MAGENTA AND MAGENTA PRODUCTION

Historically, the name Magenta has been used to refer to the mixture of the four major constituents comprising Basic Fuchsin, namely Basic Red 9 (Magenta 0), Magenta I (Rosaniline), Magenta II, and Magenta III (New fuchsin). Although samples of Basic Fuchsin can vary considerably in the proportions of these four constituents, today each of these compounds except Magenta II is available commercially under its own name. Magenta I and Basic Red 9 are the most widely available.

1. Exposure Data

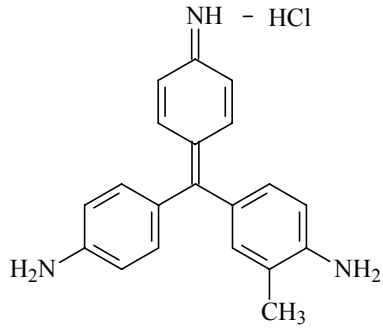
1.1 Chemical and physical data

1.1.1 *Magenta I*

(a) Nomenclature

Chem. Abstr. Serv. Reg. No.: 632–99–5
CAS Name: 4-[(4-Aminophenyl)(4-imino-2,5-cyclohexadien-1-ylidene)methyl]-2-methylbenzenamine, hydrochloride (1:1)
Synonyms: 4-[(4-Aminophenyl)(4-imino-2,5-cyclohexadien-1-ylidene)methyl]-2-methylbenzenamine, monohydrochloride; Basic Fuchsin hydrochloride; C.I. 42510; C.I. Basic Red; C.I. Basic Violet 14; C.I. Basic Violet 14, monohydrochloride; 2-methyl-4,4'-[(4-imino-2,5-cyclohexadien-1-ylidene)methylene]dianiline hydrochloride; rosaniline chloride; rosaniline hydrochloride

(b) Structural formula, molecular formula, and relative molecular mass



$C_{20}H_{19}N_3.HCl$ Rel. mol. mass: 337.85

(c) Chemical and physical properties of the pure substance

Description: Metallic green, lustrous crystals (O'Neil, 2006; Lide, 2008)
Melting-point: Decomposes above 200 °C (O'Neil, 2006; Lide, 2008)
Solubility: Slightly soluble in water (4 mg/mL); soluble in ethanol (30 mg/mL) and ethylene glycol methyl ether (30 mg/mL); insoluble in diethyl ether (Green, 1990; O'Neil, 2006; Lide, 2008)

(d) Trade names

Trade names for Magenta I include: Aizen Magenta; Aniline Red; Astra Fuchsine B; Basic Fuchsine; Basic Fuchsin; Basic Magenta; Basic Magenta E-200; C-WR Violet 8; Calcozine Fuchsine HO; Calcozine Magenta RIN; Calcozine Magenta RTN; Calcozine Magenta XX; Cerise B; Diabasic Magenta; Diamond Fuchsin; Diamond Fuchsine; Fuchsin; Fuchsin Basic; Fuchsine; Fuchsine A; Fuchsine CS; Fuchsine G; Fuchsine HO; Fuchsine N; Fuchsine RTN; Fuchsine SBP; Fuchsine Y; Magenta; Magenta DP; Magenta E; Magenta G; Magenta PN; Magenta S; Magenta Powder N; Magenta Superfine; Magenta Super Fine; Methyl Fuchsin; Mitsui Magenta; Orient Basic Magenta; RGB 20; RGN 10.

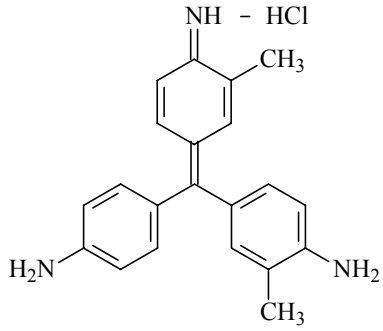
1.1.2 *Magenta II*

(a) Nomenclature

Chem. Abstr. Serv. Reg. No.: 26261–57–4
CAS Name: 4-[(4-Aminophenyl)(4-imino-3-methyl-2,5-cyclohexadien-1-ylidene)methyl]-2-methylbenzenamine, hydrochloride (1:1)
Synonyms: 4-[(4-Aminophenyl)(4-imino-3-methyl-2,5-cyclohexadien-1-ylidene)methyl]-2-methylbenzenamine, monohydrochloride; α4-(*p*-aminophenyl)-

α4-(4-imino-3-methyl-2,5-cyclohexadien-1-ylidene)-2,4-xylidine, monohydrochloride; dimethyl fuchsin

(b) Structural formula, molecular formula, and relative molecular mass



$C_{21}H_2N_3.HCl$ Rel. mol. mass: 351.87

(c) Chemical and physical properties of the pure substance

No information was available to the Working Group.

(d) Technical products and impurities

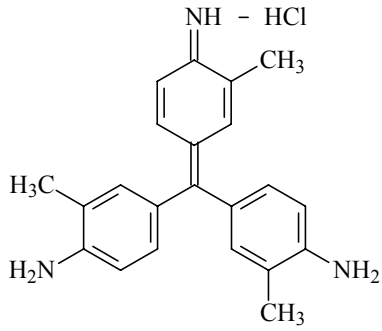
Magenta II is not available as a separate commercial product.

1.1.3 *Magenta III*

(a) Nomenclature

Chem. Abstr. Serv. Reg. No.: 3248–91–7
CAS Name: 4,4'-[(4-imino-3-methyl-2,5-cyclohexadien-1-ylidene)methylene]bis[2-methylbenzenamine], hydrochloride (1:1)
Synonyms: 4-[(4-Amino-3-methylphenyl)(4-imino-3-methyl-2,5-cyclohexadien-1-ylidene) methyl]-2-methylbenzenamine, monohydrochloride; 4-[(4-amino-m-tolyl)(4-imino-3-methylcyclohexa-2,5-dien-1-ylidene)methyl]-o-toluidine, monohydrochloride; Basic Violet 2; C.I. 42520; C.I. Basic Violet 2; C.I. Basic Violet 2, monohydrochloride; 4,4'-[(4-imino-3-methyl-2,5-cyclohexadien-1-ylidene)methylene]bis[2-methylbenzenamine], monohydrochloride

(b) Structural formula, molecular formula, and relative molecular mass



$C_{22}H_{23}N_3.HCl$ Rel. mol. mass: 365.90

(c) Chemical and physical properties of the pure substance

Description: Green crystalline powder (Green, 1990)
Solubility: Soluble in water (20 mg/mL), ethanol (20 mg/mL), and ethylene glycol methyl ether (20 mg/mL) (Green, 1990)

(d) Trade names

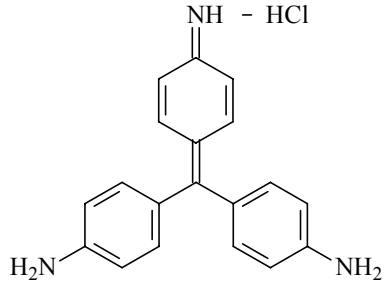
Trade names for Magenta III include: Astra New Fuchsine G; Astrazon Fuchsine GN; Calcozine New Fuchsine; Fuchsin NB; Leather Ruby HF; Magenta ABN; Neofuchsine; New Fuchsin; New Fuchsine; New Fuchsine G Crystal; New Magenta; Remacryl Magenta B.

1.1.4 *Basic Red 9 (Magenta 0)*

(a) Nomenclature

Chem. Abstr. Serv. Reg. No.: 569–61–9
CAS Name: 4,4'-[(4-imino-2,5-cyclohexadien-1-ylidene)methylene]bis[benzenamine], hydrochloride (1:1)
Synonyms: Basic Parafuchsine; Basic Red 9; C.I. 42500; C.I. Basic Red 9; C.I. Basic Red 9, monohydrochloride; *para*-Fuchsin; *para*-Fuchsine; 4,4'-[(4-imino-2,5-cyclohexadien-1-ylidene)methylene]bis[benzenamine], monohydrochloride; 4,4'-(4-iminocyclohexa-2,5-dienylidenemethylene)dianiline hydrochloride; Magenta 0; Parafuchsin; Parafuchsine; Parafuchsin hydrochloride; Para Magenta; Pararosaniline; Pararosaniline chloride; Pararosaniline hydrochloride; *para*-Rosaniline hydrochloride

(b) Structural formula, molecular formula, and relative molecular mass



$C_{19}H_{17}N_3.HCl$ Rel. mol. mass: 323.82

(c) Chemical and physical properties of the pure substance

Description: Pale violet powder (Lide, 2008)
Melting-point: 269 °C (decomposes) (Lide, 2008)
Solubility: Slightly soluble in water (3 mg/mL); soluble in ethanol (25 mg/mL) and ethylene glycol methyl ether (70 mg/mL) (Green, 1990)

(d) Trade names

Trade names for C.I. Basic Red 9 (Magenta 0) include: Calcozine Magenta N; Fuchsine DR 001; Fuchsine SP; Fuchsine SPC; and Orient Para Magenta Base.

