Health Council of the Netherlands

1,1,1-Trichloroethane

Evaluation of the carcinogenicity and genotoxicity

Gezondheidsraad
Health Council of the Netherlands

Aan de staatssecretaris van Sociale zaken en Werkgelegenheid

Onderwerp : aanbieding advies *1,1,1-Trichloroethane*
Uw kenmerk : DGV/MBO/U-932342
Ons kenmerk : U-7253/SV/fs/246-L16
Bijlagen : 1
Datum : 24 juli 2012

Geachte staatssecretaris,

Graag bied ik u hierbij het advies aan over de gevolgen van beroepsmatige blootstelling aan 1,1,1-trichloorethaan.

Dit advies maakt deel uit van een uitgebreide reeks waarin kankerverwekkende stoffen worden geclassificeerd volgens richtlijnen van de Europese Unie. Het gaat om stoffen waaraan mensen tijdens de beroepsmatige uitoefening kunnen worden blootgesteld.

Dit advies is opgesteld door een vaste subcommissie van de Commissie Gezondheid en beroepsmatige blootstelling aan stoffen (GBBS), de Subcommissie Classificatie van carcinogene stoffen. Het advies is getoetst door de Beraadsgroep Gezondheid en omgeving van de Gezondheidsraad.

Ik heb het advies vandaag ter kennisname toegezonden aan de staatssecretaris van Infrastructuur en Milieu en aan de minister van Volksgezondheid, Welzijn en Sport.

Met vriendelijke groet,

prof. dr. H. Obertop,
waarnemend voorzitter

Bezoekadres
Parnassusplein 5
2511 VX Den Haag
Telefoon (070) 340 55 08
E-mail: sr.vink@gr.nl

Postadres
Postbus 16052
2500 BB Den Haag
Telefax (070) 340 75 23
www.gr.nl

1,1,1-Trichloroethane

Evaluation of the carcinogenicity and genotoxicity

Subcommittee on the Classification of Carcinogenic Substances of
the Dutch Expert Committee on Occupational Safety,
a Committee of the Health Council of the Netherlands

to:

the State Secretary of Social Affairs and Employment

No. 2012/12, The Hague, July 24, 2012

The Health Council of the Netherlands, established in 1902, is an independent scientific advisory body. Its remit is "to advise the government and Parliament on the current level of knowledge with respect to public health issues and health (services) research..." (Section 22, Health Act).

The Health Council receives most requests for advice from the Ministers of Health, Welfare & Sport, Infrastructure & the Environment, Social Affairs & Employment, Economic Affairs, Agriculture & Innovation, and Education, Culture & Science. The Council can publish advisory reports on its own initiative. It usually does this in order to ask attention for developments or trends that are thought to be relevant to government policy.

Most Health Council reports are prepared by multidisciplinary committees of Dutch or, sometimes, foreign experts, appointed in a personal capacity. The reports are available to the public.

The Health Council of the Netherlands is a member of the European Science Advisory Network for Health (EuSANH), a network of science advisory bodies in Europe.

The Health Council of the Netherlands is a member of the International Network of Agencies for Health Technology Assessment (INAHTA), an international collaboration of organisations engaged with health technology assessment.

INAHTA

This report can be downloaded from www.healthcouncil.nl.

Preferred citation:
Health Council of the Netherlands. 1,1,1-Trichloroethane. Evaluation of the carcinogenicity and genotoxicity. The Hague: Health Council of the Netherlands, 2012; publication no. 2012/12.

ISBN: 978-90-5549-904-5

Contents

Samenvatting

Op verzoek van de minister van Sociale Zaken en Werkgelegenheid evalueert en beoordeelt de Gezondheidsraad de kankerverwekkende eigenschappen van stoffen waaraan mensen tijdens de beroepsmatige uitoefening kunnen worden blootgesteld. De evaluatie en beoordeling worden verricht door de Subcommissie Classificatie van Carcinogene Stoffen van de Commissie Gezondheid en Beroepsmatige Blootstelling aan Stoffen van de Raad, hierna kortweg aangeduid als de commissie. In het voorliggende advies neemt de commissie 1,1,1-trichloorethaan onder de loep. De stof wordt onder andere gebruikt als oplosmiddel van hechtmiddelen, voor het ontvetten in de metaal- en elektronische industrie en in de synthese van vinylideenchloride.

Op basis van de beschikbare gegevens is de commissie van mening dat de gegevens over 1,1,1-trichloorethaan niet voldoende zijn om de kankerverwekkende eigenschappen te evalueren (categorie 3).*

* Volgens het classificatiesysteem van de Gezondheidsraad (zie bijlage I).

Executive summary

At the request of the Minister of Social Affairs and Employment, the Health Council of the Netherlands evaluates and judges the carcinogenic properties of substances to which workers are occupationally exposed. The evaluation is performed by the Subcommittee on Classifying Carcinogenic Substances of the Dutch Expert Committee on Occupational Standards of the Health Council, hereafter called the Committee. In this report the Committee evaluates 1,1,1-trichloroethane. The compound is being used as a solvent for adhesives, for degreasing in metallic and electronic industries, and in the manufacture of vinylidene chloride.

The Committee is of the opinion that the available data are insufficient to evaluate the carcinogenic properties of 1,1,1-trichloroethane (category 3).*

* According to the classification system of the Health Council (see Annex I).

Chapter 1

Scope

1.1 Background

In the Netherlands, a special policy is in force with respect to occupational use and exposure to carcinogenic substances. Regarding this policy, the Minister of Social Affairs and Employment has asked the Health Council of the Netherlands to evaluate the carcinogenic properties of substances and to propose a classification (see Annex A). In addition to classifying substances, the Health Council also assesses the genotoxic properties of the substance in question. The assessment and the proposal for a classification are expressed in the form of standard sentences (see Annex I).

This report contains the evaluation of the carcinogenicity and genotoxicity of 1,1,1-trichloroethane.

1.2 Committee and procedure

The evaluation is performed by the Subcommittee on Classifying Carcinogenic Substances of the Dutch Expert Committee on Occupational Standards of the Health Council, hereafter called the Committee. The members of the Committee are listed in Annex B.

In 2012, 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 D. The Committee has taken these comments into account in deciding on the final version of the report.

1.3 Data

The evaluation and recommendation of the Committee is standardly based on scientific data, which are publicly available. The starting points of the Committees' reports are, if possible, the monographs of the International Agency for Research on Cancer (IARC). This means that the original sources of the studies, which are mentioned in the IARC-monograph, are reviewed only by the Committee when these are considered most relevant in assessing the carcinogenicity and genotoxicity of the substance in question. In the case of 1,1,1-trichloroethane, such an IARC-monograph is available, of which the summary and conclusion of IARC is inserted in Annex E.

More recently published data were retrieved from www.inchem.org, Medline, XToxline, and Chemical Abstracts. The last updated online search was in July 2011. The relevant data were included in this report.

Chapter 2

General information

2.1 Identity and physicochemical properties

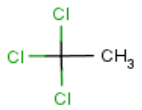
1,1,1-Trichloroethane is used as a solvent for adhesives, for degreasing in metal and electronic industries, and in the manufacture of vinylidene chloride. Other applications include: its use in pesticides; textile processing; cutting fluids; aerosols; lubricants; cutting oil formulations; drain cleaners; shoe polishes; spot cleaners; printing inks; and stain repellents.[1,2]

Occupational exposure occurs during manufacturing of 1,1,1-trichloroethane and products containing this substance. In addition, workers engaged in dry-cleaning, or degreasing processes in the metal and electronic industries, may be exposed. The general population is predominantly exposed via contaminated air, food or drinking water.[1]

The identity of 1,1,1-trichloroethane and some of its physicochemical properties are given below.*

* The data have been retrieved from the European Substance Information System (ESIS, which can be accessed via the ECB-site (http://ecb.jrc.ec.europa.eu/esis/), the Hazardous Substances Data Bank (http://toxnet.nlm.nih.gov/cgi-bin/sis/htmlgen?HSDB) and the INCHEM database of the International Programme on Chemical Safety (IPCS) (http://www.inchem.org/).

