N-NITROSODIETHANOLAMINE

This substance was considered by previous working groups, in October 1977 (IARC, 1978) and March 1987 (IARC, 1987). Since that time, new data have become available, and these have been incorporated in the monograph and taken into consideration in the evaluation.

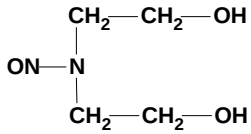
1. Exposure Data

1.1 Chemical and physical data

1.1.1 *Nomenclature*

Chem. Abstr. Serv. Reg. No.: 1116-54-7
Chem. Abstr. Name: 2,2′-(Nitrosoimino)bis[ethanol]
IUPAC Systematic Name: 2,2′-Nitrosoiminodiethanol
Synonyms: Diethanolnitrosamine; *N*,*N*-diethanolnitrosamine; *N*-nitrosobis(2-hydroxyethyl)amine; nitrosodiethanolamine; NDELA

1.1.2 *Structural and molecular formulae and relative molecular mass*



$C_4H_{10}N_2O_3$ Relative molecular mass: 134.14

1.1.3 *Chemical and physical properties of the pure substance*

(*a*) *Description*: Light yellow oil (Budavari, 1998)
(*b*) *Boiling-point*: 125 °C at 0.01 mm Hg [1.33 Pa] (Budavari, 1998)
(*c*) *Density*: 1.26 g/cm^3 (TCI America, 1999)

(*d*) *Solubility*: Miscible with water; soluble in polar solvents; insoluble in non-polar organic solvents (National Toxicology Program, 1998)
(*e*) *Volatility*: Vapour pressure, 5×10^{-5} mm Hg [0.007 Pa] at 20 °C; flash point, 11.6 °C (TCI America, 1999)
(*f*) *Stability*: Sensitive to light, especially ultraviolet light, and undergoes relatively rapid photolytic degradation (US National Toxicology Program, 1998)
(*g*) *Octanol/water partition coefficient (P)*: log P, –1.583 (TCI America, 1999)
(*h*) *Conversion factor*[1]: $mg/m^3 = 5.48 \times ppm$

1.1.4 *Technical products and impurities*

N-Nitrosodiethanolamine is not known to be a commercial product. It is available in research quantities (TCI America, 1999).

1.1.5 *Analysis*

N-Nitrosodiethanolamine can be determined in air and bulk process samples. Air samples are collected on glass fibre filters, extracted with 2-propanol and analysed by gas chromatography with thermal energy analyser detection. The limit of detection is 200 ng per sample (0.42 $\mu g/m^3$). Bulk samples can be screened for *N*-nitrosodiethanolamine by high-performance liquid chromatography (HPLC) with ultraviolet detection (Occupational Safety and Health Administration, 1990).

N-Nitrosodiethanolamine has been found in many complex matrices such as cutting and grinding fluids and cosmetics. Analysis for *N*-nitrosodiethanolamine is complicated by the matrix and a clean-up technique with derivatization is typically required before quantitation of the analyte to achieve adequate sensitivity and selectivity. Ammonium sulfamate may be added to the sample to prevent the artifactual formation of *N*-nitrosamines. Derivatives of *N*-nitrosodiethanolamine have been prepared by acylation, trifluoroacylation, trimethylsilylation and methylation. The derivatives have been analysed by gas chromatography using flame ionization and mass spectrometric detectors (Occupational Safety and Health Administration, 1990).

1.2 Production

N-Nitrosodiethanolamine is formed by the action of nitrosating agents (nitrites; 2-bromo-2-nitropropane-1,3-diol; nitrogen oxides) on diethanolamine and triethanolamine (Schmeltz & Wenger, 1979; Hoffmann *et al.*, 1982; Budavari, 1998). The rate of formation of *N*-nitrosodiethanolamine in aqueous solutions of ethanolamines is pH-, temperature- and time-dependent. The reaction was originally thought to occur only

[1] Calculated from: mg/m^3 = (relative molecular mass/24.45) × ppm, assuming a temperature of 25 °C and a pressure of 101 kPa

under acidic conditions, but it can also occur at near-neutral pH by the action of bacteria and at basic pH with appropriate catalysts. For instance, technical-grade triethanolamine (85% triethanolamine, 15% diethanolamine) yielded concentrations of *N*-nitrosodiethanolamine approximately 8- to 12-fold greater than reagent-grade triethanolamine (99% pure) under the same experimental conditions. The increase was attributed to the faster nitrosation rate of diethanolamine (10 to 20 times, depending on pH). Tertiary amines nitrosate at a slower rate than secondary amines because the reaction involves a nitrosative dealkylation rate-limiting step to yield a secondary amine which is then available for further nitrosation. Extensive analysis of representative nitrite-containing metalworking fluids for *N*-nitrosodiethanolamine suggested that the levels of *N*-nitrosodiethanolamine formed were related primarily to the amount of diethanolamine originally present (Lijinsky *et al.*, 1972; Zingmark & Rappe, 1977a,b; Rappe & Zingmark, 1978; Schmeltz & Wenger, 1979; Ong & Rutherford, 1980; Lucke & Ernst, 1993).

1.3 Use

There is no known commercial use for *N*-nitrosodiethanolamine.

1.4 Occurrence

1.4.1 *Natural occurrence*

N-Nitrosodiethanolamine is not known to occur as a natural product.

1.4.2 *Occupational exposure*

The formation of *N*-nitrosamines (mostly *N*-nitrosodiethanolamine) in cutting fluids has been documented since the mid-1970s (Zingmark & Rappe, 1977a,b). Reported concentrations in undiluted, unused fluids have varied considerably (0–30 000 ppm [mg/L]) depending, among other factors, on the presence of sodium nitrite in the fluids (see General Remarks). Concentrations in diluted fluids have been reported to be much lower (Zingmark & Rappe, 1977a,b; Fan & Fajen, 1978; Williams *et al*., 1978; Berg *et al*., 1979; Rounbehler & Fajen, 1983; Spiegelhalder *et al*., 1984, 1987; Fadlallah *et al.*, 1990; Järvholm *et al*., 1991a,b). Acidification, heat, catalysts such as formaldehyde preservatives and transition metal complexes can markedly increase the inherent tendency of diethanolamine and triethanolamine-containing alkaline cutting fluids to react with nitrite to form *N*-nitrosodiethanolamine. Alkanolamines in pH 9 cutting fluids can also react rapidly with nitric oxide in air to form *N*-nitrosodiethanolamine (Loeppky *et al*., 1983). A survey in a plant manufacturing cutting fluids and lubricant revealed concentrations of 20–500 mg/L in bulk samples from separate areas in the manufacturing process (Wolf & Young, 1977).

Fan *et al*. (1977a) reported concentrations of *N*-nitrosodiethanolamine varying between 200 and 29 900 ppm in metalworking fluids used in the United States. Analysis of some German grinding fluids containing di- and triethanolamines in combination with up to 30% nitrite showed concentrations of up to 593 mg/kg [mg/L] *N*-nitrosodiethanolamine in the original, concentrated fluid and up to 90 mg/kg in ready-to-use emulsions (Spiegelhalder *et al*., 1984). *N*-Nitrosodiethanolamine levels between 100 and 3600 mg/L were reported in some French metalworking fluids (Ducos *et al*., 1979) and Järvholm *et al*. (1991a) found a concentration of 650 ppm *N*-nitrosodiethanolamine in a diluted commercial Swedish machine-working fluid.

A recent Canadian study reported *N*-nitrosodiethanolamine concentrations in metalworking fluids to vary between 0.02 and 7.53 ppm in soluble oils, whereas it was absent in insoluble oils (Fadlallah *et al*., 1997). The same authors reported levels of *N*-nitrosodiethanolamine ranging from trace amounts to 0.193 μg/m^3 in the air of metal factories. No correlation was found between *N*-nitrosodiethanolamine concentrations in metalworking fluids and air concentrations (Fadlallah *et al*., 1996). A German study in the metal industry (1991–93) reported an arithmetic mean concentration of *N*-nitrosodiethanolamine of 0.2 μg/m^3 (N = 132, range < 0.01–3.66 μg/m^3). Analyses of metalworking fluids showed no *N*-nitrosodiethanolamine present in 50% of the fluids, while it varied between 0.1 and 16 mg/kg in the other 50% (Pfeiffer *et al.*, 1996). Another recent study from Italy reported *N*-nitrosodiethanolamine levels from traces to high concentrations (0.3–1900 mg/kg in used cutting fluids, and 0.4–31 mg/kg in new cutting fluids) in 21% of the 63 cutting fluids tested (Monarca *et al*., 1993). Keefer *et al*. (1990) reported mean concentrations of 1.5 ppm (0.5–4.5 ppm) for all six semi-synthetic fluids, 0.07 ppm (0.11–0.16 ppm) for three of six petroleum-based lubricants and 11.4 ppm (0.5–59 ppm) for five of six synthetic metalworking fluids in the United States.

Comparison of data from three studies (Williams *et al.*, 1978; 410–4150 mg/kg; Fadlallah *et al.*, 1990; synthetic, 36–95 ppm; soluble, 5–35 ppm; Fadlallah *et al.*, 1997; soluble, 0.02–7.5 ppm; none in insoluble oils) indicates that concentrations of *N*-nitrosodiethanolamine in metalworking fluids used in Canada have declined over a period of two decades.

No information was available on the number of exposed workers in the United States from the 1981–83 National Occupational Exposures Survey (NOES, 1999).

N-Nitrosodiethanolamine is easily absorbed through the skin and uptake can therefore occur both via the dermal route as well as the respiratory route. In preliminary investigations, *N*-nitrosodiethanolamine was found in the urine of metal grinders: of 264 analysed, 166 showed positive results (> 0.5 μg/kg (detection limit) with levels up to 103 μg/kg) (Spiegelhalder *et al.*, 1984). During biological monitoring of workers using cutting fluids with detectable levels of *N*-nitrosodiethanolamine, only workers working with cutting fluids containing ≥ 5 mg *N*-nitrosodiethanolamine/L excreted trace amounts of *N*-nitrosodiethanolamine in their urine (68% had values < 0.5 μg/kg (detection limit) and 32% had values between 0.6 and 2.7 μg/kg urine). Workers with low exposure and a control group were all *N*-nitrosodiethanolamine-negative (Monarca *et al*., 1996).

Järvholm *et al*. (1991a) reported levels of 19 mg/kg and < 0.02 mg/kg (detection limit) urine for two workers at the end of a shift. The first worker had considerably higher dermal exposure.

1.4.3 *Environmental occurrence*

The presence of *N*-nitrosodiethanolamine is widespread in the environment as a contaminant of cutting fluids and oils, certain pesticides, antifreeze, a broad range of consumer products (including cosmetics, lotions and shampoos) and tobacco and foods at concentrations ranging from 1 to 130 000 ppb (Fan *et al*., 1977a,b; IARC, 1978; Williams *et al*., 1978; Elder, 1980; Brunnemann *et al*., 1982–83; Loeppky *et al*., 1983; Spiegelhalder *et al*., 1984; Eisenbrand *et al*., 1991; Department of Health and Human Services, 1999).

(*a*) *Cosmetics*

Ninety-nine samples representative of 17 cosmetic products were evaluated and *N*-nitrosodiethanolamine was detected in 21 of the samples at concentrations of 50 to greater than 1000 ppb, and in traces in nine samples (Beyer *et al*., 1983).

In a study of off-the-shelf cosmetic formulations carried out by the United States Food and Drug Administration from 1978 to 1980, *N*-nitrosodiethanolamine was detected in 110 of 252 products containing triethanolamine and in 25 of 64 products not containing triethanolamine (however, the products without triethanolamine may have contained diethanolamine or monoethanolamine) (Beyer *et al*., 1983). Levels of *N*-nitrosodiethanolamine as high as 130 ppm have been reported (Edwards *et al.*, 1979).

N-Nitrosodiethanolamine at levels of 600–7386 ppb was detected in 11 samples of cosmetics in the United States which included hand creams, face creams, shampoos, cocoa butter cream, moisturizing lotion and a make-up remover (all products listed diethanolamine and/or triethanolamine as ingredients) (Tunick *et al*., 1982). *N*-Nitrosodiethanolamine was found in all seven cosmetic formulations; one with traces (< 10 ng/g) and six at levels of 41–47 000 ng/g and in 12 of 13 lotions (seven with traces < 10 ng/g and five with 14–140 ng/g) and in eight of nine hair shampoos (three with traces < 10 ng/g and five with 17–280 ng/g) in the United States (Fan *et al*., 1977b). Of 191 cosmetics analysed, 77 contained from 10 to more than 2000 ppb *N*-nitrosodiethanolamine (Elder, 1980). Westin *et al*. (1990) analysed 20 different suntan lotions in Israel and found that three were contaminated with 17–27 ppb *N*-nitrosodiethanolamine (with traces: ~ 5–10 ppb).

Levels of *N*-nitrosodiethanolamine ranging from 100 to 380 μg/kg have been found in 15 of 20 cosmetic and dermo-pharmaceutical preparations commercially available in Belgium (Rollmann *et al*., 1981). Among commercial samples of cosmetics from the German market examined during the summer of 1986, 40% were contaminated with *N*-nitrosodiethanolamine (7–2000 μg/kg). In 1987, the German Federal Health Office recommended that secondary amines no longer be used in cosmetics. Six to 18 months

later, a survey showed that 13% of commercially available cosmetic products from the German market were still contaminated with *N*-nitrosodiethanolamine (10–235 μg/kg) (Eisenbrand *et al*., 1991).

Fan *et al*. (1977b) and Brunnemann *et al.* (1982–83) calculated that women who use 2 g of cosmetics daily would absorb 0.41 μg *N*-nitrosodiethanolamine through the skin. They estimated that women using 1–2 cosmetics daily are exposed to 50–100 μg *N*-nitrosodiethanolamine from these cosmetic products daily.

After application of a cosmetic contaminated with 980 μg of *N*-nitrosodiethanolamine (77 μg/g), *N*-nitrosodiethanolamine (< 50 ng/mL [μg/kg]) was detected in human urine with a half-life greater than 13 hours (Edwards *et al.*, 1979).