1.1.5 *Analysis*

The first reports on analysis of magenta were published during the 1960s. The more recent studies have involved the use of LC/MS, in which laser-desorption electrospray ionization was employed to analyse paper and textiles for the presence of magenta. Assessments using high-resolution TLC, including an electromolecular propulsion method, have been reported (Table 1.1).

An LC-MS method involving electrospray ionization has been developed to distinguish Magenta 0 from its homologues, which have identical UV-visible absorption spectra (Huang *et al.*, 2005).

1.2 Production and use

1.2.1 *Production*

Magenta was among the first synthetic dyes to be produced, beginning in the 1850s (Bannister & Elliot, 1983). Magenta was produced commercially in England (Bannister & Olin, 1965) and in the USA (US Tariff Commission, 1922). In the United Kingdom, the process for manufacturing magenta has involved condensation of *ortho*-toluidine (see

IARC, 1982a, 1987) and formaldehyde (see IARC, 1982b, 1987) in the presence of nitrotoluene, resulting mainly in the production of Magenta III (Howe, 1977). Magenta I is prepared by the reaction of a mixture of aniline (see IARC, 1982a, 1987), *ortho*- and *para*-toluidine and their hydrochlorides with nitrobenzene or a mixture of nitrobenzene and *ortho*-nitrotoluene in the presence of ferrous chloride, ferrous oxide and zinc chloride (US National Library of Medicine, 1992). C.I. Basic Red 9 is prepared by the reaction of aniline with formaldehyde in the presence of hydrogen chloride, forming 4,4'-methylenedianiline (see IARC, 1986), which is then heated with aniline and aniline hydrochloride in the presence of nitrobenzene and ferric chloride (US National Library of Medicine, 1992).

Table 1.1. Selected methods of analysis of magenta in various matrices

Sample matrix	Sample preparation	Assay method	Detection limit	Reference
Solvent mixtures	Apply to stationary phase, develop plate	EMP-TLC	Not given	Haber (1998)
Natural and synthetic samples	Dissolve dye (10^{-3} M) in ethanol–water (4:1, v/v).	IPTLC	500ng	Misra & Gupta (2002)
Dyestuffs and textile fibres	Place dye sample or cut dyed cotton fibre in glass vials, add 2-propanol/water (4:1), heat, remove fibres, and evaporate. Dissolve residue in methanol for analysis.	LC-MS with ESI	5ppm	Huang *et al.*, (2005)

EMP, electromolecular propulsion; ESI, electrospray ionization; IPTLC, ion-pair thin-layer chromatography; LC-MS, liquid chromatography-mass spectrometry; TLC, thin-layer chromatography

Magenta III is prepared by condensation of two moles of *ortho*-toluidine with formaldehyde in nitrobenzene in the presence of iron salts to give the corresponding substituted diphenylmethane base. This base is not isolated, but undergoes an oxidative condensation with another mole of *ortho*-toluidine to produce the dye (Thetford, 2000).

No recent data were available on the production of magenta or C.I. Basic Red 9. Production data for Magenta I in the USA were last reported for 1964, when the combined production of five producers was reported as 53 tonnes (US Tariff Commission, 1965). Production of C.I. Basic Red 9 in the USA was estimated as greater than 0.9 tonnes in 1972 and 0.5 tonnes in 1975 (US National Library of Medicine, 1992).

The USEPA (2003, 2007) Inventory Update Rule regulation requires manufacturers and importers of certain chemical substances listed in the TSCA Chemical Substance Inventory to report manufacturing information (aggregate production volumes) for

chemicals manufactured (including imported) in amounts of 10 000 pounds [4500 kg] or greater (in 1986) or 25 000 pounds [11 000 kg] or greater (in 2003) at a single site. The aggregate production volume reported for Magenta I in 1990 was 10 000-500 000 pounds [4500–227 000 kg].

Available information indicates that Magenta I was produced and/or supplied in research quantities in the following countries: Canada, Germany, Hong Kong Special Administrative Region, India, the Netherlands, Switzerland, the United Kingdom, and the USA (Chemical Sources International, 2008).

Magenta III was produced and/or supplied in research quantities in the following countries: Canada, Germany, India, Japan, Switzerland, and the USA (Chemical Sources International, 2008).

Magenta 0 was produced and/or supplied in research quantities in the following countries: Germany, Hong Kong Special Administrative Region, India, Japan, the Netherlands, Switzerland, and the USA (Chemical Sources International, 2008).

1.2.2 *Use*

The general population can be exposed to magenta through a variety of uses. Under the name of Basic Violet 14, magenta is used in hair dyes and is also used in cosmetic products not intended to come in contact with mucous membranes (EU Directive 76/768/EEC). Magenta stains animal fibres directly and vegetable fibres after mordanting. Under the name of Basic Red 9, magenta is also used as a colourant in artists' paints. Magenta III is used as a thin-layer chromatography developing agent for perfluorinated organics (Williams *et al.*, 1992).

Magenta is antiseptic against gram-positive bacteria and can be used in dermatology for the treatment of pyoderma, dermatitis, intertrigo, eczema, and burns in solutions of 2% to 5% (Balabanova *et al.*, 2003). Known as Castellani's paint or magenta paint, it has been used topically since it was introduced in the 1920s (Castellani, 1968) to treat skin conditions such as fungal skin lesions (Whitwell, 1968) or infective dermatitis (Shelley, 2004). Carbol-Fuchsin solution, containing less than 1% of CI Basic Red 9, is used to treat postoperative phenol nail procedures and as a dermal first-aid antiseptic drying agent (Biogenex, 2003).

Magenta is reported to be used as a food-irradiation dosimeter in an aqueous solution of 3.13×10^{-5} mol/L (Khan & Naz, 2007) and as a meat-marking colour in New Zealand (Dacre, 1971).

1.3 Occurrence

1.3.1 *Natural occurrence*

Magenta is not known to occur as a natural product.

1.3.2 *Occupational exposure*

The only well-described groups of workers exposed to magenta include those in a dyestuffs-manufacturing plant in Ludwigshafen, Germany (Rehn, 1895), the manufacture of magenta in the British chemical industry (1910–1952) (Case & Pearson, 1954) and the manufacture of "new fuchsin" in an Italian dyestuffs factory (Rubino *et al.*, 1982; Piolatto *et al.*, 1991). No environmental or biological measurements have been reported for these plants or any other plants that have produced or are currently producing magenta.

Production of magenta may expose workers to process chemicals (e.g., aniline, *ortho*- and *para*-toluidine, and – historically – arsenic acid). Exposure to other chemicals used and produced at the same location may also occur (e.g., benzidine, 1-naphthylamine, 2-naphthylamine, auramine, aniline) (Case & Pearson, 1954).

Occupational exposure can also occur during the use of magenta as a dye intermediate and when dyeing textile fibres, fabrics and paper products (IARC, 1993). The levels of exposure to magenta have not been reported for these occupational groups.

Exposure to magenta can also occur in laboratories, where it is widely used as a biological stain (basic fuchsin dye) to stain bacteria and as a component of Schiff's reagent to detect aldehydes. Under laboratory conditions, magenta has also been produced directly on nylon fabric by a fungal strain closely related to *Phoma herbarum* (Chiba *et al.*, 2006).

The US National Exposure Survey (1981–1983) estimates the number of employees potentially exposed to Basic Red 9 as approximately 900, the largest group being laboratory technicians and medical scientists. A total of around 12 700 workers were potentially exposed to Magenta I in six industries (NIOSH, 1990).

