4,4′-METHYLENEBIS(2-CHLOROANILINE) (MOCA)

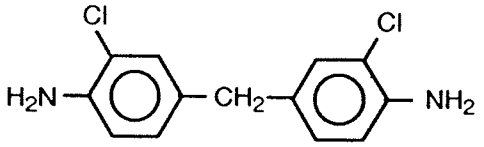
This substance was evaluated by previous working groups, in 1973 (IARC, 1974) and 1987 (IARC, 1987a). Since that time, new data have become available, and these have been incorporated into the monograph and taken into consideration in the evaluation.

1. Exposure Data

1.1 Chemical and physical data

1.1.1 *Synonyms, structural and molecular data*

Chem. Abstr. Serv. Reg. No.: 101-14-4; replaces 29371-14-0; 51065-07-7; 78642-65-6
Chem. Abstr. Name: 4,4′-Methylenebis(2-chlorobenzenamine)
IUPAC Systematic Name: 4,4′-Methylenebis(2-chloroaniline)
Synonyms: Bis(4-amino-3-chlorophenyl)methane; bis(3-chloro-4-aminophenyl)methane; 3,3′-dichloro-4,4′-diaminodiphenylmethane; MBOCA; methylenebis(3-chloro-4-aminobenzene); 4,4′-methylenebis(*ortho*-chloroaniline)



$C_{13}H_{12}Cl_2N_2$ Mol. wt: 267.16

1.1.2 *Chemical and physical properties*

From American Conference of Governmental Industrial Hygienists (1990a), unless otherwise noted

(*a*) *Description*: Light-brown (technical material) to colourless crystalline (pure compound) (Anon., 1985) solid with a faint amine-like odour

(*b*) *Melting-point*: 100–109 °C

(*c*) *Specific gravity*: 1.44 at 4 °C

(*d*) *Spectroscopy data*: Infrared, ultraviolet and nuclear magnetic resonance spectral data have been reported (Sadtler Research Laboratories, 1980; Pouchert, 1981; Sadtler Research Laboratories, 1991).

(*e*) *Solubility*: Slightly soluble in water (13.9 mg/l) (Voorman & Penner, 1986); very soluble in benzene, diethyl ether and ethanol. It is also soluble in (g/100 ml): trichloroethylene, 4.2; toluene, 7.5; ethoxyethyl acetate, 34.4; methyl ethyl ketone, 43.0; tetrahydrofuran, 55.5; dimethylformamide, 61.7; and dimethyl sulfoxide, 75.0

(*f*) *Vapour pressure*: 1.3×10^{-5} mm Hg [1.7 mPa] at 60 °C

(*g*) *Octanol/water partition coefficient (P)*: log P, 3.94 (US National Library of Medicine, 1992)

(*h*) *Conversion factor*: $mg/m^3 = 10.9 \times ppm$[1]

1.1.3 *Trade names, technical products and impurities*

Some trade names of MOCA are Bisamine S; Bisamine A, Cuamine M; Cuamine MT; Curalin M, Curalon M; Curene 442; Diamet Kh; LD 813; Millionate M; Quodorole

Commercial-grade MOCA was originally offered in flake form, for a short period in the 1960s; it is currently available as granules (prill), pellets (pastilles) and as a liquid mixture premixed with polyhydric alcohols (polyols). It is available at a purity of 99.7–99.8%, with 2-chloroaniline as a typical impurity (0.1–0.3% by weight) (PEDCo Environmental, 1984; Anon, 1985; Palmer Davis Seika, 1992; US National Library of Medicine, 1992; Ihara Chemical Industry Co., undated).

1.1.4 *Analysis*

Several methods for the analysis of MOCA in various matrices are presented in Table 1. The method of the US National Institute for Occupational Safety and Health (1990) for 4,4′-methylenedianiline can also be used for the analysis of MOCA in air.

MOCA is excreted primarily as a heat-labile glucuronide metabolite; very little is eliminated as acetylated metabolites. The latter can be hydrolysed to the parent compound by heating under acidic conditions (Linch *et al.*, 1971; Ward *et al.*, 1986; Cocker *et al.*, 1988, 1990). In order to detect unmetabolized MOCA and the glucuronide conjugate in urine samples, heat hydrolysis should be performed before analysis (Cocker *et al.*, 1990; Lowry & Clapp, 1992). MOCA detected without hydrolysis is referred to as 'free' MOCA.

1.2 Production and use

1.2.1 *Production*

The manufacture of MOCA is based on the reaction of formaldehyde (see IARC, 1982a, 1987b) and 2-chloroaniline (Anon., 1985).

MOCA was developed and marketed in the mid-1950s and was first reported to the US Tariff Commission in 1956 (US Tariff Commission, 1957). It was manufactured by two companies in the USA, both of which had ceased production by 1980. US production started at approximately 500 tonnes per year and was estimated to have reached 1500 tonnes by 1970. Since 1980, all MOCA used in the USA has been imported from Japan (Ward *et al.*, 1987) and Taiwan. There are currently approximately 100 industrial users of MOCA in the USA.

---

[1]Calculated from: mg/m^3 = (molecular weight/24.45) × ppm, assuming normal temperature (25 °C) and pressure (760 mm Hg [101.3 kPa])

Table 1. Methods for the analysis of MOCA

Sample matrix	Sample preparation	Assay procedure	Limit of detection	Reference
Air	Adsorb on silica gel; desorb with methanol	HPLC/UV	3 μg/m^3	Taylor (1977)
	Collect on glass fibre filter; extract with methanol	HPLC/ECD	100 ng/m^3	Purnell & Warwick (1981)
	Collect on acid-treated glass, extract with toluene; derivatize with heptafluorobutyric anhydride	GC/ECD	440 ng/m^3	US Occupational Safety and Health Administration (1988)
Prepolymer	Dissolve in a solution of tetrahydrofuran saturated with ammonia or of dioxane	HPLC/UV	NR	Becker *et al.* (1974)
Polymer	Extract with toluene; concentrate and react with trifluoroacetic anhydride	GC	NR	Becker *et al.* (1974)
	Extract with toluene	HPLC/UV	NR	Becker *et al.* (1974)
Soil	Filter aqueous extract; preconcentrate on reverse-phase chromatography column	HPLC/ECD	≤ 1 μg/kg	Rice & Kissinger (1981)
Urine	Adjust pH to > 12; extract with diethyl ether:hexane solution; convert to heptafluorobutyryl derivative with heptafluorobutyric anhydride	GC/ECD	1 μg/l	Eller (1985)
	Adjust pH to 9.5; extract with diethyl ether; wash, dry, concentrate and purify by TLC; extract and convert to trifluoroacetyl derivative	GC/FID	1 μg/l	Van Roosmalen *et al.* (1981)
	Clean up sample, extract with dichloromethane	HPLC/PCD	1 μg/l	Ducos *et al.* (1985); Lowry & Clapp (1992)
	Extract with acetonitrile; precolumn enrichment	HPLC/UV HPLC/ECD	20 ng 2.2 ng	Trippel-Schulte *et al.* (1986)
	Alkaline hydrolysis at 95 °C; extract with hexane	HPLC/UV	1 ng (10 μg/l)	McKerrell *et al.* (1987)
	Hydrolyse at 80 °C; extract with diethyl ether; derivatize with heptafluorobutyryl chloride	GC/MS	NR	Cocker *et al.* (1990)
	Preserve sample with citric acid; alkalize sample to pH 12; extract with diethyl ether:hexane; derivatize with pentafluoropropionic anhydride	GC/ECD	1 μg/l	US National Institute for Occupational Safety and Health (1984)
Water	Filter sample; preconcentrate on reverse-phase chromatography column	LC/ECD	≤ 1 μg/l	Rice & Kissinger (1981)

Abbreviations: HPLC/UV, high-performance liquid chromatography/ultraviolet detection; HPLC/ECD, high-performance liquid chromatography/electrochemical detection; GC/ECD, gas chromatography/electron capture detection; NR, not reported; GC/FID, gas chromatography/flame ionization detection; TLC, thin-layer chromatography; HPLC/PCD, high-performance liquid chromatography/photoconductivity detection; GC/MS, gas chromatography/mass spectrometry; LC/ECD, liquid chromatography/elctrochemical detection

Only one firm in the United Kingdom has manufactured MOCA. In 1979, about 100 tonnes of MOCA were used in about 70 factories; by 1983, only 36 industrial users of MOCA had been identified, and the number of workers exposed was about 200 (Locke, 1986). MOCA is no longer manufactured in the United Kingdom.

In 1972, world production was about 3300 tonnes (Will *et al.*, 1981). Currently, there are three major producers in the world: two in Japan and one in Taiwan. In the early 1980s, Japan produced approximately 2000 tonnes of MOCA per year; in the mid-1980s, the level was 3000 tonnes; and by the early 1990s, the level was approximately 3600 tonnes. Japan exports 40–60% of its production, approximately 900–1000 tonnes going to the USA, 450 to Europe and 450 to Southeast Asia, Canada, Brazil, South Africa and Australia. Taiwan currently produces 1000 tonnes per year, with 450 tonnes exported to Japan and 45–90 tonnes to the USA. Small quantities may also be produced in the Republic of Korea and France (Chemical Information Services, 1991).

1.2.2 *Use*

MOCA is used principally as a curing agent for polyurethane prepolymers in the manufacturing of high-performance, specialized, castable urethane rubber products (see IARC, 1982b, 1987c). Mouldings such as industrial tyres and rollers, shock absorption pads and conveyor belting are among the wide variety of uses (Anon., 1985; American Conference of Governmental Industrial Hygienists, 1990a). A use of MOCA that is specific to Japan and the Far East is as a curing agent in roofing and wood sealing.

1.3 Occurrence

1.3.1 *Natural occurrence*

MOCA is not known to occur as a natural product.

1.3.2 *Occupational exposure*

Occupational exposure to MOCA may occur through inhalation, ingestion and skin absorption: because of its low vapour, it can be inhaled as a dust when processed in a dry form; poor personal hygiene can allow contamination of the hands and ingestion while eating or smoking; however, the most likely route of exposure is thought to be by skin absorption after contact with contaminated surfaces (Linch *et al.*, 1971; Clapp *et al.*, 1991; Lowry & Clapp, 1992). Evaluations of sources of exposure have included sampling of air, work surfaces (e.g., wipe) and urine of potentially exposed workers.

Urinary levels as high as 25 000 μg/l were reported when MOCA was first produced in a full-scale commercial manufacturing plant in the USA in 1962; however, the analytical methods used were non-specific. By 1970, more specific analytical methods had been developed that involved acid hydrolysis of initial urine samples, and better work practices and engineering controls had been implemented. The average urinary level in four workers involved in pelletizing and packaging MOCA at the same plant was found by gas chromatography to be 695 μg/l (range, 70–1500 μg/l); personal air levels were generally below the detection limit of 10 $\mu g/m^3$, and the workers wore respirators. Average urinary

concentrations in workers in MOCA production were 620 μg/l in 1969 and 250 μg/l in 1970, when local ventilation, fresh daily clothing, wash-down of the operating area twice daily and use of butyl rubber gloves were introduced. The authors concluded that inhalation was not the primary route of exposure (Linch *et al.*, 1971).

The extent of exposure to MOCA was determined in 19 factories in France during the early 1980s (Ducos *et al.*, 1985). In more than 340 analyses of urine from 150 workers, MOCA was present at below the detection limit of 0.5 μg/l up to levels of 1600 μg/l. The levels of 'free' MOCA, determined by high-performance liquid chromatography (HPLC) in urine samples in specific factories are presented in Table 2, which shows dramatic decreases in urinary levels after modifications in the handling of MOCA were instituted.

Table 2. Occupational exposure to MOCA in France

Process	Year surveyed	Before or after improvements	No. of samples/ workers	Urinary concentration (μg/l)	
				Mean	Range
Production of crystal MOCA (1 plant)	1982	Before	12 workers	600	
	1983	After	53 samples	46	20–62
Blending of solid MOCA with polyol (1 plant)	1982	Before	4 workers	318	75–940
	1983	After	3 workers	5	1–9
Production of urethane elastomers from solid pellets of MOCA or in solution (2 plants)	1982	Before	22 workers	100	43–156
	1984	After	25 workers	26	18–34

From Ducos *et al.* (1985)

In a plastics manufacturing and processing plant in Germany, 49 urine samples collected randomly from workers were found (using reversed-phase HPLC with ultraviolet detection) to contain between 15 (limit of detection) and 100 μg/l of 'free' MOCA. Concentrations in work place air were not determined (Will *et al.*, 1981).

In the United Kingdom, urine samples were taken from workers in a polyurethane plastics manufacturing company every month during 1978–82 (Thomas & Wilson, 1984). The process involved melting of pelletized MOCA before blending it with liquid polymer. After improvements designed to prevent exposures, urinary concentrations of 50 nmol/mmol creatinine [about 1300 μg/l] of 'free' MOCA were gradually reduced to less than 5 nmol/mmol creatinine [130 μg/l] by 1982.

In Japan, airborne levels and urinary concentrations of MOCA were determined (by HPLC with electrochemical detection) for five workers making polyurethane elastomer products (Ichikawa *et al.*, 1990). Average airborne concentrations of 0.2–0.5 μg/m^3 were measured over one week for four workers who transferred dry MOCA into a mixing vessel and processed elastomer tubes; a worker who poured hot elastomer mix was exposed to an average air concentration of 8.9 μg/m^3. The mean urinary concentrations at the end of each working day were 2.4–64.0 μg/g creatinine [about 4–120 μg/l] for the four workers and 96.6 μg/g creatinine [200 μg/l] for the worker who poured the mix [not specified whether

'free' or hydrolysed]. There was no significant difference between preshift and postshift urinary concentrations. It was estimated that only 0.5–5% of the total exposure was due to inhalation.