(*b*) *Tobacco products*

N-Nitrosodiethanolamine has been detected in cigarette smoke at concentrations of 24–36 ng per cigarette, in smoking tobacco at concentrations up to 420 μg/kg, and in smokeless tobacco products (chewing tobacco, snuff) at concentrations up to 6800 μg/kg (Brunnemann & Hoffmann, 1981; Brunnemann *et al*., 1982–1983; IARC, 1985). The presence of *N*-nitrosodiethanolamine in tobacco and tobacco smoke was attributed, at least in part, to a maleic hydrazide–diethanolamine herbicidal combination commonly applied to tobacco to prevent sucker growth and levels of *N*-nitrosodiethanolamine have declined (< 100 ppb) in some tobaccos since the herbicide was phased out (Brunnemann & Hoffmann, 1991)

N-Nitrosodiethanolamine levels in five United Kingdom samples of oral tobacco (mean, 105 μg/kg; range, not detected–220 μg/kg), in five samples of Swedish moist snuff (mean, 19 μg/kg; range, 8–31 μg/kg), in 11 samples of Indian zarda (mean, 9.5 μg/kg; range, not detected–54 μg/kg) and in 10 samples of European nasal snuff (mean, 12 μg/kg; range, not detected–42 μg/kg) in 1987–88, and in one sample each of moist snuff (94 μg/kg) and chewing tobacco (74 μg/kg) in the United States in 1990 have been reported (Brunnemann & Hoffmann, 1991; Tricker & Preussmann, 1991). *N*-Nitrosodiethanolamine was found in five Nigerian tobacco products (16 brands analysed), at levels of 3.88–34.1 μg/kg (Okieimen & Akintola, 1985).

(*c*) *Food products*

N-Nitrosodiethanolamine was found in a number of Nigerian dairy products (1.5–7 μg/kg) and meat (0.87–1.89 mg/kg) (Okieimen & Akintola, 1985; Anucha *et al.*, 1986).

(*d*) *Miscellaneous products*

N-Nitrosodiethanolamine was found in five of 23 samples of antifreeze (used in the protection of internal combustion engines of cars and trucks) at concentrations varying from 15 to 5700 mg/L in several brands which contained both sodium nitrite and triethanolamine borate (Ducos *et al*., 1983).

1.5 Regulations and guidelines

N-Nitrosodiethanolamine is listed as a class 2 carcinogenic substance in Germany (Deutsche Forschungsgemeinschaft, 1999; substances that are considered to be carcinogenic for man because sufficient data from long-term animal studies or limited evidence from animal studies substantiated by evidence from epidemiological studies indicate that they can make a significant contribution to cancer risk). The European Commission has listed *N*-nitrosodiethanolamine as category 2 carcinogen. Germany has set an 8-h TRK (technical exposure limit) value for combined exposure to 10 *N*-nitroso compounds, including *N*-nitrosodiethanolamine, of 0.0025 mg/m^3 and a 15-min STEL of 0.01 mg/m^3 for some rubber, polyacrylonitrile and amine handling operations, and an 8-h TRK value of 0.001 mg/m^3 for all other operations (Pflaumbaum *et al.*, 1999; United Nations Environment Programme, 1999).

2. Studies of Cancer in Humans

The Working Group was not aware of any studies that specifically examined the risk of cancer among persons exposed to *N*-nitrosodiethanolamine. However, ethanolamines and sodium nitrite have been used as additives for metalworking fluids since the 1950s and together these can lead to the formation of *N*-nitrosodiethanolamine. There are three major types of metalworking fluid; straight (generally mineral oils), soluble (straight oils diluted with water and additives), and synthetic (water and additives) (see General Remarks). Ethanolamines and nitrites are additives used in both soluble and synthetic metalworking fluids (see Sections 1.3 and 1.4.2). A number of studies have examined the risk of cancer among workers exposed to metalworking fluids. Studies stating that ethanolamines and nitrites were used together as additives or which presented results for exposure to nitrosamines are described in detail below. A summary of their characteristics is presented in Table 1 and a summary of their results for specific cancer sites is presented in Table 2. Tables 4 and 5 in the monograph on diethanolamine in this volume also include information on other studies of workers exposed to soluble or synthetic metalworking fluids where ethanolamines are a likely additive. These additional studies are described in detail in the monograph on diethanolamine but are of relevance for this monograph because it is likely that some of these workers were also exposed to *N*-nitrosodiethanolamine through the use of sodium nitrite as an additive.

Järvholm *et al.* (1986) examined the risk for cancer among 219 Swedish men who had worked for at least one year on a machine in which cutting fluids containing amines (mostly triethanolamine, diethanolamine and monoethanolamine) and sodium nitrite were used. This population overlaps with a larger cohort of men employed for at least five years and any time between 1950 and 1966 in the grinding and turning departments of a company producing bearing rings, described in detail in the monograph on

Table 1. Characteristics of studies of cancer in potentially *N*-nitrosodiethanolamine-exposed workers

Study/ country	Study design	Study population	Follow-up period	Potential exposures
Järvholm *et al.* (1986) Sweden	Cohort	219 men employed for at least one year on a machine using fluids containing ethanolamines and sodium nitrite	1966–83	No sub-group or exposure analysis. Population presumed to be exposed to *N*-nitroso-diethanolamine
Park *et al.* (1988) United States	Industry-based proportionate mortality study	755 decedents; employed for 10 or more years in bearing manufacturing plant	1969–82	Both straight and soluble metalworking fluids. Different types of metalworking fluids examined in a nested case–control analysis.
Park & Mirer (1996) United States	Industry-based proportionate mortality study	1870 decedents; employed for > 2 years at two auto engine manufacturing plants 1966–87	1970–89	Population primarily exposed to soluble fluids. Different types of metalworking fluids examined in nested case-control analysis.
Sullivan *et al.* (1998) United States	Nested case–control of oesophageal cancer	53 fatal cases; 971 controls (study base: Eisen *et al.*, 1992 cohort)	1941–84	Cumulative exposure to the three types of metalworking fluids; duration of exposure to metalworking fluids and other components, incl. nitrosamines.

Table 2. Results of epidemiological studies of cohorts exposed to soluble and synthetic metalworking fluids potentially containing *N*-nitrosodiethanolamine (mortality studies)

Reference	Stomach		Oesophagus		Larynx		Leukaemia		Pancreas		All cancer		All mortality	
	Obs.	SMR/ PMR	Obs.	SMR/ PMR/OR	Obs.	SMR/ PMR/OR	Obs.	SMR/ PMR	Obs.	SMR/ PMR	Obs.	SMR/ PMR	Obs.	SMR/ PMR
Järvholm *et al*. (1986)														
≥ 1 year's exposure	NR		NR		NR		NR		NR		4	[0.5] (0.1–1.3)	29	[1.0] (0.6–1.4)
≥ 5 years' exposure	NR		NR		NR		NR		NR		3	[0.5] (0.1–1.5)	20	[0.8] (0.5–1.3)
Park *et al*. (1988)														
White males	11	2.0 (1.1–3.5)	6	1.8 (0.7–4.0)	NR		1	0.2 (0.0–1.3)	8	1.1 (0.6–2.2)	157	1.15 (1.0–1.3)	NA	(PMR study)
Park & Mirer (1996)														
Plant 1							All lymphopoietic							
White males	8	2.1 (0.9–4.1)	NR		1	0.7 (0.0–3.8)	10	1.0 (0.5–1.8)	10	1.8 (0.9–3.3)	107	1.0 (0.8–1.2)	NA	(PMR study)
Black males	2	1.2 (0.2–4.4)	NR		1	1.5 (0.0–8.4)	4	1.5 (0.4–3.9)	0	0.0 (0.0–1.9)	35	1.1 (0.8–1.4)	NA	
Plant 2														
White males	8	1.3 (0.6–2.6)	NR		4	1.7 (0.5–4.3)	15	0.9 (0.6–1.5)	11	1.2 (0.6–2.2)	199	1.1 (1.0–1.3)	NA	
Black males	5	0.8 (0.3–2.0)	NR		1	0.5 (0.0–2.8)	7	1.0 (0.4–2.1)	6	1.1 (0.4–2.4)	111	1.0 (0.9–1.2)	NA	
Sullivan *et al*. (1998) (20-year lag)														
5 years of exposure to nitrosamines			–	3.7 (1.2–11.1)										

NA, not applicable; NR, not reported; PMR, proportionate mortality ratio

diethanolamine in this volume (Järvholm & Lavenius, 1987). Mortality and cancer incidence follow-up was conducted from 1966 (10 years after the first use of fluids containing both types of additives) to 1983 and expected numbers were calculated using reference rates from the same city. There were 29 deaths (30.5 expected), four cancer deaths (7.6 expected) and seven incident cancers (13.0 expected). No notable excesses for specific cancer sites were observed, although the numbers were very small.

Park *et al*. (1988) conducted a proportionate mortality study of workers employed at a bearing-manufacturing plant in the United States. The study population included many workers employed in grinding operations using soluble metalworking fluids, at least one of which was known to contain nitrites and organic amines. The study population consisted of any person who was employed at the facility for at least 10 years and who had died between 1 January 1969 and 31 July 1982. Employment in various work areas and exposure to straight and water-based (soluble) metalworking fluids were classified based on the last job held 15 years before death using available work history records. Deaths were identified using death benefit records, pension records and local union files. Of the 768 decedents meeting the entrance criteria, 13 (2%) were excluded due to missing death certificates (755 left). Proportionate mortality ratios (PMR) were calculated using national rates. Among all deceased white males (n = 610), a somewhat greater number of cancers was observed than expected (PMR, 1.15) and excesses of oesophageal (PMR, 1.8; 95% confidence interval [CI], 0.7–4.0), stomach (PMR, 2.0; 95% CI, 1.1–3.5), rectal (PMR, 3.1; 95% CI, 1.5–5.5) and skin (PMR, 1.9; 95% CI, 0.5–4.8) cancers. An excess of stomach cancer among white women was also observed (PMR, 3.1; based on three observed cases; p = 0.15). No detailed results were presented for white women (n = 124) or the small number of non-white men (n = 20) and women (n = 2) in the population. The risk for cancer in relation to employment in various job and exposure groups was examined in a mortality odds ratio analysis (case–control study). An additional 53 decedents (7%) were excluded (702 left) from the analyses because of a lack of work history information. Mortality odds ratios were calculated using Mantel–Haenszel techniques to adjust for age. Decedents with causes of death thought to be unrelated to potential exposures at the facilities were used as controls. An excess of stomach cancer (mortality odds ratio, 6.6; p = 0.02) was associated with exposure to water-based cutting fluids. [The Working Group expressed some concern about the validity of exposure classification based on one job title and about the representativeness of the control group used.]

Park and Mirer (1996) conducted a proportionate mortality study of workers employed at two large automotive engine manufacturing plants in the United States. The study population included many workers employed in operations using soluble or synthetic metal working fluids. *N*-Nitrosodiethanolamine was detected in synthetic fluids containing nitrites and ethanolamines in 1978. Levels ranged from 1 to 4 μg/mL in used fluids and from 6 to 140 μg/mL in new, undiluted fluids. The study population consisted of any person who was employed at the facilities for at least two years some time between 1966 and 1987 and who died between 1 January 1970 and 31 December

1989, inclusive. Exposure was assessed on the basis of industrial hygiene reports and work history data. Detailed work history data were only available after 1966 and employment before 1966 was assigned to the same department as in 1966. Ranks (0, 1, 2, 4, 8) were assigned for exposure to metalworking fluids, oil smoke, solvents, coal tar and cobalt. Deaths were identified using state vital records, insurance claims and pension records and the authors estimated that over 90% of deaths were identified. Of the 1914 decedents eligible for the PMR analysis, 44 (2.3%) were excluded due to missing cause of death (1870 left). PMRs were presented only for deceased white (1170) and black (613) men employed at each facility and the total number of cancers was similar to that expected (white men—plant 1: PMR, 1.0; white men—plant 2: PMR, 1.1; black men—plant 1: PMR, 1.1; black men—plant 2: PMR, 1.0). The risk for cancer in relation to potential exposures and duration of employment in various operations was examined in a mortality odds ratio analysis. Mortality odds ratios were calculated using logistic regression and adjusting for age, sex, race, year and overall duration of employment at the facilities. Decedents with causes of death thought to be unrelated to potential exposures at the facilities were used as controls. Employment in camshaft and crankshaft grinding at plant 1, where exposure to nitrosamines had been documented, was associated with an increased risk for stomach cancer (mortality odds ratio, 5.1; 95% CI, 1.6–17, at the mean duration of the exposed cases). Exposure to soluble or synthetic metalworking fluids in grinding was found to be significantly associated with non-Hodgkin lymphoma and multiple myeloma combined (mortality odds ratio, 4.1; 95% CI, 1.1–15.4 at the mean cumulative exposure of the exposed cases). No other association with soluble or synthetic fluids was reported.

Sullivan *et al*. (1998) conducted a nested case–control study of oesophageal cancer among the members of the cohort studied by Eisen *et al*. (1992). Potential cases were 60 individuals who died of oesophageal cancer between 1941 and 1984, seven of whom were excluded due to missing work history data. Incidence density sampling was used to select 20 controls for each case matched on the basis of year of birth, plant, race and sex and 58 of these were excluded because of missing data. Extensive efforts were made to assess exposure to biocides (odds ratio, 16.0; 95% CI, 1.8–143 for a five-year exposure and a 20-year latency). It was stated that many of the same workers were exposed to both nitrosamines and biocides. Co-exposure to nitrites and ethanolamines was considered to indicate exposure to nitrosamines. Matched analyses were performed using conditional logistic regression with additional adjustment for time since hire and lagging was used to account for latency. After allowing for a 20-year latency, the incidence of oesophageal cancer was associated with duration of exposure to nitrosamines (odds ratio, 3.7; 95% CI, 1.2–11.1 for five years), to straight, synthetic and soluble fluids, as well as to nitrosamines, biocides, sulfur, asbestos, solvents and various metals. [The Working Group noted that data on tobacco smoking and alcohol drinking were not directly presented.]

