

Azobenzene; CASRN 103-33-3

Human health assessment information on a chemical substance is included in the IRIS database only after a comprehensive review of toxicity data, as outlined in the [IRIS assessment development process](#). Sections I (Health Hazard Assessments for Noncarcinogenic Effects) and II (Carcinogenicity Assessment for Lifetime Exposure) present the conclusions that were reached during the assessment development process. Supporting information and explanations of the methods used to derive the values given in IRIS are provided in the [guidance documents located on the IRIS website](#).

STATUS OF DATA FOR Azobenzene

File First On-Line 09/07/1988

Category (section)	Assessment Available?	Last Revised
Oral RfD (I.A.)	not evaluated	
Inhalation RfC (I.B.)	not evaluated	
Carcinogenicity Assessment (II.)	yes	09/07/1988

I. Chronic Health Hazard Assessments for Noncarcinogenic Effects

I.A. Reference Dose for Chronic Oral Exposure (RfD)

Substance Name — Azobenzene
CASRN — 103-33-3

Not available at this time.

I.B. Reference Concentration for Chronic Inhalation Exposure (RfC)

Substance Name — Azobenzene
CASRN — 103-33-3

Not available at this time.

II. Carcinogenicity Assessment for Lifetime Exposure

Substance Name — Azobenzene
CASRN — 103-33-3
Last Revised — 09/07/1988

Section II provides information on three aspects of the carcinogenic assessment for the substance in question; the weight-of-evidence judgment of the likelihood that the substance is a human carcinogen, and quantitative estimates of risk from oral exposure and from inhalation exposure. The quantitative risk estimates are presented in three ways. The slope factor is the result of application of a low-dose extrapolation procedure and is presented as the risk per (mg/kg)/day. The unit risk is the quantitative estimate in terms of either risk per ug/L drinking water or risk per ug/cu.m air breathed. The third form in which risk is presented is a drinking water or air concentration providing cancer risks of 1 in 10,000, 1 in 100,000 or 1 in 1,000,000. The rationale and methods used to develop the carcinogenicity information in IRIS are described in The Risk Assessment Guidelines of 1986 (EPA/600/8-87/045) and in the IRIS Background Document. IRIS summaries developed since the publication of EPA's more recent Proposed Guidelines for Carcinogen Risk Assessment also utilize those Guidelines where indicated (Federal Register 61(79):17960-18011, April 23, 1996). Users are referred to Section I of this IRIS file for information on long-term toxic effects other than carcinogenicity.

II.A. Evidence for Human Carcinogenicity

II.A.1. Weight-of-Evidence Characterization

Classification — B2; probable human carcinogen

Basis — Azobenzene induced invasive sarcomas in the spleen and other abdominal organs in male and female F344 rats following dietary administration. It is genotoxic and may be converted to benzidine, a known human carcinogen, under the acidic conditions in the stomach.

II.A.2. Human Carcinogenicity Data

None.

II.A.3. Animal Carcinogenicity Data

Sufficient. In an NCI study (1979), azobenzene was administered in the diet to F344 rats and B6C3F1 mice (50 animals/sex/dose) for 106 weeks and 105 weeks, respectively. Control groups consisted of 50 untreated animals of each sex. The dietary concentrations were 200 and 400 ppm for rats. For male mice the dietary concentrations were 200 and 400. Because of lowered body weights after 38 weeks, the initial 400 and 800 ppm concentrations for female mice were reduced, to give time-weighted concentrations of 208 and 505 ppm. In rats sarcomas of various types, including fibrosarcomas, hemangiosarcomas and osteosarcomas, were observed in the spleen and other abdominal organs at incidences that were dose-related in each sex and that were statistically significantly higher in the high-dose group than in controls in each sex. In mice no tumors were observed at any site at significantly higher incidence than in the controls for either sex.

In a carcinogenesis screening assay (Innes, 1968; Innes et al., 1969) in two hybrid strains of mice (B6C3F1 and B6AKF1) azobenzene was administered either by a single subcutaneous injection or in the diet at a concentration of 56 ppm continuously until termination of the test at 18 months. No response was observed in the subcutaneous injection study and of the four groups (including males and females of these strains) only male B6C3F1 mice responded. The incidence of hepatomas was 8/18 in the treated group compared with 8/73 in controls. These results were considered by the authors as "requiring additional evaluation".