1.3.3 *Exposure of the general population*

Magenta may be present in the waste effluents from plants where it is produced or used. Concentrations of magenta in water or soil have not been reported. Wastewater containing dyes from the textile industry is difficult to treat using conventional methods, because the dyes are stable to light and oxidizing agents, and are resistant to aerobic digestion (Rai *et al.*, 2007). Various methods for the degradation of magenta in wastewater have been developed (Li *et al.*, 1999; Yiyun & Jiepin, 2005).

1.4 Regulations and guidelines

1.4.1 *Magenta*

(a) Germany

The MAK Commission listed Magenta (basic fuchsin hydrochloride) as a substance being examined for the establishment of MAK values based on its carcinogenic effects (MAK, 2007).

(b) Japan

The Japan Society for Occupational Health (2007) follows the classification by IARC of Magenta (containing C.I. Basic Red 9) in Group 2B.

1.4.2 *Basic Red 9 (Magenta 0)*

(a) Europe

(i) *Commission Directive 2000/32/EC*

Commission Directive 2000/32/EC of 19 May 2000 adapts to technical progress for the 26th time Council Directive 67/548/EEC on the approximation of the laws, regulations and administrative provisions relating to the classification, packaging and labelling of dangerous substances (European Commission, 2000). 4,4'-(4-iminocyclohexa-2,5-dienylidenemethylene)dianiline hydrochloride (C.I. Basic Red 9) is listed in Annex I to Directive 67/548/EEC, which contains a list of dangerous substances, together with details on the classification and labelling of each substance.

(ii) *Directive 2004/37/EC*

C.I. Basic Red 9 is regulated by Directive 2004/37/EC (European Commission, 2004), which applies to activities in which workers are exposed to carcinogens or mutagens of Category 1 or 2. Rules are fixed regarding the employers' obligations of reduction and replacement, prevention and reduction of exposure, unforeseen exposure, foreseeable exposure, access to risk areas, hygiene and individual protection, information for the competent authority, information and training of workers, consultation and participation of workers, health surveillance, record keeping and limit values.

(b) Japan

The Japan Society for Occupational Health (2007) follows the classification by IARC of C.I. Basic Red 9 in Group 2B.

(c) USA

C.I. Basic Red 9 monohydrochloride is listed in the NTP *Report on Carcinogens* as *reasonably anticipated to be a human carcinogen* (NTP, 2005).

2. Studies of Cancer in Humans

2.1 Case report

Rehn (1895) was the first to report the appearance of bladder tumours in three of 45 workers involved in the manufacture of fuchsin in one factory in Germany. At the time, this process involved heating a mixture of chemicals, including toluidine, aniline

and nitrobenzene to obtain crude fuchsin, which was then purified and crystallized. The three workers had been employed for 15–29 years. One worker developed a "fibroma papillare", another an oedematous papilloma, and the third a carcinoma of the bladder.

2.2 Cohort studies

Case & Pearson (1954) surveyed workers employed for at least six months between 1910 and 1952 in the manufacture of magenta in the British chemical industry. Workers who had been exposed to benzidine or 1- or 2-naphthylamine were excluded. Bladder-cancer occurrence was determined from factory and hospital records. Deaths were identified from alphabetical lists of death certificates, and the numbers were compared with mortality rates for England and Wales for the period 1921–1952. Among 85 magenta production workers known to have been in contact with magenta, but not exposed to auramine, 1- or 2-naphthylamine or benzidine, there were five cases of bladder cancer, with exposure duration ranging from 1–19 years. Three of these cases were mentioned on death certificates, whereas only 0.13 would have been expected for the whole male population in England and Wales (SMR 23.1; $P < 0.005$). One case of bladder cancer was observed among nine subjects who had been exposed to both magenta and auramine, but no death was seen from this cause (0.02 expected). [The Working Group noted that during the manufacture of magenta exposure to other aromatic amines cannot be ruled out.]

Rubino *et al.* (1982) studied case-specific mortality of 53 male workers who had been employed for ≥ 1 month in the manufacture of 'new fuchsin' (Magenta III, Basic Violet 2, CI No. 42520) and Safranine T (Basic Red 2, CI No. 50240) during the period 1922–1970 in a factory in the province of Torino, Italy. Workers engaged in the manufacture and use of 1- or 2-naphthylamine or benzidine were excluded from the study. Subjects and their work histories were identified from factory personnel records, and the workers were followed for mortality from 1946 to 1976, as identified from factory records and from the municipal registries of their current residence. Among the 53 workers exposed, five deaths from bladder cancer were observed, while 0.08 were expected on the basis of mortality rates for Italy in 1951–76 (SMR 62.5; $P < 0.001$). The cases had been occupationally exposed to Magenta III and Safranine T for 12–40 years. The exposures also involved *ortho*-toluidine and 4,4'-methylenebis(2-methylaniline). The same cohort of workers was followed to the end of 1981 (Decarli *et al.*, 1985) and further until 1989 (Piolatto *et al.*, 1991). No additional deaths from bladder cancer were reported.

2.3 Case–control studies

A total of 512 male cases of bladder cancer and 596 hospital-based controls were studied for occupation and bladder-cancer risk during 1978–1983 in the province of Torino, Italy (Vineis & Magnani, 1985). Complete occupational histories and related information were obtained via hospital interviews. Exposures to specific chemicals,

including magenta, were estimated from ILO occupation and industry titles, using information on the industrial uses of these chemicals as described in published sources. On the basis of industrial branches in which magenta exposure could have occurred, 41 cases were classified as having been exposed before the age of 60, to give a relative risk of 1.8 (95% confidence interval, 1.1–2.9). On the basis of job titles in which exposure to magenta could have occurred, two cases were classified as having been exposed before the age of 60, with an associated relative risk of 3.0 (95% confidence interval, 0.4–20.0).

3. Studies of Cancer in Experimental Animals

3.1 Magenta

3.1.1 *Oral administration*

(a) Mouse

A group of sixty stock mice (30 males, 30 females; strain and age unspecified) received 12 mg magenta (BDH; purity not specified) in arachis oil by gastric instillation once per week for 52 weeks (total dose, 624 mg), and were kept for their life-span. Eighteen mice were used as controls. Four (20%) lymphomas and one (5%) hepatoma were found in 20 surviving treated animals, and five (25%) lymphomas were found in 18 controls (Bonser *et al.* 1956).

(b) Rat

A group of eighty Sprague Dawley rats (40 males, 40 females), 12 weeks of age, were treated by intragastric instillation with magenta (purity unspecified) dissolved in saline, twice weekly for life. Initial dose was 400 mg/kg bw per week, but due to severe toxicity the treatment had to be interrupted and continued at half the original dose level. A control group of similar size received the solvent only. Tumour incidences reported were 5% for the magenta-treated rats and 40% for the controls (males and females combined) (Ketkar & Mohr 1982).

(c) Hamster

Syrian golden hamsters (40 males, 40 females), 12 weeks of age, were treated by intragastric instillation with magenta dissolved in saline, twice weekly for life. Dose levels were 400 and 600 mg/kg bw. The 600-mg/kg bw dose was not tolerated by the animals. A control group was given the solvent only. Tumour incidences reported at the low dose were 5% (one nasal cavity fibroma and a bronchiogenic adenoma in two male hamsters; one adrenal cortex adenoma and one submandibular gland adenoma in two females), and 10% for the solvent controls (except for one adrenal cortex adenoma, all

nine other tumours in eight control animals – three males, five females – were seen at sites different from those in the treated hamsters) (Green *et al.*, 1979).