3. Studies of Cancer in Experimental Animals

3.1 Oral administration

Mouse: In a mouse lung screening bioassay, a group of 40 female A/J mice, aged 8–10 weeks, received 0.2 μmol/mL of *N*-nitrosodiethanolamine (reported to be pure by chromatography) in the drinking water for 10 weeks followed by tap water alone for the next 20 weeks. A group of 40 mice receiving tap water alone served as concurrent controls. At 30 weeks, all mice were killed and lung adenomas were counted [survival not reported]. In the control group, 40% of animals had lung tumours compared with 70% in treated animals. The number of lung tumours per mouse (mean ± SD) was 0.5 ± 0.6 in controls versus 1.4 ± 1.2 ($p < 0.01$) in treated animals (Hecht *et al*., 1989).

Rat: Groups of 10 male and 10 female Fischer 344 rats, six to seven weeks of age, were given solutions of *N*-nitrosodiethanolamine [purity not specified] in neutral deionized tap-water as the drinking water. Doses of 20 mL *N*-nitrosodiethanolamine solution (3900, 7800, 15 600, 31 250 or 62 500 ppm [mg/L]) were given to each animal on five days per week for 34 weeks. On the remaining two days, the animals were given tap-water without *N*-nitrosodiethanolamine (animals were dosed on seven days per week *ad libitum* for the first 12 weeks of the experimental period). Due to toxicity, treatment of the group given the highest concentration of *N*-nitrosodiethanolamine was discontinued. A few animals that received the 15 600 and 31 250 ppm concentrations died before week 34 [number not stated]. At week 34, all surviving animals were killed and necropsied. The incidence of hepatocellular carcinomas in the remaining four treatment groups was 100% compared with 0% in the controls. Cholangiocellular carcinomas also occurred at the higher doses (males: 0/10, 1/10, 6/10, 8/10, 10/10 and females: 0/10, 3/10, 5/10, 8/10 and 7/10 in the 0, 3900, 7800, 15 600 and 32 500 ppm groups respectively). At the 15 600- and 31 250-ppm doses, many of the induced liver carcinomas metastasized to the lungs and peritoneal cavity (Lijinsky *et al*., 1980).

Groups of 36 to 88 male Sprague-Dawley rats, 100 days old, were administered 5 mL of an aqueous solution of *N*-nitrosodiethanolamine (purity > 99%) in the drinking-water (tap-water) on five days per week for life. There were five groups dosed at concentrations of *N*-nitrosodiethanolamine calculated to provide 1.5, 6, 25, 100 or 400 mg/kg bw daily. Controls received tap-water only. All animals were allowed to die naturally or were killed when moribund. In all groups, *N*-nitrosodiethanolamine was well tolerated without any sign of acute or subchronic toxicity. During the first six months of the study, body weights of all treated animal groups did not significantly differ from those of the control group. This continued for the lifespans for the two lowest-dose groups, but in the three higher-dose groups, body weights were reduced. Survival times did not differ significantly between the two low-dose groups and the controls. In the other three groups, survival times decreased sharply with increasing daily dose. There was a dose-related increase in the incidences of hepatocellular neoplasms and neoplasms of the nasal cavity. The liver tumours were principally adenocarcinomas, with a total

incidence of 0/88, 7/72, 43/72, 33/36, 32/36 and 31/36 in the groups receiving 0, 1.5, 6, 25, 100 and 400 mg/kg bw *N*-nitrosodiethanolamine daily. The nasal cavity neoplasms were mainly squamous-cell carcinomas as well as neuro-epitheliomas of the olfactory epithelium, with total incidences of 0/88, 2/72, 0/72, 6/36, 6/36 and 1/36 in the groups receiving 0, 1.5, 6, 25, 100 and 400 mg/kg bw *N*-nitrosodiethanolamine daily (Preussmann *et al.*, 1982).

Groups of 16–20 male and 16–20 female Fischer 344 rats [age unspecified] were administered 20 mL of an aqueous solution of *N*-nitrosodiethanolamine (purity, > 99%) in the drinking-water (tap-water) (a cage of four animals shared 80 mL of solution) on five days per week for life. The doses administered and the duration of the exposure periods for the various groups were as follows: 2500 mg/L drinking-water for 45 weeks, 1000 mg/L drinking-water for 50 weeks or 400 mg/L drinking-water for 75 weeks. A control group consisted of 40 males and 40 females received tap-water *ad libitum*. All treated rats had died by week 110 and all surviving control rats were killed at 130 weeks; 12 males were killed at 95 weeks and the remainder at 130 weeks. Almost all of the treated animals were reported to have died due to tumour development, principally in the liver and nasal cavity. There was an increased incidence of hepatocellular carcinomas (females: controls, 0/40; 400 mg/L for 50 weeks, 15/16; 400 mg/L for 75 weeks, 16/16; 1000 mg/L for 50 weeks, 19/20; 2500 mg/L for 45 weeks, 20/20; males: controls, 1/28; 400 mg/L for 50 weeks, 14/16; 400 mg/L for 75 weeks, 14/16; 1000 mg/L for 50 weeks, 18/20; 2500 mg/L for 45 weeks, 20/20) together with a few cholangiocarcinomas. In almost all of the treated groups with hepatocellular carcinomas, 30–40% of the rats had lung metastases. In the nasal cavity, the principal tumours induced were adenocarcinomas, with some squamous-cell carcinomas (Lijinsky & Reuber, 1984).

Groups of male and female Fischer 344 rats, seven to eight weeks of age, received 20 mL per animal (a cage of four animals shared 80 mL of solution) of *N*-nitrosodiethanolamine [purity not specified] in deionized water at three concentrations: 28 mg/L, 64 mg/L or 160 mg/L on five days per week. The low dose was given to 39 male and 39 female rats for 100 weeks (total dose, 280 mg per rat). The medium dose was given to 20 male and 20 female rats for 50 weeks (total dose, 320 mg per rat) and 20 males and 20 females for 100 weeks (total dose, 640 mg per rat). The highest dose was given to 27 male and 27 female rats for 50 weeks (total dose, 800 mg per rat). Controls consisting of 20 males and 20 females received deionized water *ad libitum*. Animals were allowed to die naturally or were killed when moribund. The survival of treated male rats beyond 100 weeks was reduced compared with controls, but not in females. A significant increase in the incidence of liver tumours (neoplastic nodules, hepatocellular carcinomas, cholangiocarcinomas or adenomas) was observed in the groups treated with 64 mg/L *N*-nitrosodiethanolamine solution for 100 weeks and 160 mg/L for 50 weeks compared with the control group (males: controls, 4/20; 28 mg/L for 100 weeks, 6/39; 64 mg/L for 100 weeks, 11/20 [$p = 0.024$]; 64 mg/L for 50 weeks, 2/20; 160 mg/L for 50 weeks, 19/27 [$p < 0.001$]; females: controls, 1/20; 28 mg/L for

100 weeks, 10/39 [$p = 0.05$]; 64 mg/L for 100 weeks, 14/20 [$p < 0.001$]; 64 mg/L for 50 weeks, 5/20; 160 mg/L for 50 weeks, 27/27 [$p < 0.001$]) (Lijinsky & Kovatch, 1985).

Groups of 80 male Sprague-Dawley rats, 100 ± 10 days old, received *N*-nitrosodiethanolamine (purity, > 99%) in the drinking-water at doses of 0.2 (low), 0.63 (mid) or 2.0 mg/kg bw (high) on five days per week for life. On the other two days per week, all rats received normal tap-water *ad libitum*. Control animals (500 rats) received tap-water only. There were no signs of acute or subchronic toxicity at any of the dose levels. The incidence of hepatocellular neoplasms was increased (controls, 3/500; low, 2/80 ($p < 0.05$); mid, 1/80; and high, 6/80 ($p < 0.001$)). The gastrointestinal tract had a significantly increased incidence of tumours only in the low-dose group compared with the controls, with the numbers of animals affected as follows: controls, 26/500; low, 7/80 ($p < 0.05$); mid, 3/80; and high, 6/80. The authors noted that this latter increase was due to a slightly higher rate of tumours in the oral cavity and in the forestomach. In addition, there were significant increases in the incidence of tumours in the neurogenic tissue in the medium-dosed group (controls, 54/500; low 8/80; mid, 16/80 ($p < 0.05$); high, 11/80) and of the haematopoietic and lymphatic system in the low- and high-dosed groups (controls, 23/500; low, 8/80 ($p < 0.05$); mid, 5/80; high, 7/80 ($p < 0.1$) (Berger *et al.*, 1987).

A group of 20 female Fischer 344 rats, eight weeks of age, received approximately 20 mL per day on five days per week, of tap-water containing 150 mg/L *N*-nitrosodiethanolamine. On the other two days per week, tap-water alone was provided. A group of 20 control rats received tap-water alone. Treatment was for 50 weeks and surviving animals were maintained on tap-water alone. All surviving animals were killed at weeks 120–124. Survival was 18/20 in both controls and treated groups at 80 weeks and 17/20 versus 11/20 at 100 weeks. No tumours were reported in the control group but in the treated animals, 14/20 had hepatocellular carcinomas (Hecht *et al.*, 1989).

3.2 Subcutaneous administration

Hamster: Groups of 15 male and 15 female Syrian golden hamsters, 8–10 weeks of age, received either seven subcutaneous injections (twice weekly) of 2260 mg/kg bw of *N*-nitrosodiethanolamine (1/5th of the LD_{50}) in 0.5 mL saline over three weeks or 27 subcutaneous injections of 565 mg/kg bw (1/20th of the LD_{50}) over 45 weeks. Injection-site necrosis required interruption of the dosing schedule with cessation of dosing after seven applications in the high-dose group and the need for injection-free intervals of one to two weeks for the low-dose group. This latter group received a total of 27 injections over 45 weeks. Negative controls received 78 injections of 0.5 mL saline solution alone. After 78 weeks, all surviving animals were killed and necropsied, as well as animals found to be moribund during the study. Survival rates to the appearance of the first tumour (33 weeks) of the treated groups (92%) did not differ significantly from the controls (90%). The following incidences of tumours were reported: nasal tumours (mainly adenocarcinomas): males: controls, 0/15; lose-dose, 7/13; high-dose 6/15; and

females: controls, 0/12; low-dose, 5/14; and high-dose, 4/13; tracheal tumours: males: controls, 0/15; low-dose, 2/13; high-dose, 5/15; and females: controls, 0/12; low-dose, 5/14; and high-dose, 3/13 (Hilfrich *et al*., 1977). [No statistical analysis was reported].

Groups of 15 male and 15 female Syrian golden hamsters, 8–10 weeks of age, were given weekly subcutaneous injections [vehicle not specified but presumed to be saline] of 250, 500 or 1000 mg/kg bw *N*-nitrosodiethanolamine [chromatographically homogeneous] for life. The control group received saline only. *N*-Nitrosodiethanolamine was not found to be toxic, even at the highest dose tested. A few animals died of spontaneous infectious diseases early in the observation period and were excluded from the study. Average body weight in treated animals did not vary significantly from that of control animals. An increase in the incidence of tumours of the nasal cavity (adenocarcinomas) was seen in all treatment groups (males—0/15, 8/13, 8/14 and 11/15; females—0/15, 5/14, 6/15 and 11/15 in the control, low-, mid- and high-dose groups, respectively) (Pour & Wallcave, 1981).

Groups of 15 male and 15 female Syrian golden hamsters, 8–10 weeks of age, received weekly subcutaneous injections of 58, 170 or 500 mg/kg bw *N*-nitrosodiethanolamine (purified by HPLC) in 0.9% NaCl solution for 27 weeks. Groups of 10 male and 10 female hamsters received similar treatment but with only the 0.9% NaCl solution and served as controls. The experiment was terminated after 20 months when approximately 20% of animals were still alive [further details not reported]. Nasal cavity tumours occurred in males: 0/10, 0/15, 2/15, 11/15 and females: 0/8, 0/14, 2/14 and 5/15 in the control, low-, mid- and high-dose groups respectively. These tumours were described as ranging from squamous papillomas to olfactory esthesioneuroepitheliomas. Three of the latter in the high-dose female group invaded the brain. Also reported in these groups were tracheal tumours; males: 0/0, 2/15, 1/15, 3/15 and in females: 0/8, 0/14, 3/14, 4/15 in the control, low-, mid- and high-dose groups, respectively, and single laryngeal tumours in one male and one female in the high-dose group (Hoffman *et al*., 1983). [No statistical analysis was reported.]

3.3 Topical application

Hamster: Groups of 15 male and 15 female Syrian golden hamsters, 8–10 weeks of age, received topical application to the shaved back of 2.5, 8 or 25 mg *N*-nitrosodiethanolamine (purified by HPLC) three times per week in 0.5 mL acetone for 36 weeks. Groups of 10 male and 10 female hamsters received similar treatment with 0.5 mL acetone and served as controls. The experiment was terminated at 20 months when approximately 20% of animals were still alive [further details not reported]. The treatment-related tumours reported were 2/15 and 2/15 nasal cavity tumours in the male and female high-dose (25 mg) groups respectively and 4/15 tracheal tumours in the high-dose male group (Hoffman *et al*., 1983). [The Working Group noted the small group sizes and the inadequate reporting.]

3.4 Other routes of administration

Hamster: Groups of 20 male and 20 female Syrian golden hamsters, 8–10 weeks of age, received applications by swab to the oral cavity (including lips and cheek pouches) of 20 mg *N*-nitrosodiethanolamine in 0.9% saline solution (0.4 mL) three times per week for 45 weeks. Groups of 10 male and 10 female hamsters received the saline solution alone and served as controls. The experiment was terminated at 20 months when approximately 20% of animals were still alive [further details not reported]. Nasal cavity tumours were reported in 12/20 treated males and 4/18 treated females compared with none in the control group. Tracheal tumours were seen in the treated groups (4/20 males, 2/18 females) but not in controls (Hoffman *et al.*, 1983).