Monitoring of 'free' MOCA concentrations in urine, using non-specific thin-layer chromatography, was performed on a voluntary basis by several companies in the USA after 1978 (Lowry & Clapp, 1992); in 1984, the analysis was made more specific by introducing HPLC. Between 1980 and 1983, 3323 urinary samples were analysed from 54 companies: the MOCA levels exceeded 50 μg/l in 16.9% of all samples and exceeded 100 μg/l in 9.2% (Ward *et al.*, 1987). In 1985, the urinary concentration exceeded 50 μg/l in 12% of all samples tested; in 1990, 8% of all samples exceeded that level (Lowry & Clapp, 1992).

In Western Australia, urinary levels of 'free' MOCA were determined by gas chromatography in workers in five of seven companies where MOCA was used in the manufacture of polyurethane polymers from 1984 onwards (Wan *et al.*, 1989). The initial levels were at a geometric mean of 30 μg/l; after a training programme on the safe use of MOCA, the level was reduced to 10 μg/l.

Urinary levels of MOCA were reported at a plant in the USA which made cast polyurethane products (Clapp *et al.*, 1991). Of 77 samples collected and analysed by HPLC, five contained more than 50 μg/l; all were from workers who mixed pelletized MOCA (Table 3). The level of MOCA in 40 samples of personal air was below the limit of detection (~ 0.02 μg/m^3) in 88%; in the remainder, the level was less than 1 μg/m^3.

Table 3. Distribution of urinary levels of MOCA in workers in a cast polyurethane products plant in the USA

Job	No. of workers	No. of samples with mean concentration of MOCA (μg/l urine)[a]					
		ND	< 5	5–50	50–100	> 100	Total
Mixer	2	0	0	5	3	2	10
Moulder	16	10	3	22	0	0	35
Clean-up man	1	1	0	3	0	0	4
Trimmer	3	4	0	0	0	0	4
Supervisor	2	5	0	0	0	0	5
Office employee	7	14	0	0	0	0	14

From Clapp *et al.* (1991); ND, not detected

[a]Normalized to specific gravity of 1.019; limit of detection, 5 μg/l

In an accidental exposure in the USA, a 30-year old male polyurethane moulder accidentally sprayed molten MOCA (approximately 3 gallons [11.4 l]) over his upper body and extremities, where it remained for several seconds. He was wearing trousers, a shirt with rolled-up sleeves, asbestos gloves, safety glasses and a respirator. He did not ingest any MOCA, and the duration of exposure was limited by removing his clothing, showering and washing gently within approximately 45 min of the initial exposure. Analysis of his urine 4 h after the exposure showed a peak concentration of 1700 μg/l; the urinary level remained above 100 μg/l for four days (Osorio *et al.*, 1990). In a separate accident, a worker was

sprayed in the face with hot liquid MOCA, some of which entered his mouth. His eyes and face were washed immediately. The urinary concentration was 3600 μg/l (1400 μg/g creatinine) 5 h after exposure (Hosein & Van Roosmalen, 1978).

A survey of the concentrations of MOCA on work surfaces during various operations in 39 plants representing 10–20% of all the polyurethane plants using MOCA in the USA is summarized in Table 4 (PEDCo Environmental, 1984).

Table 4. Surface contaminations during some processes involving MOCA in the USA

Process	No. of facilities	No. of samples	Mean surface concentration (μg/100 cm^2)
Storage and manual transfer of solid MOCA to melting operations	18	37	847
Manual transfer of molten MOCA to mixing operation and mixing	8	9	1 650
Storage and manual transfer of liquid MOCA to mixing with other compounds	8	25	30
Manual mixing of liquid MOCA with other compounds	3	4	15 000

From PEDCo Environmental (1984)

1.3.3 *Water, sediments and soil*

Extensive environmental contamination with MOCA on several hundred hectares of land surrounding a MOCA plant was found in 1979 in Adrian, MI, USA. Levels up to several milligrams per kilogram were found in gardens and community recreation areas. MOCA was also found in the urine of factory workers and of young children living in the contaminated area (Keeslar, 1986). The concentrations in sediment samples collected from the lagoon used by the plant ranged from 1600 to 3800 ppm (mg/kg dry weight). Effluent water from the lagoon had a concentration of 250 ppb (μg/l), deep well-water from under the plant had a concentration of 1.5 ppb, and surface run-off water had a concentration of 1 ppb. Activated sludge from the sewage-treatment plant contained an estimated 18 ppm (mg/kg). MOCA was not detected in sewage-treatment plant influent or effluent water (detection limit, 0.5 ppb (μg/l)) or in the water of a river located near the plant (detection limit, 0.1 ppb) (Parris *et al.*, 1980; Verschueren, 1983; Fishbein, 1984).

MOCA is rapidly bound to the soil matrix and probably exists largely as covalent adducts. Some MOCA was metabolized by oxidation of the methylene bridge to a benzophenone derivative, 4,4′-diamino-3,3′-dichlorobenzophenone, presumably by microbial activity (Voorman & Penner, 1986).

1.4 Regulations and guidelines

Occupational exposure limits and guidelines for MOCA in some countries are presented in Table 5.

Table 5. Occupational exposure limits and guidelines for MOCA

Country	Year	Concentration (mg/m^3)	Interpretation	Classification as carcinogen
Australia		0.22 (s)	TWA	
Belgium		0.22 (s)	TWA	
Denmark				Yes
Finland		0.2 (s)	TWA	
		0.6 (s)	STEL	
France		0.22	TWA	
Germany				Yes
Italy	1978	0.22 (s)	TWA	
Mexico	1983	0.22 (s)	TWA	
Netherlands	1989	0.22 (s)	TWA	
Sweden				Yes
Switzerland	1992	0.02 (s)	TWA	
United Kingdom		0.005 (s)	TWA (MEL)	
USA				
ACGIH	1992	0.11 (s)[a]	TWA (TLV)	
NIOSH	1990	0.003	TWA (REL)	
OSHA	1990	0.22 (s)	TWA (PEL)	
Venezuela	1978	0.22 (s)	TWA	

From National Swedish Board of Occupational Safety and Health (1984); Health and Safety Executive (1985); Cook (1987); American Conference of Governmental Industrial Hygienists (ACGIH) (1990b, 1992); ILO (1991); Caisse Nationale Assurance (1992)

Abbreviations: TWA, time-weighted average; STEL, short-term exposure limit; MEL, maximum exposure level; TLV, threshold limit value; NIOSH, National Institute of Occupational Safety and Health; OSHA, Occupational Safety and Health Administration; REL, recommended exposure level; PEL, permissible exposure level; (s), skin notation

[a]Intended change to 0.11 mg/m^3 for 1992–93, with notation A_2, suspected human carcinogen (American Conference of Governmental Industrial Hygienists, 1992)

In the European Economic Community, MOCA is classified as a category 2 carcinogen, substances that should be regarded as if they were carcinogenic to man, and is labelled as R45 (may cause cancer) (Commission of the European Communities, 1967, 1991). Since MOCA may cause cancer, the Council Directive on protection of workers from risks related to exposure to carcinogens at work applies (Commission of the European Communities, 1990), which requires employers to replace carcinogenic agents or to take measures to prevent or reduce exposure.

In Germany, MOCA is classified with A_2 compounds, which are considered to have been proven to be carcinogenic only in animal experimentation but under conditions comparable to those of possible human exposure at the workplace (Deutsche Forschungsgemeinschaft, 1992). MOCA has been classified as a carcinogen in Denmark since 1976 (Arbejdstilsynet, 1977) and in the Netherlands since 1989.

The recommended maximum exposure level for MOCA in the United Kingdom was 200 μg/m^3 over 8 h, until 1979, when a control limit of 50 μg/m^3 was recommended. In 1984, after consideration of studies on MOCA, the control limit was lowered to 5 μg/m^3 (Locke, 1986). In Switzerland, the limit value for occupational exposure to MOCA was recently lowered from 0.22 mg/m^3 to 0.02 mg/m^3 (Caisse Nationale Assurance, 1992).

In the USA, the only biological exposure limit for MOCA is that of the State of California, of 100 μg/l in urine (State of California, 1992). The American Conference of Governmental Industrial Hygienists (1992) announced an intended change of the present time-weighted threshold limit value of 0.22 mg/m^3 to 0.11 mg/m^3 in 1992–93.

2. Studies of Cancer in Humans

2.1 Descriptive studies

Up to 1971, a total of 209 employees had had potential contact with MOCA in the Chambers Works, New Jersey, USA, where manufacture began in 1954 (Linch *et al.*, 1971). No case of cancer of the bladder was mentioned in the medical records of the company.

In a review, Cartwright (1983) reported that a cohort study was under way in a plant where MOCA was manufactured and where 13 new cases of bladder cancer had occurred within a period of a few years. The number was stated as being far larger than that which would be expected.

A study was undertaken in a small plant in Michigan, USA, where MOCA had been produced between 1968 and 1979. All 532 workers employed in 1968–79 and an additional 20 workers first employed in 1980 and 1981 who had had possible exposure owing to contamination of the plant site were included (Ward *et al.*, 1988, 1990). The median duration of employment was 3.2 months. The workers may have been heavily exposed, since urinary levels of MOCA several months after production had ceased were reported to have ranged up to 50 000 μg/l. The workers had not been exposed to benzidine or β-naphthylamine. Of the 552 predominantly white (89.5%) workers, 452 participated in a telephone interview in 1981 and 385 participated in a urine screening examination. Three asymptomatic bladder tumours were identified. After a 28-year-old worker was diagnosed with a noninvasive papillary transitional-cell tumour, the screening procedure was supplemented for some workers with cystoscopy. A second worker, aged 29 years, was diagnosed with a papillary bladder neoplasm, and, in a subsequent round of screening, a third, 44-year-old worker was diagnosed with a papillary transitional-cell carcinoma of the bladder. The expected number of bladder tumours could not be calculated, as no valid comparison rates of asymptomatic bladder tumours were available.

3. Studies of Cancer in Experimental Animals

3.1 Oral administration

3.1.1 *Mouse*

Groups of 25 male and 25 female HaM/ICR mice, six to eight weeks old, were fed diets containing 0, 1000 or 2000 mg/kg of diet (ppm) MOCA as the hydrochloride (97% pure) for

18 months. Surviving animals were killed 24 months after the start of the study; about 55% of the control and treated mice were still alive at 20–22 months. The effective numbers of animals at the end of the study were: males—control, 18; low-dose, 13; high-dose, 20; females—control, 20; low-dose, 21; high-dose, 14. Haemangiomas or haemangiosarcomas (mainly subcutaneous) combined occurred in 0/18 control, 3/13 low-dose and 8/20 high-dose male mice. 'Hepatomas' occurred in 0/20 control, 9/21 low-dose and 7/14 high-dose female mice ($p < 0.01$, Fisher exact test). The incidence of lymphosarcomas and reticulum-cell sarcomas was decreased in treated females. The authors stated that the incidence of vascular tumours in the high-dose animals was comparable to that in historical controls of the same strain (Russfield *et al.*, 1975).

3.1.2 *Rat*

Groups of 25 male and 25 female Wistar rats, 100 days [14 weeks] of age, were fed 0 or 1000 mg/kg of diet (ppm) MOCA [purity unspecified] in a protein-deficient diet [not otherwise specified] for 500 days [71 weeks] [total dose, 27 g/kg bw], followed by an observation period on protein-deficient diet. Animals were killed when moribund; mean survival of treated males and females was 565 days [81 weeks] and 535 days [76 weeks], respectively, and mean survival of male and female controls on a similar diet was 730 days [104 weeks]. Of the 25 treated males, 23 died with tumours; 'hepatomas' occurred in 22/25 [$p < 0.001$, Fisher exact test], and lung tumours (mainly carcinomas) in 8/25 [$p = 0.002$, Fisher exact test]. Among the treated females, 20 rats died with tumours; 'hepatomas' occurred in 18/25 [$p < 0.001$ Fisher exact test], and lung tumours were observed in 5/25 [$p = 0.025$, Fisher exact test]. No 'hepatoma' or lung tumour was observed among control animals (Grundmann & Steinhoff, 1970).

Groups of 25 male Charles River CD-1 rats, six to eight weeks old, were administered diets containing 0, 500 or 1000 mg/kg of diet (ppm) MOCA as the hydrochloride (97% pure) for 18 months. All surviving animals were killed 24 months after the start of the study; about 55% of the control and treated animals were still alive at 20–22 months. The effective numbers were: 22 control, 22 low-dose and 19 high-dose animals. 'Hepatomas' occurred in 0/22 control, 1/22 low-dose and 4/19 high-dose rats [$p < 0.05$, Cochran-Armitage trend test] (Russfield *et al.*, 1975). [The Working Group noted the small number of animals used in the study.]