Chemical name	:	1,1,1-trichloroethane
CAS registry number	:	71-55-6
EC/EINECS number	:	200-756-3
Synonyms	:	methylchloroform, chloroethene
Molecular formula	:	CH_3 -CCl_3
Structural formula	:	



Colour and physical state	:	colourless liquid
Melting	:	-30.4 °C
Boiling point		74 °C
Molecular weight	:	133.40
Relative density of vapour/ air mixture at 20 °C	:	4.6
Vapour pressure	:	13.3 kPa at 20 °C
Solubility	:	slightly soluble in water (0.07 g/100 ml at 20 °C); soluble in acetone, benzene, carbon tetrachloride, methanol, ethanol and diethyl ether
Conversion factors (25 °C, 760 mm Hg)	:	1 ppm = 0.18 mg/m^3 1 mg/m^3 = 5.46 x ppm
EU classification		H332 - Harmful if inhaled (Based on Regulation (EC) No. 1272/2008 of the European Parliament of the Council on Classification, labelling, and packaging of substances and mixtures; 16 December 2008).

2.2 IARC conclusion

In 1999, IARC concluded that there is inadequate evidence for the carcinogenicity of 1,1,1-trichloroethane in experimental animals and humans. Therefore, according to the IARC guidelines, 1,1,1-trichloroethane was considered to be not classifiable as to its carcinogenicity to humans (Group 3).[2]

Chapter 3

Carcinogenicity studies

3.1 Observations in humans

The relevant epidemiological studies are summarised below and presented in a table in Annex F.

Cohort studies

In a Finnish cohort study of cancer incidence and exposure to halogenated hydrocarbons, 2,050 male and 1,924 female workers were followed up from 1967 to 1992.[3] Exposure to 1,1,1-trichloroethane was assessed in 140 men, and 131 women, by biological monitoring. Seventeen cases of cancer were seen in the exposed workers (standardized incidence ratio (SIR) of 1.6; 95% CI 0.9-2.5). No statistically significant increases were found for cancer of the pancreas, lung, cervix, kidney and non-Hodgkin lymphomas. Statistically significant increases of cancer were observed for the nervous system (SIR of 6.1; 95% CI 1.25-17.7), and multiple myeloma (SIR of 16.0; 95% CI 1.9-57.7). However, as these results are based on only three observed cases for nervous system tumours, and two cases (both females) for multiple myeloma, and furthermore co-exposure to other solvents was noted, no conclusions can be made based on this study.

Another cohort study involved 14,457 workers at an aircraft maintenance facility, which have been potentially exposed to many solvents, such as

1,1,1-trichloroethane.[4] Exposure to 1,1,1-trichloroethane was assessed on a qualitative (ever or never exposed) basis. Subjects had been working at least for one year at the facility between 1952 and 1956, and were followed up to 1982. The only increased standardized mortality ratio (SMR) observed was for multiple myeloma in female workers (SMR of 56.6 (95% CI 6.85-204.5)), although this increase was based on only two cases.

This cohort has been updated by Blair et al. (1998), extending follow up to the end of 1990.[5] Again, an increased risk of multiple myeloma in women was found (RR of 13.2 (95% CI 2.2-80.4)). However, this increase is based on the same two cases of multiple myeloma as no additional cases were reported during the extended follow up.

No conclusions can be drawn based on this cohort, as it only involves a limited number of multiple myeloma cases, and most workers were exposed to a number of solvents subsequently or simultaneously.

Case-control studies

The most recent case-control study involved the relationship between multiple myeloma, and the exposure to several chlorinated solvents, including 1,1,1-trichloroethane.[6] One-hundred-eighty-one cases were matched with 481 population controls, which were selected for an ongoing non-Hodgkin lymphoma case-control study. Occupational histories and information on jobs with likely 1,1,1-trichloroethane exposure were obtained by in-person interviews. Exposure to 1,1,1-trichloroethane was associated with an increased OR of 1.8 (95% CI 1.1-2.9) for subjects ever exposed versus non-exposed. When occupations with low confidence for 1,1,1-trichloroethane exposure were included in the unexposed category, the OR increased to 2.2 (95% CI 1.1-4.4). The association found in this study is difficult to interpret, as information bias cannot be excluded. Also, there was no trend observed between the incidence of multiple myeloma, and cumulative exposure (10-year lagged and unlagged) or exposure duration to 1,1,1-trichloroethane.

In Montreal, Canada, a population-based case-control study was conducted to study the association between several types of cancer following exposure to 293 workplace substances.[7] Approximately one per cent of the study subjects (3,730 cancer patients, and 533 age-stratified control from the general population) had ever been exposed to 1,1,1-trichloroethane. For most types of cancer examined (oesophagus, stomach, colon, rectum, pancreas, prostate, bladder, skin melanoma, lymphoma) no indication of excess risk was found. For lung cancer,

in French Canadians, the OR was 3.5 (90% CI 1.0-12.0; 7 cases exposed at any level). For kidney cancer among the whole population, the OR was 2.4 (90% CI 1.0-6.0; 4 cases exposed at any level). The workers included in this study have been exposed to multiple solvents.

Another population-based case-control study evaluated the risk of pancreatic cancer in residents from 24 US states (based on death certificates) exposed to solvents, including 1,1,1-trichloroethane.[8] Of the total of 63,097 cases 5,866 had a low, medium or high probability of exposure to 1,1,1-trichloroethane. For each case, four controls were matched by state, race, gender and 5-year age group. An increased odds ratio for pancreatic cancer was found for black men (OR of 2.9 for group with high probability of exposure; 95% CI 1.2-7.5; based on 8 exposed cases only) on exposure to 1,1,1-trichloroethane. No positive association was found for this group with chlorinated hydrocarbons or organic solvents combined. No conclusion could be drawn due to limitations of the study, as no information on duration of employment, cigarette smoking, socioeconomic status and other lifestyle factors was available.

In a population-based case-control study by Dosemeci et al. (1999) among workers residential in Minnesota, US, and exposed to several organic solvents, including 1,1,1-trichloroethane, no increased risk of renal cell carcinoma was found (OR of 1.26; 95% CI 0.6-2.8; n=13).[9] Of the total of 438 renal cell cancer cases and 687 controls, 66 and 74 had been exposed to 1,1,1-trichloroethane. Odds ratios were adjusted for: age; smoking; hypertension status; use of diuretics; anti-hypertension drugs; and, body mass index, for men and women separately. As all cases who died (35%) were excluded from the analysis to avoid using next-of-kin interviews, a potential survival bias confounded the study. Also data on exposure and duration of employment were limited, as only current and usual jobs were assessed.

A case-control study among white males on the risk of astrocytic brain cancer was carried out in three US states with prominent workforce representation in the petroleum refining, and chemical manufacturing industries.[10] Based on death certificates, 300 cases and 320 matched controls were included, whereas occupational information was retrieved from next-of-kin. No increased risk was found for exposure to 1,1,1-trichloroethane, although a trend was found for the average intensity of exposure for 21+ years (expressed by an OR of 1.6 (95% CI 0.9-3.1) for low-medium intensity exposure compared to an OR of 3.7 (95% CI 0.7-27.9) for high intensity exposure (Chi for trend 2.28 ($p<0.05$)). As the

population for high intensity exposure was only based on six cases and two controls, the Committee considers this result uncertain. Also in this study, most workers were exposed to multiple other solvents for which trends were observed.

