CI ACID ORANGE 3

1. Exposure Data

1.1 Chemical and physical data

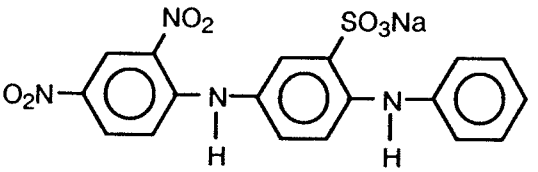
1.1.1 *Synonyms, structural and molecular data*

Chem. Abstr. Serv. Reg. No.: 6373-74-6; replaces 74968-36-8

Chem. Abstr. Name: 5-[(2,4-Dinitrophenyl)amino]-2-(phenylamino)benzene-sulfonic acid, monosodium salt

Colour Index No.: 10385

Synonyms: 2-Anilino-5-(2,4-dinitroanilino)benzenesulfonic acid, monosodium salt; sodium 4-(2,4-dinitroanilino)diphenylamine-2-sulfonate



$C_{18}H_{13}N_4O_7S.Na$ Mol. wt: 452.39

1.1.2 *Chemical and physical properties*

(*a*) *Description*: Dark orange-brown microcrystals (US National Toxicology Program, 1988)

(*b*) *Spectroscopy data*: Infrared and nuclear magnetic resonance spectral data have been reported (US National Toxicology Program, 1988).

(*c*) *Solubility*: Very soluble in water (> 2% w/w) and ethanol (Society of Dyers and Colourists, 1971).

1.1.3 *Trade names, technical products and impurities*

Some trade names are: Acid Fast Yellow AG; Acid Fast Yellow E 5R; Acid Leather Light Brown G; Acid Orange 3; Acid Yellow E; Airedale Yellow E; Amido Yellow E; Amido Yellow EA; Amido Yellow EA-CF; Anthralan Yellow RRT; Coranil Brown H EPS; Derma Fur Yellow RT; Derma Yellow P; Dimacide Yellow N-5RL; Duasyn Acid Yellow RRT; Elbenyl Orange A-3RD; Erio Fast Yellow AE; Erio Fast Yellow AEN; Erio Yellow AEN; Erionyl Yellow E-AEN; Fast Light Yellow E; Fenalan Yellow E; Heliacid Light Yellow 4R; Intranyl Orange T-4R; Kiton Fast Yellow A; Lanaperl Yellow Brown GT; Light Fast Yellow

ES; Lissamine Fast Yellow AE; Lissamine Fast Yellow AES; Lissamine Yellow AE; Multacid Yellow 3R; Multicuer Brown MPH; Nailamide Yellow Brown E-L; Nylocrom Yellow 3R; Nylomine Acid Yellow B-RD; Nylosan Yellow E-3R; Polan Yellow E-3R; Sellacid Yellow AEN; Solanile Yellow E; Sulfacid Light Yellow 5RL; Superian Yellow R; Tectilon Orange 3GT; Tertracid Light Yellow 2R; Unitertracid Light Yellow RR; Vondacid Fast Yellow AE; Vondacid Light Yellow AE; Xylene Fast Yellow ES.

A technical product was reported to contain approximately 65–67% CI Acid Orange 3 (US National Toxicology Program, 1988), together with approximately 16% sodium chloride and 19% sodium sulfate.

1.1.4 *Analysis*

No data were available to the Working Group.

1.2 Production and use

1.2.1 *Production*

CI Acid Orange 3 was first synthesized by Schmidlin in 1911 and has been produced commercially since the 1920s. It is prepared by the condensation of 1-chloro-2,4-dinitrobenzene with 5-amino-2-anilinobenzenesulfonic acid (Society of Dyers and Colourists, 1971).

At present, approximately 900 kg of CI Acid Orange 3 are used annually in the USA, according to industry estimates.

1.2.2 *Use*

CI Acid Orange 3 has been used as a dye in semi-permanent hair colouring products since the late 1950s. These products are generally shampooed into the hair, lathered and then allowed to remain in contact with the hair and scalp for 30–45 min. At the concentrations (up to 0.2%) used in these preparations, CI Acid Orange 3 is in solution (Frenkel & Brody, 1973; US National Toxicology Program, 1988). It is also used to dye textiles (US Environmental Protection Agency, 1990).

1.3 Occurrence

1.3.1 *Natural occurrence*

CI Acid Orange 3 is not known to occur as a natural product.

1.3.2 *Occupational exposure*

At one site where upholstery fabric was printed and powder acid dyes were used for dyeing nylon fibres, an industrial hygiene study was conducted to estimate dust exposures during weighing of the powder dyes. The principal dyes handled were CI Acid Black 107, CI Acid Brown 298 and CI Acid Orange 3. Spectrophotometric estimates of the average

airborne concentration of active colourant were 10 $\mu g/m^3$ using personal filters and < 0.01 $\mu g/m^3$ using area filters (US Environmental Protection Agency, 1990).