Groups of 50 males and 50 female Charles River CD rats, 36 days [5 weeks] of age were administered 0 (control) or 1000 mg/kg of diet (ppm) MOCA ($\sim$ 95% pure) in a standard diet (23% protein) for life. The average duration of the experiment was 560 days [80 weeks] for treated males, 548 days [78 weeks] for treated females, 564 days [80 weeks] for male controls and 628 days [89 weeks] for female controls. Six animals from each group were sacrificed at one year for interim evaluation. Lung adenocarcinomas occurred in 21/44 ($p < 0.05$, χ^2 test) treated males and 27/44 ($p < 0.05$, χ^2 test) treated females. An additional squamous-cell carcinoma of the lung was observed in one treated male and one treated female. No lung tumour was observed among control animals. Lung adenomatosis, considered to be a preneoplastic lesion, developed in 14/44 treated males and 11/44 treated females and in 1/44 male controls and 1/44 female controls ($p < 0.05$). Pleural mesotheliomas occurred in 4/44 treated males and 2/44 treated females; no such tumour was observed among controls.

Hepatocellular adenomas and hepatocellular carcinomas occurred in 3/44 and 3/44 treated males and in 2/44 and 3/44 treated females, respectively, but not in controls. Ingestion of MOCA resulted in a lower incidence of pituitary tumours in treated females than in controls (1/44 *versus* 12/44) (Stula *et al.*, 1975).

In the same study, another 25 males and 25 females were administered 0 (control) or 1000 ppm MOCA (~ 95% pure) in a low-protein diet (7%) for 16 months. Six animals from each group were sacrificed at one year for interim evaluation. The average duration of the experiment was 400 days [57 weeks] for treated males, 423 days [60 weeks] for treated females, 384 days [55 weeks] for control males and 466 days [66 weeks] for control females. Lung adenocarcinomas occurred in 5/21 treated males ($p < 0.05$, χ^2 test) and 6/21 females ($p < 0.05$, χ^2 test); no such tumour developed in 21 untreated male or female controls. Hepatocellular adenomas occurred in 5/21 treated males ($p < 0.05$, χ^2 test) and 2/21 treated females; hepatocellular carcinomas were observed in 11/21 treated males ($p < 0.05$, χ^2 test) and 1/21 treated females; no hepatocellular tumour was observed among 21 untreated males or females. Fibroadenomas of the mammary gland occurred in 1/21 treated and 7/21 control female rats ($p < 0.05$). Mammary gland adenocarcinomas developed in 6/21 treated female rats and in 0/21 untreated females ($p < 0.05$, χ^2 test) (Stula *et al.*, 1975).

Groups of 100, 100, 75 and 50 male Charles River CD rats, 35 days [5 weeks] of age, were fed either a 'protein-adequate' (27%) diet containing 0, 250, 500 or 1000 mg/kg of diet (ppm) MOCA (industrial grade [purity unspecified]) or a 'protein-deficient' (8%) diet containing 0, 125, 250 and 500 ppm MOCA for 18 months followed by a 32-week observation period. Animals were sacrificed at 104 weeks. Administration of MOCA was associated with decreased survival in both groups: mean survival time (weeks) was: 'protein-adequate' diet: control, 89; low-dose, 87; mid-dose, 80 ($p < 0.01$); high-dose, 65 ($p < 0.001$); 'protein-deficient' diet: control, 87; low-dose, 81; mid-dose, 79; high-dose, 77 ($p < 0.05$). The numbers of rats on the 'protein-adequate' diet still alive at week 104 were: control, 20/100; low-dose, 14/100; mid-dose, 10/75; and high-dose, 0/50 (at 84 weeks, there were six surviving rats). The numbers of animals on the 'protein-deficient' diet still alive at week 104 were: control, 34/100; low-dose, 22/100; mid-dose, 14/75; and high-dose, 5/50. MOCA induced several tumour types in both groups; the incidences of the predominant tumours are shown in Table 6. Dose-related increases in the incidences of lung tumours, mammary adenocarcinomas, Zymbal gland carcinomas and hepatocellular carcinomas were observed in both experiments. The highest tumour incidence was observed in the lung. An increased incidence of haemangiosarcomas was observed only in the group on the 'protein-deficient' diet. In groups given 500 ppm MOCA, tumour incidence was generally lower in those fed 'protein-deficient' diet, but hepatocellular carcinomas and Zymbal gland carcinomas occurred at a higher incidence in this group (18 and 12%) than in the 'protein-adequate' group (4 and 7%). The incidence of pituitary adenomas decreased with increasing concentration of MOCA in the 'protein-adequate' diet, perhaps because of decreased survival in the treated groups (Kommineni *et al.*, 1978).

Table 6. Percentages of male rats with tumours at specific sites after feeding of MOCA in diets with different protein contents

Dietary protein	MOCA (ppm)	No. of rats autopsied	Lung adenocarcinomas	All lung tumours	Mammary adenocarcinomas	Zymbal gland carcinomas	Hepatocellular carcinomas	Haemangiosarcomas	Pituitary adenomas[a]
Adequate (27%)	0	100	0	1	1	1	0	2	42
	250	100	14***	23***	5	8*	3	4	36
	500	75	27***	37***	11**	7	4	4	25*
	1000	50	62***	70***	28***	22***	36***	0	4***
Deficient (8%)	0	100	0	0	0	0	0	1	23
	125	100	3	6**	1	0	0	2	16
	250	75	9**	15***	4	5*	0	5	12*
	500	50	16***	26***	6*	12***	18***	8*	20

From Kommineni *et al.* (1978); *, $p < 0.05$; **, $p < 0.01$; ***, $p < 0.001$

[a]Includes pituitary adenocarcinomas (0–2 per group)

3.1.3 *Dog*

A group of six female beagle dogs, approximately one year old, were administered a daily dose of 100 mg MOCA (~ 90%, ~ 10% polyamines with a three-ring structure and ~ 0.9% *ortho*-chloroaniline) by capsule on three days a week for six weeks, then on five days a week for up to nine years. A further group of six females served as untreated controls. One treated dog died early, at 3.4 years of age, because of intercurrent infection; the other animals were killed between 8.3 and nine years. Transitional-cell carcinomas of the urinary bladder occurred in four of five treated dogs, and a composite tumour (transitional-cell carcinoma/adenocarcinoma) of the urethra developed in one dog. No such tumour was observed among six untreated controls ($p < 0.025$, Fisher exact test) (Stula *et al.*, 1977).

3.2 Subcutaneous administration

Rat

In a study reported as a short communication, groups of 17 male and 17 female Wistar rats [age unspecified] were injected subcutaneously with 500 or 1000 mg/kg bw MOCA (94% pure) as a suspension in saline either once a week or at longer time intervals for 620 days [88 weeks] (total dose, 25 g/kg bw). The rats were fed a laboratory diet with a normal protein content. The mean observation period was 778 days [111 weeks]. A total of 22 animals developed 29 malignant tumours. Hepatocellular carcinomas occurred in 9/34 [$p < 0.0042$, Fisher exact test], and malignant lung tumours (six adenocarcinomas, one carcinoma) were observed in 7/34 [$p < 0.016$, Fisher exact test]. A malignant subcutaneous tumour [unspecified] was found in one rat [sex unspecified]. Among 25 male and 25 female untreated controls (mean observation period, 1040 days [148 weeks]), a total of 13 malignant tumours, including one lung tumour, developed; no hepatocellular carcinoma was observed (Steinhoff & Grundmann, 1971). [The Working Group noted the inadequate reporting of the experiment.]

4. Other Relevant Data

4.1 Absorption, distribution, metabolism and excretion

4.1.1 *Humans*

Biological monitoring of workers exposed to MOCA in factories where polyurethane plastics were manufactured showed levels of MOCA in urine that ranged from 1 to 1000 nmol/mmol creatinine (0.1–110 μM; 27–27 000 μg/l), with averages ranging from 5 to 50 nmol MOCA/mmol creatinine at different sampling periods (Thomas & Wilson, 1984; Cocker *et al.*, 1988; Edwards & Priestly, 1992). Exposure appeared to occur *via* both inhalation and skin absorption (Linch *et al.*, 1971; Ward *et al.*, 1986) (see also section 1.3.2). Assuming that MOCA levels in urine represent about 1% of the total absorbed and that spot urine sampling is indicative of 24-h urine collection (1.3 l urine per day), the calculated internal dose corresponding to 100 μg/l urine is 13 mg per day (Ward *et al.*, 1986); the internal

doses of workers therefore range from 3.5 to 3500 mg per day [calculated by the Working Group from the data given above]. Studies on percutaneous absorption of MOCA through cultured neonatal foreskin showed rapid, time-dependent absorption (Chin *et al.*, 1983).

In the individual who was sprayed accidentally with molten MOCA (see p. 276), pharmacokinetic analyses (one-compartment model) indicated a biological half-time of 23 h, with 94% elimination from the body in four days; 35% of the parent MOCA present in the urine was excreted as conjugates (Osorio *et al.*, 1990). High levels of MOCA (3.6 mg/l [13 μM]) were also found in the urine of another worker accidentally sprayed with MOCA (Hosein & Van Roosmalen, 1978; see pp. 276–277) 4 and 11 h after exposure. By 17 and 20 h, the levels had decreased to 0.03–0.06 mg/l [0.1–0.2 μM].

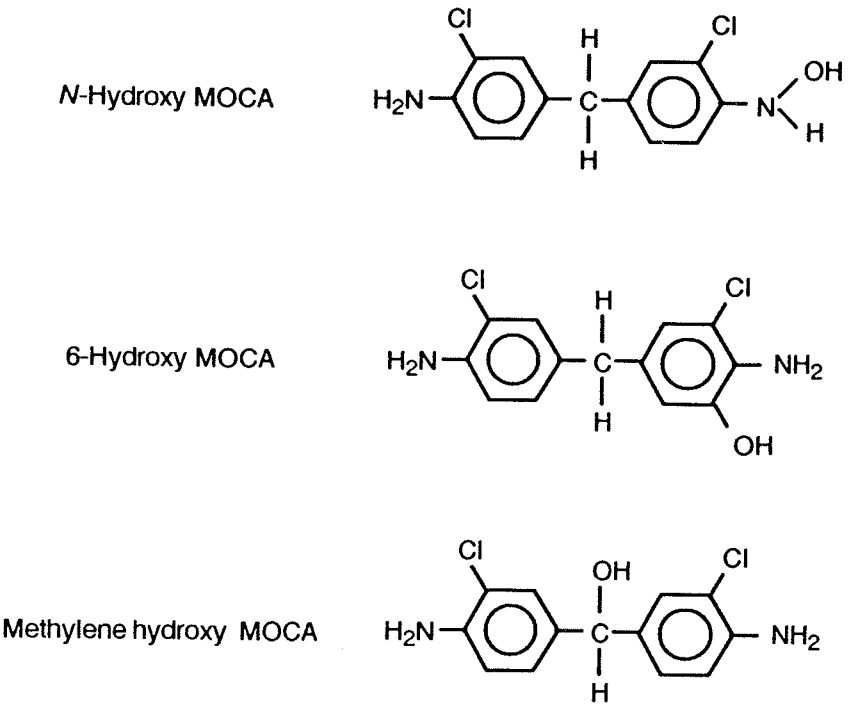
Urinary metabolites of MOCA detected in humans (Cocker *et al.*, 1990) include its *N*-acetyl derivative (1–9% of urinary MOCA in 10/23 individuals) (Cocker *et al.*, 1988) and its *N*-glucuronide (levels two- to three-fold higher than those of MOCA) (Cocker *et al.*, 1990). Urinary thioethers were not detected (Edwards & Priestly, 1992).

4.1.2 *Experimental systems*

Oxidative metabolism of [methylene-^{14}C]- and [aniline-^{14}C]MOCA (58 and 10.9 mCi/mmol, respectively [radiochemical purity unspecified]), by human liver microsomes has been demonstrated *in vitro*, resulting in the formation of its *N*-hydroxy(4-amino-4′-hydroxylamino-3,3′-dichlorodiphenylmethane), 6-hydroxy(5-hydroxy-4,4′-diamino-3,3′-dichlorodiphenyl methane) and [methylene or C$^{4,4'}$]hydroxy(4,4′-diamino-3,3′-dichlorobenzhydrol) derivatives (see Fig. 1) (Morton *et al.*, 1988). In a survey of liver microsomes from 22 individuals, the rate of *N*-oxidation of [methylene-^{14}C]MOCA (57 mCi/mmol; radiochemical purity, > 95%) varied by eight fold; *N*-hydroxy-MOCA was always the major metabolite, accounting for 81–94% of all the oxidation products (Butler *et al.*, 1989). Using antibodies and other inhibitors, substrate–activity correlations and purified or recombinant enzymes, cytochrome P450 3A4 was identified as the major enzyme that catalyses MOCA *N*-oxidation in human liver; a minor role for cytochrome P450 2A6 was shown (Yun *et al.*, 1992). It has been suggested that human and dog urinary bladder explant cultures metabolize MOCA, on the basis of the apparent covalent binding of ^{3}H-MOCA (30 Ci/mmol; radiochemical purity, 97%) to DNA (Shivapurkar *et al.*, 1987).