Another population-based case-control study examined the association between maternal exposure to occupational solvents, including 1,1,1-trichloroethane, and childhood acute lymphoblastic leukaemia (ALL).[11] The study included 790 cases and 790 controls. The cases were up to seven times more exposed to 1,1,1-trichloroethane then controls, taking into account an exposure period of 2 years before pregnancy up to giving birth. Exposure was associated with an increased incidence of childhood ALL in their offspring. This increase however, was not statistically significant (OR of 7.55; 95% CI 0.92-61.97).

Additional studies have been referred to by US EPA in which no association was found between occupational exposure to 1,1,1-trichloroethane, and some types of cancer. These include oesophageal cancer or stomach cancer in workers at an industrial facility in California (Garland, 1987; Garabrant, 1986; Cited by US EPA).[12]

Also, US EPA reported on an environmental study in which no significant correlation was found between the release of 1,1,1-trichloroethane and age-adjusted incidence of childhood brain tumours in 26 Florida counties (Mulla, 1996; cited by US EPA).[12]

3.2 Carcinogenicity studies in animals

Several carcinogenicity studies have been conducted, in rats as well as in mice (Annex F).

Inhalation

Quast et al. conducted chronic bioassays in Fischer 344 rats and B6C3F1 mice, exposed to 1,1,1-trichloroethane by inhalation (both species 80/sex/dose).[13] The animals were exposed to 1,1,1-trichloroethane (at concentrations of 0, 820, 2,700 or 8,200 mg/m^3 (0, 150, 500 or 1,500 ppm), 6 hr/day, 5 days/week for 24 months. Interim sacrifices of 10 animals/sex/group were conducted after 6, 12 and 18 months of exposure. In this study, clinical signs of toxicity, mortality, haematology, serum chemistry, urinalysis endpoints (rats only), body weight, organ weights, gross pathology, and histopathology were comprehensively assessed.

Rats showed no statistically significant difference in survival between exposed and control groups; body weight was decreased in females at the two higher exposure concentrations. A statistically significantly increased number of rats with *bilateral* testicular interstitial cell neoplasm was noted. However, no dose dependency was observed, nor the total number of male rats with testicular interstitial cell tumours was increased. As Fischer 344 rats show a high background incidence of this tumour type, the Committee considers this finding not to be related to the treatment.

In mice, no differences in body weight and survival were observed for the exposed groups compared to controls. In females, a statistical significant increase of adenoma or cystadenoma of the lacrimal/Harderian gland (7 at 8,200 mg/m^3 compared to 3 in the controls). As this was only observed at the highest dose level, and no dose response was noted, the Committee does not consider the observed effect to be toxicologically relevant.

Rampy et al. (as cited in [13,14]) administered 1,1,1-trichloroethane to rats (96/sex/dose) via inhalation at a concentration of 4,700 and 9,500 mg/m^3 (875 or 1,750 ppm), for 6 hrs/day, 5 days/week, for 12 months, followed by observation up to 30 months. The authors stated that the tumour incidences in treated rats were comparable to controls. The Committee notes that only an abstract has been published and several details on methods and results are lacking, hampering the interpretation of the study.

Oral administration

Technical-grade 1,1,1-trichloroethane was administered by gavage at 500 mg/kg bw per day (in olive oil) to male and female Sprague-Dawley rats (50/sex for control groups, and 40/sex for the exposure groups), 4-5 days/week for 104 weeks.[15] A complete necropsy was performed on all animals. Exposure did not affect survival, whereas a reduction of body weight was only observed in female rats from 80 weeks of exposure onwards.

An increase in leukaemias/lymphomas was found in treated males and females. The incidences of leukaemia were 3/50 and 9/40 for control and treated males, and 1/50 and 4/40 for control and treated females, respectively. These involved mainly immunoblastic lymphosarcomas in the lung.

The Committee considers the findings in this study inconclusive, due to inherent limitations of the experimental design (only one dose and one species was assessed) and incomplete analysis and reporting of results.

In a bioassay conducted by the National Cancer Institute, technical grade 1,1,1-trichloroethane was administered in corn oil by gavage to 50 male and 50 female Osborne-Mendel rats, and B6C3F1 mice, 5 days per week for 78 weeks.[16]

Rats, exposed to 750 mg/kg bw or 1,500 mg/kg bw, showed a high mortality rate during the whole study. Average weight gain in male rats appeared to be treatment related during the first year of the study. During study, in the treatment groups, increasing numbers of females and to a lesser extent, males, showed urine staining of the abdominal fur.

In mice, exposed to average dose of 2,807 mg/kg bw or 5,615 mg/kg bw*, a decreased body weight was observed in both sexes. A reduced survival was noted in females of the high-dose group in the first year. Male mice showed also high early mortality rate in both control and treated groups.

Several types of tumours were observed both for rats and mice. Incidences and types in the exposure group were similar to those observed in the untreated controls. However, no conclusion can be made from this study due to the very low survival at study termination. A high incidence of chronic pneumonia was present in all control and treated rats and mice of both sexes, and was considered by the authors as the cause for the high incidence of early death.

* Consecutive exposure to 2,000 mg/kg bw for 10 weeks, 2,500 mg/kg bw for 10 weeks and 3,000 mg/kg bw for 58 weeks; or 4,000 mg/kg bw for 10 weeks, 5,000 mg/kg bw for 10 weeks and 6,000 mg/kg bw for 58 weeks, respectively.

Chapter 4

Genotoxicity

The summary in this chapter is primarily based on the information given in IARC (1999) and ATSDR.[2,17] In vitro and in vivo genotoxicity data are summarized below and presented in a table in Annex H.

4.1 In vitro assays

Both positive and negative findings have been reported in in vitro genotoxicity assays. Negative results might be attributed to low exposure levels, as for most of these studies it is not clear to the Committee whether measures were taken to prevent evaporation of the highly volatile 1,1,1-trichloroethane. However, also for those assays reported to be conducted using a desiccator (thereby preventing evaporation), both positive as well as negative results have been observed. Importantly, it is not exactly clear whether technical or purified 1,1,1 trichloroethane has been tested. As technical 1,1,1-trichloroethane contains (suspected) genotoxic stabilizers such as 1,2-epoxybutane and 1,4-dioxane, no conclusions can be drawn from these positive results.

Prokaryotic cell systems

1,1,1-Trichloroethane has been extensively tested in several bacterial mutagenicity assays, in which also inconsistent results have been obtained.

Ames tests have shown predominantly positive results in strains TA100 and TA1535, both with and without metabolic activation.[18-27] A positive result was found for TA104 with metabolic activation.[27] One out of six tests with TA98 was also positive (with and without metabolic activation), as was the only test with TA97.[18,19,21,22,27,28] Tests with TA1537 and TA1538 were all negative.[18,19,21,22,24] Two mutation tests with *S. typhimurium* showed negative results.[29,30] One of the three reverse mutation tests with *Escherichia coli* was positive without metabolic activation.[18,19,28] Legault et al. tested 1,1,1-trichloroethane in four different bacterial assays, namely the Ames plate incorporation assay; the fluctuation test with TA98 and TA100; the SOS Chromotest (*E.coli* PQ37); and, the Mutatox assay (*V. fischeri* M169).[31] All results were negative. Furthermore, 1,1,1-Trichloroethane did not induce an SOS response in the *umu* test with tester strain *S. typhimurium* TA1535/pSK1002.[32]

Tests conducted with *S. cerevisiae* for mutations, gene conversions, and induction of DNA damage or aneuploidy, were all negative.[33-38] An intra-chromosomal recombination assay in *S. cerevisiae* showed equivocal results.[39] The fungus *A. nidulans* showed no genetic crossing-over or aneuploidy without metabolic activation.[40]

Mammalian cell systems

Mixed results have been observed in mammalian genotoxicity assays. Assays for the induction of micronuclei in cytochalasin B-induced binucleate cells of human lymphoblastoid cell lines of varying metabolic activity (AHH-1 with CYP1A1 activity, h2E1 with CYP2E1, and MCL-5 with multiple CYP activities), were all positive.[41] An increase of cells with chromosomal aberrations was found in Chinese hamster ovary cells without metabolic activation, but not with metabolic activation.[42] Additionally, two sister chromatid exchange tests have been performed in Chinese hamster ovary cells. One was negative with metabolic activation and the other was inconclusive, either with or without metabolic activation.[42,43]