4. Other Data Relevant to an Evaluation of Carcinogenicity and its Mechanisms

4.1 Absorption, distribution, metabolism and excretion

4.1.1 *Humans*

Dermal absorption of *N*-nitrosodiethanolamine was studied in a 40-year-old man following application of 12.7 g of a facial cream containing 77 μg/g *N*-nitrosodiethanolamine [980 μg]. The cream was applied for 7.75 h (after which it was removed by washing) to a 2090-cm^2 area of skin on the chest and back and covered by a plastic undershirt. About 2% (17.3 μg) of the applied dose was found in the urine at 21.5 h. *N*-Nitrosodiethanolamine was excreted in urine at a high rate (0.98–1.24 μg/h) both before (at 4–7.75 h) and after the cream was removed (at 7.75–11 h), and somewhat more slowly (0.80 μg/h) 13 h after removal of the cream. The half-life of *N*-nitrosodiethanolamine appeared to be long enough [no specific figure given] to allow accumulation of the compound in subjects exposed on a daily basis (Edwards *et al.*, 1979).

In 264 metal grinders analysed for urinary excretion of *N*-nitrosodiethanolamine, 166 showed positive results (> 0.5 μg/kg (L)) with levels up to 103 μg/kg (L) urine. The amount of *N*-nitrosodiethanolamine in grinding fluids (0.04–90 mg/kg) was predictive of the amounts detected in workers' urine. Data from quantitative excretion studies (24-h urines) indicated accumulation of *N*-nitrosodiethanolamine during a working week (Spiegelhalder *et al.*, 1984, 1987).

The excretion of *N*-nitrosodiethanolamine and thioethers in urine (for the first four hours of the shift on Thursday) was studied in 62 male subjects of whom 25 had high exposure to *N*-nitrosodiethanolamine from metal factories using cutting fluids with an *N*-nitrosodiethanolamine content of up to 1900 mg/kg (L), and in 37 control male subjects. Only the workers using cutting fluids with *N*-nitrosodiethanolamine concentrations of ≥ 5 mg/L (high exposure) excreted detectable amounts in their urine

(0.6–2.7 ng/g). These workers had also a higher mean value of urinary thioethers than the low-exposure and control subjects. Smoking did not affect the excretion of *N*-nitrosodiethanolamine but increased the mean value of the biomarkers (D-glucaric acid, thioethers and number of sister chromatid exchanges per metaphase) (Monarca *et al*., 1993, 1996).

4.1.2 *Experimental systems*

(*a*) *Absorption*

Percutaneous penetration of *N*-nitrosodiethanolamine was measured using cryopreserved human trunk skin and three vehicle formulations (isopropyl myristate, sunscreen cream or a 10% shampoo) containing *N*-nitroso[^{14}C]diethanolamine. The absorption rate of a low dermal dose (10 μg/cm^2) of *N*-nitrosodiethanolamine was a linear function of the concentration (0.06, 0.2 or 0.6 μg/μL) applied to the skin. The peak rates for the isopropyl myristate and shampoo vehicles were seen within five hours and for the sunscreen somewhat later. Total 48-h absorption ranged from 35 to 65% of the dose and was formulation-dependent (isopropyl myristate > shampoo ≥ sunscreen). A total absorption of 4–6 μg/cm^2 was estimated to equate to an applied *N*-nitrosodiethanolamine dose of 10 μg/cm^2. When applied as a large 'infinite' dose (0.5 mg/cm^2), total *N*-nitrosodiethanolamine absorption (4–35% of the applied dose) followed a different rank order (shampoo ≥ isopropyl myristate > sunscreen), probably due to the barrier-damaging properties of the vehicles. The permeability coefficient for isopropyl myristate was 3.5×10^{-3} cm/h (Franz *et al*., 1993).

Percutaneous absorption of *N*-nitrosodiethanolamine through human skin was studied in diffusion cells to determine the permeability constants for water (5.5×10^{-6} cm/h), propylene glycol (3.2×10^{-6} cm/h), a popular lotion (an oil in water emulsion) (6.2×10^{-5} cm/h) and isopropyl myristate (1.1×10^{-3} cm/h). The *N*-nitrosodiethanolamine membrane (stratum corneum):vehicle partition coefficients obtained were 1.8 for water, 1.0 for propylene glycol and 230 for isopropyl myristate. The skin penetration of *N*-nitrosodiethanolamine was greatly increased when lipoidal formulations (isopropyl myristate) were used. The use of cosmetic products over large areas of the skin and for long periods of time (e.g., sun-tanning lotion) results in the greatest absorption of *N*-nitrosodiethanolamine (Bronaugh *et al*., 1981).

Dermal penetration of *N*-nitroso[^{14}C]diethanolamine (4 μg/cm^2) *in vivo* was investigated by applying skin lotion and acetone to 3–15 cm^2 of the skin of monkeys (the abdomen) and pigs (the back) for a 24-h contact time. The skin penetration capacity was greater in monkeys (23% in skin lotion, 34% in acetone) than in pigs (4% in skin lotion, 11.5% in acetone) and the permeability was greater from acetone than skin lotion (Marzulli *et al*., 1981).

Rats were treated with *N*-nitrosodiethanolamine to compare the absorption through the skin with that from the stomach. Skin painting with *N*-nitrosodiethanolamine in water (20 mg/100 μL) and in cutting oil (25 mg/25 μL) yielded ≤ 25 μg/mL

blood levels of *N*-nitrosodiethanolamine. In contrast, a 50 mg dose of *N*-nitrosodiethanolamine applied undiluted to the skin or given in 500 μL water by gavage yielded blood levels of 130–220 μg/mL and 150–190 μg/mL after 1 h, respectively. From 20 to 30% of the *N*-nitrosodiethanolamine applied undiluted or by gavage was recovered in the 24-h urine (Sansone *et al*., 1980; Lijinsky *et al*., 1981).

(*b*) *Body distribution*

Whole-body autoradiography of male Sprague-Dawley rats given intravenous injections (2.7 mg/kg bw) of *N*-nitroso[^{14}C]diethanolamine showed an even distribution in most tissues except for tissue-bound radioactivity that was localized in the liver and the nasal olfactory mucosa. A lower level of labelling in the central nervous system probably indicated that *N*-nitrosodiethanolamine penetrated the blood–brain barrier poorly, while higher labelling in the kidney and urinary bladder may reflect elimination of *N*-nitroso[^{14}C]diethanolamine in urine (Löfberg & Tjälve, 1985).

Oral administration of *N*-nitroso[^{14}C]diethanolamine to Osborne-Mendel rats resulted in rapid distribution throughout the entire body, with little difference between the test doses of 0.5 and 50 mg/kg bw. Radioactivity was highest in tissues at 8 h and detectable up to one week. After a single topical dose, *N*-nitroso[^{14}C]diethanolamine (0.5 or 50 mg/kg bw in acetone) was slowly absorbed over one week and distributed as in the orally dosed rats. In most tissues, the radioactivity was highest after four to seven days (Lethco *et al*., 1982).

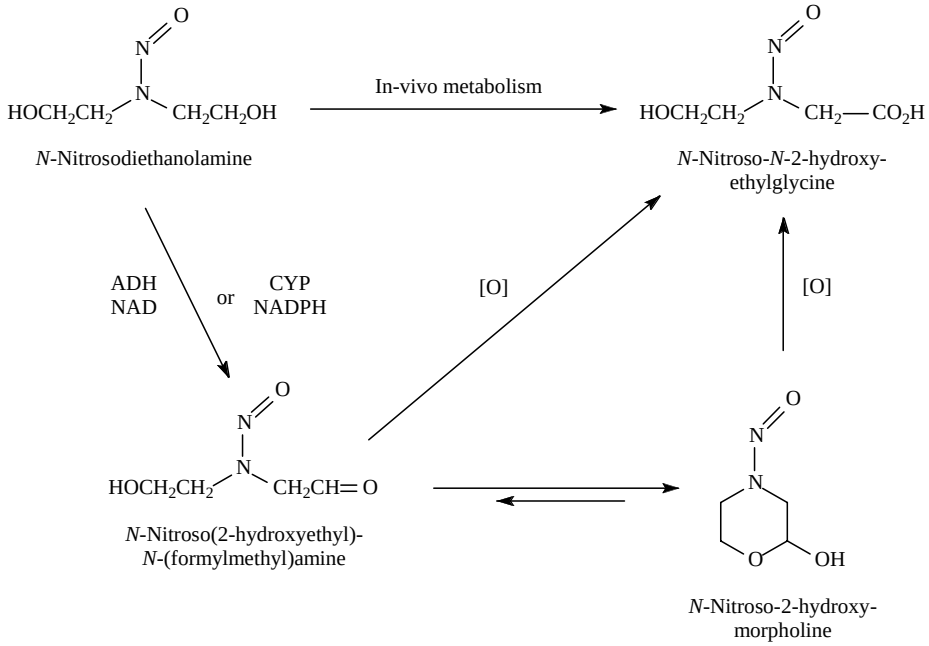
A three-compartment model has been described for the toxicokinetics of *N*-nitrosodiethanolamine studied in CD-COBS rats after a low intravenous dose (5 mg/kg bw). Blood levels of *N*-nitrosodiethanolamine reflected the levels in the liver, suggesting that the liver may not accumulate *N*-nitrosodiethanolamine. The overall elimination rate corresponded to a half-life of 5.77 h (Airoldi *et al*., 1984a).

(*c*) *Metabolism*

The metabolism of *N*-nitrosodiethanolamine has been reviewed (Eisenbrand & Janzowski, 1994; Loeppky, 1999).

The β-oxidation pathway (Figure 1) is the major route for the metabolic disposition of *N*-nitrosodiethanolamine in rodents (Bonfanti *et al*., 1986, 1987). The conversion of *N*-nitrosodiethanolamine to *N*-nitroso-*N*-2-hydroxyethylglycine [*N*-nitroso(2-hydroxyethyl)-*N*-carboxymethyl)amine] proceeds through formation of *N*-nitroso(2-hydroxyethyl)-*N*-(formylmethyl)amine, which exists in equilibrium with its cyclic hemiacetal form, *N*-nitroso-2-hydroxymorpholine. The latter is produced in rat liver preparations as well as by alcohol dehydrogenase (ADH)-mediated oxidation (Airoldi *et al*., 1984b; Eisenbrand *et al*., 1984; Hecht, 1984; Loeppky *et al*., 1987). The β-oxidation pathway shown with S9 fraction from rodent livers was preferentially catalysed by ADH/aldehyde dehydrogenase (Airoldi *et al*., 1983, 1984b; Eisenbrand *et al*., 1984; Bonfanti *et al*., 1986; Denkel *et al*., 1987) rather than by microsomal monooxygenases (Knasmüller *et al*., 1986; Loeppky, 1999). Some of the metabolic discrepancies in

Figure 1. Proposed metabolism of *N*-nitrosodiethanolamine by the β-oxidation pathway



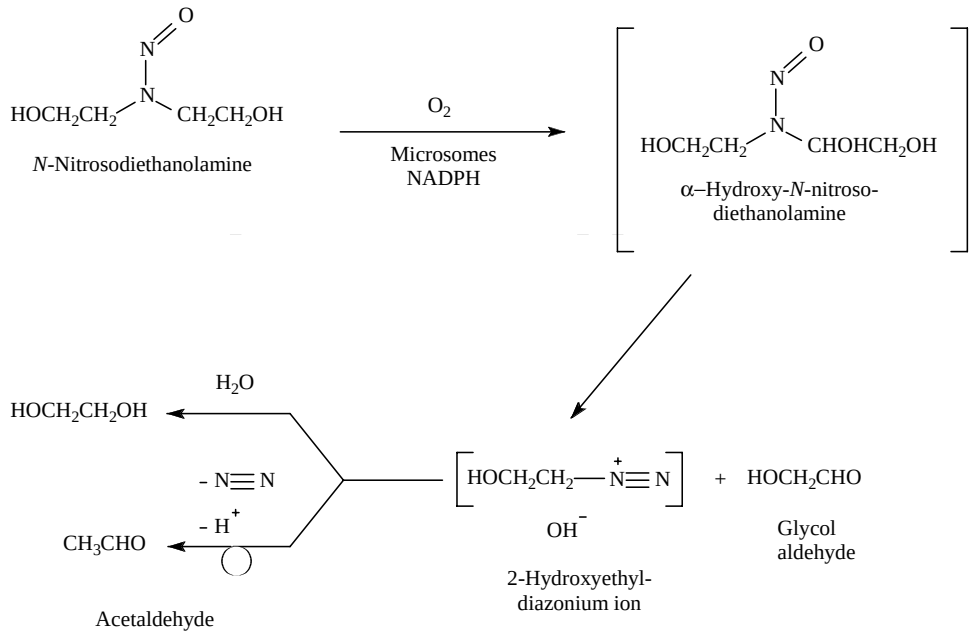
From Loeppky (1999)
ADH, alcohol dehydrogenase; CYP, cytochrome P450; NAD, nicotinamide-adenine dinucleotide; NADPH, nicotinamide-adenine dinucleotide phosphate (reduced form)

bioactivation of *N*-nitrosodiethanolamine may be explained by impurities in commercial *N*-nitrosodiethanolamine products or by vehicles (such as dimethyl sulfoxide) that are inhibitory (Mori *et al*., 1987; Henn *et al*., 1989).

The α-oxidation pathway of *N*-nitrosodiethanolamine metabolism (Figure 2) leads to the formation of an α-hydroxynitrosamine that rapidly decomposes, producing glycol aldehyde, acetaldehyde, ethylene glycol and molecular nitrogen. The latter is assumed to arise from a reactive (2-hydroxyethyl)diazonium ion, which probably is responsible for the formation of 2-hydroxyethylated adducts in DNA (Scherer *et al*., 1991; Loeppky *et al*., 1998; Loeppky, 1999).

The metabolism of *N*-nitrosodiethanolamine by α-hydroxylation, which is a cytochrome P450 (CYP)-mediated pathway, was not detected in liver preparations from uninduced male Fischer 344 rats (Farrelly *et al*., 1984, 1987). The existence of α-hydroxylation was proved later, notably by the formation of glycol aldehyde in liver microsomes from rats pretreated with CYP2E1 inducers. The microsomal metabolism of *N*-nitrosodiethanolamine was slower by α-oxidation than by β-oxidation (Loeppky, 1999). Bioactivation tests of *N*-nitrosodiethanolamine in V79 Chinese hamster cells showed that cytotoxicity was observed only in cells transfected with human CYP2E1 but not in cells expressing CYP2B1 or in the controls (Janzowski *et al*., 1996; Loeppky, 1999).