3.2 CI Basic Red 9 (*para*-magenta)

3.2.1 *Oral administration*

(a) Mouse

Groups of 50 male and 50 female B6C3F$_1$ mice, 6–10 weeks of age, were given a diet containing 0, 500 or 1000 mg/kg (ppm) CI Basic Red 9 for 103 weeks and were killed at 110–115 weeks of age. Two lots of the test chemical were used, with purities of 93 and 99% (water was the major impurity). Mean body weights of treated mice were lower than those of controls throughout the study. At the end of the experiment, 42/50 (84%) control; 32/50 (64%) low-dose; 35/50 (70%) high-dose males and 31/50 (62%) control, 12/50 (24%) low-dose, and 6/50 (12%) high-dose females were alive ($P < 0.001$). In male mice, CI Basic Red 9 caused a dose-related increase in the incidence of hepatocellular carcinomas (10/50 (20%) control; 20/50 (40%) low-dose; 27/50 (54%) high-dose; $P < 0.001$, incidental tumour trend test). The incidence of hepatocellular adenomas was 22/50 (44%) control, 21/50 (42%) low-dose, and 17/50 (34%) high-dose. The combined incidence of liver tumours was 29/50 (58%) control, 37/50 (74%) low-dose, and 41/50 (82%) high-dose ($P = 0.005$, incidental tumour trend test). In female mice, the compound caused a dose-related increase in the incidence of hepatocellular carcinomas (3/49 (6.1%) control; 19/50 (38%) low-dose; 37/49 (75%) high-dose; $P < 0.001$, Cochran-Armitage trend test). The incidence of hepatocellular adenomas was 2/49 (4%) control, 18/50 (36%) low-dose and 4/49 (8%) high-dose ($P < 0.001$, Fischer exact test). The combined incidence of liver tumours in females was 5/49 (10%) control, 35/50 (70%) low-dose, and 41/49 (84%) high-dose ($P < 0.001$, Cochran-Armitage trend test). An increase in the incidence of benign and malignant adrenal phaeochromocytomas (combined) was found in females (1/48 (2%) control; 8/47 (17%) low-dose; 8/45 (18%) high-dose; $P = 0.015$, Cochran-Armitage trend test) (NTP, 1986).

(b) Rat

Groups of 40 male and 40 female Sprague Dawley rats, 12 weeks of age, were treated intragastrically twice a week with 0 or 600 mg/kg bw CI Basic Red 9 [purity unspecified] in 0.9% saline. The dose of 600 mg/kg was found to be toxic and, after 12 weeks, treatment was discontinued for one week; after a further six weeks, half of the original dose (300 mg/kg bw) was used for the remaining treatment, for life. Average survival times were 104 weeks for control males, 70 weeks for treated males, 92 weeks for control females and 69 weeks for treated females. No treatment-related increase in the incidence of tumours was observed in rats of either sex (Ketkar & Mohr, 1982). [The Working Group noted the poor survival in the treated groups and the inadequate reporting of the study.]

Groups of 50 male and 50 female Fischer 344/N rats, 6–7 weeks of age, were given a diet containing 0, 1000 or 2000 mg/kg (ppm) (males) and 0, 500 or 1000 ppm (females) CI Basic Red 9 for 103 weeks and were killed at 110–113 weeks of age. Two lots of the test chemical were used, with purities of 93 and 99% (water was the major impurity). Increased mortality was seen in high-dose males and females, and at the end of the experiment, 36/50 (72%) control, 29/50 (58%) low-dose and 0/50 high-dose males and 37/50 (74%) control, 35/50 (70%) low-dose, and 14/50 (28%) high-dose females were still alive. CI Basic Red 9 caused significant increases in the incidences of benign and malignant tumours at various sites in both males and females (Table 3.1) (NTP, 1986).

Table 3.1. Trends in tumour incidences at specific sites in Fischer 344/N rats fed diets containing CI Basic Red 9

Tumour site and type	Control	Low-dose	High-dose	*P* (trend)[a]
Male				
Dose (mg/kg diet)	0	1000	2000	
Skin				
Squamous-cell carcinoma	0/50 -	1/50 (2%)	10/50 (20%)	< 0.001
Trichoepithelioma	0/50 -	0/50 -	7/50 (14%)	= 0.001
Sebaceous adenoma	0/50 -	0/50 -	5/50 (10%)	= 0.006
Subcutis				
Fibroma	2/50 (4%)	20/50 (40%)	16/50 (32%)	< 0.001
Zymbal gland				
Carcinoma	1/50 (2%)	1/50 (2%)	13/50 (26%)	< 0.001
Thyroid gland				
Follicular adenoma	0/49 -	0/46	9/44 (20%)	< 0.001
Follicular carcinoma	0/49 -	5/46 (11%)	18/44 (41%)	< 0.001
Combined	0/49 -	5/46 (11%)	25/44 (57%)	< 0.001
Liver				
Hepatocellular neoplastic nodule	5/50 (10%)	14/50 (28%)	6/50 (12%)	= 0.447
Hepatocellular carcinoma	0/50 -	2/50 (4%)	8/50 (16%)	= 0.001
Combined	5/50 (10%)	15/50 (30%)	14/50 (28%)	= 0.021
Females				
Dose (mg/kg diet)	0	500	1000	
Subcutis				
Fibroma	0/50 -	15/50 (30%)	10/50 (20%)	= 0.005
Zymbal gland				
Carcinoma	0/50 -	2/50 (4%)	7/50 (14%)	= 0.003
Thyroid				
Follicular adenoma	0/47 -	0/48 -	4/50 (8%)	= 0.017
Follicular carcinoma	0/47 -	2/48 (4%)	2/50 (4%)	> 0.05
Combined	0/47 -	2/48 (4%)	6/50 (12%)	= 0.009

From NTP (1986)
[a] Cochran-Armitage trend test

(c) Hamster

Syrian golden hamsters (40 males, 40 females), 12 weeks of age, were treated by intragastric instillation with CI Basic Red 9 dissolved in saline, twice weekly for life. Dose levels were 300 and 600 mg/kg bw. The high dose was not tolerated by the animals. Tumour incidences reported at the low dose were 6% (one adrenal cortex adenoma, one tracheal papillary polyp and two intestinal adenocarcinomas, all in one male hamster; one tracheal papillary polyp, one subcutaneous fibrosarcoma and two intestinal adenocarcinomas in four females), and 10% for the solvent controls (except for one adrenal cortex adenoma, all nine other tumours in eight control animals – three males, five females – were seen at sites different from those in the treated hamsters) (Green *et al.*, 1979).

3.2.2 *Subcutaneous administration*

(a) Rat

Twenty BD III rats received 10 mg *para*-magenta as a 1% aqueous solution by subcutaneous injection once per week for 65 weeks (total dose, 650 mg). The first local sarcoma appeared at 10 months (total dose, 370 mg); six more were observed in subsequent months in 12 surviving animals. The spontaneous incidence of sarcomas in these rats was < 0.5% (Druckrey *et al.* 1956). [The Working Group noted the lack of use of controls and the lack of adequate description of the experiment.]

4. Mechanistic and Other Relevant Data

4.1 Absorption, distribution, metabolism, elimination

No data were available to the Working Group.

4.2 Genetic and related effects

4.2.1 *Magenta* (see Tables 4.1 and 4.2)

[The data reported in the Table are the results of experiments with commercial preparations of magenta; in most cases, the exact composition of the products tested and the degree of purity is not known. In the experiments with Magenta I the test compound seems to be better identified, but also in this case its degree of purity is not reported.]

Magenta came out negative in the prophage-induction test, both in the absence and the presence of metabolic activation (Speck *et al.*, 1978). It was mutagenic in *Salmonella typhimurium* strains TA98, TA100, and TA1535, but only in the presence of metabolic activation. The effect was observed with TA98 (Mortelmans *et al.*, 1986), TA100

Table 4.1. Genetic and related effects of Magenta and Magenta 1

Test system	Result[a]		Dose (HID/LED)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Magenta				
Prophage induction/SOS/strand breaks/x-links	–	–	500 µg/ml	Speck *et al.* (1978)
S. typhimurium TA100, reverse mutation	–	+	50 µg/plate	Mortelmans *et al.* (1986)
S. typhimurium TA100, reverse mutation	–	+	167 µg/plate	Dunkel *et al.* (1984)
S. typhimurium TA100, reverse mutation	NT	+	5 µg/plate	Yamaguchi (1988)
S. typhimurium TA100, TA98, reverse mutation	–	–	1000/5000 µg/plate	Japan Chemical Industry Ecology (JETOC) (1996)
S. typhimurium TA1535, TA1538, reverse mutation	–	–	125 µg/plate	Rosenkranz & Poirier (1979)
S. typhimurium TA1535, reverse mutation	–	+	17 µg/plate	Mortelmans *et al.* (1986)
S. typhimurium TA1535, TA1537, TA1538, TA98, reverse mutation	–	–	167 µg/plate	Dunkel *et al.* (1984)
S. typhimurium TA1535, TA1537, TA1538, reverse mutation	–	–	200/5000 µg/plate	Japan Chemical Industry Ecology (JETOC) (1996)
S. typhimurium TA1537, reverse mutation	–	–	17 µg/plate	Mortelmans *et al.* (1986)
S. typhimurium TA98, reverse mutation	–	+	166 µg/plate	Mortelmans *et al.* (1986)