In male beagle-type mongrel dogs, ^{14}C-MOCA (58 mCi/mmol [radiochemical purity unspecified]) was applied to 25 cm^2 of shaved skin in 0.5 ml acetone or was injected intravenously in 0.5 ml propylene glycol at total doses of 10 mg per dog. By 24 h, urine collected through surgically implanted catheters contained 1.3% of the administered percutaneous dose (0.4% of which was unchanged MOCA) and 45% of the intravenous dose (0.54% of which was unchanged MOCA). Following intravenous injection, the time-course of disappearance of MOCA from the blood was rapid, with an apparent volume of distribution of 244 l and biphasic half-times of 0.09 and 0.70 h. After skin application, no radiolabel was measured in blood up to 24 h later. Biliary excretion was 0.62% of the dose after percutaneous administration and 32% after intravenous injection; none was unchanged MOCA. Tissue distribution was 10–20 times greater after intravenous dosing and was highest in liver, kidney, fat and lung tissues (Manis *et al.*, 1984). As with other carcinogenic aromatic amines, the major urinary metabolite of MOCA in dogs, accounting for 75% of the

Fig. 1. Oxidative metabolism of MOCA by liver microsomes



From Chen *et al.* (1989)

urinary radiolabel, was the sulfate conjugate of the *ortho*-hydroxy metabolite, 6-hydroxy-MOCA (5-hydroxy-3,3′-dichloro-4,4′-diaminodiphenyl methane-5-sulfate) (Manis & Braselton, 1984). The same major metabolite was detected in dog liver and kidney slices incubated with ^{14}C-MOCA [58 mCi/mmol, methylene-labelled; 10.9 mCi/mmol, aniline-labelled; radiochemical purity, > 99%]. An unknown glucoside and three glucuronide metabolites were also observed; and apparent covalent binding to DNA was measured, which was greater in liver than kidney (Manis & Braselton, 1986).

In female LAC:Porton rats, [methylene-^{14}C]MOCA (8.3 mCi/mmol [radiochemical purity unspecified]) was mixed with unlabelled MOCA in solutions of glycerol formol–arachis oil and given by intraperitoneal (1, 13 or 100 mg/kg bw) or oral administration (10 mg/kg bw). The urine contained 23–41% of the radiolabel after 48 h, of which 1–2% was unchanged MOCA, while faeces contained 60–69% of the administered dose. At least nine metabolites were observed in the urine. Tissue distribution of radiolabel after 48 h was highest in the liver, then fat > kidney, small intestine (Farmer *et al.*, 1981).

^{14}C-MOCA (58 mCi/mmol; radiochemical purity, > 98%) was injected intravenously to female Sprague-Dawley rats at a dose of 0.5 mg/kg bw in ethanol:Tween 80:water. After 48 h, 21 and 73% of the dose was excreted in urine and faeces, respectively. The levels of radiolabel were highest in liver, then in lung, kidney, fat and adrenal gland (Tobes *et al.*, 1983).

Male Sprague-Dawley rats given [methylene-^{14}C]MOCA (58 mCi/mmol; radiochemical purity, 96%) by gavage (11–12 mg/kg bw in propylene glycol) excreted 16.5% of the dose in

the urine (0.25% as unchanged MOCA) and 70% in faeces after 72 h. Up to 2.54% was excreted in urine (0.008% as unchanged MOCA) and 2.11% in faeces after application of 2.5 mg MOCA in acetone to shaved skin (Groth *et al.*, 1984). In 30-day old male Charles River CD rats, administration of [methylene-^{14}C]MOCA (4–7 mCi/mmol; radiochemical purity, ~ 93%) by gavage in dimethyl sulfoxide:water (60:40) at a dose of 5.5–5.6 mg per rat resulted in 16–27% of the label being excreted into urine ($\leq$ 0.2% as unchanged MOCA) and 32–50% in faeces after 24 h. The major biliary metabolite was identified as MOCA *N*-glucuronide (Morton *et al.*, 1988). Male Wistar rats given unlabelled recrystallized MOCA at a dose of 125 or 250 mg/kg intraperitoneally in peanut oil daily for five days excreted 'free' MOCA at a level of 1–6.5 μmol/mmol creatinine in urine 24 h after the last dose (Edwards & Priestly, 1992).

The half-times of MOCA in whole blood, lymphocytes, urinary bladder and liver of male Sprague-Dawley rats ranged from 4 to 17 days after a single oral dose (281 μmol [75 mg/kg bw]) of [methylene-^{14}C]MOCA (42.4 μCi/ml; radiochemical purity, > 99.5%) in corn oil. The order of covalent binding to DNA was liver > bladder > lymphocytes. Similar results were obtained after dermal application, except that adduct formation was approximately 100-fold less (Cheever *et al.*, 1988, 1990). Multiple oral doses of 7.5 mg/kg bw for up to 28 days induced a linear increase in globin binding and half-times that were comparable to those seen after a single oral dose of 75 mg/kg; tissue levels of MOCA were highest in the liver, kidney and lung. Induction of cytochromes P450 by phenobarbital resulted in a three-fold increase in globin binding but a slight decrease in binding to liver. Intraperitoneal treatment resulted in three-fold higher binding levels in liver, globin and whole blood (Cheever *et al.*, 1991). In a similar study with male Sprague-Dawley rats and English guinea-pigs, intraperitoneal injection of ^{14}C-MOCA (58 mCi/mmol [radiochemical purity unspecified]) in propylene glycol:dimethyl sulfoxide:saline (4:4:2) at 0.5–50 mg/kg to rats and subcutaneous injection of 5–500 mg/kg to rats and 4–100 mg/kg to guinea-pigs resulted in a nearly linear, dose-related increase in haemoglobin binding. β-Naphthoflavone but not phenobarbital pretreatment of rats was found to increase MOCA–haemoglobin adduct formation *in vivo* (Chen *et al.*, 1991). In female Wistar rats dosed orally with 1, 3.8, 4.3, 66 and 134 mg/kg [aniline-^{14}C]MOCA (58 mCi/mmol; radiochemical purity, > 95%) or unlabelled MOCA in ethanol:propylene glycol (1:4), MOCA was bound in decreasing amounts to DNA, RNA and protein of lung, liver and kidney; 0.19% of the dose was bound to haemoglobin and 0.026% to serum albumin after 24 h. Alkaline hydrolysis of MOCA-bound haemoglobin released free MOCA (Sabbioni & Neumann, 1990), indicating the presence of a sulfinamide adduct derived from *N*-hydroxy-MOCA in the circulation and its oxidative conversion to a nitroso derivative in the erythrocytes. This assumption was confirmed by reactions of haemoglobin with either *N*-hydroxy-MOCA or its nitroso derivative *in vitro* and by the observation of high levels of haemoglobin binding after intravenous administration of *N*-hydroxy-MOCA (Chen *et al.*, 1991).

Oxidative metabolism of [methylene-^{14}C]- and [aniline-^{14}C]MOCA (58 and 10.9 mCi/mmol; radiochemical purity, > 99% (Chen *et al.*, 1989) [or radiochemical purity unspecified (Morton *et al.*, 1988)]) by rat, dog and guinea-pig liver microsomes has been demonstrated *in vitro*, resulting in formation of the *N*-hydroxy and 6-hydroxy derivatives. In rat and guinea-pig liver microsomes, *N*-hydroxy-MOCA appeared to be the major metabolite, whereas 6-

hydroxy-MOCA was predominant in dog liver microsomes. In rats, a methylene-hydroxy derivative was also found.

Using inducers, specific inhibitors and purified enzymes, cytochromes P450 2B1 ($P450_{PB-B}$) and P450 2B2 ($P450_{PB-D}$) were identified as the major enzymes that catalyse MOCA *N*-oxidation in rat liver; a minor role for cytochromes P450 1A2 ($P450_{ISF-G}$), P450 2C11 ($P450_{UT-A}$) and P450 1A1 ($P450_{BNF-B}$) was shown (Butler *et al.*, 1989).

N-Glucuronidation of MOCA has been demonstrated *in vitro* with uridine diphosphoglucuronic acid-fortified liver microsomes from polychlorinated biphenyl-induced rats (Cocker *et al.*, 1990). MOCA was metabolized rapidly by *Bacillus megaterium* and a *Nocardiopsis* species to *N*-acetyl, *N,N'*-diacetyl, *N*-hydroxy-*N*-acetyl and *N*-hydroxy-*N,N'*-diacetyl metabolites (Yoneyama & Matsumura, 1984) (see Fig. 2).

4.2 Toxic effects

4.2.1 *Humans*

Medical surveillance of workers with known exposure to MOCA revealed no acute toxicity; the methaemoglobinaemia syndrome, seen with exposures to other aromatic amines, was not observed (Linch *et al.*, 1971). The individual who was sprayed with three gallons of molten MOCA (see p. 276) had an initial 'mild sunburn' sensation on the arms, but no further symptom was found in a two-week follow-up period. Renal and liver function tests were normal, and methaemoglobinemia, haematuria and proteinuria were not observed (Osorio *et al.*, 1990). The initial responses in the worker sprayed in the face with MOCA (see pp. 276–277) were conjunctivitis, a burning sensation in the eyes and face and nausea (Hosein & Van Roosmalen, 1978).

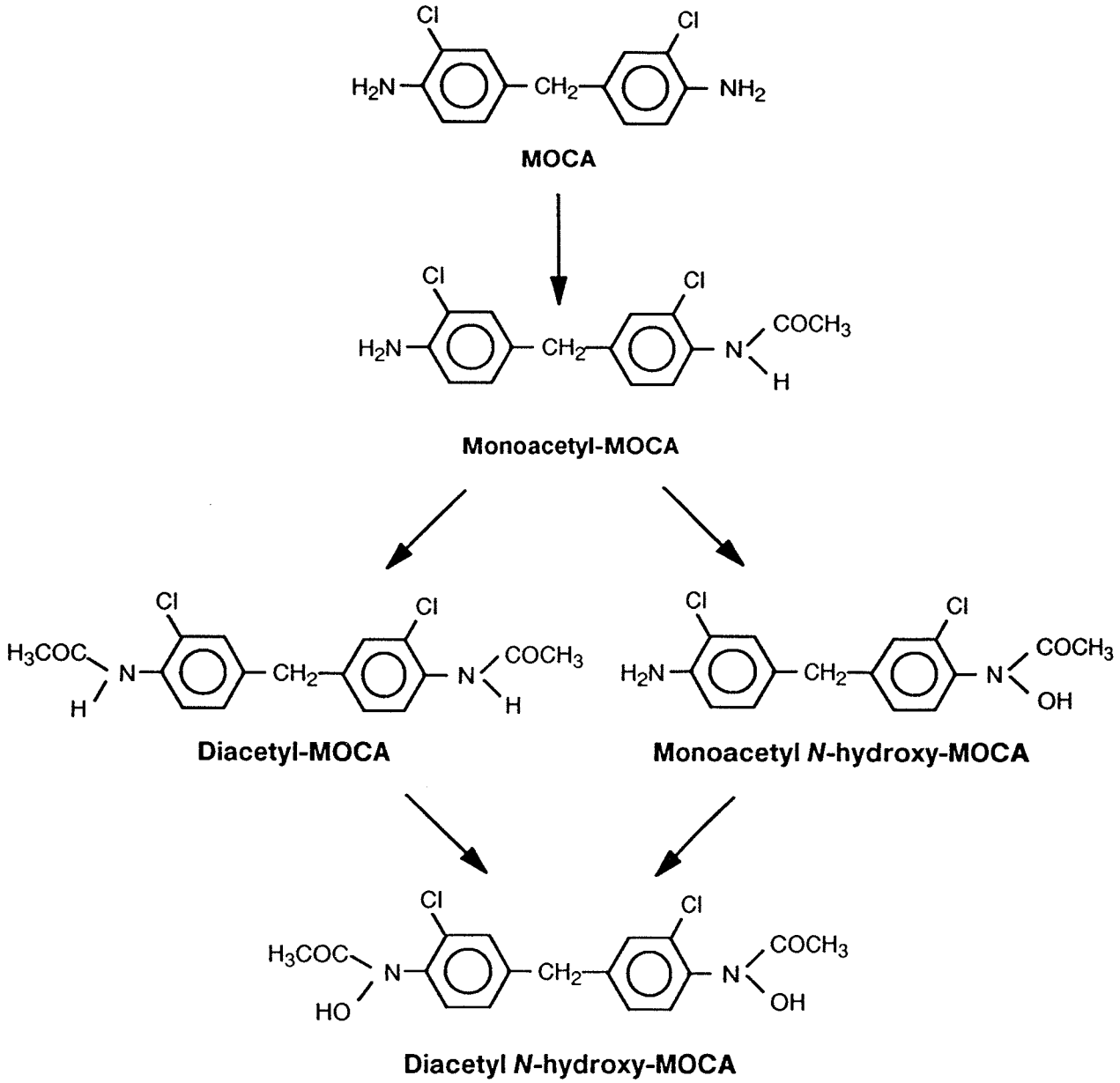
4.2.2 *Experimental systems*

In a nine-year chronic study in dogs (Stula *et al.*, 1977; see p. 283), elevated levels of plasma glutamic-pyruvic transaminase were noted during the first and last two years of treatment, accompanied by urinary changes indicative of genitourinary cancer after seven years.

MOCA also induces enzymes involved in drug metabolism and cell proliferation. Single intraperitoneal injections of technical-grade MOCA (purity, 90–100%) to male Sprague-Dawley rats at doses of 0.4–100 mg/kg bw in dimethyl sulfoxide resulted in dose-dependent increases in the levels of microsomal epoxide hydratase, ethoxyresorufin *O*-deethylase, ethoxycoumarin *O*-deethylase and glutathione *S*-transferase, but a decrease in aldrin epoxidase activity (Wu *et al.*, 1989a). Ornithine decarboxylase, which regulates polyamine synthesis and cell division and is increased by tumour promoters, was strongly induced in male Sprague-Dawley rats 12 h after intraperitoneal injection of 75 mg/kg bw MOCA in corn oil; the level returned to control values after 42 h (Savage *et al.*, 1992).