Figure 2. Proposed metabolism of *N*-nitrosodiethanolamine by the α-oxidation pathway



From Loeppky (1999)
NADPH, nicotinamide-adenine dinucleotide phosphate (reduced form)

α-Deuteration reduces the α-hydroxylation of *N*-nitrosodiethanolamine by a primary isotope effect, causing a decrease in the formation of glycol aldehyde and 2-hydroxyethyldiazonium ion as well as a corresponding decrease in DNA alkylation. As a result of β-deuteration, α-hydroxylation is enhanced through metabolic switching, leading to an increased rate of DNA alkylation (Loeppky *et al.*, 1998). α-Hydroxylation of *N*-nitroso-2-hydroxymorpholine, which could lead to DNA alkylation by the formation of either glyoxal or 2-hydroxyethyldiazonium ion, is a pathway supported both by the lack of carcinogenicity (Hecht *et al.*, 1989) and by the formation of hydroxyethylguanine adducts in rats exposed to *N*-nitroso-2-hydroxymorpholine (Chung & Hecht, 1985; Loeppky, 1999).

DNA base deamination studies employing *N*-nitroso-2-hydroxymorpholine and calf thymus DNA indicate that *N*-nitroso-2-hydroxymorpholine may deaminate the primary amino groups in DNA via nitroso transfer reactions *in vivo* (Loeppky *et al.*, 1994).

The role of CYP2E1 in α-oxidation of *N*-nitrosodiethanolamine was probed by using the deuterated isotopomers *N*-nitroso[α-D_4]diethanolamine and *N*-nitroso-[β-D_4]diethanolamine. *N*-Nitrosodiethanolamine and *N*-nitroso[β-D_4]diethanolamine were equally cytotoxic to human CYP2E1-transfected V79 cells, while *N*-nitroso-[α-D_4]diethanolamine was not. Significant DNA single-strand break levels were

observed in these cells for *N*-nitrosodiethanolamine and *N*-nitroso[β-D_4]diethanolamine but not for *N*-nitroso[α-D_4]diethanolamine. A kinetic deuterium isotope effect of 2.6 for V_{max}/K_m was observed for the horse liver ADH-mediated oxidation of *N*-nitroso[β-D_4]diethanolamine to *N*-nitroso-2-hydroxymorpholine, while k_H/k_D for *N*-nitroso[α-[D_4]diethanolamine was 1.05. These data suggest that the α- and β-hydroxylations of *N*-nitrosodiethanolamine are mediated by CYP2E1 and liver ADH, respectively (Loeppky *et al*., 1998).

Little evidence has been presented specifically for *N*-nitrosodiethanolamine to support the hypothesis that nitrosoamines could be activated by a process involving sulfation (Sterzel & Eisenbrand, 1986; Loeppky *et al*., 1987; Michejda *et al*., 1994; Loeppky *et al*., 1998).

Experiments *in vitro* showed that the liver and the nasal mucosa were capable of forming $^{14}CO_2$ from *N*-nitroso[^{14}C]diethanolamine (Löfberg & Tjälve, 1985).

(*d*) *Excretion*

About 60–90% of *N*-nitrosodiethanolamine administered orally, intravenously, cutaneously or intratracheally to rats was excreted unchanged in the urine (Preussmann *et al*., 1978, 1981; Lethco *et al*., 1982; Spiegelhalder *et al*., 1982, 1984). In some studies, the amount of *N*-nitrosodiethanolamine recovered in 24-h urine was only 3–30% of the dose (Lijinsky *et al*., 1981; Lethco *et al*., 1982; Airoldi *et al*., 1984a). Only 2–8% of the dose was found in the faeces and ≤ 2% was exhaled as $^{14}CO_2$ after oral or intravenous administration of *N*-nitroso[^{14}C]diethanolamine (Lethco *et al*., 1982; Löfberg & Tjälve, 1985; Farrelly *et al*., 1987). The amounts of *N*-nitrosodiethanolamine that were excreted in a conjugated form (glucuronide/sulfate) appeared to be marginal in CD-COBS and Fischer 344 rat urine (Bonfanti *et al*., 1985; Farrelly *et al*., 1987).

Excretion rates of *N*-nitrosodiethanolamine in urine (24 h) seemed not to be dose-dependent in rats following epicutaneous, intratracheal or oral administration of *N*-nitrosodiethanolamine at dose ranges of 0.03–300 mg per animal (Spiegelhalder *et al*., 1982), 100–1000 mg/kg bw (Preussmann *et al*., 1978) or 0.5–50 mg/kg bw (Lethco *et al*., 1982).

N-Nitrosomorpholine and *N*-nitrosodiethanolamine are both converted *in vivo* to *N*-nitroso-*N*-2-hydroxyethylglycine, which is excreted in rodent urine. The recovery of *N*-nitroso-*N*-2-hydroxyethylglycine in 24-h urine was lower in rats (8%) than in mice or hamsters (11–14%) dosed intraperitoneally with *N*-nitrosodiethanolamine (5 mg/kg bw), which was also found in urine of all the species (Bonfanti *et al*., 1986). Biliary excretion (a minor route of elimination) and enterohepatic recycling of *N*-nitrosodiethanolamine and its metabolite *N*-nitroso-*N*-2-hydroxyethylglycine has been shown in rats after intravenous administration of 5 mg/kg bw *N*-nitrosodiethanolamine (Bonfanti *et al*., 1985).

N-Nitrosodiethanolamine has been detected in urine (1–150 μg per animal) of male Sprague-Dawley rats given nitrite in their drinking water following skin treatment with diethanolamine (100–400 mg per rat) (Preussmann *et al*., 1981).

4.2 Toxic effects

No data were available to the Working Group.

4.3 Reproductive and developmental effects

No data were available to the Working Group.

4.4 Genetic and related effects

4.4.1 *Humans*

One small study (Garry *et al*., 1986) examined peripheral blood samples from 11 tool and die workers and seven non-manufacturing workers (controls) for frequencies of sister chromatid exchanges and chromosomal aberrations. No significant differences were observed for either parameter.

In a larger study (Fuchs *et al*., 1995; Oesch *et al*., 1995), peripheral blood from 65 male metal workers from seven small–medium plants in central West Germany was examined for DNA single-strand breaks (primary, including reversible DNA damage). There was a significant correlation between the increased level of DNA damage in workers and the concentration of *N*-nitrosodiethanolamine in the air of the workplace, but a concomitant effect of other genotoxic agents in the environment could not be excluded. [The Working Group noted the absence of a control group.]

4.4.2 *Experimental systems* (see Table 3 for references)

N-Nitrosodiethanolamine was mutagenic to *Salmonella typhimurium* in most assays in the presence of exogenous metabolic activation systems and in some assays in the absence of such systems.

In two studies, *N*-nitrosodiethanolamine was not mutagenic to *Escherichia coli* K-12/343/113 in the absence of exogenous metabolic activation. When exogenous metabolic activation preparations from rats or mice were included, mutations were induced at one of three loci tested, i.e. *gal*$^+$. These two studies also reported that *N*-nitrosodiethanolamine induced mutations in *E. coli* K12/343/113 in animal-mediated assays in male and female mouse livers and spleens. In one of these studies, pyrazole, an ADH-blocking agent, did not completely abolish this mutagenicity, indicating that metabolic activation pathways not requiring ADH are involved in *N*-nitrosodiethanolamine mutagenicity.

In a single study in *Drosophila melanogaster* with a wide dose range (0.01–2.0 M applied topically on late embryos and newly hatched larvae), *N*-nitrosodiethanolamine was not mutagenic in germ cells by the classic sex-linked recessive lethal test, apart from a very weak effect after very high exposure. In contrast, *N*-nitrosodiethanolamine did induce genetic effects in somatic cells at much lower doses (0.01–0.26 M). In a single

Table 3. Genetic and related effects of *N*-nitrosodiethanolamine

Test system	Result[a]		Dose[b] (LED or HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Salmonella typhimurium TA98, TA100, TA1530, TA1538, reverse mutation	–	–	NR	Gilbert *et al.* (1979)
Salmonella typhimurium TA 100, reverse mutation	+	+[c]	6300 µg/plate	Hesbert *et al.* (1979)
Salmonella typhimurium TA1535, reverse mutation	+	NR	1260 µg/plate	Hesbert *et al.* (1979)
Salmonella typhimurium TA100, TA1530, TA1535, TA1537, TA1538, TA98, reverse mutation	–	–	10 000 µg/plate	Gilbert *et al.* (1981)
Salmonella typhimurium TA100, TA1535, reverse mutation	–	+[d]	5000 µg/plate	Prival *et al.* (1982)
Salmonella typhimurium TA100, reverse mutation	–	+[e]	3350 µg/plate	Eisenbrand *et al.* (1984)
Salmonella typhimurium TA98, TA100, reverse mutation	+	+[f]	13 400 µg/plate	Dahl (1985)
Salmonella typhimurium TA100, reverse mutation	(+)	(+)[g]	20 000 µg/plate	Mori *et al.* (1987)
Salmonella typhimurium TA1535, reverse mutation	–	–[h]	1000 µg/plate	Lijinsky & Andrews (1983)
Salmonella typhimurium TA1535, reverse mutation	–	–	6700 µg/plate	Eisenbrand *et al.* (1984)
Escherichia coli K-12/343/113, forward mutation (VAL^R)	NR	–[i]	20 000	Kerklaan *et al.* (1981)
Escherichia coli K-12/343/113, mutation (gal^+)	–	+[i,k]	2680	Knasmüller *et al.* (1986)
Drosophila melanogaster unstable *zeste/white* mutation assay (somatic)	+	NT	34 800[l]	Fahmy & Fahmy (1984)
Drosophila melanogaster unstable *zeste/white* mutation assay (germ)	–	NT	268 000[l]	Fahmy & Fahmy (1984)
Drosophila melanogaster unstable *white-ivory-16* mutation assay (somatic)	+	NT	34 800[l]	Fahmy & Fahmy (1984)
Drosophila melanogaster unstable *white-ivory-16* mutation assay (germ)	–	NT	268 000[l]	Fahmy & Fahmy (1984)
DNA single-strand breaks, primary Sprague-Dawley rat hepatocytes *in vitro*	+	NT	1675	Denkel *et al.* (1986)
DNA amplification, CO631 Chinese hamster embryo cells	–	NT	1340	Denkel *et al.* (1986)
DNA single-strand breaks, primary rat hepatocytes *in vitro*	+	NT	3350	Pool *et al.* (1990)
DNA single-strand breaks, primary hamster hepatocytes *in vitro*	+	NT	1675	Pool *et al.* (1990)
DNA single-strand breaks, primary pig hepatocytes *in vitro*	+	NT	1675	Pool *et al.* (1990)
Sister chromatid exchange, human lymphocytes *in vitro*	+	NT	3570	Dittberner *et al.* (1988)
Sister chromatid exchange, human lymphocytes *in vitro*	NT	+[n]	1675 µg/tube[m]	Henn *et al.* (1989)

Table 3 (contd)

Test system	Result[a]		Dose[b] (LED or HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Sister chromatid exchange, human lymphocytes *in vitro*	NT	+[o]	840 µg/tube[m]	Henn *et al.* (1989)
Chromosomal aberrations, human lymphocytes *in vitro*	(+)	NT	13 800	Dittberner *et al.* (1988)
Chromosomal aberrations, human lymphocytes *in vitro*	NT	–[n]	8380 µg/tube[m]	Henn *et al.* (1989)
Chromosomal aberrations, human lymphocytes *in vitro*	NT	–[o]	4190 µg/tube[m]	Henn *et al.* (1989)
Micronucleus formation, human lymphocytes *in vitro*	+	NT	8770	Dittberner *et al.* (1988)
DNA single strand breaks in Namalva, human lymphoblastoid cell line	–	NT	40 240	Scherer *et al.* (1991)
DNA single strand breaks in rat (Sprague Dawley) or human kidney cells	–	NT	4290	Robbiano *et al.* (1996)
DNA single strand breaks in rat (Sprague Dawley) or human kidney cells	–	NT	6700	Brendler *et al.* (1992)
DNA strand breaks, single-cell gel electrophoresis assay, human mucosal cells of the upper aerodigestive tract *in vitro*	+	NT	6700	Harréus *et al.* (1999)
Host-mediated assay, *Escherichia coli* K-12/343/113 forward mutation (VAL^R) in female Swiss albino mouse livers *in vivo*	+		60.4 ip × 1; 3 h	Kerklaan *et al.* (1981)
Host-mediated assay, *Escherichia coli* forward mutation (*gal*$^+$) in male Swiss albino mouse livers *in vivo*	+		750 s.c. × 1; 3 h	Knasmüller *et al.* (1986)
Host-mediated assay, *E. coli* K-12/343/113 forward mutation (VAL^R) in male Swiss albino mouse liver and spleen *in vivo*	+		30 ip × 1; 3 h	Knasmüller *et al.* (1986)
Host-mediated assay, *E. coli* K-12 *uvr*B/*rec*A and *uvr*$^+$/*rec*$^+$ (differential DNA repair) in male Swiss albino mouse liver, lung and kidney *in vivo*	+		600 po × 1; 2 h	Knasmüller *et al.* (1993)
DNA single-strand breaks, Sprague-Dawley rat liver *in vivo*	+		50.3 po × 1	Denkel *et al.* (1986)
DNA single-strand breaks, male Wistar rat liver *in vivo*	+		100 po × 1	Sterzel & Eisenbrand (1986)
DNA strand breaks, male Sprague-Dawley rat liver *in vivo*	+		1.03 po × 1	Brambilla *et al.* (1987)
Unscheduled DNA synthesis, male Fischer 344 rat hepatocytes *in vivo*	–		1000 po × 1; 2 h/12 h	Mirsalis *et al.* (1989)

Table 3 (contd)

Test system	Result[a]		Dose[b] (LED or HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Unscheduled DNA synthesis, male $B6C3F_1$ mouse hepatocytes *in vivo*	–		600 po × 1; 2 h/12 h	Mirsalis *et al.* (1989)
Micronucleus assay in *Pleurodeles waltl* (newt) larvae red blood cells *in vivo*	+		12.5 µg/mL; 12 d	L'Haridon *et al.* (1993)
Micronucleus formation, BALB/c mice *in vivo*	–		10 000 ip × 1	Gilbert *et al.* (1981)
Chromosomal aberrations, BALB/c mice *in vivo*	–		10 000 ip × 1	Gilbert *et al.* (1981)

[a] +, positive; (+), weak positive; –, negative; NT, not tested
[b] LED, lowest effective dose; HID, highest ineffective dose; in-vitro tests, µg/mL; in-vivo tests, mg/kg bw/day; po, oral; ip, intraperitoneal; d, day
[c] Mouse S9
[d] Hamster S9; inactive with rat S9
[e] Activation with NAD/ADH pH 7.4 and pH 9.5; higher doses were toxic.
[f] Nasal, lung and liver S9 from rat and rabbit
[g] Commercial grade; a purified sample was not mutagenic in the absence of S9.
[h] Rat or hamster liver S9
[i] Rat or mouse liver S9
[k] No mutation leading to arginine prototrophy or 5-methyl-DL-tryptophan resistance were detected.
[l] Topical application on embryos and larvae
[m] Volume not clearly stated
[n] Activation with alcohol dehydrogenase from yeast
[o] Activation with alcohol dehydrogenase from horse liver

study in larvae of the newt *Pleurodeles waltl*, exposure to *N*-nitrosodiethanolamine increased the frequency of micronuclei in a dose-dependent manner at concentrations from 12.5 to 50 ppm (93–373 μM (to larvae, feeding and renewal daily for 12 days).