Mouse lymphoma tests showed no increase in the mutant frequency in the *tk* locus of L5178Y cells without metabolic activation, but in two out of three tests with metabolic activation the result was inconclusive.[44-46]

Cultured rat hepatocytes exposed to 1,1,1-trichloroethane vapour at concentrations of 0.1-2.5% (non-stabilized), and 0.1-5% (stabilized; not analytically confirmed) for 15 hours, did not increase unscheduled DNA synthesis.[25] Hasspieler et al. tested 1,1,1-trichloroethane in a DNA single-strand break assay and in an UDS-DNA repair assay, in a human hepatic cell line,

HepG2. The DNA single-strand assay was negative at 25-500 µg/µL, while the UDS-DNA repair assay was positive at concentrations exceeding 25 µg/µL.[47]

Several studies are available for which their relevance to mutagenicity (or carcinogenicity) is unclear. Cell transformation assays in BALB/c-3T3 mouse cells, Fischer rat embryo cells or SA7/Syrian hamster embryo cells, only tested without metabolic activation, were all positive.[48-50] 1,1,1-Trichloroethane was also found to bind to calf thymus DNA, RNA or protein when incubated with rat or mouse liver microsomes, although at much lower rates/exposure levels than measured for 1,1,2-trichloroethane, 1,1-dichloroethane, and other haloethanes, as was stated by the authors.[51]

4.2 In vivo assays

Three micronucleus tests in mice are available; in all three negative results have been observed.

One bone marrow micronucleus assay has been conducted using male and female NMRI mice. Animals were administrated 1,1,1-trichloroethane at a dose of 100, 266, or 2,000 mg/kg bw i.p. (4 animals per dose), at 0 and 24 h. Bone marrow smears were prepared 30 hours after the last treatment. Control animals were treated with vehicle only. No significant increase in micronucleated polychromatic erythrocytes was found.[20]

Tsuchimoto and Matter reported on the ability of 1,1,1-trichloroethane to induce micronucleated erythrocytes in CD-1 mice.[52] Animals received two intraperitoneal doses of 0.008, 0.016 and 0.032 mL/kg bw (equivalent to doses of 11, 21 and 42 mg/kg bw, assuming the administration of pure 1,1,1-trichloroethane), for two consecutive days. Six hours after the last dose was applied, the animals was killed and bone marrow smears were analysed. No increase in cells with micronuclei were noted. Despite the claim of the authors that the doses applied represent one-eighth, one-fourth and one-half of the LD_{50}, the Committee notes that no data on acute toxicity via the intraperitoneal route are available, and the doses applied in this study appear to be far below the doses applied by Gocke et al. (1981).[20]

A micronucleus assay with a modified test protocol was applied by Salamone et al. (1981), involving multiple sampling at one dose rather than a single sample at multiple doses.[53] B6C3F1 hybrid mice were injected i.p. with 1,1,1-trichloroethane at 0 and 24 hours, at a dose which was reported as '80% of the LD_{50}' (not further specified). Samples were subsequently taken at 48, 72 and 96 hours after the last injection. A significant increase of cells with polychromatic micronuclei

were observed after 72 hours. A confirmation test at doses of 40, 60, and 80% of the LD_{50}, however, could not confirm this positive finding. The interpretation of this study is hampered by the fact that the actual doses were not specified.

DNA, RNA and protein binding of 1,1,1-trichloroethane *in vivo* was studied in six male Wistar rats and twelve male BALB/c mice. Radiolabeled compound was injected intraperitoneally and animals were killed 22 hours later. Similar results were obtained for both species; binding to isolated DNA, RNA and proteins was reported as low in general, with a slightly higher labeling in kidney DNA and RNA.[51]

1,1,1-Trichloroethane was included in a screen for inducing spermhead abnormalities in mice, as an indicator test for germ cell mutagenicity. Five male BALB/c mice received intraperitoneal 5 daily concentrations of 1,1,1-trichloroethane in a volume of 0.1, 0.25, 0.5, 1.0, 2.0 mL in corn oil for 5 weeks. The exact dose was not specified but reported as equivalents of the LD_{50}; the highest dose was recorded as a lethal dose. No abnormal sperm morphology was noted.[54]

Sex-linked recessive lethal mutations were not found in *D. melanogaster*.[20]

4.3 Carcinogenic mechanism of action

For 1,1,1-trichloroethane, no general view on a possible mode of action for genotoxic and carcinogenic activity is published in literature.

Chapter 5

Classification

5.1 Evaluation of data on carcinogenicity and genotoxicity

Although a number of epidemiological studies is available, none of these studies provide basis for conclusions on the potential carcinogenic properties of 1,1,1-trichloroethane. In some studies associations were found, however, all of these studies have been compromised by methodological limitations, such as lack of statistical power and multiple exposures to other solvents – some of which are known or suspected to be carcinogens themselves.

Animal carcinogenicity studies are available. Inhalation studies did not reveal evidence for carcinogenicity. For the oral route, an increased incidence of leukemia (mainly immunoblastic lymphoma) was noted in rats of both sexes, whereas another study showed no increased incidence of tumours in rats or mice. The Committee, however, considers both oral studies of insufficient quality to assess the carcinogenicity of 1,1,1-trichloroethane.

Conflicting results have been obtained for *in vitro* genotoxicity testing. Both negative, and positive or equivocal results have been obtained in bacterial and mammalian assays covering either gene mutations, structural or numerical chromosome aberrations. In contrast, no DNA damage (recombinagenic effects, aneuploidy) was reported in yeast and no mutations were found in the sex-linked recessive *Drosophila* test. The high volatility of 1,1,1-trichloroethane does not appear to provide a sound explanation for the conflicting results that are also observed in enclosed systems. Conflicting results might be attributed to the presence of genotoxic additives, as commercially available 1,1,1-trichloroethane

is known to contain stabilisers such as nitromethane, 1,4-dioxane and butylene oxide.[14,17]

In vivo, no clastogenicity is observed in mice exposed to 1,1,1-trichloroethane.[20,52,53]

As conflicting results have been obtained *in vitro*, and no information is available related to the potential to induce gene mutation *in vivo*, no conclusion can be drawn on the genotoxicity of 1,1,1-trichloroethane. It can, therefore, not be excluded that 1,1,1-trichloroethane possesses a low genotoxic potential.

5.2 Recommendation for classification

The Committee is of the opinion that the available data are insufficient to evaluate the carcinogenic properties of 1,1,1-trichloroethane (category 3).*

* According to the classification system of the Health Council (see Annex I).

References

1 Environmental Health Criteria: 1,1,1-Trichloroethane. 1992: 136. Internet: http://www.inchem.org/documents/ehc/ehc/ehc136.htm.

2 1,1,1-Trichloroethane. IARC Monogr Eval Carcinog Risks Hum 1999; 71: 881-903.

3 Antilla A, Pukkala E, Sallmen M, Hernberg S, Hemminki K. Cancer incidence among Finnish workers exposed to halogenated hydrocarbons. J Occup Environ Med 1995; 37(7): 797-806.

4 Spirtas R, Stewart PA, Lee JS, Marano DE, Forbes CD, Grauman DJ et al. Retrospective cohort mortality study of workers at an aircraft maintenance facility. I. Epidemiological results. British Journal of Industrial Medicine 1991; 48: 515-530.

5 Blair A, Hartge P, Stewart PA, McAdams M, Lubin J. Mortality and cancer incidence of aircraft maintenance workers exposed to trichloroethylene and other organic solvents and chemicals: extended follow up. Occup Environ Med 1998; 55(3): 161-171.