Table 4.1 (contd)

Test system	Result[a]		Dose (HID/LED)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
E. coli WP2 *uvrA*, reverse mutation	–	–	166 µg/plate	Dunkel *et al.* (1984)
E. coli WP2 *uvrA*, reverse mutation	–	–	1000/5000 µg/plate	Japan Chemical Industry Ecology (JETOC) (1996)
S. cerevisiae, homozygosis	–	–	300 µg/ml	Simmon (1979b)
Unscheduled DNA synthesis, rat primary hepatocytes	–	NT	1 µg/ml	Williams *et al.* (1982)
Cell transformation, SHE, clonal assay	–	NT	1 µg/ml	Pienta *et al.* (1977)
Magenta 1				
S. typhimurium TA100, reverse mutation	–	+	17 µg/ml	Mortelmans *et al.* (1986)
S. typhimurium TA1535, TA1537, reverse mutation	–	–	16.5 µg/plate	Mortelmans *et al.* (1986)
S. typhimurium TA98, reverse mutation	–	+	166 µg/plate	Mortelmans *et al.* (1986)

[a] +, positive; –, negative
HID, highest ineffective dose; LED, lowest effective dose; NT, not tested

Table 4.2. Genetic and related effects of CI Basic Red 9 (para-rosaniline)

Test system	Result[a]		Dose (LED/HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Prophage induction/SOS repair test, DNA strand breaks, cross-links or related damage	–	NT	500 µg/ml	Speck *et al.* (1978)
Escherichia coli pol A^+/pol A^-W3110-P3478 differential toxicity (liquid suspension)	+	NT	20 µg/ml	Rosenkranz & Poirier (1979)
Escherichia coli WP2/WP67/CM871, differential toxicity	+	–	155 µg/ml	De Flora *et al.* (1984a)
Salmonella typhimurium TA100, TA1535, TA1537, TA1538, TA98, TA1586, reverse mutation	–	–	250 µg/plate	Simmon (1979a)
Salmonella typhimurium TA100, T1535, TA1537, reverse mutation	–	–	1000 µg/plate	Bonin *et al.* (1981)
Salmonella typhimurium TA100, TA1535, TA1537, TA1538, TA98, reverse mutation	–	–	1070 µg/plate	De Flora (1981)
Salmonella typhimurium TA100, reverse mutation	–	+	167 µg/plate	Dunkel *et al.* (1984)
Salmonella typhimurium TA100, reverse mutation	–	+	17 µg/plate	Mortelmans *et al.* (1986)
Salmonella typhimurium TA102, reverse mutation	–	–	4 µg/ml	De Flora *et al.* (1984b)
Salmonella typhimurium TA1535, TA1538, reverse mutation	–	–	125 µg/plate	Rosenkranz & Poirier (1979)
Salmonella typhimurium TA1535, TA1537, TA1538, reverse mutation	–	–	167 µg/plate	Dunkel *et al.* (1984)

Table 4.2 (contd)

Test system	Result[a]		Dose (LED/HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Salmonella typhimurium TA1535, reverse mutation	–	(+)	500 µg/plate	Mortelmans *et al.* (1986)
Salmonella typhimurium TA1537, reverse mutation	–	–	167 µg/plate	Mortelmans *et al.* (1986)
Salmonella typhimurium TA1538, reverse mutation	–	+	1000/320 µg/plate	Bonin *et al.* (1981)
Salmonella typhimurium TA98, reverse mutation	–	(+)	167 µg/plate	Dunkel *et al.* (1984)
Salmonella typhimurium TA98, reverse mutation	NT	+	0.2 µg/ml	Arni *et al.* (1985)
Salmonella typhimurium TA98, reverse mutation	–	+	50 µg/plate	Mortelmans *et al.* (1986)
Salmonella typhimurium TA97, reverse mutation	–	(+)	1600 µg/plate	De Flora *et al.*(1984b)
Escherichia coli exclusive of strain K12, forward mutation	+	(+)	5000 µg/plate	Hayes *et al.* (1984)
Escherichia coli WP2 *uvrA*, reverse mutation	–	–	167 µg/plate	Dunkel *et al.* (1984)
Saccharomyces cerevisiae, homozygosis by mitotic recombination	–	–	300 µg/plate	Simmon (1979b)
Unscheduled DNA synthesis, rat primary hepatocytes	–	NT	1 µg/ml	Williams *et al.* (1982)
Unscheduled DNA synthesis, rat primary hepatocytes	+	NT	2.2 µg/ml	NTP (1986)
Unscheduled DNA synthesis, rat primary hepatocytes	–	NT	3.24 µg/ml	Kornbrust & Barfknecht (1984)

Table 4.2 (contd)

Test system	Result[a]		Dose (LED/HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Unscheduled DNA synthesis, Syrian hamster primary hepatocytes	+	NT	3.24 µg/ml	Kornbrust & Barfknecht (1984)
Gene mutation, mouse lymphoma L5178Y cells, *Tk* locus	+	+	1 µg/ml	Mitchell *et al.* (1988)
Gene mutation, mouse lymphoma L5178Y cells, *Tk* locus	?	–	7.5/100 µg/ml	Myhr & Caspary (1988)
Gene mutation, Chinese hamster CHL/IU cell line with *gpt* shuttle vector	–	NT	20 µg/ml	Yamada *et al.* (2000)
Sister chromatid exchange, Chinese hamster cells *in vitro*	–	–	15 µg/ml	Anderson *et al.* (1990)
Cromosomal aberrations, Chinese hamster cells *in vitro*	–	–	50 µg/ml	Anderson *et al.* (1990)
Cromosomal aberrations, Syrian hamster embryo cells *in vitro*	+	NT	0.29 µg/ml	Hagiwara *et al.* (2006)
Cromosomal aberrations, Chinese hamster cells CHL/IU *in vitro*	–	NT	20 µg/ml	Yamada *et al.* (2000)
Cell transformation, BALB/c3T3 mouse cells	+	NT	0.04 µg/ml	Dunkel et al. (1981)
Cell transformation, Syrian hamster embryo cells, clonal assay	–	NT	1 µg/ml	Pienta *et al.* (1977)

Table 4.2 (contd)

Test system	Result[a]		Dose (LED/HID)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
Cell transformation, Syrian hamster embryo cells, clonal assay	–	+	2 μg/ml	Pienta & Kawalek (1981)
Cell transformation, RLV/Fischer rat embryo cells	+	NT	1.4 μg/ml	Dunkel et al. (1981)
Urine from mouse and rat, microbial mutagenicity	+	+	120 mg/kg x 2 po	Lawlor *et al.* (1987)
Host-mediated assay, *Salmonella typhimurium* in mice	–		1600 mg/kg x 1 po	Simmon *et al.* (1979)
Host-mediated assay, *Salmonella typhimurium* in mice	–		1600 mg/kg x 1 im	Simmon *et al.* (1979)
Host-mediated assay, *Saccharomyces cerevisiae* in mice	–		1600 mg/kg x 1 po	Simmon *et al.* (1979)

[a] +, positive; (+), weakly positive; –, negative; ?, inconclusive (variable response in several experiments within an adequate study)
HID, highest ineffective dose; LED, lowest effective dose; NT, not tested

(Dunkel *et al.*, 1984; Mortelmans *et al.*, 1986; Yamaguchi, 1988), and TA1535 (Mortelmans *et al.*, 1986). Magenta was negative with TA1537 (Dunkel *et al.*, 1984; Mortelmans *et al.*, 1986; JETOC, 1996), TA98 and TA100 (JETOC, 1996) and TA1538 (Rosenkranz and Poirier, 1979; Dunkel *et al.*, 1984; JETOC, 1996). In both the absence and the presence of metabolic activation, it did not induce reverse mutations in *Escherichia coli* (Dunkel *et al.*, 1984; JETOC, 1996) or homozygosis with *Saccharomyces cerevisiae* (Simmon, 1979b). In cultured, non-human mammalian cells, magenta did not induce unscheduled DNA synthesis in primary rat hepatocytes (Williams *et al.*, 1982) and, tested only in the absence of metabolic activation, failed to induce morphological transformation in Syrian hamster embryo cells (Pienta *et al.*, 1977). Mutagenic activity of Magenta I in bacteria was similar to that of magenta; it showed a positive result, but only in the presence of metabolic activation, in *Salmonella typhimurium* TA98 and TA100, and gave a negative response in TA1535 and TA1537 (Mortelmans *et al.*, 1986).