DNA single-strand breaks were induced *in vitro* by *N*-nitrosodiethanolamine in rat, hamster and pig hepatocytes. However, no DNA single-strand breaks were found in Chinese hamster embryo cells, in a human lymphoblastoid cell line or in rat and human kidney cells *in vitro*.

In one study using cultured human lymphocytes from two female donors, sister chromatid exchanges, chromosomal aberrations and micronuclei were found at increased frequencies after exposure to *N*-nitrosodiethanolamine without exogenous metabolic activation. Dose-dependent increases were detected for all three end-points. In another study, the effect of ADH/NAD from horse liver and yeast was tested for induction of chromosomal aberrations and sister chromatid exchanges by *N*-nitrosodiethanolamine in human lymphocyte cultures. No significantly higher levels of numerical or structural chromosomal aberrations were seen, but the sister chromatid exchange frequency was significantly increased in a dose-dependent manner at dose levels which were significantly lower than those employed in the first study.

A study using the Comet assay in mucosal cells isolated from human biopsies of the upper aerodigestive tract indicated that *N*-nitrosodiethanolamine caused significant DNA damage in oral cavity epithelia and also in the mucosa of the pharynx and larynx.

Induction of DNA single-strand breakage in rat liver after in-vivo exposure to *N*-nitrosodiethanolamine was demonstrated in three studies and dose-dependent effects were shown. In one of these studies, the DNA strand-breaking potential of *N*-nitrosodiethanolamine was found to be abolished by inhibition of sulfotransferase by 2,6-dichloro-4-nitrophenol. Unscheduled DNA synthesis was not detected in rats or mice in an in-vivo/in-vitro hepatocyte DNA repair assay after treatment with *N*-nitrosodiethanolamine. A single study in mice exposed *in vivo* to *N*-nitrosodiethanolamine did not find any significant induction of structural or numerical chromosomal aberrations or micronuclei in bone-marrow cells.

4.5 Mechanistic considerations

Evidence from biological and synthetic model studies suggests that the metabolic activation of *N*-nitrosodiethanolamine may involve enzymatic oxidation at both α and β carbons (Eisenbrand & Janzowski, 1994; Loeppky *et al*., 1998; Park *et al*., 1998; Loeppky, 1999). *N*-Nitrosodiethanolamine has been found to be mutagenic in *Salmonella typhimurium*, mainly in the presence of metabolic activation systems. Eisenbrand *et al*. (1984) reported an activation mechanism for *N*-nitrosodiethanolamine by alcohol dehydrogenase and also investigated several of the metabolites generated during these processes. All were direct-acting mutagens in *S. typhimurium* TA100 (Denkel *et al*., 1986). Cytotoxicity in mammalian cells appears to be CYP2E1-dependent (Janzowski *et al*., 1996; Loeppky, 1999).

5. Summary of Data Reported and Evaluation

5.1 Exposure data

N-Nitrosodiethanolamine is a contaminant formed by the action of nitrites on ethanolamines in a wide range of products including metalworking fluids, pesticides, antifreeze and personal care products. Occupational exposure by inhalation and dermal contact may occur from water-diluted metalworking fluids contaminated with *N*-nitrosodiethanolamine. General population exposure is possible through contact with a variety of personal care products and the use of some tobacco products. Contamination levels in both metalworking fluids and personal care products have considerably decreased since the 1980s.

5.2 Human carcinogenicity data

Four studies showed inconsistent increases in cancer mortality or incidence at various sites among workers using metalworking fluids containing ethanolamines and sodium nitrite. Only one of them attempted indirectly to estimate exposure to nitrosamines, showing an increased risk for oesophageal cancer with increasing duration of exposure, but there was concomitant exposure to biocides, also associated with an increased risk for oesophageal cancer in this study.

5.3 Animal carcinogenicity data

N-Nitrosodiethanolamine was tested for carcinogenicity by addition to drinking-water in six studies in rats. It was also tested in hamsters by subcutaneous injection in three studies and in single studies by topical or buccal administration. In rats, it consistently produced liver tumours (principally hepatocellular carcinomas). It also produced adenocarcinomas and squamous-cell carcinomas of the nasal cavity. In hamsters, *N*-nitrosodiethanolamine consistently induced adenocarcinomas of the nasal cavity.

In a mouse lung screening assay, *N*-nitrosodiethanolamine increased the incidence and multiplicity of lung tumours.

5.4 Other relevant data

N-Nitrosodiethanolamine is metabolized *in vivo* slowly and only to a small extent, being principally eliminated unchanged in human and rodent urine. Bioactivation of *N*-nitrosodiethanolamine is associated with α- and β-hydroxylation pathways involving the enzymes CYP2E1 and alcohol dehydrogenase, resulting in DNA adduct formation.

Two studies have examined the potential genotoxic hazard of occupational exposure to *N*-nitrosodiethanolamine. The larger one, measuring DNA damage, indicated an association between single-strand breakage and the presence of *N*-nitrosodiethanolamine in

the air of the workplace, but the effects of other exposures could not be excluded. The small study measuring chromosome damage in tool-room workers did not find any significant effect.

N-Nitrosodiethanolamine is mutagenic to bacteria. Studies using cultured cells *in vitro* found induction of DNA single-strand breakage after exposure to *N*-nitrosodiethanolamine in human buccal cells and in rat, hamster and pig hepatocytes. Chromosomal damage was detected in human lymphocytes without exogenous metabolic activity in one study; sister chromatid exchange frequency alone was increased and was detected at lower doses in another study with an exogenous metabolic system including alcohol dehydrogenase.

5.5 Evaluation

There is *inadequate evidence* in humans for the carcinogenicity of *N*-nitrosodiethanolamine.

There is *sufficient evidence* in experimental animals for the carcinogenicity of *N*-nitrosodiethanolamine.

Overall evaluation

N-Nitrosodiethanolamine is *possibly carcinogenic to humans (Group 2B).*

6. References

Airoldi, L., Bonfanti, M., Benfenati, E., Tavecchia, P. & Fanelli, R. (1983) Identification of an acidic metabolite of *N*-nitrosodiethanolamine isolated from rat urine. *Biomed. Mass Spectrom.*, **10**, 334–337

Airoldi, L., Macri, A., Bonfanti, M., Bonati, M. & Fanelli, R. (1984a) Kinetics and bioavailability of *N*-nitrosodiethanolamine after intravenous and cutaneous administration to rats. *Food chem. Toxicol.*, **22**, 133–138

Airoldi, L., Bonfanti, M., Fanelli, R., Bove, B., Benfenati, E. & Gariboldi, P. (1984b) Identification of a nitrosamino aldehyde and a nitrosamino acid resulting from β-oxidation of *N*-nitrosodiethanolamine. *Chem.-biol. Interact.*, **51**, 103–113

Anucha, T.C.A., Okieimen, F.E. & Ajibola, M.M. (1986) Contamination of meat products by trace quantities of nitrosodiethanolamine (NDELA). *Bull. environ. Contam. Toxicol.*, **36**, 392–395

Berg, H., Fallentin, B., Haunsoe, N., Meyer Henius, U., Klove Johansen, A., Moeslund, J., Rietz, B. & Schneider, T. (1979) *Omdannelser I Køle-smøremidler under Brug*, Copenhagen, Teknologisk Institut

Berger, M.R., Schmähl, D. & Zerban, H. (1987) Combination experiments with very low doses of three genotoxic *N*-nitrosamines with similar organotrophic carcinogenicity in rats. *Carcinogenesis*, **8**, 1635–1643

Beyer, K.H., Jr, Bergfeld, W.F., Berndt, W.O., Boutwell, R.K., Carlton, W.W., Hoffmann, D.K. & Schroeder, A.L. (1983) Final report on the safety assessment of triethanolamine, diethanolamine and monoethanolamine. *J. Am. Coll. Toxicol.*, **2**, 183–235

Bonfanti, M., Castelli, M.G., Fanelli, R. & Airoldi, L. (1985) Enterohepatic recycling of *N*-nitrosodiethanolamine in rats. *Food chem. Toxicol.*, **23**, 1011–1013

Bonfanti, M., Magagnotti, C., Fanelli, R. & Airoldi, L. (1986) Comparative in vitro and in vivo β-oxidation of *N*-nitrosodiethanolamine in different animal species. *Chem.-biol. Interact.*, **59**, 203–210

Bonfanti, M., Magagnotti, C., Fanelli, R. & Airoldi, L. (1987) β-Oxidation of *N*-nitrosodiethanolamine in different animal species *in vitro* and *in vivo*. In: Bartsch, H., O'Neill, I. & Schulte-Hermann, R., eds, *The Relevance of N-Nitroso Compounds to Human Cancer. Exposures and Mechanisms* (IARC Scientific Publications No. 84), Lyon, IARC*Press*, pp. 91–93

Brambilla, G., Carlo, P., Finollo, R. & Sciabà, L. (1987) Dose–response curves for liver DNA fragmentation induced in rats by sixteen *N*-nitroso compounds as measured by viscometric and alkaline elution analyses. *Cancer Res.*, **47**, 3485–3491

Brendler, S.Y., Tompa, A., Hutter, K.F., Preussmann, R. & Pool-Zobel, B.L. (1992) *In vivo* and *in vitro* genotoxicity of several *N*-nitrosamines in extrahepatic tissues of the rat. *Carcinogenesis*, **13**, 2435–2441

Bronaugh, R.L., Congdon, E.R. & Scheuplein, R.J. (1981) The effect of cosmetic vehicles on the penetration of *N*-nitrosodiethanolamine through excised human skin. *J. invest. Dermatol.*, **76**, 94–96

Brunnemann, K.D. & Hoffmann, D. (1981) Assessment of the carcinogenic *N*-nitrosodiethanolamine in tobacco products and tobacco smoke. *Carcinogenesis*, **2**, 1123–1127

Brunnemann, K.D. & Hoffmann, D. (1991) Decreased concentrations of *N*-nitrosodiethanolamine and *N*-nitrosomorpholine in commercial tobacco products. *J. agric. Food Chem.*, **39**, 207–208

Brunnemann, K.D., Hecht, S.S. & Hoffmann, D. (1982–1983) N-Nitrosamines; environmental occurrence, in vivo formation and metabolism. *J. Toxicol. clin. Toxicol.*, **19**, 661–688

Budavari, S., ed. (1998) *The Merck Index*, 12th Ed., Version 12:2, Whitehouse Station, NJ, Merck & Co. [CD-ROM]

Chung, F.-L. & Hecht, S.S. (1985) Formation of the cyclic 1,N^2-glyoxal-deoxyguanosine adduct upon reaction of *N*-nitroso-2-hydroxymorpholine with deoxyguanosine. *Carcinogenesis*, **6**, 1671–1673

Dahl, A.R. (1985) Mutagenicity of some dialkylnitrosamines, cyclic nitrosamines and N,N-diethanolnitrosamine in *Salmonella typhimurium* with rat and rabbit nasal, lung and liver S9 homogenates. *Mutat. Res.*, **158,** 141–147

Denkel, E., Pool, B.L., Schlehofer, J.R. & Eisenbrand, G. (1986) Biological activity of N-nitrosodiethanolamine and of potential metabolites which may arise after activation by alcohol dehydrogenase in *Salmonella typhimurium*, in mammalian cells, and in vivo. *J. Cancer Res. clin. Oncol.*, **111**, 149–153

Denkel, E., Sterzel, W. & Eisenbrand, G. (1987) In-vivo activation of *N*-nitrosodiethanolamine and other *N*-nitroso-2-hydroxyalkylamines by alcohol dehydrogenase and sulfotransferase. In: Bartsch, H., O'Neill, I. & Schulte-Hermann, R., eds, *The Relevance of N-Nitroso Compounds to Human Cancer. Exposures and Mechanisms* (IARC Scientific Publications No. 84), Lyon, IARC*Press*, pp. 83–86

Department of Health and Human Services (1999) *8th Report on Carcinogens. 1998 Summary*, Research Triangle Park, NC, National Toxicology Program, pp. 158–159

Deutsche Forschungsgemeinschaft (1999) *List of MAK and BAT Values 1999*, Report No. 35, Weinheim, Wiley-VCH, pp. 82-116

Dittberner, U., Eisenbrand, G. & Zankl, H. (1988) Cytogenetic effects of N-nitrosodiethanolamine (NDELA) and NDELA-monoacetate in human lymphocytes. *J Cancer Res. clin. Oncol.*, **114**, 575–578

