4,4’-Methylene bis (2-chloroaniline)

Health based calculated occupational cancer risk values

Aan de Staatssecretaris van Sociale Zaken en Werkgelegenheid

Onderwerp : Aanbieding advies
Uw kenmerk : DGV/BMO-U-932542
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Bij brief van 3 december 1993, nr DGV/BMO-U-932542, verzocht de Staatssecretaris van Welzijn, Volksgezondheid en Cultuur namens de Minister van Sociale Zaken en Werkgelegenheid om gezondheidskundige advieswaarden af te leiden ten behoeve van de bescherming van beroepsmatig aan stoffen blootgestelde personen.

Per 1 januari 1994 heeft mijn voorganger daartoe een commissie ingesteld die de werkzaamheden voortzet van de Werkgroep van Deskundigen (WGD). De WGD was een door genoemde minister ingestelde adviescommissie.

Hierbij bied ik u - gehoord de Beraadsgroep Gezondheid en Omgeving - een publicatie van de commissie aan over 4,4'-methyleen bis (2-chlooraniline).
Deze publicatie heb ik heden ter kennisname aan de Minister van Volksgezondheid Welzijn en Sport en aan de Minister van Volkshuisvesting Ruimtelijke Ordening en Milieubeheer gestuurd.

w.g.
prof. dr JJ Sixma

4,4’-Methylene bis (2-chloroaniline)

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Dutch Expert Committee on Occupational Standards,
a committee of the Health Council of the Netherlands

to

the Minister and State Secretary of Social Affairs and Employment

No. 2000/09OSH, The Hague, 6 September 2000

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Samenvatting

Op verzoek van de Minister van Sociale Zaken en Werkgelegenheid schat de Commissie WGD van de Gezondheidsraad het extra kankerrisico bij beroepsmatige blootstelling aan stoffen die door de Europese Unie of door de Commissie WGD als genotoxisch kankerverwekkend zijn aangemerkt. In dit rapport maakt zij zo'n schatting voor 4,4'-methyleen bis (2-chlooraniline). Zij heeft daarbij gebruik gemaakt van de methode die is beschreven in het rapport 'Berekening van het risico op kanker' (1995/06WGD) (Dec95).

Naar schatting van de commissie is de extra kans op kanker voor 4,4'-methyleen bis (2-chlooraniline):

- 4×10^{-5} bij 40 jaar beroepsmatige blootstelling aan 0.02 mg/m^3
- 4×10^{-3} bij 40 jaar beroepsmatige blootstelling aan 2 mg/m^3

Executive summary

On request of the Minister of Social Affairs and Employment the Dutch Expert Committee on Occupational Standards (DECOS), a committee of the Health Council of the Netherlands, estimates the additional lifetime cancer risk associated with occupational exposure to substances that have been classified by the European Union or the DECOS as genotoxic carcinogen. In this report the committee presents such estimates for 4,4'-methylene bis (2-chloroaniline). It has used the method described in the report 'Calculating cancer risks due to occupational exposure to genotoxic carcinogens' (1995/06WGD) (Dec95).

The committee estimated that the additional lifetime cancer risk for 4,4'-methylene bis (2-chloroaniline) amounts to:

- 4×10^{-5} for 40 years of occupational exposure to 0.02 mg/m^3
- 4×10^{-3} for 40 years of occupational exposure to 2 mg/m^3

Chapter 1

Scope

1.1 Background

In the Netherlands, occupational exposure limits for chemical substances are set using a three-step procedure. In the first step, a scientific evaluation of the data on the toxicity of the substance is made by the Dutch Expert Committee on Occupational Standards (DECOS), a committee of the Health Council of the Netherlands, on request of the Minister of Social Affairs and Employment (annex A). This evaluation should lead to a health-based recommended exposure limit for the concentration of the substance in air. Such an exposure limit cannot be derived if the toxic action cannot be evaluated using a threshold model, as is the case for substances with genotoxic carcinogenic properties.

In this case an exposure-response relationship is recommended for use in regulatory standard setting, ie. the calculation of so-called health-based calculated occupational cancer risk values (HBC-OCRVs). The committee calculates HBC-OCRVs for compounds which are classified as genotoxic carcinogens by the European Union or by the present committee.

For the establishment of the HBC-OCRV's the committee generally uses a linear extrapolation method, as described in the committee's report 'Calculating cancer risk due to occupational exposure to genotoxic carcinogens' (1995/06WGD). The linear model to calculate occupational cancer risk is used as a default method, unless scientific data would indicate that using this model is not appropriate.

