/m^3)
1,3-propanesultone	3.1×10^{-1} per mg/kg bw	(1.1×10^{-1} per mg/m^3)
4-aminobiphenyl	2.6×10^{-1} per mg/kg bw	(4.1×10^{-1} per mg/m^3)
1,1-DMH	7.1×10^{-2} per mg/kg bw	(1.1×10^{-1} per mg/m^3)
1,2-dibromoethane		
potassium bromate	5.2×10^{-2} per mg/kg bw	(1.8×10^{-2} per mg/m^3)
3,3'-dichlorobenzidine	4.5×10^{-2} per mg/kg bw	(1.8×10^{-2} per mg/m^3)
4,4'-methylene bis		
acrylonitrile		
(2-chloroaniline)	2.3×10^{-2} per mg/kg bw	(8.1×10^{-3} per mg/m^3)
styrene oxide	1.7×10^{-2} per mg/kg bw	(6.0×10^{-3} per mg/m^3)
1,2-dichloroethane	4.2×10^{-3} per mg/kg bw	(6.7×10^{-3} per mg/m^3)
5-nitroacenaphthene	1.6×10^{-3} per mg/kg bw	(1.3×10^{-3} per mg/m^3)

* To convert mg/kg bw into mg/m^3 the standard values as published by Gold et al., 1984 were used (Appendix 4-1, p. 91, of this report).

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