In primary cultures of rat hepatocytes, MOCA induced dose-dependent leakage of two intracellular enzymes, lactate dehydrogenase and glutamic-oxaloacetic transaminase (McQueen & Williams, 1982).

Fig. 2. Microbial metabolism of MOCA



From Yoneyama and Matsumura (1984)

4.3 Reproductive and prenatal effects

No data were available to the Working Group.

4.4 Genetic and related effects

4.4.1 *Humans*

Exfoliated urothelial cells recovered from urine samples (Osorio *et al.*, 1990) provided by the worker accidentally sprayed with molten MOCA (see p. 276) at different times

following exposure (up to 430 h) had a single, major DNA adduct, shown by ^{32}P-post-labelling and thin-layer chromatography to co-chromatograph with the known major *N*-hydroxy-MOCA–DNA adduct, *N*-(deoxyadenosine-8-yl)-4-amino-3-chlorobenzyl alcohol. The adduct was detected in cells from urine collected up to 98 h after initial exposure, but not thereafter (Kaderlik *et al.*, 1993). This finding is in agreement with the calculated biological half-time for MOCA in urine, 23 h, and the prediction that 94% of an initial dose will be eliminated within 96 h (Osorio *et al.*, 1990).

An increased frequency of sister chromatid exchange was seen in peripheral lymphocytes from a small number of workers exposed to MOCA in polyurethane manufacture (Edwards & Priestly, 1992).

4.4.2 *Experimental systems* (see also Table 7 and Appendices 1 and 2)

MOCA caused prophage induction in *Escherichia coli* and differential toxicity in *Bacillus subtilis rec*-deficient strains. It was mutagenic to *Salmonella typhimurium*, *Escherichia coli* and at the *tk* locus in mouse lymphoma L5178Y cells, but not to *Saccharomyces cerevisiae*. MOCA caused aneuploidy in *S. cerevisiae* but gave equivocal results with regard to gene conversion and did not induce mitotic crossing over in the same organism. It induced mutation in *Drosophila melanogaster* and unscheduled DNA synthesis in primary cultures of hepatocytes from mice, rats and Syrian hamsters. Sister chromatid exchange but not chromosomal aberration was induced in Chinese hamster ovary cells; and neither sister chromatid exchange nor chromosomal aberration was induced in human cells (abstract). MOCA induced cell transformation in mammalian cells and inhibited gap-junctional intercellular communication in cultured rat liver cells.

MOCA induced sister chromatid exchange in lymphocytes of rats treated *in vivo*. It formed adducts with DNA in cultured canine and human bladder cells, in the livers of rats treated topically or by intraperitoneal administration *in vivo* and in lung, liver and kidney following oral administration to rats. One of three HPLC peaks of an enzymatic digest of DNA derived from rats treated *in vivo* was identified tentatively as *N*-(deoxyadenosin-8-yl)-4-amino-3-chlorobenzyl alcohol (Silk *et al.*, 1989). Reaction of *N*-hydroxy[methylene-^{14}C]MOCA with DNA *in vitro* resulted in the formation of two major adducts, which were identified by mass spectroscopy as *N*-(deoxyadenosin-8-yl)-4-amino-3-chlorobenzyl alcohol and *N*-(deoxyadenosin-8-yl)-4-amino-3-chlorotoluene. The same adducts were formed *in vivo*: In rats given a single dose of 95 μmol/kg bw [methylene-^{14}C]MOCA by gavage, DNA adducts were found after 24 h at 7 pmol/mg DNA in liver, 2 pmol/mg in lung and 0.5 pmol/mg in kidney (Segerbäck & Kadlubar, 1992). MOCA also binds to RNA and proteins, including haemoglobin, in rats treated *in vivo*.

N-Hydroxy-MOCA was mutagenic to *S. typhimurium* TA98 and TA100 in the absence of an exogenous metabolic activation system but did not inhibit intercellular communication in cultured WB-F344 rat liver epithelial cells. Other MOCA metabolites, *ortho*-hydroxy-MOCA, 4-amino-3,3′-dichloro-4′-nitrosodiphenylmethane (mononitroso derivative) and di(3-chloro-4-nitrosophenyl)methane (dinitroso derivative), were not mutagenic to *S. typhimurium* TA98 or TA100. The mutagenic activity of the mononitroso derivative towards TA100, however, appeared to be masked by its toxicity (Kuslikis *et al.*, 1991).

Table 7. Genetic and related effects of MOCA

Test system	Result		Dose[a] (LED/HID)	Reference
	Without exogenous metabolic system	Wirh exogenous metabolic system		
PRB, Prophage λ induction, *Escherichia coli*	0	+	1000.0000	Thomson (1981)
BSD, *Bacillus subtilis rec* strains, differential toxicity	+	+	1000.0000	Kada (1981)
SAF, *Salmonella typhimurium*, forward mutation	0	+	50.0000	Bridges *et al.* (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	0	+	50.0000	McCann *et al.* (1975)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	16.0000	Baker & Bonin (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	12.5000	Brooks & Dean (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	0.0000	Garner *et al.* (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	10.0000	Hubbard *et al.* (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	0.0000	Ichinotsubo *et al.* (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	25.0000	MacDonald (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	12.5000	Martire *et al.* (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	25.0000	Nagao & Takahashi (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	25.0000	Simmon & Shepherd (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	25.0000	Venitt & Crofton-Sleigh (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	17.0000[b]	Haworth *et al.* (1983)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	25.0000	Cocker *et al.* (1985)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	12.5000	Hesbert *et al.* (1985)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	6.7000	Cocker *et al.* (1986)
SA0, *Salmonella typhimurium* TA100, reverse mutation	0	+	3.3000	Kugler-Steigmeier *et al.* (1989)
SA0, *Salmonella typhimurium* TA100, reverse mutation	0	+	12.5000	Wu *et al.* (1989b)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	–	500.0000	Richold & Jones (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	–	+	6.3000	Rowland & Severn (1981)
SA0, *Salmonella typhimurium* TA100, reverse mutation	0	?	0.0000	Trueman (1981)
SA5, *Salmonella typhimurium* TA1535, reverse mutation	–	–	0.0000	Baker & Bonin (1981)
SA5, *Salmonella typhimurium* TA1535, reverse mutation	–	–	1000.0000	Brooks & Dean (1981)
SA5, *Salmonella typhimurium* TA1535, reverse mutation	–	–	500.0000	Richold & Jones (1981)
SA5, *Salmonella typhimurium* TA1535, reverse mutation	–	–	1000.0000	Rowland & Severn (1981)
SA5, *Salmonella typhimurium* TA1535, reverse mutation	–	–	0.0000	Simmon & Shepherd (1981)
SA5, *Salmonella typhimurium* TA1535, reverse mutation	0	+	0.0000	Trueman (1981)

Table 7 (contd)

Test system	Result		Dose[a] (LED/HID)	Reference
	Without exogenous metabolic system	Wirh exogenous metabolic system		
SA5, *Salmonella typhimurium* TA1535, reverse mutation	–	–	167.0000	Haworth *et al.* (1983)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	–	–	0.0000	Baker & Bonin (1981)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	–	–	1000.0000	Brooks & Dean (1981)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	–	–	0.0000	Martire *et al.* (1981)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	–	–	0.0000	Nagao & Takahashi (1981)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	–	–	500.0000	Richold & Jones (1981)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	–	–	167.0000	Haworth *et al.* (1983)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	–	–	1000.0000	Rowland & Severn (1981)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	–	–	0.0000	Simmon & Shepherd (1981)
SA7, *Salmonella typhimurium* TA1537, reverse mutation	0	–	1250.0000	Trueman (1981)
SA8, *Salmonella typhimurium* TA1538, reverse mutation	–	–	0.0000	Simmon & Shepherd (1981)
SA8, *Salmonella typhimurium* TA1538, reverse mutation	–	–	0.0000	Baker & Bonin (1981)
SA8, *Salmonella typhimurium* TA1538, reverse mutation	–	–	1000.0000	Brooks & Dean (1981)
SA8, *Salmonella typhimurium* TA1538, reverse mutation	–	–	500.0000	Richold & Jones (1981)
SA8, *Salmonella typhimurium* TA1538, reverse mutation	0	–	1250.0000	Trueman (1981)
SA8, *Salmonella typhimurium* TA1538, reverse mutation	–	+	40.0000	Gatehouse (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	+	0.0000	Baker & Bonin (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	+	0.0000	Brooks & Dean (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	+	0.0000	Garner *et al.* (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	–	0.0000	Hubbard *et al.* (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	–	0.0000	Ichinotsubo *et al.* (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	+	25.0000	MacDonald (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	+	0.0000	Martire *et al.* (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	+	0.0000	Nagao & Takahashi (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	–	500.0000	Richold & Jones (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	–	1000.0000	Rowland & Severn (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	+	0.0000	Simmon & Shepherd (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	0	+	0.0000	Trueman (1981)
SA9, *Salmonella typhimurium* TA98, reverse mutation	–	+	25.0000	Venitt & Crofton-Sleigh (1981)

Table 7 (contd)

Test system	Result		Dose[a] (LED/HID)	Reference
	Without exogenous metabolic system	Wirh exogenous metabolic system		
SA9, *Salmonella typhimurium* TA98, reverse mutation	0	+	0.0000	Rao *et al.* (1982)
SA9, *Salmonella typhimurium* TA98, reverse mutation	-	+	50.0000	Haworth *et al.* (1983)
SA9, *Salmonella typhimurium* TA98, reverse mutation	0	+	167.0000	Kugler-Steigmeier *et al.* (1989)
SA9, *Salmonella typhimurium* TA98, reverse mutation	0	+	25.0000	Wu *et al.* (1989b)
EC2, *Escherichia coli* WP2, reverse mutation	0	-	0.0000	Matsushima *et al.* (1981)
ECW, *Escherichia coli* WP2 uvrA, reverse mutation	-	-	250.0000	Matsushima *et al.* (1981)
ECR, *Escherichia coli* WP2 uvrA (pKM101), reverse mutation	-	+	10.0000	Matsushima *et al.* (1981)
ECW, *Escherichia coli* WP2 *uvr*A, reverse mutation	-	-	0.0000	Gatehouse (1981)
ECW, *Escherichia coli* WP2 *uvr*A, reverse mutation	-	+	25.0000	Venitt & Crofton-Sleigh (1981)
SCG, *Saccharomyces cerevisiae*, gene conversion	-	-	167.0000	Jagannath *et al.* (1981)
SCG, *Saccharomyces cerevisiae*, gene conversion	+	0	100.0000	Sharp & Parry (1981)
SCH, *Saccharomyces cerevisiae*, homozygosis	-	-	100.0000	Kassinova *et al.* (1981)
SCR, *Saccharomyces cerevisiae*, reverse mutation	-	-	889.0000	Mehta & von Borstel (1981)
SCN, *Saccharomyces cerevisiae*, aneuploidy	+	0	50.0000	Parry & Sharp (1981)
DMM, *Drosophila melanogaster*, somatic mutation	(+)		1335.0000	Kugler-Steigmeier *et al.* (1989)
URP, Unscheduled DNA synthesis, rat primary hepatocytes	+	0	2.6700	McQueen *et al.* (1981)
URP, Unscheduled DNA synthesis, rat primary hepatocytes	+	0	100.0000	Williams *et al.* (1982)
URP, Unscheduled DNA synthesis, rat primary hepatocytes	+	0	2.6700	Mori *et al.* (1988)
UIA, Unscheduled DNA synthesis, mouse primary hepatocytes	+	0	13.3500	McQueen *et al.* (1981)
UIA, Unscheduled DNA synthesis, Syrian hamster primary hepatocytes	+	0	2.6700	McQueen *et al.* (1981)
G5T, Gene mutation, mouse lymphoma L5178Y cells *in vitro*	-	+	42.0000	Mitchell *et al.* (1988)
G5T, Gene mutation, mouse lymphoma L5178Y cells *in vitro*	-	+	5.0000	Myhr & Caspary (1988)
SIC, Sister chromatid exchange, Chinese hamster ovary cells *in vitro*	-	-	10.0000	Perry & Thomson (1981)
SIC, Sister chromatid exchange, Chinese hamster ovary cells *in vitro*	+[c]	(+)[c]	50.0000	Galloway *et al.* (1985)
CIC, Chromosomal aberrations, Chinese hamster ovary cells *in vitro*	-[d]	-[d]	300.0000	Galloway *et al.* (1985)

Table 7 (contd)