In the next phase of the three-step procedure, the Social and Economic Council advises the Minister of Social Affairs and Employment on the feasibility of using the HBC-

OCRVs as regulatory occupational exposure limits. In the final step of the procedure the Minister sets the official occupational exposure limits.

1.2 Committee and procedure

The present document contains the derivation of HBC-OCRVs for 4,4'-methylene bis (2-chloroaniline) by the committee. The members of the committee are listed in Annex B. The first draft of this report was prepared by MI Willems, from the TNO Nutrition and Food Research Institute in Zeist, by contract with the Ministry of Social Affairs and Employment.

In 1998, the President of the Health Council released a draft of the report for public review. The individuals and organisations that commented on the draft are listed in annex C. The committee has taken these comments into account in deciding on the final version of the report.

Chapter 2

4,4’-Methylene bis (2-chloroaniline)

2.1 Introduction

The carcinogenicity of 4,4’-methylene bis (2-chloroaniline) (CAS no. 101-14-4) has been evaluated by IARC (IARC74, IARC87), ACGIH (ACG91) and DFG (Gre95). According to IARC there is sufficient evidence for carcinogenicity to animals, but inadequate evidence for carcinogenicity to humans (IARC87). The European Union has classified 4,4’-methylene bis (2-chloroaniline) as a category 2 carcinogen.

This evaluation of the carcinogenicity was based on a review by IARC (IARC74, IARC87). In addition, literature was retrieved from online databases Medline, Toxline and Cancerlit covering the period 1975 to 1996

2.2 Carcinogenicity studies and selection of study suitable for risk estimation in the occupational situation

The available epidemiological data do not allow quantitative risk assessment for 4,4’-methylene bis (2-chloroaniline).

Table 1 (Annex D) summarizes the carcinogenicity studies with experimental animals. 4,4’-methylene bis (2-chloroaniline) induces neoplasms in livers and lungs of rats (Gru70; Stu71, Stu75), rats and mice (Rus75), pulmonary adenomas and adenocarcinomas, mammary adenocarcinomas, Zymbal gland carcinomas and hepatocellular carcinomas in male rats (Kom78) and bladder cancer in dogs (Stu77). Subcutaneous admini-

stration of 4,4'-methylene bis(2-chloroaniline) in mice resulted in malignant primary lung tumours and liver cell carcinomas (see Table 1, Ste71).

Since no inhalation studies are available, oral studies are used to calculate the carcinogenic activity in experimental animals.

2.3 Carcinogenic activity in experimental animals, lifetime low-dose exposure

Four of the studies listed in Table 1 (Annex D) met the criteria for estimation of the carcinogenic potency viz. the rat studies of Grundmann and Steinhoff (Gru70), Stula *et al*. (Stu71, Stu75), Kommineni *et al*. (Kom78) and the mice study of Russfield *et al*. (Rus75). The committee is of the opinion that the available data do not indicate that the use of the linear model is not appropriate.

Below the calculations are given for each of these studies (DEC95a).

Grundmann and Steinhoff (Gru70)

The number of male and female rats with malignant tumours is used as starting point to calculate the incidence per mg/kg body weight per day. Taking 27 g as total intake of the test substance per kg body weight and 890 and 835 days (average 863 days) as exposure period for males and females, respectively, the intake of the test substance amounts to 31.3 mg per kg bw per day taken males and females together.

The incidence of tumour-bearing animals per mg test substance/kg bw/day (lifespan conditions, assuming a linear dose response relationship) is calculated as follows:

$$I^*_{dose} = \frac{I_e - I_c}{C \times (X_{po}/L) \times (X_{pe}/L) \times \text{exposure hours per day}/24 \times \text{exposure days per week}/7}$$

$$= \frac{43/50 - 0/50}{31.3 \times 863/1000 \times 863/1000 \times 24/24 \times 7/7} = 3.7 \times 10^{-2}\ [\text{mg/kg/d}]^{-1}$$

Stula *et al.* (Stu71, Stu75)

To calculate the incidence per mg 4,4'-methylene bis (2-chloroaniline)/kg bw/day the observed number of animals (male and female rats) showing lung adenocarcinomas are used. Assuming an average daily intake of 45 g of food per kg body weight for male and

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* I =the carcinogenic activity attributable to the exposure to the substance per unit daily dose under lifespan conditions assuming a linear dose response relationship
Ie and Ic = incidence of tumour bearing animals or tumours in exposed and control amnimals, respectively,
Xpo = exposure period, Xpe = experimental period
and L = standard lifespan for the animals in question (L rat is assumed to be 1000 days)

female rats, the dose rate of 1000 ppm corresponds to 1000 x 0.045 = 45 mg/kg bw/day (DEC95).