These data indicate that in the presence of an exogenous system of activation, both magenta and Magenta I are mutagenic in bacteria.

4.2.2 *CI Basic Red 9 (*para-*rosaniline)* (see Table 4.2)

[In the large majority of the studies reported in the Table, information on the degree of purity of CI Direct Red 9, a common constituent of magenta, is lacking; commercial products were used.]

In the absence of metabolic activation, CI Basic Red 9 came out negative in the prophage-induction test (Speck *et al.*, 1978), but induced repairable DNA damage in two *E. coli* differential toxicity assays (Rosenkranz & Poirier, 1979; De Flora *et al.*, 1984a). It was more often non-mutagenic in *Salmonella typhimurium* (Simmon 1979a; Bonin *et al.*, 1981; De Flora, 1981). A positive response was obtained only in the presence of metabolic activation with TA98 (Dunkel *et al.*, 1984; Arni *et al.*, 1985; Mortelmans *et al.*, 1986), with TA100 (Dunkel *et al.*, 1984; Mortelmans *et al.*, 1986), with TA1535 (Mortelmans *et al.*, 1986), with TA1538 (Bonin *et al.*, 1981), and with TA97 (De Flora *et al.*, 1984b). The response was negative with TA1537 (Mortelmans *et al.*, 1986), with TA102 (De Flora *et al.*, 1984b) and TA1586 (Simmon, 1979a). CI Basic Red 9 induced forward mutation in *Escherichia coli* in both the absence and the presence of metabolic activation, but not reverse mutation (Hayes *et al.*, 1984). In a study with *Saccharomyces cereviasiae*, CI Basic Red 9 did not induce homozygosis by mitotic recombination (Simmon, 1979b).

In cultured, non-human mammalian cells, both positive and negative results were obtained. CI Basic Red 9 induced unscheduled DNA synthesis in Syrian hamster hepatocytes (Kornbrust & Barfknecht, 1984) and in one study with rat primary hepatocytes (NTP, 1986) and was negative in two other studies in rats (Williams *et al.*, 1982; Kornbrust & Barfknecht, 1984). Mutation at the thymidine kinase (*Tk*) locus in mouse lymphoma cells was induced in both the absence and the presence of metabolic

activation in only one of two studies (Mitchell *et al.*, 1988), and a negative result was obtained in a different gene-mutation assay (Myhr & Caspary, 1988; Yamada *et al.*, 2000). Similarly, chromosomal aberrations were induced in one study (Hagiwara *et al.*, 2006), but not in two others (Anderson *et al.*, 1990; Yamada *et al.*, 2000), and sister chromatid exchange was not observed (Anderson *et al.*, 1990). Variable responses were obtained in four assays for cell transformation: two positive (Dunkel *et al.*, 1981) and two negative (Pienta *et al.*, 1977; Pienta & Kawalek, 1981) results in the absence of metabolic activation, and one positive result in the presence of an exogenous metabolic system (Pienta & Kawalek, 1981). Oral administration of CI Basic Red 9 to mice or rats resulted in urine that was mutagenic to bacteria (Lawlor *et al.*, 1987), but in a host-mediated assay the dye did not induce mutation in *Salmonella typhimurium* recovered from the peritoneal cavity of mice (Simmon *et al.*, 1979).

The inconsistency in the genotoxicity data could be attributed to the different grades of purity of the material employed.

5. Summary of Data Reported

5.1 Exposure data

Historically, the name magenta has been used to refer to a mixture of the four major constituents comprising Basic Fuchsin, namely Basic Red 9, Magenta I, Magenta II and Magenta III. Today all except Magenta II are available commercially under their own names. Magenta I and Basic Red 9 are the most widely available.

Magenta has not been reported to occur as such in nature. Under the name of Basic Violet 14 (Magenta I), magenta is used in hair dyes and in cosmetic products not intended to come in contact with mucous membranes. Basic Red 9 is used as colourant in artist's paints.

Magenta has antiseptic properties; it has been used in dermatology since the 1920s under the name of Castellani's paint or magenta paint. Carbol-Fuchsin solution, which contains Basic Red 9, is used topically as a first-aid antiseptic drying agent. It is also widely used in laboratories as a biological stain.

Magenta is used as a food-irradiation dosimeter and was used as a meat-marking colour in New Zealand.

Occupational exposure to magenta can occur during its production, during its use as a dye intermediate, when dyeing textile fibres, fabrics and paper products, and in laboratories.

Production of magenta may involve exposure to process chemicals such as aniline, *ortho*- and *para*-toluidine, and – historically – arsenic acid. Exposure to other chemicals used and produced at the same location may also occur (e.g., benzidine, 1-naphthylamine, 2-naphthylamine, auramine, aniline).

Magenta may be present in the waste effluents from plants where it is produced or used.

5.2 Human carcinogenicity data

One case report from Germany and two small cohort studies from the United Kingdom and Italy have reported an increased risk for bladder cancer among workers engaged in the manufacture of magenta. These cohort studies excluded workers exposed to benzidine and β-naphthylamine. One case–control study from Italy has shown an increased risk for bladder cancer associated with occupational exposure to magenta. Taken together, these studies indicate that excess bladder cancer risks are caused by the production of magenta, but co-exposures preclude a similar evaluation for magenta itself.

5.3 Animal carcinogenicity data

No adequate study was available to evaluate the carcinogenicity of magenta in experimental animals.

CI Basic Red 9 (Magenta 0) was tested for carcinogenicity in one study in mice and in one study in rats by oral administration in the diet, and in one study in rats by subcutaneous administration. After oral administration, the compound induced hepatocellular carcinomas in male and female mice and in male rats; adrenal gland phaeochromocytomas in female mice; benign and malignant skin tumours in male rats; and subcutaneous fibromas, thyroid gland follicular-cell tumours, and Zymbal gland carcinomas in male and female rats. Subcutaneous administration to rats resulted in a high incidence of local sarcomas. A study in hamsters was found inadequate for evaluation.

5.4 Other relevant data

There are no data on the toxicokinetics of magenta or CI Basic Red 9.

Few data are available on the mutagenicity and genotoxicity of magenta and Magenta I. Both were mutagenic in *S. typhimurium* TA100 and TA 98 (magenta in two out of four experiments) in the presence of an exogenous metabolic activation system, but they were inactive in all other strains tested. Magenta did not induce homozygosis in *S. cerevisiae*, unscheduled DNA synthesis (UDS) in primary hepatocytes or cell transformation of SHE cells.

The data for CI Basic Red 9 are also inconsistent. In the mutagenicity tests in *Salmonella*, it was always negative in the absence of an exogenous system of activation, while in the presence of bioactivation, the results were clearly positive only in strain TA100 (two out of three experiments). Similar conflicting results were obtained in different tests in *E. coli*.

In mammalian cells *in vitro*, CI Basic Red 9 was positive for UDS induction in one of three experiments in rat hepatocytes and in one study with hamster hepatocytes. In a

recent test for chromosomal aberrations in Syrian hamster embryo cells, the compound was positive, in the absence of activation, at a very low dose (0.29 microg/mL), while it was negative for the same test in two different lines of Chinese hamster cells. Similar conflicting results were obtained in other tests *in vitro*.

Mutagenicity was detected, both in the absence and in the presence of metabolic activation, in the urine of mice and rats fed with a diet containing CI Basic Red 9. However, the host-mediated assay with *Salmonella* and *S. cerevisiae* in mice, in the absence of a system of activation, was consistently negative.

6. Evaluation

6.1 Cancer in humans

There is *sufficient evidence* in humans for the carcinogenicity of magenta production. Magenta production causes bladder cancer in humans.

There is *inadequate evidence* in humans for the carcinogenicity of magenta.

6.2 Cancer in experimental animals

There is *sufficient evidence* in experimental animals for the carcinogenicity of CI Basic Red 9 (para-magenta).

There is *inadequate evidence* in experimental animals for the carcinogenicity of magenta.

6.3 Overall evaluation

Magenta production is *carcinogenic to humans (Group 1)*.