Test system	Result		Dose[a] (LED/HID)	Reference
	Without exogenous metabolic system	Wirh exogenous metabolic system		
SHL, Sister chromatid exchanges, human leukocytes	-	-	0.0000	Ho *et al.* (1979); abstr.
CHL, Chromosomal aberrations, human leukocytes	-	-	0.0000	Ho *et al.* (1979); abstr.
TCS, Cell transformation in Syrian hamster kidney BHK cells	+	+	13.7000	Daniel & Dehnel (1981)
TCS, Cell transformation in Syrian hamster kidney BHK cells	0	+	2.5000	Styles (1981)
TRR, Cell transformation, RLV/Fischer rat embryo cells	+	0	1.0000	Dunkel *et al.* (1981)
TBM, Cell transformation in BALB/c 3T3 mouse cells	+	0	0.2000	Dunkel *et al.* (1981)
SVA, Sister chromatid exchange, rat lymphocytes *in vivo*	+		125.0000 × 6 mg/kg, ip	Edwards & Priestly (1992)
MVM, Micronucleus test, mouse bone marrow *in vivo*	+		32.0000 × 2 ip	Salamone *et al.* (1981)
BID, Binding (covalent) to DNA in human bladder cells *in vitro* (^{32}P post-labelling)	+	0	0.0300	Stoner *et al.* (1988)
BID, Binding (covalent) to DNA in dog bladder cells *in vitro* (^{32}P post-labelling)	+	0	0.0300	Stoner *et al.* (1988)
BVD, Binding (covalent) to rat liver DNA *in vivo* (tritium label)	+		24.0000 × 1 ip	Silk *et al.* (1989)
BVD, Binding (covalent) to rat lung, liver and kidney DNA *in vivo* (^{14}C-label)	+		25.0000 × 1 po	Segerbäck & Kadlubar (1992)
BVD, Binding (covalent) to rat liver DNA *in vivo* (^{14}C-label)	+		75.0000 × 1 po	Cheever *et al.* (1990)
BVD, Binding (covalent) to rat liver DNA in vivo (^{14}C-label)	+		75.0000 skin	Cheever *et al.* (1990)
BVD, Binding (covalent) to rat liver DNA *in vivo*	+		1.4300 × po	Kugler-Steigmeier *et al.* (1989)
BVD, Binding (covalent) to rat lung DNA *in vivo*	+		1.4300 × po	Kugler-Steigmeier *et al.* (1989)
BVP, Binding (covalent) to rat globin *in vivo* (^{14}C-label)	+		75.0000 × 1 po	Cheever *et al.* (1990)
BVP, Binding (covalent) to rat lung, liver and kidney RNA and protein *in vivo*	+		1.0000 × 1 po	Sabbioni & Neumann (1990)
ICR, Inhibition of cell–cell communication in WB-F344 rat liver epithelial cells *in vitro*	+	0	2.0000	Kuslikis *et al.* (1991)

+, positive; (+), weakly positive; -, negative; 0, not tested

[a]In-vitro tests, μg/ml; in-vivo tests, mg/kg bw; 0.0000, not given

[b]Positive in two laboratories; in a third laboratory, negative with rat S9, weakly positive with hamster S9

[c]Positive in one laboratory; negative in another

[d]Tested in two laboratories

5. Summary of Data Reported and Evaluation

5.1 Exposure data

4,4′-Methylenebis(2-chloroaniline) (MOCA) was introduced in the mid-1950s in the production of high-performance polyurethane mouldings. It is used in many countries, with a total worldwide production of several thousand tonnes per year; it is used as a curing agent for roofing and wood sealing in Japan and the Far East. There was considerable occupational exposure by cutaneous absorption in the early years of use of MOCA, as revealed by urine analysis, but exposure has decreased with the implementation of control measures. Extensive environmental contamination is known to have occurred in a large area surrounding at least one factory, prior to the introduction of controls.

5.2 Human carcinogenicity data

Three asymptomatic cases of cancer of the urinary bladder (two in men under the age of 30 among 552 workers) were identified in a factory where MOCA was produced and where screening for this cancer was undertaken in a subgroup. Although this finding suggests an excess, expected numbers could not be calculated.

5.3 Animal carcinogenicity data

MOCA was tested for carcinogenicity by oral administration in the diet in mice in one study, in rats of each sex in two studies, in male rats in a further two studies using normal and low-protein diets and in capsules in female dogs. It was also tested by subcutaneous administration to rats in one study. Oral administration of MOCA increased the incidence of liver tumours in female mice. In a series of experiments in which rats were fed either standard or low-protein diets, it induced liver-cell tumours and malignant lung tumours in males and females in one study, a few liver-cell tumours in male rats in another, lung adenocarcinomas and hepatocellular tumours in males and females in a third and malignant lung tumours, mammary gland adenocarcinomas, Zymbal gland carcinomas and hepatocellular carcinomas in a fourth. Oral administration of MOCA to female beagle dogs produced transitional-cell carcinomas of the urinary bladder and urethra. Subcutaneous administration to rats produced hepatocellular carcinomas and malignant lung tumours.

5.4 Other relevant data

MOCA forms adducts with DNA, both *in vitro* and *in vivo*. One of the two major adducts, *N*-(deoxyadenosin-8-yl)-4-amino-3-chlorobenzyl alcohol, was found in rat tissues; it also co-chromatographed with a DNA adduct from urothelial cells recovered from the urine of a worker in the polyurethane industry who was accidentally exposed to a high dose of MOCA. An increased frequency of sister chromatid exchange was seen in a small number of workers exposed to MOCA.

MOCA induced DNA damage in prokaryotes, cultured mammalian and human cells and in animals treated *in vivo*. Gene mutation was induced in bacteria and cultured mammalian cells, but not in yeast. Equivocal results for mitotic recombination were obtained in yeasts. Aneuploidy was induced in yeast and sister chromatid exchange, transformation and inhibition of intercellular communication in cultured mammalian cells. Micronuclei were induced in the bone marrow of mice treated *in vivo*, and sister chromatid exchange was induced in the bone marrow of rats treated *in vivo*.

MOCA is comprehensively genotoxic. Furthermore, (i) rats, dogs and humans metabolize MOCA to *N*-hydroxy-MOCA by hepatic cytochromes P450; (ii) DNA adducts are formed by reaction with *N*-hydroxy-MOCA, and MOCA is genotoxic in bacteria and mammalian cells; (iii) the same major MOCA–DNA adduct is formed in the target tissues for carcinogenicity in animals (rat liver and lung; dog urinary bladder) as that found in urothelial cells from a man with known occupational exposure to MOCA.

5.5 Evaluation[1]

There is *inadequate evidence* in humans for the carcinogenicity of 4,4′-methylenebis(2-chloroaniline) (MOCA).

There is *sufficient evidence* in experimental animals for the carcinogenicity of 4,4′-methylenebis(2-chloroaniline) (MOCA).

Overall evaluation[2]

4,4′-Methylenebis(2-chloroaniline) (MOCA) is *probably carcinogenic to humans (Group 2A)*.

6. References

American Conference of Governmental Industrial Hygienists (1990a) Notice of intended changes—4,4′-methylenebis(2-chloroaniline), perfluoroiso-butylene, and triethanolamine. *Appl. occup. environ. Hyg.*, **5**, 798–804

American Conference of Governmental Industrial Hygienists (1990b) *Guide to Occupational Exposure Values—1990*, Cincinnati, OH, p. 12

American Conference of Governmental Industrial Hygienists (1992) *Annual Reports of the Committees on Threshold Limit Values and Biological Exposure Indices*, Cincinnati, OH, p. 1

Anon. (1985) International toxicity update. *Dangerous Prop. ind. Mater. Rep.*, **5**, 30–36

Arbejdstilsynet (Labor Inspection) (1977) *Liste over Hygiejniske Graensevaerdier 1977* (List of limit values 1977), Copenhagen

Baker, R.S.U. & Bonin, A.M. (1981) Study of 42 coded compounds with the *Salmonella*/mammalian microsome assay. *Prog. Mutat. Res.*, **1**, 249–260

---

[1]For definition of the italicized terms, see Preamble, pp. 26–30.

[2]Overall evaluation 2A and not 2B on the basis of supporting evidence from other relevant data

Becker, J.W., Blackwell, J., Caruso, P.P., Pugh, T.L. & Yeager, F.W. (1974) Determination of unreacted 'MOCA' in polyether-based polyurethane systems. *Rubber World*, **170**, 57–62, 80

Bridges, B.A., MacGregor, D., Zeiger, E., Bonin, A., Dean, B.J., Lorenzo, F., Garner, R.C., Gatehouse, D., Hubbard, S., Ichinotsubo, D., MacDonald, D., Martire, G., Matsushima, T., Mohn, G., Nagao, M., Richold, M., Rowland, I., Simmon, V., Skopek, T., Truman, R. & Venitt, S. (1981) Summary report on the performance of bacterial mutation assays. *Prog. Mutat. Res.*, **1**, 49–67

Brooks, T.M. & Dean, B.J. (1981) Mutagenic activity of 42 coded compounds in the *Salmonella*/-microsome assay with preincubation. *Prog. Mutat. Res.*, **1**, 261–270

Butler, M.A., Guengerich, F.P. & Kadlubar, F.F. (1989) Metabolic oxidation of the carcinogens 4-aminobiphenyl and 4,4′-methylene-bis(2-chloroaniline) by human hepatic microsomes and by purified rat hepatic cytochrome P-450 monooxygenases. *Cancer Res.*, **49**, 25–31

Caisse Nationale Assurance (1992) *Valeurs Limites d'Exposition aux postes de travail 1992*, Lucerne, Division Médecine du Travail

Cartwright, R.A. (1983) Historical and modern epidemiological studies on populations exposed to *N*-substituted aryl compounds. *Environ. Health Perspectives*, **49**, 13–19

Cheever, K.L., Richards, D.E., Weigel, W.W., Begley, K.B., Savage, R.E., Jr & Daniel, F.B. (1988) Macromolecular adduct formation by 4,4′-methylene-bis(2-chloroaniline) in adult male rat. *Scand. J. Work Environ. Health*, **14** (Suppl. 1), 57–59

Cheever, K.L., Richards, D.E., Weigel, W.W., Begley, K.B., DeBord, D.G., Swearengin, T.F. & Savage, R.E., Jr (1990) 4,4′-Methylene-bis(2-chloroaniline)(MOCA): comparison of macromolecular adduct formation after oral or dermal administration in the rat. *Fundam. appl. Toxicol.*, **14**, 273–283

Cheever, K.L., DeBord, D.G. & Swearengin, T.F. (1991) 4,4′-Methylene bis(2-chloroaniline) (MOCA): the effect of multiple oral administration, route, and phenobarbital induction on macromolecular adduct formation in the rat. *Fundam. appl. Toxicol.*, **16**, 71–80

Chemical Information Services (1991) *Directory of World Chemical Producers 1992/93*, Dallas, TX, p. 79

Chen, T.H., Kuslikis, B.I. & Braselton, W.E., Jr (1989) Hydroxylation of 4,4′-methylenebis(2-chloroaniline) by canine, guinea pig, and rat liver microsomes. *Drug Metab. Disposition*, **17**, 406–413

Chen, T.H., Kuslikis, B.I. & Braselton, W.E., Jr (1991) Unlabeled hemoglobin adducts of 4,4′-methylenebis(2-chloroaniline) in rats and guinea pigs. *Arch. Toxicol.*, **65**, 177–185

Chin, B., Tobes, M.C. & Han, S.S. (1983) Absorption of 4,4′-methylenebis[2-chloroaniline] by human skin. *Environ. Res.*, **32**, 167–178

Clapp, D.E., Piacitelli, G.M., Zaebst, D.D. & Ward, E. (1991) Assessing exposure to 4,4′-methylene bis(2-chloroaniline) (MBOCA) in the workplace *Appl. occup. environ. Hyg.*, **6**, 125–130

Cocker, J., Boobis, A.R., Gibson, J.F. & Davies, D.S. (1985) The metabolic activation of 4,4′-methylenebis(2-chlorobenzeneamine) to a bacterial mutagenic by hepatic postmitochondrial supernatant from human and other species. *Environ. Mutag.*, **7**, 501–509

Cocker, J., Boobis, A.R. & Davies, D.S. (1986) Routes of activation of 4,4′-methylenebis(2-chloroaniline) and 4,4′-methylenedianiline to bacterial mutagens. *Food chem. Toxicol.*, **24**, 755–756

Cocker, J., Boobis, A.R. & Davies, D.S. (1988) Determination of the *N*-acetyl metabolites of 4,4′-methylene dianiline and 4,4′-methylene-bis(2-chloroaniline) in urine. *Biomed. environ. Mass Spectrom.*, **17**, 161–167

Cocker, J., Boobis, A.R., Wilson, H.K. & Gompertz, D. (1990) Evidence that a β-*N*-glucuronide of 4,4′-methylenebis (2-chloroaniline) (MbOCA) is a major urinary metabolite in man: implications for biological monitoring. *Br. J. ind. Med.*, **47**, 154–161

Commission of the European Communities (1967) Council Directive of 16 August 1967 on the approximation of the laws, regulations and administrative provisions relating to the classification, packaging and labelling of dangerous substances. *Off. J. Eur. Commun.*, **L196**, 1

Commission of the European Communities (1990) Council Directive of 28 June 1990 on the protection of workers from the risks related to exposure to carcinogens at work (6th individual Directive within the meaning of Article 16 of Directive 89/391/EEC). *Off. J. Eur. Commun.*, **L196**, 1–7

Commission of the European Communities (1991) Twelfth Adaptation to Technical Progress of Council Directive 67/548/EEC of 16 August 1967. *Off. J. Eur. Commun.*, **L180A**, 918

Cook, W.A. (1987) *Occupational Exposure Limits—Worldwide*, Akron, OH, American Industrial Hygiene Association, pp. 146, 199

Daniel, M.R. & Dehnel, J.M. (1981) Cell transformation test with baby hamster kidney cells. *Prog. Mutat. Res.*, **1**, 626–637

Deutsche Forschungsgemeinschaft (1992) *MAK- and BAT-Values List 1992. Maximum Concentrations at the Workplace (MAK) and Biological Tolerance Values (BAT) for Working Materials* (Report No. 28), Weinheim, VCH Verlagsgesellschaft, p. 49