According to the equation noted above, the estimated incidence of tumour-bearing animals per mg/kg bw/day (lifespan conditions) amounts to

$$\frac{48/88 - 0/88}{45 \times 726/1000 \times 726/1000 \times 24/24 \times 7/7} = 2.3 \times 10^{-2}\ [\text{mg/kg/d}]^{-1}$$

Russfield *et al*. (Rus75)

The number of male and female mice with vascular tumours in the 0.2 % group is used as starting point to calculate the incidence per mg/kg body weight per day. Assuming that the daily food consumption amounts to 120 and 130 g/kg body weight per day in males and females, respectively, the intake of the test substance amounts to 250 mg/kg bw/day taken males and females together.

The incidence of tumour-bearing animals per mg per kg body weight /day (lifespan conditions). according to the equation above amounts to

$$\frac{14/34 - 1/38}{250 \times 546/750 \times 730/750 \times 24/24 \times 7/7} = 2.2 \times 10^{-3}\ [\text{mg/kg/d}]^{-1}$$

Kommineni *et al*. (Kom78)

To calculate the incidence per mg 4,4’-methylene bis (2-chloroaniline)/kg bw/day the observed number of animals (male rats) showing lung adenocarcinomas are used. At the dose level of 250 ppm a significant increase in the number of males with lung adenocarcinomas was observed. Assuming an average daily intake of 40 g of food per kg body weight for male rats, the dose rate of 250 ppm corresponds to 250 x 0.040 = 10 mg/kg bw/day (DEC95).

The incidence of tumour-bearing animals per mg/kg bw/day (lifespan conditions) amounts to:

$$\frac{14/100 - 0/100}{10 \times 546/1000 \times 728/1000 \times 24/24 \times 7/7} = 3.5 \times 10^{-2}\ [\text{mg/kg/d}]^{-1}$$

Conclusion

The highest calculated cancer incidence in the above studies, i.e. 3.7 x 10^{-2} per mg/kg bw/day from the study of Grundmann and Steinhoff (Gru70), is used as starting point for quantitative risk estimation in humans.

2.4 Health risk to humans

To estimate the additional lifetime risk of cancer in humans under lifespan conditions on the basis of results in animal experiments, it is assumed that no difference exists between experimental animals and man with respect to toxicokinetics, mechanism of tumour induction, target, susceptibility etc, unless specific information is available which justifies a different approach. Furthermore, it is assumed that the average man lives 75 years, weights 70 kg and is exposed 24 hours per day 7 days/week, 52 weeks per year for lifetime.

2.5 Calculation of the HBC-OCRV

To estimate the additional lifetime risk of cancer in humans under workplace conditions, it is assumed that the average man lives 75 years, is exposed 8 hours per day, five days a week, 48 weeks a year, for 40 years, and inhales 10 m^3 air per 8 hour-working day. Using as starting point the estimated incidence of 3.7 x 10^{-2} per mg/kg bw/day, the additional lifetime cancer risk per mg/m^3 under occupational conditions, the HBC-OCRV, amounts to:

$$\text{HBC-OCRV} = 3.7x10^{-2}\ x\frac{40y}{75y}x\frac{48w}{52w}x\frac{5d}{7d}x\frac{10m^3}{70kg} = 1.9 \times 10^{-3}[mg/m^3]^{-1}$$

Based on the HBC-OCRV of 2 x 10^{-3} per mg/m^3 the reference additional lifetime cancer risk amounts to:

- 4 x 10^{-5} for 40 years of exposure to 0.02 mg/m^3
- 4 x 10^{-3} for 40 years of exposure to 2 mg/m^3.

2.6 Existing occupational exposure limits

Table 2 summarizes the occupational exposure limits established by the regulatory authorities of Germany, United Kingdom and Sweden, and by USA-ACGIH. No occupational exposure limits have been established in The Netherlands, Germany and Sweden.

Table 2 Occupational exposure limits for 4,4'-methylene bis (2-chloroaniline).

country	level		time relation	notations	ref.
	ppm	mg/m^3			
The Netherlands[a]	-	-	-	-	ISZW95
Germany[b]	-	(0.02)	-	skin	DFG96
UK	-	0.005 (MEL)	8-h TWA	skin	HSE95
Sweden[c]	-	-	-	-	NBO93
USA-ACGIH[d]	0.01	0.11	8-h TWA	skin	ACG96

[a] In the Netherlands, this compound is listed as a carcinogen.