Ducos, P., Limasset, J.-C., Maire, C., Lafontaine, M., Truy, S., Hesbert, A., Lemonnier, M. & Cavelier, M.C. (1979) [*N*-Nitrosodiethanolamine in synthetic cutting fluids]. *Cah. Notes doc.*, **95**, 249–257 (in French)

Ducos, P., Maire, C, Limasset, J.C. & Gaudin, R. (1983) N-Nitrosodiethanolamine in anti-freeze. *Environ. Res*., **31**, 95–99

Edwards, G.S., Peng, M., Fine, D.H., Spiegelhalder, B. & Kann, J. (1979) Detection of N-nitrosodiethanolamine in human urine following application of a contaminated cosmetic. *Toxicol. Lett.*, **4**, 217–222

Eisen, E.A., Tolbert, P.E., Monson, R.R. & Smith, T.J. (1992) Mortality studies of machining fluid exposure in the automobile industry. I: A standardized mortality ratio analysis. *Am. J. ind. Med.*, **22**, 809–824

Eisenbrand, G. & Janzowski, C. (1994) Potential mechanism of action of nitrosoamines with hydroxy, oxo, or carboxy groups. In: Loeppky, R.N. & Michejda, C.J., eds, *Nitrosamines and Related N-Nitroso Compounds. Chemistry and Biochemistry* (ACS Symposium Series No. 553), Washington DC, American Chemical Society, pp. 179–194

Eisenbrand, G., Denkel, E. & Pool, B. (1984) Alcohol dehydrogenase as an activating enzyme for *N*-nitrosodiethanolamine (NDELA): in vitro activation of NDELA to a potent mutagen in *Salmonella typhimurium*. *J. Cancer Res. clin. Oncol*., **108**, 76–80

Eisenbrand, G., Blankart, M., Sommer, H. & Weber, B. (1991) *N*-Nitrosoalkanolamines in cosmetics. In: O'Neil, I.K., Chen, J. & Bartsch, H., eds, *Relevance to Human Cancer of N-Nitroso Compounds, Tobacco Smoke and Mycotoxins* (IARC Scientific Publications No. 105), Lyon, IARC*Press*, pp. 238–241

Elder, R.L. (1980) Final report on the safety assessment for 2-bromo-2-nitropropane-1,3-diol. *J. environ. Pathol. Toxicol.*, **4**, 47

Fadlallah, S., Cooper, S.F., Fournier, M., Drolet, D. & Perreault, G. (1990) Determination of *N*-nitroso compounds in metal working fluids. *J. chromatogr. Sci*., **28**, 517–523

Fadlallah, S., Cooper S.F., Perrault, G., Truchon, G. & Lesage, J. (1996) N-Nitroso compounds in the ambient air of metal factories using metal-working fluids. *Bull. environ. Contam. Toxicol*., **57**, 867–874

Fadlallah, S., Cooper, S.F., Perrault, G., Truchon, G. & Lesage, J. (1997) Presence of *N*-nitroso-diethanolamine contamination in Canadian metal-working fluids. *Sci. total Environ*., **186**, 197–204

Fahmy, M.J. & Fahmy, O.G. (1984) Genetic activity of the carcinogen *N*-nitrosodiethanol-amine (NDELA) on unstable mutant alleles of the *white* locus in *Drosophila melanogaster*. *Teratog. Carcinog. Mutag*., **4**, 437–447

Fan, S. & Fajen, J. (1978) *Survey for N-Nitroso Compounds at Chrysler Forge and Axle Plant, Detroit, Michigan. Thermo Electron Research Center, Waltham, Massachusetts*, Cincinnati, OH, National Institute for Occupational Safety and Health

Fan, T.Y., Morrison, J., Rounbehler, D.P., Ross, R., Fine, D.H., Miles, W. & Sen, N.P. (1977a) Nitrosodiethanolamine in synthetic cutting fluids: A part-per-hundred impurity. *Science*, **196**, 70–71

Fan, T.Y, Goff, U., Song, L., Fine, D.H., Arsenault, G.P. & Biemann, K. (1977b) N-Nitrosodiethanolamine in cosmetics, lotions and shampoos. *Food Cosmet. Toxicol*., **15**, 423–430

Farrelly, J.G., Stewart, M.L. & Lijinsky, W. (1984) The metabolism of nitrosodi-*n*-propylamine, nitrosodiallylamine and nitrosodiethanolamine. *Carcinogenesis*, **5**, 1015–1019

Farrelly, J.G., Thomas, B.J. & Lijinsky, W. (1987) Metabolism and cellular interactions of *N*-nitrosodiethanolamine. In: Bartsch, H., O'Neill, I. & Schulte-Hermann, R., eds, *The Relevance of N-Nitroso Compounds to Human Cancer. Exposures and Mechanisms* (IARC Scientific Publications No. 84), Lyon, IARC*Press*, pp. 87–90

Franz, T.J., Lehman, P.A., Franz, S.F., North-Root, H., Demetrulias, J.L., Kelling, C.K., Moloney, S.J. & Gettings, S.D. (1993) Percutaneous penetration of *N*-nitrosodiethanolamine through human skin (*in vitro*): comparison of finite and infinite dose applications from cosmetic vehicles. *Fundam. appl. Toxicol.*, **21**, 213–221

Fuchs, J., Burg, J., Hengstler, J.G., Bolm-Audorff, U. & Oesch, F. (1995) DNA damage in mononuclear cells of metal workers exposed to N-nitrosodiethanolamine in synthetic cutting fluids. *Mutat. Res.*, **342**, 95–102

Garry,V.F, Jacobs, D.R., Kreiger, R.A., Nelson, R.L., Loeppky, R. & Harkins, M.E. (1986) Integration of laboratory and epidemiologic studies to evaluate genotoxic exoposure in tool and die workers. *Prog. clin. biol. Res.*, **207**, 183–193

Gilbert, P., Rollmann, B., Rondelet, J., Mercier, M. & Poncelet, F. (1979) Mutagenicity of N-nitrosodiethanolamine and its acetyl-derivatives. *Arch. int. Physiol. Biochim.*, **87**, 813–814

Gilbert, P., Fabry, L., Rollmann, B., Lombart, P., Rondelet, J., Poncelet, F., Leonard, A. & Mercier, M. (1981) Mutagenicity of *N*-nitrosodiethanolamine and its acetyl derivatives. *Mutat. Res*., **89**, 217–228

Harréus, U., Schmezer, P., Kuchenmeister, F. & Maier, H. (1999) [Genotoxic effects in human epithelial cells of the upper aerodigestive tract]. *Laryngorhinootologie*, **78**, 176–181 (in German)

Hecht, S.S. (1984) N-Nitroso-2-hydroxymorpholine, a mutagenic metabolite of N-nitrosodiethanolamine. *Carcinogenesis*, **5**, 1745–1747

Hecht, S.S., Lijinsky, W., Kovatch, R.M., Chung, F.-L. & Saavedra, J.E. (1989) Comparative tumorigenicity of *N*-nitroso-2-hydroxymorpholine, *N*-nitrosodiethanolamine and *N*-nitrosomorpholine in A/J mice and F344 rats. *Carcinogenesis*, **10**, 1475–1477

Henn, I., Eisenbrand, G. & Zankl, H. (1989) Increased mutagenicity of *N*-nitrosodiethanolamine in human lymphocyte cultures after activation by alcohol dehydrogenase. *J. Cancer Res. clin. Oncol.*, **115**, 445–448

Hesbert, A., Lemonnier, M. & Cavelier, C. (1979) Mutagenicity of N-nitrosodiethanolamine on *Salmonella typhimurium*. *Mutat. Res.*, **68**, 207–210

Hilfrich, J., Schmeltz, I. & Hoffmann, D. (1977) Effects of *N*-nitrosodiethanolamine and 1,1-diethanolhydrazine in Syrian golden hamsters. *Cancer Lett.*, **4**, 55–60

Hoffmann, D., Brunneman, K.D., Rivenson, A. & Hecht, S.S. (1982) *N*-Nitrosodiethanolamine: analysis, formation in tobacco products and carcinogenicity in Syrian Golden hamsters. In: Bartsch, H., O'Neill, I.K., Castegnaro, M. & Okada, M., eds, *N-Nitroso Compounds: Occurrence and Biological Effects* (IARC Scientific Publications No. 41), Lyon, IARC*Press*, pp. 299–302

Hoffmann, D., Rivenson, A., Adams, J.D., Juchatz, A., Vinchkoski, N. & Hecht, S.S. (1983) Effects of route of administration and dose on the carcinogenicity of *N*-nitrosodiethanolamine in the Syrian golden hamster. *Cancer Res.*, **43**, 2521–2524

IARC (1978) *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans*, Vol. 17, *Some* N-*Nitroso Compounds*, Lyon, IARC*Press*, pp. 77–82

IARC (1985) *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans*, Vol. 37, *Tobacco Habits Other than Smoking: Betel-Quid and Areca-Nut Chewing; and Some Related Nitrosamines*, Lyon, IARC*Press*, pp. 73–74

IARC (1987) *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans*, Suppl. 7, *Overall Evaluations of Carcinogenicity: An Update of IARC Monographs Volumes 1 to 42*, Lyon, IARC*Press*, p. 67

Janzowski, C., Scharner, A. & Eisenbrand, G. (1996) Activation of nitrosamines by cytochrome P450 isoenzymes expressed in transfected V79 cells (Abstract). *Eur. J. Cancer Prev.*, **5**, S154

Järvholm, B. & Lavenius, B. (1987) Mortality and cancer morbidity in workers exposed to cutting fluids. *Arch. environ. Health*, **42**, 361–366

Järvholm, B., Lavenius, B. & Sällsten, G. (1986) Cancer morbidity in workers exposed to cutting fluids containing nitrites and amines. *Br. J. ind. Med.*, **43**, 563–565

Järvholm, B., Zingmark, P.-A. & Österdahl, B.-G. (1991a) High concentration of *N*-nitrosodiethanolamine in a diluted commercial cutting fluid. *Am. J. ind. Med.*, **19**, 237–239

Järvholm, B., Zingmark, P.-A. & Österdahl, B.-G. (1991b) N-Nitrosodiethanolamine in commercial cutting fluids without nitrites. *Ann. occup. Hyg.*, **35**, 659–663

Keefer, L.K., Goff, U., Stevens, J. & Bennett, E.O. (1990) Persistence of *N*-nitrosodiethanolamine contamination in American metal-working lubricants. *Food chem. Toxicol.*, **28**, 531–534

Kerklaan, P., Mohn, G. & Bouter, S. (1981) Comparison of the mutagenic activity of dialkylnitrosamines in animal-mediated and *in vitro* assays using an *Escherichia coli* indicator. *Carcinogenesis*, **2**, 909–914

Knasmüller, S., Stehlik, G. & Mohn, G. (1986) Studies on the metabolic activation of diethanolnitrosamine in animal-mediated and in vitro assays using *Escherichia coli* K-12 343/113 as an indicator. *J. Cancer Res. clin.Oncol.*, **112**, 266–271

Knasmüller, S., Huber, W. & Schulte-Hermann, R. (1993) Prevention of genotoxic effects by dietary constituents in various organs of mice treated with nitrosamines. In: Bronzetti, G., Hayatsu, H., De Flora, S., Waters, M.D. & Shankel, D.N., eds, *Antimutagenesis and Anticarcinogenesis Mechanisms III*, New York, Plenum Press, pp. 447–457

L'Haridon, J., Fernandez, M., Ferrier, V. & Bellan, J. (1993) Evaluation of the genotoxicity of *N*-nitrosoatrazine, *N*-nitrosodiethanolamine and their precursors *in vivo* using the newt micronucleus test. *Water Res.*, **27**, 855–862

Lethco, E.J., Wallace, W.C. & Brouwer, E. (1982) The fate of *N*-nitrosodiethanolamine after oral and tropical administration to rats. *Food chem. Toxicol.*, **20**, 401–406

Lijinsky, W. & Andrews, A.W. (1983) The superiority of hamster liver microsomal fraction for activating nitrosamines to mutagens in *Salmonella typhimurium*. *Mutat. Res.*, **111**, 135–144

Lijinsky, W. & Kovatch, R. (1985) Induction of liver tumours in rats by nitrosodiethanolamine at low doses. *Carcinogenesis*, **6**, 1679–1681

Lijinsky, W. & Reuber, M.D. (1984) Dose response study with *N*-nitrosodiethanolamine in F344 rats. *Food chem. Toxicol.*, **22**, 23–36

Lijinsky, W., Keefer, L., Conrad, E. & Van de Bogart, R. (1972) Nitrosation of tertiary amines and some biological implications. *J. natl Cancer Inst.*, **49**, 1239–1249

Lijinsky, W., Reuber, M.D. & Manning, W.B. (1980) Potent carcinogenicity of nitrosodiethanolamine in rats. *Nature*, **288**, 589–590

Lijinsky, W., Losikoff, A.M. & Sansone, E.B. (1981) Penetration of rat skin by *N*-nitrosodiethanolamine and *N*-nitrosomorpholine. *J. natl Cancer Inst.*, **66**, 125–127

Lijinsky, W., Saavedra, J.E. & Kovatch, R.M. (1988) Carcinogenesis and nucleic acid alkylation by some oxygenated nitrosamines in rats and hamsters. *Chem.-biol. Interact.*, **66**, 37–47

Loeppky, R.N. (1999) The mechanism of bioactivation of *N*-nitrosodiethanolamine. *Drug Metab. Rev*., **31**, 175–193

Loeppky, R.N., Hansen, J.J. & Keefer, L.K. (1983) Reducing nitrosamine contamination in cutting fluids. *Food chem. Toxicol*., **5**, 607–613

Loeppky, R.N., Tomasik, W., Eisenbrand, G. & Denkel, E. (1987) α-Nitrosaminoaldehydes: highly reactive metabolites. In: Bartsch, H., O'Neill, I. & Schulte-Hermann, R., eds, *The Relevance of N-Nitroso Compounds to Human Cancer. Exposures and Mechanisms* (IARC Scientific Publications No. 84), Lyon, IARC*Press*, pp. 94–99