On the basis of a survey conducted in the USA between 1981 and 1983, the US National Institute for Occupational Safety and Health estimated that a total of 22 238 workers, including 14 728 women, may have been exposed to CI Acid Orange 3 in beauty salons (US National Library of Medicine, 1992).

1.4 Regulations and guidelines

No data were available to the Working Group.

2. Studies of Cancer in Humans

No data were available to the Working Group.

3. Studies of Cancer in Experimental Animals

3.1 Oral administration

3.1.1 *Mouse*

Groups of 50 male and 50 female B6C3F_1 mice, eight to nine weeks of age, were given 0, 125 or 250 (males) or 0, 250 or 500 (females) mg/kg bw CI Acid Orange 3 (90% pure, $\leq$ 1% impurities other than water and acetone) by gavage in corn oil on five days per week for 103 weeks and were sacrified at 112–113 weeks of age. Mean body weights of high-dose mice were 5–11% lower than those of vehicle controls after week 74. Survival at the end of the study was: males—control, 38/50; low-dose, 25/50; and high-dose, 26/50; females—control, 23/50; low-dose, 23/50; and high-dose, 24/50. Treatment-related nephrotoxicity occurred in animals of each sex. Survival of males was reduced after week 100; this was attributed to the nephrotoxicity. There was no significant increase in the incidence of any tumour. Epithelial hyperplasia of the urinary bladder was observed in one low-dose and three high-dose females. One low-dose female had a squamous-cell carcinoma of the urinary bladder (US National Toxicology Program, 1988).

3.1.2 *Rat*

Groups of 50 male and 50 female Fischer 344/N rats, six to seven weeks of age, were given 0, 375 or 750 mg/kg bw CI Acid Orange 3 (90% pure, $\leq$ 1% impurities other than water and acetone) by gavage in corn oil on five days per week for 103 weeks and were sacrificed at 110–112 weeks of age. Survival of high-dose male rats (after week 33) and high-dose female rats (after week 14) was significantly lower than that of vehicle controls and was attributed to nephrotoxicity. Final survival rates were: males—control, 36/50; low-dose, 30/50; and high-dose 0/50 ($p < 0.001$); females—control, 43/50; low-dose, 34/50; and high-dose, 7/50

($p < 0.001$). The primary cause of death in treated animals was a spectrum of non-neoplastic renal lesions. Renal pelvic epithelial hyperplasia was seen in 0/50 control, 6/50 low-dose and 13/50 high-dose males and 0/50 control, 2/50 low-dose and 13/50 high-dose females. Transitional-cell carcinomas of the renal pelvis were observed only in high-dose female rats (6/50; $p < 0.001$, life table test) at 87–104 weeks; 18 animals were still alive at 87 weeks. No significant increase in the incidence of tumours at any site was reported in male rats, but, because of a marked reduction in survival, the high-dose group was considered to be inadequate for an assessment of carcinogenic activity (US National Toxicology Program, 1988).

4. Other Relevant Data

4.1 Absorption, distribution, metabolism and excretion

No data were available to the Working Group.

4.2 Toxic effects

4.2.1 *Humans*

No data were available to the Working Group.

4.2.2 *Experimental systems*

In 14-day studies, groups of five Fischer 344/N rats of each sex received 0, 94, 187, 375, 750 or 1500 mg/kg bw CI Acid Orange 3 (purity, 90%) and groups of five B6C3F$_1$ mice of each sex received 0, 62, 125, 250, 500 or 1000 mg/kg bw CI Acid Orange 3 in corn oil by gavage for 14 consecutive days. No compound-related toxic effect was observed (US National Toxicology Program, 1988).

In 13-week studies, groups of 10 Fischer 344/N rats of each sex received 0, 94, 187, 375, 750 or 1500 mg/kg bw CI Acid Orange 3 and groups of 10 B6C3F$_1$ mice of each sex received 0, 250, 500, 1000 or 2000 mg/kg bw in corn oil by gavage on five days per week. Dose-related kidney lesions were observed in rats and mice of each sex, including variable degrees of degeneration and necrosis of epithelial cells in the proximal convoluted tubules, increased basophilia of tubular epithelium and granular casts in the tubules. A marked reduction in survival was observed in female rats that received the highest dose (US National Toxicology Program, 1988).

In the two-year studies in Fischer 344/N rats and B6C3F$_1$ mice described above, the primary cause of death was dose-related kidney lesions (US National Toxicology Program, 1988).

Acid Orange 3 was present at a low concentration (0.2%) in semi-permanent hair colouring formulations evaluated in a 13-week study of dermal toxicity in rabbits (Burnett *et al.*, 1976) and in a two-year feeding study in dogs (Wernick *et al.*, 1975), described in detail on p. 97. No treatment-related toxicity was detected in either study. [The Working Group

noted that the dose of each component of the formulation was very low and unlikely to have been toxic.]