Ducos, P., Maire, C. & Gaudin, R. (1985) Assessment of occupational exposure to 4,4′-methylene-bis(2-chloroanilione) 'MOCA' by a new sensitive method for biological monitoring. *Int. Arch. occup. environ. Health*, **55**, 159–167

Dunkel, V.C., Pienta, R.J., Sivak, A. & Traul, K.A. (1981) Comparative neoplastic transformation responses of BALB/3T3 cells, Syrian hamster embryo cells, and Rauscher murine leukemia virus-infected Fischer 344 rat embryo cells to chemical carcinogens. *J. natl Cancer Inst.*, **67**, 1303–1315

Edwards, J.W. & Priestly, B.G. (1992) Biological and biological-effect monitoring of workers exposed to 4,4′-methylene-bis(2-chloroaniline). *Hum. exp. Toxicol.*, **11**, 229–236

Eller, P.M., ed. (1985) *NIOSH Manual of Analytical Methods*, 3rd ed., Suppl. 1 (DHHS (NIOSH) Publ. No. 84-100), Washington DC, US Government Printing Office, pp. 8302-1—8302-4

Farmer, P.B., Rickard, J. & Robertson, S. (1981) The metabolism and distribution of 4,4′-methylene-bis(2-chloroaniline) (MBOCA) in rats. *J. appl. Toxicol.*, **1**, 317–322

Fishbein, L. (1984) Aromatic amines. In: Hutzinger, O., ed., *The Handbook of Environmental Chemistry*, Vol. 3, Part C, Berlin, Springer-Verlag, pp. 1–40

Galloway, S.M., Bloom, A.D., Resnick, M., Margolin, B.H., Nakamura, F., Archer, P. & Zeiger, E. (1985) Development of a standard protocol for in vitro cytogenetic testing with Chinese hamster ovary cells: comparison of results for 22 compounds in two laboratories. *Environ. Mutag.*, **7**, 1–51

Garner, R.C., Welch, A. & Pickering, C. (1981) Mutagenic activity of 42 coded compounds in the *Salmonella*/microsome assay. *Prog. Mutat. Res.*, **1**, 280–284

Gatehouse, D. (1981) Mutagenic activity of 42 coded compounds in the 'microtiter' fluctuation test. *Prog. Mutat. Res.*, **1**, 376–386

Groth, D.H., Weigel, W.W., Tolos, W.P., Brewer, D.E., Cheever, K.L. & Burg, J.R. (1984) 4,4′-Methylene-bis-*ortho*-chloro-aniline (MBOCA): absorption and excretion after skin application and gavage. *Environ. Res.*, **34**, 38–54

Grundmann, E. & Steinhoff, D. (1970) Liver and lung tumours after administration of 3,3′-dichloro-4,4′-diaminodiphenylmethane to rats (Ger.). *Z. Krebsforsch.*, **74**, 28–39

Haworth, S., Lawlor, T., Mortelmans, K., Speck, W. & Zeiger, E. (1983) *Salmonella* mutagenicity test results for 250 chemicals. *Environ. Mutag.*, **1**, 3–142

Health and Safety Executive (1985) *Occupational Exposure Limits 1987*, London, Her Majesty's Stationery Office

Hesbert, A., Bottin, M.C. & De Ceaurriz, J. (1985) Mutagenicity of 4,4′-methylene-bis(2-chloroaniline) 'MOCA' and its *N*-acetyl derivatives in *S. typhimurium*. *Int. Arch. occup. environ. Health*, **55**, 169–174

Ho, T., Hardigree, A.A., Larimer, F.W., Nix, C.E., Rao, T.K., Tipton, S.C. & Epler, J.L. (1979) Comparative mutagenicity study of potentially carcinogenic industrial compounds (Abstract Ea-10). *Environ. Mutag.*, **1**, 167–168

Hosein, H.R. & Van Roosmalen, P.B. (1978) Acute exposure to methylene-bis-ortho chloroaniline (MOCA). *Am. ind. Hyg. Assoc. J.*, **39**, 496–497

Hubbard, S.A., Green, M.H.L., Bridges, B.A., Wain, A.J. & Bridges, J.W. (1981) Fluctuation test with S9 and hepatocyte activation. *Prog. Mutat. Res.*, **1**, 361–370

IARC (1974) *IARC Monographs on the Evaluation of Carcinogenic Risk of Chemicals to Humans*, Vol. 4, *Some Aromatic Amines, Hydrazine and Related Substances,* N-*Nitroso Compounds and Miscellaneous Alkylating Agents*, Lyon, pp. 65–71

IARC (1982a) *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans*, Vol. 29, *Some Industrial Chemicals and Dyestuffs*, Lyon, pp. 345–389

IARC (1982b) *IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans*, Vol. 28, *The Rubber Industry*, Lyon

IARC (1987a) *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans*, Suppl. 7, *Overall Evaluations of Carcinogenicity: An Updating of* IARC Monographs *Volumes 1 to 42*, Lyon, pp. 246–247

IARC (1987b) *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans*, Suppl. 7, *Overall Evaluations of Carcinogenicity: An Updating of* IARC Monographs *Volumes 1 to 42*, Lyon, pp. 211–216

IARC (1987c) *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans*, Suppl. 7, *Overall Evaluations of Carcinogenicity: An Updating of* IARC Monographs *Volumes 1 to 42*, Lyon, pp. 332–334

Ichikawa, Y., Yoshida, M., Okayama, A., Hara, I. & Morimoto, K. (1990) Biological monitoring for workers exposed to 4,4′-methylenebis (2-chloroaniline). *Am. ind. Hyg. Assoc. J.*, **51**, 5–7

Ichinotsubo, D., Mower, H. & Mandel, M. (1981) Mutagen testing of a series of paired compounds with the Ames *Salmonella* testing system. *Prog. Mutat. Res.*, **1**, 298–301

Ihara Chemical Industry Co. (undated) *Data Sheet: Cuamine-M (4,4′-Methylene bis(2-chloroaniline))*, Tokyo

ILO (1991) *Occupational Exposure Limits for Airborne Toxic Substances*, 3rd ed. (Occupational Safety and Health Series No. 37), Geneva, International Labour Office, pp. 128–129

Jagannath, D.R., Vultaggio, D.M. & Brusick, D.J. (1981) Genetic activity of 42 coded compounds in the mitotic gene conversion assay using *Saccharomyces cerevisiae* strain D4. *Prog. Mutat. Res.*, **1**, 456–467

Kada, T. (1981) The DNA-damaging activity of 42 coded compounds in the rec-assay. *Prog. Mutat. Res.*, **1**, 175–182

Kaderlik, K.R., Talaska, G., DeBord, D.G., Osorio, A.M. & Kadlubar, F.F. (1993) 4,4′-Methylene-bis(2-chloroaniline)–DNA adduct analysis in human exfoliated urothelial cells by ^{32}P-postlabelling. *Cancer Epidemiol. Biomarkers Prev.*, **2**, 63–69

Kassinova, G.V., Kovaltsova, S.V., Marfin, S.V. & Zakharov, I.A. (1981) Activity of 40 coded compounds in differential inhibition and mitotic crossing-over assays in yeast. *Prog. Mutat. Res.*, **1**, 434–455

Keeslar, F.L. (1986) The removal and control of methylene bis orthochloroaniline in residential and industrial areas of Adrian, Michigan. In: *1986 Hazardous Material Spills—Conference Proceedings. Preparedness, Prevention, Control and Cleanup of Releases, May 5–8, 1986, St Louis, Missouri* (NIOSH-00165082), Cincinatti, OH, National Institute for Occupational Safety and Health

Kommineni, C., Groth, D.H., Frockt, I.J., Voelker, R.W. & Stanovick, R.P. (1978) Determination of the tumorigenic potential of methylene-bis-orthochloroaniline. *J. environ. Pathol. Toxicol.*, **2**, 149–171

Kugler-Steigmeier, M.E., Friederich, U., Graf, U., Lutz, W.K., Maier, P. & Schlatter, C. (1989) Genotoxicity of aniline derivatives in various short-term tests. *Mutat. Res.*, **211**, 279–289

Kuslikis, B.I., Trosko, J.E. & Braselton, W.E., Jr (1991) Mutagenicity and effect on gap-junctional intercellular communication of 4,4′-methylenebis(2-chloroaniline) and its oxidized metabolites. *Mutagenesis*, **6**, 19–24

Linch, A.L., O'Connor, G.B., Barnes, J.R., Killian, A.S., Jr & Neeld, W.E., Jr (1971) Methylene-bis-*ortho*-chloroaniline (MOCA): evaluation of hazards and exposure control. *Am. ind. Hyg. Assoc. J.*, **32**, 802–819

Locke, J. (1986) Fixing exposure limits for toxic chemicals in the UK—some case studies. *Sci. total Environ.*, **51**, 237–260

Lowry, L.K. & Clapp, D.E. (1992) Urinary 4,4′-methylenebis(2-chloroaniline) (MBOCA): a case study for biological monitoring. *Appl. occup. environ. Hyg.*, **7**, 1–6

MacDonald, D.J. (1981) *Salmonella*/microsome tests on 42 coded chemicals. *Prog. Mutat. Res.*, **1**, 285–297

Manis, M.O. & Braselton, W.E., Jr (1984) Structure elucidation and in vitro reactivity of the major metabolite of 4,4′-methylenebis(2-chloroaniline) (MBOCA) in canine urine. *Fundam. appl. Toxicol.*, **4**, 1000–1008

Manis, M.O. & Braselton, W.E., Jr (1986) Metabolism of 4,4′-methylenebis(2-chloroaniline) by canine liver and kidney slices. *Drug Metab. Disposition*, **14**, 166-174

Manis, M.O., Williams, D.E., McCormack, K.M., Schock, R.J., Lepper, L.F., Ng, Y.-C. & Braselton, W.E. (1984) Percutaneous absorption, disposition, and excretion of 4,4′-methylenebis(2-chloroaniline) in dogs. *Environ. Res.*, **33**, 234–245

Martire, G., Vricella, G., Perfumo, A.M. & De Lorenzo, F. (1981) Evaluation of the mutagenic activity of coded compounds in the *Salmonella* test. *Prog. Mutat. Res.*, **1**, 271–279

Matsushima, T., Takamoto, Y., Shirai, A., Sawamura, M. & Sugimura, T. (1981) Reverse mutation test on 42 coded compounds with the *E. coli* WP2 system. *Prog. Mutat. Res.*, **1**, 387–395

McCann, J., Choi, E., Yamasaki, E. & Ames, B.N. (1975) Detection of carcinogens as mutagens in the *Salmonella*/microsome test: assay of 300 chemicals. *Proc. natl Acad. Sci. USA*, **72**, 5135–5139

McKerrell, P.J., Saunders, G.A. & Geyer, R. (1987) Determination of 4,4′-methylenebis(2-chloroaniline) in urine by high-performance liquid chromatography. *J. Chromatogr.*, **408**, 399–401

McQueen, C.A. & Williams, G.M. (1982) Cytotoxicity of xenobiotics in adult rat hepatocytes in primary culture. *Fundam. appl. Toxicol.*, **2**, 139–144

McQueen, C.A., Maslansky, C.J., Crescenzi, S.B. & Williams, G.M. (1981) The genotoxicity of 4,4′-methylenebis-2-chloroaniline in rat, mouse and hamster hepatocytes. *Toxicol. appl. Pharmacol.*, **58**, 231–235

Mehta, R.D. & von Borstel, R.C. (1981) Mutagenic activity of 42 encoded compounds in the haploid yeast reversion assay, strain XV185-14C. *Prog. Mutat. Res.*, **1**, 414–423

Mitchell, A.D., Rudd, C.J. & Caspary, W.J. (1988) Evaluation of the L5178Y mouse lymphoma cell mutagenesis assay: intralaboratory results for sixty-three coded chemicals tested at SRI International. *Environ. mol. Mutag.*, **12** (Suppl. 13), 37–101

Mori, H., Yoshimi, N., Sugie, S., Iwata, H., Kawai, K., Mashizu, N. & Shimizu, H. (1988) Genotoxicity of epoxy resin hardeners in the hepatocyte primary culture/DNA repair test. *Mutat. Res.*, **204**, 683–688

Morton, K.C., Lee, M.-S., Siedlik, P. & Chapman, R. (1988) Metabolism of 4,4′-methylenebis-2-chloroaniline (MOCA) by rats *in vivo* and formation of *N*-hydroxy MOCA by rat and human liver microsomes. *Carcinogenesis*, **9**, 131–139

Myhr, B.C. & Caspary, W.J. (1988) Evaluation of the L5178Y mouse lymphoma cell mutagenesis system: intralaboratory results for 63 coded chemicals tested at Litton Bionetics, Inc. *Environ. mol. Mutag.*, **12** (Suppl. 13), 103–194

Nagao, M. & Takahashi, Y. (1981) Mutagenic activity of 42 coded compounds in the *Salmonella*/-microsome assay. *Prog. Mutat. Res.*, 1, 302–313

National Swedish Board of Occupational Safety and Health (1984) *Hygieniska Gränsvärden* (Hygiene limit values), Stockholm, p. 32

Osorio, A.M., Clapp, D., Ward, E., Wilson, H.K. & Cocker, J. (1990) Biological monitoring of a worker acutely exposed to MBOCA. *Am. J. ind. Med.*, **18**, 577–589