6 Gold LS, Stewart PA, Milliken K, Purdue M, Severson R, Seixas N et al. The relationship between multiple myeloma and occupational exposure to six chlorinated solvents. Occup Environ Med 2011; 68(6): 391-399.

7 Risk factors for cancer in the workplace. Siemiatycki, J. Risk factors for cancer in the workplace. Boca Raton: CRC Press; 1991.

8 Kernan G, Ji B-T, Dosemeci M et al. Occupational risk factors for pancreatic cancer: a case-control study based on death certificates from 24 U.S. states. Am J Ind Med 1999; 36: 260-270.

9 Dosemeci M, Cocco P, Chow W-H. Gender differences in risk of renal cell carcinoma and occupational exposures to chlorinated aliphatic hydrocarbons. Am J Ind Med 1999; 36: 54-59.

10 Heineman E, Cocco P, Gomez M et al. Occupational Exposure to chlorinated aliphatic hydrocarbons and risk of astrocytic brain cancer. American Journal of Industrial medicine 1994; 26: 155-169.

11 Infante-Rivard C, Siemiatycki J, Lakhani R et al. Maternal exposure to occupational solvents and childhood leukemia. Environ Health Perspect 2005; 113(6): 787-792.
12 Toxicological review of 1,1,1-trichloroethane. Washington DC: US EPA; 2007. Internet: www.epa.gov/iris.
13 Quast J, Calhoun L, Frauson LE. 1,1,1-Trichloroethane formulation: a chronic inhalation toxicity and oncogenicity study in Fischer 344 rats and B6C3F1 mice. Fundam Appl Toxicol 1988; 11: 611-625.
14 1,1,1-Trichloroethane. IARC Monogr Eval Carcinog Risks Hum 1979; 20: 515-531.
15 Maltoni C, Cotti G, Patella V. Results of long-term carcinogenicity bioassays on Sprague-Dawley rats of methyl chloroform, administered by ingestion. Acta Oncol 1986; 7(2): 101-117.
16 National Cancer Institute. Bioassay of 1,1,1-trichloroethane for possible carcinogenicity (CAS No. 71-55-6). Washington DC: US Department of Health, Education & Welfare; 1977: 3.
17 Agency for Toxic Substances and Disease Registry (ATSDR). Toxicological Profile for 1,1,1-Trichloroethane. 2006.
18 Falck K, Partanen P, Sorsa M et al. Mutascreen, an automated bacterial mutagenicity assay. Mutat Res 1985; 150: 119-125.
19 Gatehouse D. Mutagenic activity of 42 coded compounds in the 'microtiter' fluctuation test. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. de Serres FJ & Ashby J, eds. [1], 376-386. 1981. Amsterdam Elsevier.
20 Gocke E, King M.-T, Eckhardt K.& Wild D. Mutagenicity fo cosmetics ingredients licensed by the European communities. Mutat Res 1981; 90: 91-109.
21 Haworth S, Lawlor T, Mortelmans K et al. Salmonella mutagenicity test results for 250 chemicals. Environ Mutag 1983; 5 (suppl. 1): 3-142.
22 Nestmann ER, Lee EGH, Matula TI et al. Mutagenicity of constituents identified in pulp an dpaper mill effluents using the Salmonella/mammalian-micorsome assay. Mutat Res 1980; 79: 203-212.
23 Nestmann ER, Otson R, Kowbel DJ, Bothwell PD, Harrington TR. Mutagenicity in a modified Salmonella assay of fabric-protecting products containing 1,1,1-trichloroethane. Environ Mutagen 1984; 6(1): 71-80.
24 Richold M and Jones E. Mutagenic activity of 42 coded compounds in the Salmonella/microsome assay. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. De Serres FJ and Ashby J, eds. [1], 314-322. 1981. Amsterdam Elsevier.
25 Shimada T, Swanson AF, Leber P et al. Activities of chlorinated ethane and ethylene compounds in the Salmonella/rat microsome mutagenesis and rat hepatocyte/DNA repair assays under vapor phase exposure conditions. Cell Biol Toxicol 1985; 1: 159-179.
26 Simmon VF, Kauhanen K, Tardiff RG. Mutagenic activity of chemicals identified in drinking water. Progress in Genetic Toxicology: Developments in Toxicology and Environmental Science, Vol. 2, 249-258. 1977. Amsterdam Elsevier.
27 Strobel K and Grummt T. Aliphatic and aromatic halocarbons as potential mutagens in drinking water. III. Halogenated ethanes and ethenes. Toxicol Environ Chem 1987; 15: 101-128.

28 Norpoth K, Reisch A, Heinecke A. Biostatistics of Ames-test data. In: Short-Term Test Systems for Detecting Carcinogens. Norpoth KH and Garner RC, eds, New York, Springer-Verlag.; 1980: 312-322.

29 Roldan-Arjona T, Garcia-Pedrajas MD, Luque-Romero FL et al. An association betwen mutagenicity of the Ara test of Salmonella typhimurium and carcinogenicity in rodents for 16 halogenated aliphatic hydrocarbons. Mutagenesis 1991; 6: 199-205.

30 Skopek TR, Andon BM, Kaden DA and Thilly WG. Mutagenic activity of 42 coded compounds using 8-azaguanine resistance as a genetic marker in Salmonella typhimurium. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. de Serres FJ and Ashby J, eds. [1], 371-375. 1981. Amsterdam Elsevier.

31 Legault R, Blaise C, Rokosh D et al. Comparative assessment of the SOS chromotest kit and the mutatox test with the Salmonella plate incorporation (Ames test) and fluctuation tests for screening genotoxic agents. Environ Toxicol Water Qual 1994; 9: 45-57.

32 Nakamura S, Oda Y.Shimada T et al. SOS-inducing activity of chemical carcinogens and mutagens in Salmonella typhimurium TA1535/pSK1002: examination with 151 chemicals. Mutat Res 1987; 192: 239-246.

33 Jagannath DR, Vultaggio DM and Brusick DJ. Genetic activity of 42 coded compounds in the mitotic gene conversion assay using Saccharomyces cerevisiae strain D4. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. de Serres F.J.& Ashby J., eds. [1], 456-467. 1981. Amsterdam Elsevier.

34 Mehta RD and Von Borstel RC. Mutagenic activity of 42 encoded compounds in the haploid yeast reversion assay, strain XV185-14C. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. De Serres F.J.& Ashby J., eds. [1], 414-423. 1981. Amsterdam Elsevier.

35 Parry JM and Sharp DC. Induction of mitotic aneuploidy in the yeast strain D6 by 42 coded compounds. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. de Serres FJ and Ashby J, eds. [1], 468-480. 1981. Amsterdam Elsevier.

36 Sharp DC and Parry JM. Use of repair-deficient strains of yeast to assay the activity of 40 coded compounds. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. De Serres FJ and Ashby J, eds. [1], 502-516. 1981. Amsterdam Elsevier.

37 Whittaker SG, Zimmermann FK, Dicus B et al. Detection of induced mitotic chromosome loss in Saccharomyces cerevisiae - An interlaboratory assessment of 12 chemicals. Mutat Res 1990; 241: 225-242.

38 Zimmerman FK and Scheel I. Induction of mitotic gene conversion in strain D7 of Saccharomyces cerevisiae by 42 coded chemicals. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. De Serres FJ and Ashby J, eds. [1], 481-490. 1981. Amsterdam Elsevier.

39 Brennan R and Schiestl RH. Chloroform and carbon tetrachloride induce intrachromosomal recombination and oxidative free radicals in Saccharomyces cerevisiae. Mutat Res 1998; 397: 271-278.

40 Crebelli R, Benigni R, Franekic J et al. Induction of chromosome malsegregation by halogenated organic solvents in Aspergillus nidulans: unspecific or specific mechanism? Mutat Res 1988; 201: 401-411.