[b] In Germany, this compound is classified as a category A2 carcinogen. DFG category A carcinogens are not assigned a health-based occupational exposure limit, but a so called TRK-value (TRK = Technische Richtkonzentrationen), a concentration feasible with currently available technical means. TRK-values are given in brackets.

[c] In Sweden, this compound is placed under section 9 (carcinogen) and may only be handled by permission of the Labour Inspectorate.

[d] Classified as A2 carcinogen: suspected human carcinogen.

The lowest occupational exposure limit settled by these countries amounts to 0.005 mg/m^3 (UK, HSE95). In the United Kingdom, it was concluded that considering the carcinogenicity data, a threshold could not be identified and that consequently a maximum exposure limit (MEL)* (0.005 mg/m^3) was appropriate. This concentration is a factor 400 lower than the concentration leading to an additional cancer risk of 4×10^{-3} (2 mg/m^3) and a factor 4 lower than the concentration leading to an additional cancer risk of 4×10^{-5} (0.02 mg/m^3).

* In setting a Maximum Exposure Limit (MEL), not only the protection of the health of the employee is considered, also socio-economic factors are taken into account. A cost benefit assessment is prepared to assist the considerations of these. In practice, MELs have been most often allocated to carcinogens, respiratory sensitizers and to other substances for which no threshold level of exposure for the effects can be identified and for which there is no doubt about the seriousness of the hazard(s) posed by the substance.

For the committee,
The Hague, 6 September 2000

dr ASAM van der Burght, scientific secretary	Prof. dr GJ Mulder, chairman

References

ACG91 American Conference of Governmental Industrial Hygienists (ACGIH). 4,4'-Methylene bis(2-chloroaniline). In: Documentation of the Threshold Limit Values and Biological Exposure Indices. 6th ed. Cincinnati, OH, USA: ACGIH, 1991: 988-95.

ACG96 American Conference of Governmental Industrial Hygienists (ACGIH). 1996. TLVs(R) and BEIs(R).Threshold Limit Values for chemical substances and physical agents. Biological Exposure Indices. Cincinnati OH, USA: ACGIH, 1996: 27.

DEC95 Health Council of the Netherlands: Dutch Expert Committee on Occupational Standards (DECOS). Calculating cancer risk. The Hague, The Netherlands: Health Council of the Netherlands, 1995; pub no 1995/06WGD.

DFG96 Deutsche Forschungsgemeinschaft (DFG): Senatskommission zur Prüfung gesundheitsschädlicher Arbeitsstoffe. MAK- und BAT-Werte-Liste 1996. Maximale Arbeitsplatzkonzentrationen und biologische Arbeitsstofftoleranzwerte. Weinheim, FRG: VCH Verlagsgesellschaft mbH, 1996: 72, 108, 129 (Mitteilung 32).

Gre95 Greim H, ed. 4,4'-Methylen-bis(2-chloranilin). In: Gesundheitsschädliche Arbeitsstoffe. Toxikologisch-arbeitsmedizinische Begründungen von MAK-Werten (Maximale Arbeitsplatz-Konzentrationen). 1st - 21st ed. Weinheim, FRG: VCH Verlagsgesell-schaft mbH, 1995.

Gru70 Grundmann E, Steinhoff D. Leber- und Lungentumoren nach 3,3'-Dichlor-4,4'-diaminodiphenylmethan bei ratten. Z Krebsforsch 1970; 74: 28-39.

HSE95 Health and Safety Executive (HSE). Occu uffolk), UK: HSE Books, 1995: 27 (Guidance note 40/95).

IARC74 International Agency for Research on Cancer (IARC). 4,4'Methylene bis (2-chloroaniline).Some aromatic amines, hydrazine and related substances, N-nitroso compounds and miscellaneous alkylating agents. Lyon, France: IARC, 1974: 65-71. In: IARC monographs on the evaluation of carcinogenic risk of chemicals to man, Vol 4.

IARC87 International Agency for Research on Cancer (IARC). 4,4'Methylene bis (2-chloroaniline) (MOCA). Overall evaluations of carcinogenicity: an updating of IARC monographs. Lyon, France: IARC, 1987: 246-7 In: IARC monographs on the evaluation of carcinogenic risks to humans, Volumes 1 to 42; Suppl 7.