Palmer Davis Seika (1992) *Material Safety Data Sheet: Bis Amine A (4,4′-Methylene-bis(2-chloroaniline))*, Fort Washington, NY

Parris, G.E., Diachenko, G.W., Entz, R.C., Poppiti, J.A., Lombardo, P., Rohrer, T.K. & Hesse, J.L. (1980) Waterborne methylene bis(2-chloroaniline) and 2-chloroaniline contamination around Adrian, Michigan. *Bull. environ. Contam. Toxicol.*, **24**, 497–503

Parry, J.M. & Sharp, D. (1981) Induction of mitotic aneuploidy in the yeast strain D6 by 42 coded compounds. *Prog. Mutat. Res.*, **1**, 468–480

PEDCo Environmental (1984) *Analysis of Workplace Monitoring Data for MOCA* (Contract No. 68-02-3935), Washington DC, US Environmental Protection Agency, Office of Pesticides and Toxic Substances

Perry, P.E. & Thomson, E.J. (1981) Evaluation of the sister chromatid exchange method in mammalian cells as a screening system for carcinogens. *Prog. Mutat. Res.*, **1**, 560–569

Pouchert, C.J. (1981) *The Aldrich Library of Infrared Spectra*, 3rd ed., Milwaukee, WI, Aldrich Chemical Co., p. 730

Purnell, C.J. & Warwick, C.J. (1981) Method 3—Analysis of 3,3′-dichloro- 4,4′-diaminodiphenylmethane (MOCA) and 2-chloroaniline (OCA) in air. In: Egan, H., Fishbein, L., Castegnaro, M., O'Neill, I.K. & Bartsch, H., eds, *Environmental Carcinogens: Selected Methods of Analysis*, Vol. 4, *Some Aromatic Amines and Azo Dyes in the General and Industrial Environment* (IARC Scientific Publications No. 40), Lyon, IARC, pp. 133–140

Rao, T.K., Dorsey, G.F., Allen, B.E. & Epler, J.L. (1982) Mutagenicity of 4,4'-methylenedianiline derivatives in the *Salmonella* histidine reversion assay. *Arch. Toxicol.*, **49**, 185–190

Rice, J.R. & Kissinger, P.T. (1981) Method 4—Determination of benzidine, 3,3′-dimethoxybenzidine, 4-aminobiphenyl, 3,3′-dichlorobenzidine and 4,4′-methylene-bis(2-chloroaniline) [MOCA] in soil samples, surface water and groundwater by liquid chromatography with electrochemical detection. In: Egan, H., Fishbein, L., Castegnaro, M., O'Neill, I.K. & Bartsch, H., eds, *Environmental Carcinogens Selected Methods of Analysis*, Vol. 4, *Some Aromatic Amines and Azo Dyes in the General and Industrial Environment* (IARC Scientific Publications No. 40), Lyon, IARC, pp. 141–151

Richold, M. & Jones, E. (1981) Mutagenic activity of 42 coded compounds in the *Salmonella*/-microsome assay. *Prog. Mutat. Res.*, **1**, 314–322

Rowland, I. & Severn, B. (1981) Mutagenicity of carcinogens and noncarcinogens in the *Salmonella*/-microsome test. *Prog. Mutat. Res.*, **1**, 323–332

Russfield, A.B., Homburger, F., Boger, E., Van Dongen, C.G., Weisburger, E.K. & Weisburger, J.H. (1975) The carcinogenic effect of 4,4′-methylene-bis(2-chloroaniline) in mice and rats. *Toxicol. appl. Pharmacol.*, **31**, 47–54

Sabbioni, G. & Neumann, H.-G. (1990) Quantification of haemoglobin binding of 4,4′-methylenebis-(2-chloroaniline) (MOCA) in rats. *Arch. Toxicol.*, **64**, 451–458

Sadtler Research Laboratories (1980) *Sadtler Standard Spectra, 1980 Cumulative Index*, Philadelphia, PA

Sadtler Research Laboratories (1991) *Sadtler Standard Spectra, 1981–1991 Supplementary Index*, Philadelphia, PA

Salamone, M.F., Heddle, J.A. & Katz, M. (1981) Mutagenic activity of 41 compounds in the in vivo micronucleus assay. *Prog. Mutat. Res.*, **1**, 686–697

Savage, R.E., Jr, Weigel, W.W. & Krieg, E.F., Jr (1992) Induction of ornithine decarboxylase activity by 4,4′-methylene bis(2-chloroaniline) in the rat. *Cancer Lett.*, **62**, 63–68

Segerbäck, D. & Kadlubar, F.F. (1992) Characterization of 4,4′-methylenebis(2-chloroaniline)–DNA adducts formed *in vivo* and *in vitro*. *Carcinogenesis*, **13**, 1587–1592

Sharp, D.C. & Parry, J.M. (1981) Induction of mitotic gene conversion by 41 coded compounds using the yeast culture JD1. *Prog. Mutat. Res.*, **1**, 491–501

Shivapurkar, N., Lehman, T.A., Schut, H.A.J. & Stoner, G.D. (1987) DNA binding of 4,4′-methylene-bis(2-chloroaniline) (MOCA) in explant cultures of human and dog bladder. *Cancer Lett.*, **38**, 41–48

Silk, N.A., Lay, J.O., Jr & Martin, C.N. (1989) Covalent binding of 4,4′-methylenebis(2-chloroaniline) to rat liver DNA *in vivo* and of its *N*-hydroxylated derivative to DNA *in vitro*. *Biochem. Pharmacol.*, **38**, 279–287

Simmon, V.F. & Shepherd, G.F. (1981) Mutagenic activity of 42 coded compounds in the *Salmonella*/-microsome assay. *Prog. Mutat. Res.*, **1**, 333–342

State of California (1992) *General Industry Safety Orders*, Title 8, No. 5215, *4,4′-Methylenebis(2-chloroaniline)*, Sacramento, CA, pp. 886–890

Steinhoff, D. & Grundmann, E. (971) Carcinogenic effect of 3,3′-dichloro-4,4′-diaminodiphenylmethane in rats (Ger.). *Naturwissenschaften*, **58**, 578

Stoner, G.D., Shivapurkar, N.M., Schut, H.A.J. & Lehman, T.A. (1988) DNA binding and adduct formation of 4,4′-methylene-bis(2-chloroaniline) (MOCA) in explant cultures of human and dog bladder. In: King, C.M., Romano, L.J. & Schuetzle, D., eds, *Carcinogenic and Mutagenic Responses to Aromatic Amines and Nitroarenes*, Amsterdam, Elsevier, pp. 237–240

Stula, E.F., Sherman, H., Zapp, J.A., Jr & Clayton, J.W., Jr (1975) Experimental neoplasia in rats from oral administration of 3,3′-dichlorobenzidine, 4,4′-methylene-bis(2-chloroaniline), and 4,4′-methylene-bis(2-methylaniline). *Toxicol. appl. Pharmacol.*, **31**, 159–176

Stula, E.F., Barnes, J.R., Sherman, H., Reinhardt, C.F. & Zapp, Z.A., Jr (1977) Urinary bladder tumors in dogs from 4,4′-methylene-bis(2-chloroaniline) (MOCA®). *J. environ. Pathol. Toxicol.*, **1**, 31–50

Styles, J.A. (1981) Activity of 42 coded compounds in the BHK-21 cell transformation test. *Prog. Mutat. Res.*, **1**, 638–646

Taylor, D.G. (1977) *NIOSH Manual of Analytical Methods*, 2nd ed., Vol. 1, Cincinnati, OH, National Institute for Occupational Safety and Health, pp. 236-1—236-9

Thomas, J.D. & Wilson, H.K. (1984) Biological monitoring of workers exposed to 4,4′-methylenebis (2-chloroaniline) (MOCA). *Br. J. ind. Med.*, **41**, 547–551

Thomson, J.A. (1981) Mutagenic activity of 42 coded compounds in the lambda induction assay. *Prog. Mutat. Res.*, **1**, 224–235

Tobes, M.C., Brown, L.E., Chin, B. & Marsh, D.D. (1983) Kinetics of tissue distribution and elimination of 4,4′-methylene bis(2-chloroaniline) in rats. *Toxicol. Lett.*, **17**, 69–75

Trippel-Schulte, P., Zeiske, J. & Kettrup, A. (1986) Track analysis of selected benzidine and diaminodiphenylmethane derivatives in urine by means of liquid chromatography using precolumn sample preconcentration, UV and electrochemical detection. *Chromatographia*, **22**, 138–146

Trueman, R.W. (1981) Activity of 42 coded compounds in the *Salmonella* reverse mutation test. *Prog. Mutat. Res.*, **1**, 343–350

US National Institute for Occupational Safety and Health (1984) *Manual of Analytical Methods*, 3rd ed., *Method 8302* (NIOSH (HEW) Pub. No. 84-100), Cincinatti, OH, pp. 8302-1—8302-4

US National Institute for Occupational Safety and Health (1990) *Manual of Analytical Methods*, 4th ed., *Method 5029* (NIOSH (HEW) Pub. No. 90-100), Cincinatti, OH, pp. 5029-1—5029-5

US National Library of Medicine (1992) *Hazardous Substances Data Bank* (HSDB No. 2629), Bethesda, MD

US Occupational Safety and Health Administration (1988) *o-Dianisidine; 4,4′-Methylenebis(2-chloroaniline) (MOCA); o-Tolidine (Method No. 71)* Salt Lake City, UT

US Tariff Commission (1957) *Synthetic Organic Chemicals, United States Production and Sales, 1956* (Report No. 200, Second Series), Washington DC, US Government Printing Office, p. 74

Van Roosmalen, P.B., Klein, A.L. & Drummond, I. (1981) Method 8—Determination of 3,3′-dichloro-4,4′-diaminodiphenylmethane (MOCA) in urine. In: Egan, H., Fishbein, L., Castegnaro, M., O'Neill, I.K. & Bartsch, H., eds, *Environmental Carcinogens Selected Methods of Analysis*, Vol. 4, *Some Aromatic Amines and Azo Dyes in the General and Industrial Environment* (IARC Scientific Publications No. 40), Lyon, IARC, pp. 183–191

Venitt, S. & Crofton-Sleigh, C. (1981) Mutagenicity of 42 coded compounds in a bacterial assay using *Escherichia coli* and *Salmonella typhimurium*. *Prog. Mutat. Res.*, **1**, 351–360

Verschueren, K. (1983) *Handbook of Environmental Data on Organic Chemicals*, 2nd ed., New York, Van Nostrand Reinhold, p. 846

Voorman, R. & Penner, D. (1986) Fate of MBOCA [4,4′-methylene-bis(2-chloro- aniline)] in soil. *Arch. environ. Contam. Toxicol.*, **15**, 595–602

Wan, K.C., Dare, B.R. & Street, N.R. (1989) Biomedical surveillance of workers exposed to 4,4′-methylene-bis(2-chloroaniline) (MBOCA) in Perth, Western Australia. *J. R. Soc. Health*, **5**, 159–167

Ward, E., Clapp, D., Tolos, W. & Groth, D. (1986) Efficacy of urinary monitoring for 4,4′-methylene-bis(2-chloroaniline). *J. occup. Med.*, **28**, 637–642

Ward, E., Smith, A.B. & Halperin, W. (1987) 4,4′-Methylenebis (2-chloroaniline): an unregulated carcinogen. *Am. J. ind. Med.*, **12**, 537–549

Ward, E., Halperin, W., Thun, M., Grossman, H.B., Fink, B., Koss, L., Osorio, A.M. & Schulte, P. (1988) Bladder tumors in two young males occupationally exposed to MBOCA. *Am. J. ind. Med.*, **14**, 267–272

Ward, E., Halperin, W., Thun, M., Grossman, H.B., Fink, B., Koss, L., Osorio, A.M. & Schulte, P. (1990) Screening workers exposed to 4,4′-methylene bis(2-chloroaniline) for bladder cancer by cystoscopy. *J. occup. Med.*, **32**, 865–868

Will, W., Gossler, K., Raithel, H.J. & Schaller, K.H. (1981) Quantitative determination of 4,4′-methylene-bis(2-chloraniline) (MOCA®) in the urine by high-pressure liquid chromatography. *Arbeitsmed. Sozialmed. Praventivmed.*, **16**, 201–203

Williams, G.M., Laspia, M.F. & Dunkel, V.C. (1982) Reliability of the hepatocyte primary culture/DNA repair test in testing of coded carcinogens and noncarcinogens. *Mutat. Res.*, **97**, 359–370

Wu, K., Leslie, C.L. & Stacey, N.H. (1989a) Effects of mutagenic and non-mutagenic aniline derivatives on rat liver drug-metabolizing enzymes. *Xenobiotica*, **19**, 1275–1283

Wu, K., Bonin, A.M., Leslie, C.L., Baker, R.S.U. & Stacey, N.H. (1989b) Genotoxicity and effects on rat liver drug-metabolizing enzymes by possible substitutes for 4,4′-methylene bis(2-chloroaniline). *Carcinogenesis*, **10**, 2119–2122

Yoneyama, K. & Matsumura, F. (1984) Microbial metabolism of 4,4′-methylene- bis(2-chloroaniline). *Arch. environ. Contam. Toxicol.*, **13**, 501–507

Yun, C.-H., Shimada, T. & Guengerich, F.P. (1992) Contributions of human liver cytochrome P450 enzymes to the *N*-oxidation of 4,4′-methylene-bis(2-chloroaniline). *Carcinogenesis*, **13**, 217–222