41 Doherty AT, Ellard S, Parry EM, Parry JM. An investigation into the activation and deactivation of chlorinated hydrocarbons to genotoxins in metabolically competent human cells. Mutagenesis 1996; 11(3): 247-274.

42 Galloway SM, Armstrong MJ, Reuben C. Chromosome aberrations an dsister chromatid exchanges in Chinese hamster ovary cells: Evaluations of 108 chemicals. Environ Mol Mutag 1987; 10 (suppl 10): 1-175.

43 Perry PE, Thomson EJ. Evaluation of the sister chromatid exchange method in mammalian cells as a screening system for carcinogens. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. De Serres FJ and Ashby J eds. [1], 560-569. 1981. Amsterdam Elsevier.

44 Mitchell AD, Rudd CJ and Caspary WJ. Evaluation of the L5178Y mouse lymphoma cell mutagenesis assay: intralaboratory results for sixty-three coded chemicals tested at SRI International. Environ Mol Mutag 1988; 12 (suppl 13): 37-101.

45 Myhr BC and Caspary WJ. Evaluation of the L5178Y mouse lymphoma cell mutagenesis assay: Intralaboratory results for sixty-three coded chemicals tested at Litton Bionetics, Inc. Environ Mol Mutag 1988; 12 (suppl 13): 103-194.

46 Tennant RW, Stasiewicz S and Spalding JW. Comparison of multiple parameters of rodent carcinogenicity and in vitro genetic toxicity. Environ Mutag 1986; 8: 205-227.

47 Hasspieler B, Haffner D, Stelljes M, Adeli K. Toxicological assessment of industrial solvents using human cell bioassays: assessment of short-term cytotoxicity and long-term genotoxicity potential. Toxicology and Industrial Health 2006; 22: 301-315.

48 Hatch GG, Mamay PD, Ayer ML et al. Chemical enhancement of viral transformation in Syrian hamster embryo cells by gaseous and volatile chlorinated methanes and ethanes. Cancer Res 1983; 43: 1945-1950.

49 Price PJ, Hassett CM and Mansfield JI. Transforming activities of trichloroethylene and proposed industrial alternatives. In Vitro 1978; 14: 290-293.

50 Tu AS, Murray TA, Hatch KM et al. In vitro transformation of BALB/c-3T3 cells by chlorinated ethanes and ethylenes. Cancer Lett 1985; 28: 85-92.

51 Turina MP, Collaci A, Grilli S et al. Short-term tests of genotoxicity for 1,1,1-trichloroethane. Res Comm Chem Pathol Pharmacol 1977; 52: 305-320.

52 Tsuchimoto T, Matter BE. Activity of coded compounds in the micronucleus test. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. de Serres FJ and Ashby J, eds. [1], 705-711. 1981. Amsterdam Elsevier.

53 Salamone MF, Heddle JA and Katz M. Mutagenic activity of 41 compounds in the in vivo micronucleus assay. In: Evaluation of Short-Term Tests for Carcinogens. Report of the International Collaborative Program. De Serres FJ and Ashby J, eds. [1], 686-697. 1981. Amsterdam Elsevier.

54 Topham JC. Do induced sperm-head abnormalities in mice specifically identify mammalian mutagens rather than carcinogens? Mutat Res 1980; 74: 379-387.

55 Health Council of the Netherlands. Guideline to the classification of carcinogenic compounds. Health Council of the Netherlands, The Hague; 2010: publication no. A10/07E.

A	Request for advice
B	The Committee
C	Submission letter (in English)
D	Comments on the public review draft
E	IARC Monograph
F	Human data
G	Animal data
H	Genotoxicity data
I	Carcinogenic classification of substances by the Committee

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

- R.A. Woutersen, *chairman*
  Toxicologic Pathologist; TNO Innovation for Life, Zeist; Professor of Translational Toxicology, Wageningen University and Research Centre, Wageningen
- J. van Benthem
  Genetic Toxicologist, National Institute for Public Health and the Environment, Bilthoven
- P.J. Boogaard
  Toxicologist, SHELL International BV, The Hague
- G.J. Mulder
  Emeritus Professor of Toxicology, Leiden University, Leiden
- Ms M.J.M. Nivard
  Molecular Biologist and Genetic Toxicologist, Leiden University Medical Center, Leiden
- G.M.H. Swaen
  Epidemiologist, Dow Chemicals NV, Terneuzen
- E.J.J. van Zoelen
  Professor of Cell Biology, Radboud University Nijmegen, Nijmegen
- S.R. Vink, *scientific secretary*
  Health Council of the Netherlands, The Hague

The Health Council and interests

Members of Health Council Committees are appointed in a personal capacity because of their special expertise in the matters to be addressed. Nonetheless, it is precisely because of this expertise that they may also have interests. This in itself does not necessarily present an obstacle for membership of a Health Council Committee. Transparency regarding possible conflicts of interest is nonetheless important, both for the chairperson and members of a Committee and for the President of the Health Council. On being invited to join a Committee, members are asked to submit a form detailing the functions they hold and any other material and immaterial interests which could be relevant for the Committee's work. It is the responsibility of the President of the Health Council to assess whether the interests indicated constitute grounds for non-appointment. An advisorship will then sometimes make it possible to exploit the expertise of the specialist involved. During the inaugural meeting the declarations issued are discussed, so that all members of the Committee are aware of each other's possible interests.

Annex **C**

Submission letter (in English)

Subject : Submission of the advisory report *1,1,1-Trichloroethane*
Our reference : DGV/MBO/U-932342
Your Reference : U-7258/SV/fs/246-N16/E
Enclosed : 1
Date : 24 July 2012

Dear State Secretary,

I hereby submit the advisory report on the effects of occupational exposure to *1,1,1-Trichloroethane.*

This advisory report is part of an extensive series in which carcinogenic substances are classified in accordance with European Union guidelines. This involves substances to which people can be exposed while pursuing their occupation.

The advisory report was prepared by the Subcommittee on Classifying Carcinogenic Substances, a permanent subcommittee of the Health Council's Dutch Expert Committee on Occupational Safety (DECOS). The advisory report has been assessed by the Health Council's Standing Committee on Health and the Environment.

I have today sent copies of this advisory report to the State Secretary of Infrastructure and the Environment and to the Minister of Health, Welfare and Sport, for their consideration.

Yours sincerely,

(signed)
Prof. H. Obertop,
Acting President

Annex **D**

Comments on the public review draft

A draft of the present report was released in 2012 for public review. The following organisations and persons have commented on the draft document:

- Ms V. Gálvez Pérez, Instituto Nacional de Seguridad e Higiene en el Trabajo, Madrid, Spain
- Mr T.J. Lentz, National Institute of Occupational Safety and Health, Cincinnati, the USA

Annex **E**

IARC Monograph

VOL: 71 (1999) (p.1295)

Summary of Data Reported and Evaluation

Exposure data

1,1,1-Trichloroethane is a solvent. It has been detected in waste-, ground-, drinking- and ambient water as well as in ambient and urban air.

Human carcinogenicity data

An increased risk for central nervous system and multiple myeloma was reported from a cohort study of workers exposed to 1,1,1-trichloroethane in Finland. These findings were not confirmed by two case-control studies carried out in the United States and Canada, while an increased risk for cancer of the lung and kidney was shown in the Canadian study.

Animal carcinogenicity data

1,1,1-Trichloroethane was tested for carcinogenicity by oral administration in rats in two experiments and in mice in one experiment. Although leukaemia was

seen in both sexes of rats in one study and a few liver tumours occurred in male mice, the results of these studies were considered to be inadequate for evaluation. 1,1,1-Trichloroethane was tested by inhalation in rats in two experiments and in mice in one experiment. No chemically related increase in tumour incidence was observed in either rats or mice in one adequate study. Another inhalation study was considered to be inadequate.