ISZW95 Inspectiedienst van het Ministerie van Sociale Zaken en Werkgelegenheid (I-SZW). De Nationale MAC-lijst 1995. The Hague, The Netherlands: Sdu Servicecentrum Uitgeverijen, 1995: 28, 42, 63 (pub no P145).

Kom78 Kommineni C, Groth DH, Frockt IJ, *et al*. Determination of the tumorigenic potential of methylene-bis-orthochloroaniline. J Environ Pathol Toxicol 1978; 2: 149-71.

NBO93 National Board of Occupational Safety and Health (NBOSH). Occupational exposure limits. Solna, Sweden: NBOSH, 1993: 74 (Ordinance AFS 1993/9).

Rus75 Russfield AB, Homburger F, Boger E, *et al*. The carcinogenic effect of 4,4'-methylene-bis-(2-chloroaniline) in mice and rats. Toxicol Appl Pharmacol 1975; 31: 47-54.

Stu71 Stula EF, Sherman H, Zapp JA, *et al*. Experimental neoplasia in ChR-CD rats with the oral administration of 3,3'-dichlorobenzidine, 4,4'-methylene-bis(2-chloroaniline) and 4,4'-methylene-bis-(2-methylaniline). Toxicol Appl Pharmacol 1971; 19: 380-1.

Stu75 Stula EF, Sherman H, Zapp JA, *et al*. Experimental neoplasia in rats from oral administration of 3,3'-dichlorobenzidine, 4,4'-methylene-bis(2-chloroaniline) and 4,4'-methylene-bis-(2-methylaniline). Toxicol Appl Pharmacol 1975; 31: 159-76.

Stu77 Stula EF, Barnes JR, Sherman H, *et al*. Urinary bladder tumors in dogs from 4,4'-methylene-bis(2-chloroaniline). J Environ Pathol Toxicol 1977; 1: 31-50.

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A	Request for advice
B	The committee
C	Comments on the public draft
D	Animal studies

Annexes

Annex **A**

Request for advice

In a letter dated October 11, 1993, ref DGA/G/TOS/93/07732A, to, the State Secretary of Welfare, Health and Cultural Affairs, the Minister of Social Affairs and Employment wrote:

Some time ago a policy proposal has been formulated, as part of the simplification of the governmental advisory structure, to improve the integration of the development of recommendations for health based occupation standards and the development of comparable standards for the general population. A consequence of this policy proposal is the initiative to transfer the activities of the Dutch Expert Committee on Occupational Standards (DECOS) to the Health Council. DECOS has been established by ministerial decree of 2 June 1976. Its primary task is to recommend health based occupational exposure limits as the first step in the process of establishing Maximal Accepted Concentrations (MAC-values) for substances at the work place.

In an addendum, the Minister detailed his request to the Health Council as follows:

The Health Council should advice the Minister of Social Affairs and Employment on the hygienic aspects of his policy to protect workers against exposure to chemicals. Primarily, the Council should report on health based recommended exposure limits as a basis for (regulatory) exposure limits for air quality at the work place. This implies:

- A scientific evaluation of all relevant data on the health effects of exposure to substances using a criteria-document that will be made available to the Health Council as part of a specific request for advice. If possible this evaluation should lead to a health based recommended exposure limit, or, in

the case of genotoxic carcinogens, a 'exposure versus tumour incidence range' and a calculated concentration in air corresponding with reference tumour incidences of 10^{-4} and 10^{-6} per year.

- The evaluation of documents review the basis of occupational exposure limits that have been recently established in other countries.
- Recommending classifications for substances as part of the occupational hygiene policy of the government. In any case this regards the list of carcinogenic substances, for which the classification criteria of the Directive of the European Communities of 27 June 1967 (67/548/EEG) are used.
- Reporting on other subjects that will be specified at a later date.

In his letter of 14 December 1993, ref U 6102/WP/MK/459, to the Minister of Social Affairs and Employment the President of the Health Council agreed to establish DECOS as a Committee of the Health Council. The membership of the Committee is given in annex B.