4.3 Reproductive and developmental effects

4.3.1 *Humans*

No data were available to the Working Group.

4.3.2 *Experimental systems*

No data were available to the Working Group on the reproductive and developmental effects of CI Acid Orange 3 alone. This compound was present at low concentrations in semi-permanent hair colouring formulations evaluated in a study of fertility and reproductive performance in rats (0.2%) (Wernick *et al.*, 1975; see p. 99), in a study of heritable translocation in rats (0.06%) (Burnett *et al.*, 1981; see p. 104) and in studies of teratogenesis in rats (0.2%) (Wernick *et al.*, 1975; Burnett *et al.*, 1976) and rabbits (0.2%) (Wernick *et al.*, 1975) (see p. 100). No treatment-related adverse effect was detected. [The Working Group noted that the dose of each component of the formulation was very low and unlikely to be toxic.]

4.4 Genetic and related effects

4.4.1 *Humans*

No data were available to the Working Group.

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

CI Acid Orange 3 was mutagenic to *Salmonella typhimurium* in the presence and absence of metabolic activation.

Table 1. Genetic and related effects of CI Acid Orange 3

Test system	Result		Dose (LED/HID) (μg/ml)	Reference
	Without exogenous metabolic system	With exogenous metabolic system		
SA0, *Salmonella typhimurium* TA100, reverse mutation	+	+	167.0000	Zeiger *et al.* (1988)
SA5, *Salmonella typhimurium* TA1535, reverse mutation	–	–	500.0000	Zeiger *et al.* (1988)
SA9, *Salmonella typhimurium* TA98, reverse mutation	+	+	50.0000	Zeiger *et al.* (1988)
SA5, *Salmonella typhimurium* TA97, reverse mutation	(+)	(+)	167.0000	Zeiger *et al.* (1988)

+, positive; (+), weakly positive; –, negative

5. Summary of Data Reported and Evaluation

5.1 Exposure data

CI Acid Orange 3 is used to a limited extent as a dye in semi-permanent hair colouring products and in the dyeing of textiles.

5.2 Human carcinogenicity data

No data were available to the Working Group.

5.3 Animal carcinogenicity data

CI Acid Orange 3 was tested for carcinogenicity by gavage in one study in mice and in one study in rats. In mice, there was no significant increase in the incidence of tumours. A significant increase in the incidence of transitional-cell carcinomas of the renal pelvis was observed in female rats given the high dose. The data on high-dose male rats could not be evaluated owing to their poor survival.

5.4 Other relevant data

CI Acid Orange 3 caused renal toxicity in rats and mice. It was mutagenic to bacteria.

5.5 Evaluation[1]

There is *inadequate evidence* in humans for the carcinogenicity of CI Acid Orange 3.

There is *limited evidence* in experimental animals for the carcinogenicity of CI Acid Orange 3.

Overall evaluation

CI Acid Orange 3 *is not classifiable as to its carcinogenicity to humans (Group 3).*

6. References

Burnett, C., Goldenthal, E.I., Harris, S.B., Wazeter, F.X., Strausburg, J., Kapp, R. & Voelker, R. (1976) Teratology and percutaneous toxicity studies on hair dyes. *J. Toxicol. environ. Health*, **1**, 1027–1040

Burnett, C., Loehr, R. & Corbett, J. (1981) Heritable translocation study on two hair dye formulations. *Fundam. appl. Toxicol.*, **1**, 325–328

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

Frenkel, E.P. & Brody, F. (1973) Percutaneous absorption and elimination of an aromatic hair dye. *Arch. environ. Health*, **27**, 401–404

Society of Dyers and Colourists (1971) *Colour Index*, 3rd ed., Vol. 4, Bradford, Yorkshire, p. 4007

US Environmental Protection Agency (1990) *Textile Dye Weighing Monitoring Study* (EPA Report No. EPA-560/5-90-009), Washington DC, Office of Toxic Substances

US National Library of Medicine (1992) *Registry of Toxic Effects of Chemical Substances* (RTECS No. DB5042500), Bethesda, MD

US National Toxicology Program (1988) *Toxicology and Carcinogenesis Studies of CI Acid Orange 3 (CAS No. 6373-74-6) in F344/N Rats and B6C3F$_1$ Mice (Gavage Studies)* (NTP Technical Report 335; NIH Publ. No. 89-2591), Research Triangle Park, NC

Wernick, T., Lanman, B.M. & Fraux, J.L. (1975) Chronic toxicity, teratology and reproduction studies with hair dyes. *Toxicol. appl. Pharmacol.*, **32**, 450–460

Zeiger, E., Anderson, B., Haworth, S., Lawlor, T. & Mortelmans, K. (1988) *Salmonella* mutagenicity tests: IV. Results from the testing of 300 chemicals. *Environ. Mol. Mutag.*, **11** (Suppl. 12), 1–158