In a multistage study for γ-glutamyltranspeptidase (γ-GT)-positive foci in the liver of male rats, neither single administration of 1,1,1-trichloroethane by gavage after a two-thirds partial hepatectomy followed by treatment with phenobarbital (initiation study) nor repeated administration of 1,1,1-trichloroethane by gavage after two-thirds partical hepatectomy and initiation with N-nitrosodiethylamine (promotion study) increased the number of (γ-GT)-positive foci.

Other relevant data

Absorption of 1,1,1-trichloroethane vapour is mainly through the respiratory tract. It is rapidly eliminated form blood. Metabolism plays a minor role in this process, more than 90% being eliminated unchanged, both in exposed people and rodents. The main metabolites are trichloroethanol, trichloroacetic acid and carbon dioxide.

1,1,1-Trichloroethane is neurotoxic and hepatotoxic, following exceptionally high exposure concentrations of people and also in rodents. No structural damage has been reported in reproductive toxicity studies in rats and mice, but delayed development, particularly of neurological attributes, has been reported in one study with mice.

1,1,1-Trichloroethane covalently bound to DNA, RNA and protein in mice and rats buth did not induce micronuclei or abnormal sperm head morphology in mice in vivo. It induced chromosomal aberrations and cell transformation in mammalian cell cultures and it showed inconclusive evidence of sister chromatid exchange induction. It did not induce unscheduled DNA synthesis or gene mutation in mammalian cells in vitro. 1,1,1-Trichloroethane did not cause mutation in plants or sex-linked mutation in *Drosophila*. It did not induce DNA

damage, gene conversion, mutation or aneuploidy in yeast or genetic crossing-over or aneuploidy in fungi, but it was mutagenic to some bacterial strains.

Evaluation

There is *inadequate evidence* for the carcinogenicity of 1,1,1-trichloroethane in humans. There is *inadequate evidence* for the carcinogenicity of 1,1,1-trichloroethane in experimental animals.

Overall evaluation

1,1,1-Trichloroethane is not classifiable as to its carcinogenicity to humans (Group 3).

Annex **F**

Human studies

Study design and population information	Exposure information	Exposure duration	Follow-up	Tumour type; relative risk (95% CI)	Ref.
Cohort; Finnish workers; 140 male and 131 female	No data;	0 - 8 y	8-17 y	Cancer of the nervous system (3 cases): SIR 6.1 (95% CI 1.25-17.7); multiple myeloma (2 cases): SIR 16.0 (95% CI 1.9-57.7)	3
Cohort; employees working at Hill Air Force Base; n = 14,457	No data	> 1 y	>1 y	Multiple myeloma mortality (2 cases): SMR 56.6 (95% CI 6.9-204.5)	4
				Multiple myeloma mortality (2 cases): SMR 13.2 (95% CI 2.2-80.4)	5
Case-control; 181 cases and 481 controls	Four exposure categories up to 60,000 (ppm*y) (10-y lagged and unlagged)	Exposure categories up to 45 years of exposure	--	Multiple myeloma: No significant increased ORs for exposure categories separately. OR for 'ever' exposed subjects 1.8 ((95% CI 1.1-2.9) (primary analysis) or 2.2 (95% CI 1.1-4.4) (reanalysis)	6

Case-control; 300 cases and 320 controls from population in 3 US states with prominent workforce representation in the petroleum refining and chemical manufacturing industries	Low to high exposure combined	Ever exposed, 2-20 y or 21+ y	--	Astrocytic brain cancer: OR 1.2 (95% CI 0.9-1.8) for ever exposed; OR 1.1 (95% CI 0.7-1.7) for 2-20 years exposed; OR 1.8 (95% CI 1.0-3.3) for 21+ years exposed; Chi for trend 1.87 ($p<0.05$)	[10]
	low-medium or high intensity of exposure	21+ y	--	astrocytic brain cancer; OR 1.6 (95% CI 0.9-3.1) for low-medium intensity exposure; OR 3.7; (95% CI 0.7-27.9) for high intensity exposure; Chi for trend 2.28 ($p<0.05$)	
Case-control; 5,866 cases and 252,386 controls from 24 US states	Probability of low, medium or high exposure	No data	--	pancreatic cancer mortality; OR 2.9 (95% CI 1.2-7.5) for black males with high probability of exposure (8 cases exposed)	[8]
Case-control; 438 cases and 687 controls	No data	No data	--	renal cell cancer (n=13); OR 1.26 (95% CI 0.6-2.8)	[9]
Case-control; 3,730 cases and 533 controls	Any level	No data	--	Lung cancer (French Canadians; n=7); OR 3.5 (90% CI 1.0-12.0); kidney cancer (whole population; n=4) 2.4 (90% CI 1.0-6.0)	[7]
Case-control; Quebec, Canada; 790 cases and 790 controls	Maternal exposure	No data	--	Acute lymphoblastic leukemia in offspring; OR 7.55 (95% CI 0.92-61.97)	[11]
Case-control; 14,067 employees at Rohr plant (control: total US white and nonwhite population and San Diego County population	No data	No data	No data	No increased risk of esophageal cancer	[12]
Case-control; 14 (esophageal cancer) and 8 (stomach cancer) cases, 56 and 32 controls for esophageal and stomach cancer, resp.	No data	No data	No data	No increased risk of esophageal or stomach cancer	[12]

Annex **G**

Animal studies

species	dose	freq	sex (no./ group)	X_{po}	X_{pe}	no. survivors	no. animals with tumours	comments/ specified tumours	Ref.
Inhalation studies									
Fischer 344 rat	0, 820, 2,700 and 8,200 mg/m^3 (0, 150, 500 and 1,500 ppm)[a]	6 hr/d; 5 d/w	M/F (50)	24 months	24 months	not significantly < control (range 50-60%, read from graph)	bilateral, benign, primary interstitial cell tumour of testes: 36/50, 30/50, 38/50. 45/50[b]	unilateral or unilateral and bilateral combined was not increased	13
B6C3F1 mouse	0, 820, 2,700 and 8,200 mg/m^3 (0, 150, 500 and 1,500 ppm)[a]	6 hr/d; 5 d/w	M/F (50)	24 months	24 months	not significantly < control (range 50-60%, read from graph)	lacrimal/ Harderian gland: M 8/50, 8/49, 5/50, 4/50; F 3/50, 1/50, 2/50, 7/50[c]	adenoma and cystadenoma combined; historical control incidence: 4-12%	13
Sprague-Dawley rat	0, 4,700 and 9,500 mg/m^3 (0, 875 and 1,750 ppm	6 hrs/d; 5 d/w	M/F (96)	12 months	30 months	no data	incidence of neoplasms comparable to control		13,14

Oral studies

Sprague-Dawley rat	0 and 500 mg/kg bw/d in olive oil (gavage)[d]	4-5 d/w	M/F (50 control; 40 treated)	104 weeks	141 weeks	not significantly < control	leukemia/ lymphoma: M 3/50, 9/40, F 1/50. 4/40 (no statistical analysis)	mainly immuno-blastic lympho-sarcomas in the lung	15
Osborne-Mendel rat	0, 750 or 1,500 mg/kg bw/d in corn oil (gavage)	5 d/w	M/F (20 control; 50 treated)	78 weeks	110 weeks	M 0/20, 0/50, 0/50, F 3/20, 2/50, 1/50	incidence and type similar to control	high incidence of chronic murine pneumonia present in all control and treated rats (M/F) was probably cause for high incidence of early deaths	16
B6C3F1 mouse	0; Low dose: 2,000, 2,500 and 3,000 mg/kg bw/d for 10, 10 and 58 w, resp. High dose: 4,000, 5,000 and 6,000 mg/kg bw/d for 10, 10 and 58 w, resp. (gavage)	5 d/w	M/F (20 control; 50 treated)	78 weeks	90 weeks	M 10/20, 19/50, 15/50; F 19/20, 41/50, 30/50	incidence and type of neoplasms similar to control	high incidence of chronic murine pneumonia present in all control and treated mice (M/F) was probably cause for high incidence of early deaths	16