Loeppky, R.N., Lee, M.P. & Müller, S. (1994a) Modification of DNA by α-nitrosamino aldehydes. In: Hemminki, K., Dipple, A., Shuker, D.E.G., Kadlubar, F.F., Segerbäck, D. & Bartsch, H., eds, *DNA Adducts: Identification and Biological Significance* (IARC Scientific Publications No. 125), Lyon, IARC*Press*, pp. 429–432

Loeppky, R.N., Bao, Y.T., Bae, J., Yu, L. & Shevlin, G. (1994b) Blocking nitrosamine formation. Understanding the chemistry of rapid nitrosation. In: Loeppky, R.N. & Michejda, C.J., eds, *Nitrosamines and Related N-Nitroso Compounds. Chemistry and Biochemistry* (ACS Symposium Series 553), Washington DC, American Chemical Society, pp. 52–65

Loeppky, R.N., Fuchs, A., Janzowski, C., Humberd, C., Goelzer, P., Schneider, H. & Eisenbrand, G. (1998) Probing the mechanism of the carcinogenic activation of *N*-nitrosodiethanolamine with deuterium isotope effects: in vivo induction of DNA single-strand breaks and related in vitro assays. *Chem. Res. Toxicol.*, **11**, 1556–1566

Löfberg, B. & Tjälve, H. (1985) Autoradiography of [^{14}C]*N*-nitrosodiethanolamine in Sprague-Dawley rats. *Cancer Lett.*, **26**, 129–137

Lucke, W.E. & Ernst, J.M. (1993) Formation and precursors of nitrosamines in metalworking fluids. *Lubr. Eng.*, **49**, 271–275

Marzulli, F.N., Anjo, D.M. & Maibach, H.I. (1981) In vivo skin penetration studies of 2,4-toluenediamine, 2,4-diaminoanisole, 2-nitro-*p*-phenylenediamine, *p*-dioxane and *N*-nitrosodiethanolamine in cosmetics. *Food Cosmet. Toxicol.*, **19**, 743–747

Michejda, C.J., Koepke, S.R., Kroeger-Koepke, M.B. & Hernandez, L. (1994) Activation of β-hydroxyalkylnitrosamines. Evidence for involvement of a sulfotransferase. In: Pignatelli, M., Loeppky, R.N. & Michejda, C.J., eds, *Nitrosamines and Related N-Nitroso Compounds. Chemistry and Biochemistry* (ACS Symposium Series 553), Washington DC, American Chemical Society, pp. 195–210

Mirsalis, J.C., Tyson, C.K., Steinmetz, K.L., Loh, E.K., Hamilton, C.M., Bakke, J.P. & Spalding, J.W. (1989) Measurement of unscheduled DNA synthesis and S-phase synthesis in rodent hepatocytes following in vivo treatment: testing of 24 compounds. *Environ. mol. Mutag.*, **14**, 155–164

Monarca, S., Scassellati Sforzolini, G., Spiegelhalder, B., Pasquini, R. & Fatigoni, C. (1993) Monitoring nitrite, *N*-nitrosodiethanolamine, and mutagenicity in cutting fluids used in the metal industry. *Environ. Health Perspect.*, **101**, 126–128

Monarca, S., Scassellati-Sforzolini, G., Donato, F., Angeli, G., Spiegelhalder, B., Fatigoni, C. & Pasquini, R. (1996) Biological monitoring of workers exposed to *N*-nitrosodiethanolamine in the metal industry. *Environ. Health Perspect.*, **104**, 78–82

Mori, Y., Yamazaki, H. & Konishi, Y. (1987) Mutagenicity of *N*-nitrosodiethanolamine in the Salmonella/microsome test. *Mutat. Res.*, **192**, 91–94

National Toxicology Program (1998) *Eighth Report on Carcinogens—1998 Summary*, Research Triangle Park, NC, Department of Health and Human Services, Public Health Service, pp. 158–159

NOES (1999) *National Occupational Exposure Survey 1981-83*. Unpublished data as of July 1990, Cincinnati OH, Department of Health and Human Services, Public Health Service, Centers for Disease Control, National Institute for Occupational Safety and Health

Occupational Safety and Health Administration (1990) *OSHA Analytical Methods Manual*, 2nd Ed., *Part 1—Organic Substances*, Vol. 2, *Methods 29-54*, Salt Lake City, UT, United States Department of Labor [Method 31]

Oesch, F., Hengstler, J.G., Arand, M. & Fuchs J. (1995) Detection of primary DNA damage: applicability to biomonitoring of genotoxic occupational exposure and in clinical therapy. *Pharmacogenetics*, **5**, S118–S122

Okieimen, F.E. & Akintola, E.O. (1985) Contamination of dairy and tobacco products by trace quantities of nitrosodiethanolamine (NDELA). *Bull. environ. Contam. Toxicol.*, **35**, 466–470

Ong, J.T.H. & Rutherford, B.S. (1980) Some factors affecting the rate of *N*-nitrosodiethanolamine formation from 2-bromo-2-nitropropane-1,3-diol and ethanolamines. *J. Soc. Cosmet. Chem.*, **31**, 153–159

Park, R.M. & Mirer, F.E. (1996) A survey of mortality at two automotive engine manufacturing plants. *Am. J. ind. Med.*, **30**, 664–673

Park, R.M., Wegman, D.H., Silverstein, M.A., Maizlish, N.A. & Mirer, F.E. (1988) Causes of death among workers in a bearing manufacturing plant. *Am. J. ind. Med.*, **13**, 569–580

Park, M., Gu, F. & Loeppky, R.N. (1998) The synthesis of highly reactive, multi-functional α,β-epoxy- and α-acetoxy-nitrosamines. *Tetrahedron Lett.*, **39**, 1287–1290

Pfeiffer, W., Breuer, D., Blome, H., Deininger, Ch., Hahn, J.-U., Kleine, H., Nies, E., Pflaumbaum, W., Stockmann, R., Willert, G. & Sonnenschein, G. (1996) *Kühlschmierstoffe* (BIA-Report, 7/96). Sankt Augustin, Germany, Hauptverband der gewerblichen Berufsgenossenschaften (in German)

Pflaumbaum, W., Blome, H., Kleine, H., Smala, T. & Schmidt, M. (1999) *Grenzwerte*. BIA-Report, Sankt Augustin, Hauptverband der gewerblichen Berufsgenossenschaften (HVBG) (in German)

Pool, B.L., Eisenbrand, G., Preussmann, R., Schlehofer, J.R., Schmezer, P., Weber, H. & Wiessler, M. (1986) Detection of mutations in bacteria and of DNA damage and amplified DNA sequences in mammalian cells as a systematic test strategy for elucidating biological activities of chemical carcinogens. *Food chem. Toxicol.*, **24**, 685–691

Pool, B.L, Brendler, S.Y., Liegibel, U.M., Tompa, A. & Schmezer, P. (1990) Employment of adult mammalian primary cells in toxicology: In vivo and in vitro genotoxic effects of environmentally significant N-nitrosodialkylamines in cells of the liver, lung and kidney. *Environ. mol. Mutag.*, **15**, 24–35

Pour, P. & Wallcave, L. (1981) The carcinogenicity of *N*-nitrosodiethanolamine, an environmental pollutant, in Syrian hamsters. *Cancer Lett.*, **14**, 23–27

Preussmann, R., Würtele, G., Eisenbrand, G. & Spiegelhalder, B. (1978) Urinary excretion of *N*-nitrosodiethanolamine administered orally to rats. *Cancer Lett.*, **4**, 207–209

Preussmann, R., Spiegelhalder, B., Eisenbrand, G., Würtele, G. & Hofmann, I. (1981) Urinary excretion of *N*-nitrosodiethanolamine in rats following its epicutaneous and intratracheal administration and its formation in vivo following skin application of diethanolamine. *Cancer Lett.*, **13**, 227–231

Preussmann, R., Habs, M., Habs, H. & Schmähl (1982) Carcinogenicity of *N*-nitrosodiethanolamine in rats at five different dose levels. *Cancer Res.*, **42**, 5167–5171

Prival, M.J., Sheldon, A.T., Jr & Popkin, D. (1982) Evaluation, using *Salmonella typhimurium*, of the mutagenicity of seven chemicals found in cosmetics. *Food chem. Toxicol.*, **20**, 427–432

Rappe, C. & Zingmark, P.A. (1978) Formation of *N*-nitrosamines in cutting fluids. In: Walker, E.A., Castegnaro, M., Griciute, L. & Lyle, R.E., eds, *Environmental Aspects of* N-*Nitroso Compounds* (IARC Scientific Publications No. 19), Lyon, IARC*Press*, pp. 213–237

Robbiano, L., Mereto, E., Corbu, C. & Brambilla, G. (1996) DNA damage induced by seven *N*-nitroso compounds in primary cultures of human and rat kidney cells. *Mutat. Res.*, **368**, 41–47

Rollmann, B., Lombart, P., Rondolet, J. & Mercier, M. (1981) Determination of *N*-nitrosodiethanolamine in cosmetics by gas chromatography with electron capture detection. *J. Chromatogr.*, **206**, 158–163

Rounbehler, D.P. & Fajen, J.M. (1983) *N-Nitrosocompounds in the Factory Environment* (Contract No. 210-77-0100), Cincinnati, OH, National Institute for Occupational Safety and Health

Sansone, E.B., Losikoff, A.M. & Lijinsky, W. (1980) The penetration of rat skin by some nitrosamines of environmental importance. In: Walker, E.A., Griciute, L., Castegnaro, M. & Börzsönyi, M., eds, N-*Nitroso Compounds: Analysis, Formation and Occurrence* (IARC Scientific Publications No. 31), Lyon, IARC*Press*, pp. 705–716

Scherer, G., Ludeke, B., Kleihues, P., Loeppky, R.N. & Eisenbrand, G. (1991) Mutagenicity, DNA damage and DNA adduct formation by *N*-nitroso-2-hydroxyalkylamine and corresponding aldehydes. In: O'Neill, I.K., Chen, J. & Bartsch, H., eds, *Relevance to Human Cancer of* N-*Nitroso Compounds, Tobacco Smoke and Mycotoxins* (IARC Scientific Publications No. 105), Lyon, IARC*Press*, pp. 339–342

Schmeltz, I. & Wenger, A. (1979) 2-Bromo-2-nitropropane-1,3-diol as a nitrosating agent for diethanolamine: a model study. *Food Cosmet. Toxicol.*, **17**, 105–109

Spiegelhalder, B., Eisenbrand, G. & Preussmann, R. (1982) Urinary excretion of *N*-nitrosoamines in rats and humans. In: Bartsch, H., O'Neill, I.K., Castegnaro, M. & Okada, M., eds, N-*Nitroso Compounds: Occurrence and Biological Effects* (IARC Scientific Publications No. 41), Lyon, IARC*Press*, pp. 443–449

Spiegelhalder, B., Preussmann, R. & Hartung, M. (1984) Biological monitoring in the metal working industry. In: O'Neill, I.K., von Borstel, R.C., Miller, C.T., Long, J. & Bartsch, H., eds, *N-Nitroso Compounds: Occurrence, Biological Effects and Relevance to Human Cancer* (IARC Scientific Publications No. 57), Lyon, IARC*Press*, pp. 943–946

Spiegelhalder, B., Müller, J., Drasche, H. & Preussmann, R. (1987) *N*-nitrosodiethanolamine excretion in metal grinders. In: Bartsch, H., O'Neill, I.K. & Schulte-Hermann, R., eds, *The Relevance of N-Nitroso Compounds to Human Cancer: Exposures and Mechanisms* (IARC Scientific Publications No. 84), Lyon, IARC*Press*, pp. 550–552

Sterzel, W. & Eisenbrand, G. (1986) *N*-nitrosodiethanolamine is activated in the rat to an ultimate genotoxic metabolite by sulfotransferase. *J. Cancer Res. clin.Oncol.*, **111**, 20–24

Sullivan, P.A., Eisen, E.A., Woskie, S.R., Kriebel, D., Wegman, D.H., Hallock, M.F., Hammond, S.K., Tolbert, P.E., Smith, T.J. & Monson, R.R. (1998) Mortality studies of metalworking fluid exposure in the automobile industry: VI. A case–control study of esophageal cancer. *Am. J. ind. Med.*, **34**, 36–48

TCI America (1999) *Material Safety Data Sheet: Diethanolnitrosamine*, Portland, OR

Tricker, A.R. & Preusmann, R. (1991) Occurrence of and exposure to *N*-nitroso compounds in tobacco. In: O'Neill, I.K., Chen, J. & Bartsch, H., eds, *Relevance to Human Cancer of N-Nitroso Compounds, Tobacco Smoke and Mycotoxins* (IARC Scientific Publications No. 105), Lyon, IARC*Press*, pp. 493–495

Tunick, M., Veale, H.S. & Harrington, G.W. (1982) Qualitative detection of *N*-nitrosodiethanolamine in cosmetic products. *Food Cosmet. Toxicol.*, **20**, 473–474

United Nations Environment Programme (1999) Recommendations and Legal Mechanisms [http://dbserver.irptc.unep.ch:8887/iprtc/owa/lg.get_search]

Westin, J.B., Spiegelhalder, B., Preussmann, R. & Shani, J. (1990) Assay of suntan lotions for the carcinogenic, non-volatile *N*-nitrosoamine *N*-nitrosodiethanolamine. *Cancer Lett.*, **50**, 157–160

Williams, D.T., Benoit, F. & Muzika, K. (1978) The determination of *N*-nitrosodiethanolamine in cutting fluids. *Bull. environ. Contam. Toxicol.*, **20**, 206–211

Wolf, M. & Young, R. (1977) *Survey for N-Nitroso Compounds at White and Bagley Company, Worcester, Massachusetts*, Cincinnati, OH, National Institute for Occupational Safety and Health

Zingmark, P.A. & Rappe, C. (1977a) On the formation of *N*-nitrosodiethanolamine in a grinding fluid concentrate after storage. *Ambio*, **6**, 237–238

Zingmark, P.A. & Rappe, C. (1977b) On the formation of *N*-nitrosodiethanolamine from a grinding fluid under simulated gastric conditions. *Ambio*, **5**, 80–81