Annex **B**

The Committee

- GJ Mulder, *chairman*
  professor of toxicology; Leiden University, Leiden
- RB Beems
  toxicologic pathologist; National Institute of Public Health and the Environment, Bilthoven
- PJ Borm
  toxicologist; Heinrich Heine Universität Düsseldorf (Germany)
- JJAM Brokamp, *advisor*
  Social and Economic Council, The Hague
- VJ Feron,
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- DJJ Heederik
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- G de Jong
  occupational physician; Shell International Petroleum Maatschappij, The Hague
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  occupational physician; BMD Akers bv, Amsterdam
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- H Roelfzema, *advisor*
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  occupational hygienist; KLM Health Safety & Environment, Schiphol and professor of working conditions, Free University, Amsterdam
- GMH Swaen
  epidemiologist; Maastricht University, Maastricht
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  toxicologist; DOW Europe, Horgen (Switzerland)
- F de Wit
  occupational physician; Labour Inspectorate, Arnhem
- CA Bouwman, *scientific secretary*
  Health Council of the Netherlands, Den Haag
- ASAM van der Burght, *scientific secretary*
  Health Council of the Netherlands, Den Haag

The first draft of the present advisory report was prepared by M Willems, from the Department of Occupational Toxicology of the TNO Nutrition and Food Research Institute, by contract with the Ministry of Social Affairs and Employment.

Secretarial assistance: J Toet.
Lay-out: J van Kan.

Annex **C**

Comments on the public draft

A draft of the present report was relased in 1998 for public review. The following organisations and persons have commented on the draft document:

- WF ten Berge, DSM, Heerlen

Annex **D**

Animal studies

See table on the next page.

Table 1 Carcinogenicity studies with 4,4'-methylene bis (2-chloroaniline).

authors	species	exposure characte-ristics	dose	exposure and experimental period	findings
Gru70	rat (25/ sex/group)	low protein diet	0.1% (total 27 g/kg bw for male and female)	Xpo =Xpe =lifespan lifespan: male: 890 days (average survival 565 days); female: 835 days (average survival 535 days).	*controls*: 0/50, no malignant tumours (2 female animals had mammary adenomas). *Treatment group:* male: 23/25 with malignant tumours (1 primary lung tumour, 7 liver + primary lung, 15 liver). female: 20/25 with malignant tumours (15 liver, 3 liver + primary lung, 2 primary lung)
Russfield *et al*. (1973), in I-ARC74; Rus75	rat[a] (25 males/ group)	diet	0.05 and 0.1%	Xpo = 18 months Xpe = 24 months	hepatomas male: 0/22, 1/22, 4/19 in 0, 0.05 and 0.1% group, respectively remark: the incidences were not statistically significant different from controls
Kom78	rat[b] (males)	diet	0, 250, 500, 1000 ppm	Xpo = 18 months Xpe = 24 months	in the 0, 250, 500 and 1000 ppm groups, respectively: lung adenocarcinomas 0/100, 14/100[c], 20/75[c], 31/50[c] all primary lung neoplasms 1/100, 23/100[c], 28/75[c], 35/50[c] -mammary adenocarcinomas 1/100, 5/100, 8/75[c], 14/50[c] -zymbal gland carcinomas 1/100, 8/100[c], 5/75, 11/50[c] -hepatocellular carcinomas 0/100, 3/100, 3/75, 18/50[c]
Stu71, Stu75	rat[a] (50/ sex/group)	diet	0, 1000 ppm	Xpo = Xpe =lifespan lifespan: average survival time male 560[d] days and 548[d] days for female (survival time controls male 564 days, for female 628 days)	*control group*: lung adenomatosis: male1/44, female 1/44. *Treatment group*: lung adenomatosis: male14/44, female 11/44. lung adenocarcinoma: male21/44, female 27/44
Stu77	dog[e] (6 females/ group)	oral (capsules)	0 and 100 mg/day (8-15 mg/kg bw/d)	Xpo = Xpe = 9 years treatment: first six weeks, 3 days/week, then 5 days/week	five treated dogs showed malignant nodules in the bladder after 9 years
Rus75	mouse (25/sex/ group)	diet	0, 0.1%, 0.2%	Xpo = 18 months Xpe = 25 months	vascular tumours male: 0/18, 3/13, 8/20; female: 1/20, 0/21, 6/14. Hepatomas male: 3/18, 3/13, 4/20; female: 0/20, 9/21, 7/14 in 0, 0.1 and 0.2% group, respectively
Grundmann & Steinhoff (1971), in IARC74	rat (17/sex/ group)	subcutaneous once a week	total dose: 25 mg/kg bw	Xpo = 620 days	*controls:* 13 tumours in 50 control rats (no malignant lung or liver cell carcinomas). Test: 9 rats with liver cell carcinomas, 7 rats with primary lung tumours

[a] ChR-CD-1rat

[b] Sprague-Dawley rats

[c] Statistically significant different from controls

[d] Range of days on test: male 152-733 days; female 224-719 days

[e] Beagle dogs