M = male; F = female; freq = frequency; X_{po}= duration of exposure; X_{pe}= duration of the experiment.

a production grade 1,1,1-trichloroethane was used containing 5% stabilizers: butylene oxide, t-amyl alcohol, methyl butynol, nitroethane, and nitromethane; <1% minor impurities

b Cochran-Armitage test; α = 0.02 (2-sided)

c Cochran-Armitage test; α = 0.05 (1-sided)

d Technical-grade 1,1,1-trichloroethane was used with maximum level of stabilizers and impurities: 3.8% 1,4-dioxane, 0.47% 1,2-epoxybutane, 0.27% nitromethane, <1 ppm N-methylpyrrole, 100 ppm chloroform, 250 ppm carbon tetrachloride, 426 ppm 1,1-dichloroethane, 2.3 ppm 1,2-dichloroethane, 41.8 ppm 1,2,3-trichloroethane, 398 ppm 1,1-dichloroethylene, 50 ppm 1,2-dichloroethylene trans, 200 ppm trichloroethylene, 475 ppm tetrachloroethylene.

Annex **H**

Genotoxicity data

In vitro and in vivo genotoxicity data are presented below. If some information was given on restriction of the volatile compound, it is presented under exposure conditions. Results from references cited in IARC were copied from IARC. [2]

Test system	Exposure conditions	Dose[a] (LED or HID)	Result [b]		Ref.
			exogenous metabolic activation		
			without	with	
In vitro					
Prophase, induction, SOS response, strand-breaks or cross-links		666	–	–	32
Salmonella typhimurium					
forward mutation		1,000	NT	–	30
forward mutation (*Ara* test)		3,75	–	–	29
TA100, reverse mutation	desiccator	70	+	+	26
TA100, reverse mutation		up to 5,000	–	–	18,21
TA100, reverse mutation	in desiccator, vapour exposure	NG	+	+	22
TA100, reverse mutation	desiccator	144	–	(+)	20
TA100, reverse mutation		150	+	+	23
TA100, reverse mutation	vapour exposure	$266 \bullet 10^3$ mg/m^3	–	–	25

TA100, reverse mutation		500	+	–	27
TA100, reverse mutation		3,300	–	NT	31
TA104, reverse mutation		5	–	+	27
TA1535, reverse mutation	vapour	NG	+	+	22
TA1535, reverse mutation		up to 5,000	–	–	18,19,21,24
TA1535, reverse mutation	desiccator	144	+	+	20
TA1535, reverse mutation		80	+	+	23
TA1535, reverse mutation	vapour exposure	$266 \bullet 10^3$ mg/m^3	–	–	25
TA1537, reverse mutation	vapour	1,000	–	–	22
TA1537, reverse mutation		up to 5,000	–	–	24 18,21
TA1538, reverse mutation	vapour	1,000	–	–	22
TA1538, reverse mutation		up to 5,000	–	–	18,24
TA98, reverse mutation	vapour	1,000	–	–	22
TA98, reverse mutation		134	–	NT	28
TA98, reverse mutation		3,300	–	NT	31
TA98, reverse mutation		up to 5,000	–	–	18,19,21
TA98, reverse mutation		5	+	+	27
TA97, reverse mutation		5	+	+	27
Escherichia coli WP2 uvrA		268	NT	+	28
Escherichia coli WP2 uvrA		up to 1,000	–	–	18,19
Escherichia coli PQ37, SOS Chromotest, DNA damage		1,000	–	NT	31
Vibrio fischeri M169, Mutatox test, DNA damage		4,800	–	NT	31
Saccharomyces cerevisiae, differential toxicity		750	–	–	36
Saccharomyces cerevisiae D4, gene conversion		125	–	–	33
Saccharomyces cerevisiae JD1, gene conversion		750	–	–	36
Saccharomyces cerevisiae D7, gene conversion		2,600	NT	–	38
Aspergillus nidulans, strain P1 crossing-over	sealed tube	1,300	–	NT	40
Saccharomyces cerevisiae XV185-14C, reverse mutation		1,488	–	–	34
Saccharomyces cerevisiae D6, aneuploidy	sealed bottle	500	–	–	35
Saccharomyces cerevisiae D61.M, aneuploidy		6,000	–	NT	37
Aspergillus nidulans strain P1, aneuploidy	sealed tube	1,300	–	NT	40
Drosophila melanogaster, Basc strain, sex-linked recessive lethal mutations		3,335 µg/mL feed	–		20

Unscheduled DNA synthesis, rat primary hepatocytes	vapour exposure	133	–	NT	25
Human hepatic cell line, HepG2, DNA single-strand breaks		500	–		47
Human hepatic cell line, HepG2, DNA repair		50	+		47
Gene mutation, mouse lymphoma L5178Y cells, *tk* locus		NG/536	–	?	45,46
Gene mutation, mouse lymphoma L5178Y cells, *tk* locus		680	–	–	44
Sister chromatid exchange, Chinese hamster ovary cells		10	NT	–	43
Sister chromatid exchange, Chinese hamster ovary cells	?	1,000	?	?	42
Chromosomal aberrations, Chinese hamster ovary cells	?	160	+	–	42
Cell transformation, BALB/c-3T3 cells	glass incubation chamber	4	+	NT	50
Cell transformation, Fischer rat embryo cells		13	+	NT	49
Cell transformation, SA7/Syrian hamster embryo cells	vapour exposure	$11 \bullet 10^3$ mg/m^3	+	NT	48
Binding (covalent) to calf thymus DNA, rat/mouse RNA or protein		7.6	NT	+	51
In vivo					
Micronucleus test, NMRI mouse bone marrow		2x 2,000 ip	–		20
Micronucleus test, B6C3F1 mouse bone marrow		2x 67 ip	–		53
Micronucleus test, CD-1 mouse bone marrow		2x 43 ip	–		52
Binding (covalent) to DNA, RNA or protein, male Wistar rat and BALB/c mouse liver, kidney, lung and stomach		1.2 ip	+ (DNA(+))		51
Sperm morphology, mice		5x 1,340 ip	–		54

[a] LED = lowest effective dose; HID = highest ineffective dose; in vitro tests: µg/mL; in vivo tests: mg/kg bw/d; NG = not given; ip = intraperitoneal

[b] + = positive; (+) = weakly positive; – = negative; NT = not tested; ? = inconclusive

Annex I

Carcinogenic classification of substances by the Committee

The Committee expresses its conclusions in the form of standard phrases:

Category	Judgement of the Committee (GR_{GHS})	Comparable with EU Category	
		67/548/EEC before 12/16/2008	EC No 1272/2008 as from 12/16/2008
1A	The compound is known to be carcinogenic to humans. • It acts by a stochastic genotoxic mechanism. • It acts by a non-stochastic genotoxic mechanism. • It acts by a non-genotoxic mechanism. • Its potential genotoxicity has been insufficiently investigated. Therefore, it is unclear whether the compound is genotoxic.	1	1A
1B	The compound is presumed to be as carcinogenic to humans. • It acts by a stochastic genotoxic mechanism. • It acts by a non-stochastic genotoxic mechanism. • It acts by a non-genotoxic mechanism. • Its potential genotoxicity has been insufficiently investigated. Therefore, it is unclear whether the compound is genotoxic.	2	1B
2	The compound is suspected to be carcinogenic to man.	3	2
(3)	The available data are insufficient to evaluate the carcinogenic properties of the compound.	Not applicable	Not applicable
(4)	The compound is probably not carcinogenic to man.	Not applicable	Not applicable

Source: Health Council of the Netherlands. Guidline to the classification of carcinogenic compounds. The Hague: Health Council of the Netherlands, 2010; publication no. A10/07E.[55]