7. References

Anderson BE, Zeiger E, Shelby MD *et al.* (1990). Chromosome aberration and sister chromatid exchange test results with 42 chemicals. *Environ Mol Mutagen*, 16 Suppl 18;55–137. doi:10.1002/em.2850160505 PMID:2091924

Arni P, Dollenmeier P, Müller D (1985). Automated modification of the Ames test with COBAS Bact. *Mutat Res*, 144:137–140. doi:10.1016/0165-7992(85)90129-0 PMID:3903483

Balabanova M, Popova L, Tchipeva R (2003). Dyes in dermatology. *Clin Dermatol*, 21:2–6. doi:10.1016/S0738-081X(02)00330-9 PMID:12609582

Bannister DW, Elliot J (1983) Triphenylmethane and related dyes. In: Mark HF, Othmer DF, Overberger CG, Seaborg GT & Grayson N eds, *Kirk-Othmer Encyclopedia of Chemical Technology*, 3rd ed., Vol. 23, New York, John Wiley & Sons, pp. 399–412.

Bannister DW, Olin AD (1965) Dyes and dye intermediates. In Kirk RE & Othmer DF eds, *Encyclopedia of Chemical Techology*, 2nd Ed, Vol. 7, New York, John Wiley & Sons, p. 464.

Biogenex (2003) *Safety data sheet Carbol-fuchsin solution.* Available at: http://www.biogenex.com

Bonin AM, Farquharson JB, Baker RS (1981). Mutagenicity of arylmethane dyes in Salmonella. *Mutat Res*, 89:21–34. doi:10.1016/0165-1218(81)90127-0 PMID:6165887

Bonser GM, Clayson DB, Jull JW (1956). The induction of tumours of the subcutaneous tissues, liver and intestine in the mouse by certain dye-stuffs and their intermediates. *Br J Cancer*, 10:653–667. PMID:13426377

Case RA, Pearson JT (1954). Tumours of the urinary bladder in workmen engaged in the manufacture and use of certain dyestuff intermediates in the British chemical industry. II. Further consideration of the role of aniline and of the manufacture of auramine and magenta (fuchsine) as possible causative agents. *Br J Ind Med*, 11:213–216. PMID:13182161

Castellani A (1968). Magenta paint. *Lancet*, 292:287. doi:10.1016/S0140-6736(68)92397-0.

Chemical Sources International (2008) ChemSources database. [http://db2.chemsources.com]

Chiba S, Tsuyoshi N, Fudou R *et al.* (2006). Magenta pigment produced by fungus. *J Gen Appl Microbiol*, 52:201–207. doi:10.2323/jgam.52.201 PMID:17116968

Dacre JC (1971). Carcinogenic compounds on edible meat in New Zealand. *N Z Med J*, 73:74–77. PMID:5279918

De Flora S (1981). Study of 106 organic and inorganic compounds in the Salmonella/microsome test. *Carcinogenesis*, 2:283–298. doi:10.1093/carcin/2.4.283 PMID:7023727

De Flora S, Camoirano A, Zanacchi P, Bennicelli C (1984b). Mutagenicity testing with TA97 and TA102 of 30 DNA-damaging compounds, negative with other Salmonella strains. *Mutat Res*, 134:159–165. PMID:6390191

De Flora S, Zanacchi P, Camoirano A *et al.* (1984a). Genotoxic activity and potency of 135 compounds in the Ames reversion test and in a bacterial DNA-repair test. *Mutat Res*, 133:161–198. PMID:6374443

Decarli A, Peto J, Piolatto G, La Vecchia C (1985). Bladder cancer mortality of workers exposed to aromatic amines: analysis of models of carcinogenesis. *Br J Cancer*, 51:707–712. PMID:3994914

Druckrey H, Nieper HA, Lo HW (1956). Carcinogene Wirkung von Parafuchsin in Injektionsversuch an Ratten. *Naturwissenschaften*, 43:543–544. doi:10.1007/BF00629375.

Dunkel VC, Pienta RJ, Sivak A, Traul KA (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 compounds. *J Natl Cancer Inst*, 67:1303–1312. PMID:6947113

Dunkel VC, Zeiger E, Brusick D *et al.* (1984). Reproducibility of microbial mutagenicity assays: I. Tests with Salmonella typhimurium and Escherichia coli using a standardized protocol. *Environ Mutagen*, 6 Suppl 2;1–251. doi:10.1002/em.2860060702 PMID:6394312

European Commission (2000) Commission Directive 2000/32/EC of 19 May 2000 adapting to technical progress for the 26th time Council Directive 67/548/EEC on the approximation of the laws, regulations and administrative provisions relating to the classification, packaging and labelling of dangerous substances(*)

European Commission (2004) Directive 2004/37/EC of the European Parliament and of the Council of 29 April 2004 on the protection of workers from the risks related to exposure to carcinogens or mutagens at work (Sixth individual Directive within the meaning of Article 16(1) of Council Directive 89/391/EEC) (codified version)

Green FJ (1990) *The Sigma-Aldrich Handbook of Stains, Dyes and Indicators*, Milwaukee, WI, Aldrich Chemical Co., pp. 126–129, 508.

Green U, Holste J, Spikermann AR (1979). A comparative study of the chronic effects of magenta, paramagenta, and phenyl-beta-naphthylamine in Syrian golden hamsters. *J Cancer Res Clin Oncol*, 95:51–55. doi:10.1007/BF00411109 PMID:500768

Haber N (1998). Electromolecular propulsion (EMP): a rapid, simple method for analyzing dyes used in microscopy. *Biotech Histochem*, 73:59–70. doi:10.3109/10520299809140508 PMID:9605620

Hagiwara M, Watanabe E, Barrett JC, Tsutsui T (2006). Assessment of genotoxicity of 14 chemical agents used in dental practice: ability to induce chromosome aberrations in Syrian hamster embryo cells. *Mutat Res*, 603:111–120. PMID:16406784

Hayes S, Gordon A, Sadowski I, Hayes C (1984). RK bacterial test for independently measuring chemical toxicity and mutagenicity: short-term forward selection assay. *Mutat Res*, 130:97–106. PMID:6371505

Howe JR (1977). Is there a cancer risk in the laboratory use of magenta and related dyes? *Lab Pract*, 26:87–91.

Huang M, Russo R, Fookes BG, Sigman ME (2005). Analysis of fiber dyes by liquid chromatography mass spectrometry (LC-MS) with electrospray ionization: Discriminating between dyes with indistinguishable UV-visible absorption spectra. *J Forensic Sci*, 50:526–534. doi:10.1520/JFS2004223.

IARC (1982a). Some aromatic amines, anthraquinones and nitroso compounds, and inorganic fluorides used in drinking-water and dental preparations. *IARC Monogr Eval Carcinog Risk Chem Hum*, 27:1–341. PMID:6955259

IARC (1982b). Some industrial chemicals and dyestuffs. *IARC Monogr Eval Carcinog Risk Chem Hum*, 29:1–398. PMID:6957379

IARC (1986). Some chemicals used in plastics and elastomers. *IARC Monogr Eval Carcinog Risk Chem Hum*, 39:1–403.

IARC (1987). Overall evaluations of carcinogenicity: an updating of IARC Monographs volumes 1 to 42. *IARC Monogr Eval Carcinog Risks Hum Suppl*, 7:1–440. PMID:3482203

IARC (1993). Occupational exposures of hairdressers and barbers and personal use of hair colourants; some hair dyes, cosmetic colourants, industrial dyestuffs and aromatic amines. *IARC Monogr Eval Carcinog Risks Hum*, 57:1–427.

Japan Chemical Industry Ecology (JETOC) (1996) *Mutagenicity test data of existing chemical substances based on the toxicity investigation of the industrial safety and health Law.*, Japan Chemical Industry Ecology - (JETOC), 1–592.

Japan Society for Occupational Health (2007). Recommendation of occupational exposure limits (2007–2008). *J Occup Health*, 49:328–344. PMID:17708058

Ketkar MB, Mohr U (1982). The chronic effects of magenta, paramagenta and phenyl-beta-naphthylamine in rats after intragastric administration. *Cancer Lett*, 16:203–206. doi:10.1016/0304-3835(82)90062-3 PMID:6812944

Khan HM, Naz S (2007). Aqueous solution of basic fuchsin as food irradiation dosimeter. *Nucl Sci Tech*, 18:141–144. doi:10.1016/S1001-8042(07)60035-9.

Kornbrust DJ, Barfknecht TR (1984). Comparison of rat and hamster hepatocyte primary culture/DNA repair assays. *Environ Mutagen*, 6:1–11. doi:10.1002/em.2860060102 PMID:6692797

Lawlor TE, Haworth SR, Lilja HS *et al.* (1987). Detection of mutagenic activity in the urine of rodents treated with p-rosaniline. *Environ Mutagen*, 9:69–78. doi:10.1002/em.2860090108 PMID:3542521

Li X, Liu G, Zhao J (1999). Two competitive primary processes in the photodegradation of cationic triarylmethane dyes under visible irradiation in TiO2 dispersions. *New J Chem*, 23:1193–1196. doi:10.1039/a906765e.

Lide DR, editor (2008) *CRC Handbook of Chemistry and Physics*, 98th Ed., Boca Raton, FL, CRC Press, pp. 3–322, 3–410.

MAK (2007) *List of MAK and BAT Values 2007*. Commission for the Investigation of Health Hazards of Chemical Compounds in the Work Area, Report No. 43, Deutsche Forschungsgemeinschaft, Weinheim, Wiley-VCH Verlag GmbH & Co. KGaA.

Misra AK, Gupta U (2002). Plain and ion-pair thin-layer chromatography of synthetic dyes on an admixture of silica gel G and barium sulfate in an aqueous ethanol system. *J Chromatogr Sci*, 40:297–303. PMID:12049159

Mitchell AD, Rudd CJ, Caspary WJ (1988). Evaluation of the L5178Y mouse lymphoma cell mutagenesis assay: intralaboratory results for sixty-three coded chemicals tested at SRI International. *Environ Mol Mutagen*, 12 Suppl 13;37–101. PMID:3416841

Mortelmans K, Haworth S, Lawlor T *et al.* (1986). Salmonella mutagenicity tests: II. Results from the testing of 270 chemicals. *Environ Mutagen*, 8 Suppl 7;1–119. doi:10.1002/em.2860080802 PMID:3516675

Myhr BC, Caspary WJ (1988). Evaluation of the L5178Y mouse lymphoma cell mutagenesis assay: intralaboratory results for sixty-three coded chemicals tested at Litton Bionetics, Inc. *Environ Mol Mutagen*, 12 Suppl 13;103–194. PMID:3416838

NIOSH (1990) *National Occupational Exposure Survey*. National Institute for Occupational Safety and Health

NTP (1986). NTP Toxicology and Carcinogenesis Studies of C.I. Basic Red 9 Monohydrochloride (Pararosaniline) (CAS No. 569–61–9) In F344/N Rats and B6C3F1 Mice (Feed Studies). *Natl Toxicol Program Tech Rep Ser*, 285:1–228. PMID:12748695

NTP (2005) C.I. Basic Red 9 Monohydrochloride CAS No. 569–61–9, *Report on Carcinogens*, Eleventh Edition; U.S. Department of Health and Human Services, Public Health Service, National Toxicology Program, available at: http://ntp.niehs.nih.gov/ntp/roc/eleventh/profiles/s046ciba.pdf

O'Neil MJ, editor (2006) *The Merck Index*, 14th Ed., Whitehouse Station, NJ, Merck & Co., Inc., p. 980.

Pienta RJ, Kawalek JC (1981). Transformation of hamster embryo cells by aromatic amines. *Natl Cancer Inst Monogr*, 58:243–251. PMID:7341981

Pienta RJ, Poiley JA, Lebherz WB 3rd (1977). Morphological transformation of early passage golden Syrian hamster embryo cells derived from cryopreserved primary cultures as a reliable in vitro bioassay for identifying diverse carcinogens. *Int J Cancer*, 19:642–655. doi:10.1002/ijc.2910190508 PMID:863543

Piolatto G, Negri E, La Vecchia C *et al.* (1991). Bladder cancer mortality of workers exposed to aromatic amines: an updated analysis. *Br J Cancer*, 63:457–459. PMID:2003988

Rai HS, Singh S, Cheema PP *et al.* (2007). Decolorization of triphenylmethane dye-bath effluent in an integrated two-stage anaerobic reactor. *J Environ Manage*, 83:290–297. doi:10.1016/j.jenvman.2006.03.003 PMID:16814454

Rehn L (1895). [Bladder Tumour in fuchsin workers]. *Arch Klin Chir*, 50:588–600.

Rosenkranz HS, Poirier LA (1979). Evaluation of the mutagenicity and DNA-modifying activity of carcinogens and noncarcinogens in microbial systems. *J Natl Cancer Inst*, 62:873–892. PMID:372656

Rubino GF, Scansetti G, Piolatto G, Pira E (1982). The carcinogenic effect of aromatic amines: an epidemiological study on the role of o-toluidine and 4,4′-methylene bis (2-methylaniline) in inducing bladder cancer in man. *Environ Res*, 27:241–254. doi:10.1016/0013-9351(82)90079-2 PMID:7084156

Shelley WB (2004). Memoir of a dermatologist's residency, 1946–1949. *J Am Acad Dermatol*, 51:674–677. doi:10.1016/S0190-9622(03)00862-4 PMID:15389218

Simmon VF (1979a). In vitro mutagenicity assays of chemical carcinogens and related compounds with Salmonella typhimurium. *J Natl Cancer Inst*, 62:893–899. PMID:372657

Simmon VF (1979b). In vitro assays for recombinogenic activity of chemical carcinogens and related compounds with Saccharomyces cerevisiae D3. *J Natl Cancer Inst*, 62:901–909. PMID:372658

Simmon VF, Rosenkranz HS, Zeiger E, Poirier LA (1979). Mutagenic activity of chemical carcinogens and related compounds in the intraperitoneal host-mediated assay. *J Natl Cancer Inst*, 62:911–918. PMID:372659

Speck WT, Santella RM, Rosenkranz HS (1978). An evaluation of the prophage lambda induction (inductest) for the detection of potential carcinogens. *Mutat Res*, 54:101–104. PMID:353543

Thetford D (2000) Triphenylmethane and Related Dyes. In: *Kirk-Othmer Encyclopedia of Chemical Technology*, Online Edition, New York, John Wiley & Sons, Inc., 21 pp.

US National Library of Medicine (1992) *Hazardous Substances Data Bank* (HSDB Record Nos. 2952 and 6192), Bethesda MD.

US Tariff Commission (1922) *Census of Dyes and Other Synthetic Organic Chemicals, 1921* (Tariff information Series No. 26), Washington DC, US Government Printing Office, p. 66.

US Tariff Commission (1965) *Synthetic Organic Chemicals, United States Production and Sales, 1964* (TC Publication 167), Washington DC, US Government Printing Office, pp. 18, 98.

USEPA (2003) Non-confidential production volume information for chemicals under the 1986–2002 Inventory Update Rule (IUR). Washington, DC [http://www.epa.gov/oppt/iur/tools/data/2002-vol.html]

USEPA (2007) Non-confidential 2006 Inventory Update Rule (IUR) records by chemical including manufacturing, processing and use information. Washington, DC [http://cfpub.epa.gov/iursearch/index.cfm?s=chem&err=t]

Vineis P, Magnani C (1985). Occupation and bladder cancer in males: a case-control study. *Int J Cancer*, 35:599–606. doi:10.1002/ijc.2910350506 PMID:3997281

Whitwell G (1968). Magenta paint (Letter to the Editor). *Lancet*, 292:110. doi:10.1016/S0140-6736(68)90396-6.

Williams GM, Laspia MF, Dunkel VC (1982). Reliability of the hepatocyte primary culture/DNA repair test in testing of coded carcinogens and noncarcinogens. *Mutat Res*, 97:359–370. PMID:7144803

Williams EL, Grosjean E, Grosjean D (1992). Exposure of Artists' Colorants to Airborne Formaldehyde. *Studies in Conservation*, 37:201–210. doi:10.2307/1506347.

Yamada T, Odawara K, Kaneko H (2000). Concurrent detection of gene mutations and chromosome aberrations induced by five chemicals in a CHL/IU cell line incorporating a gpt shuttle vector. *Mutat Res*, 471:29–36. PMID:11080658

Yamaguchi T (1988). Adsorption of carcinogenic and/or mutagenic pigments on DNA-binding sepharose. *Agric Biol Chem*, 52:845–847.

Yiyun C, Jiepin Y (2005). Effect of polyamidoamine dendrimers in decolorising triarylmethane dye effluent. *Coloration Technology*, 121:72–75. doi:10.1111/j.1478-4408.2005.tb00254.x.