II.A.4. Supporting Data for Carcinogenicity

Azobenzene was positive in a reverse mutation plate incorporation assay in Salmonella (Ames test) upon addition of hepatic extracts for metabolism. Azobenzene causes DNA single strand breaks with <30% cytotoxicity by the alkaline elution/rat hepatocyte assay (Sina et al., 1983) and produces unscheduled DNA synthesis in human lymphocytes (Gaudin et al., 1971). It should be noted that azobenzene may be converted non-enzymatically under the acidic conditions in the stomach to the known human carcinogen benzidine.

II.B. Quantitative Estimate of Carcinogenic Risk from Oral Exposure

II.B.1. Summary of Risk Estimates

Oral Slope Factor — 1.1E-1 per (mg/kg)/day

Drinking Water Unit Risk — 3.1E-6 per (ug/L)

Extrapolation Method — Linearized multistage procedure, extra risk

Drinking Water Concentrations at Specified Risk Levels:

Risk Level	Concentration
E-4 (1 in 10,000)	3E+1 ug/L
E-5 (1 in 100,000)	3E+0 ug/L
E-6 (1 in 1,000,000)	3E-1 ug/L

II.B.2. Dose-Response Data (Carcinogenicity, Oral Exposure)

Tumor Type — abdominal cavity sarcomas

Test animals — rats/F344, female

Route — diet

Reference — NCI, 1979

Administered Dose (ppm)	Human Equivalent Dose (mg/kg)/day	Tumor Incidence
0	0	0/20
200	1.53	5/50
400	3.06	21/50

II.B.3. Additional Comments (Carcinogenicity, Oral Exposure)

The slope factor calculated from data on male rats is $7.3E-2$ per (mg/kg)/day using 0.375 kg for the rat body weight. A body weight of 0.25 kg was used for female rats.

The unit risk should not be used if the water concentration exceeds $3E+3$ ug/L, since above this concentration the unit risk may not be appropriate.

II.B.4. Discussion of Confidence (Carcinogenicity, Oral Exposure)

This study is of adequate design. A sufficient number of animals were treated at the maximum tolerated dose by an appropriate route for a period approximating their natural lifespan. The response did not vary markedly between sexes.

II.C. Quantitative Estimate of Carcinogenic Risk from Inhalation Exposure

II.C.1. Summary of Risk Estimates

Inhalation Unit Risk — $3.1E-5$ per (ug/cu.m)

Extrapolation Method — linearized multistage procedure, extra risk

Air Concentrations at Specified Risk Levels:

Risk Level	Concentration
E-4 (1 in 10,000)	3 ug/cu.m
E-5 (1 in 100,000)	$3E-1$ ug/cu.m
E-6 (1 in 1,000,000)	$3E-2$ ug/cu.m

II.C.2. Dose-Response Data for Carcinogenicity, Inhalation Exposure

Calculated from oral data in Section II.B.2.

II.C.3. Additional Comments (Carcinogenicity, Inhalation Exposure)

The unit risk should not be used if the air concentration exceeds 3E+2 ug/cu.m, since above this concentration the unit risk may not be appropriate.

II.C.4. Discussion of Confidence (Carcinogenicity, Inhalation Exposure)

While the study design is adequate for a feeding bioassay, these data are less than optimal for the assessment risk related to exposure by inhalation in the absence of route-comparative pharmacokinetic information.

II.D. EPA Documentation, Review, and Contacts (Carcinogenicity Assessment)

II.D.1. EPA Documentation

Source Document — U.S. EPA, 1985

The 1985 Health and Environmental Effects Profile for Azobenzene received OHEA review.

II.D.2. EPA Review (Carcinogenicity Assessment)

Agency Work Group Review — 02/03/1988

Verification Date — 02/03/1988

Screening-Level Literature Review Findings — A screening-level review conducted by an EPA contractor of the more recent toxicology literature pertinent to the cancer assessment for Azobenzene conducted in September 2002 did not identify any critical new studies. IRIS users who know of important new studies may provide that information to the IRIS Hotline at hotline.iris@epa.gov or (202)566-1676.

II.D.3. EPA Contacts (Carcinogenicity Assessment)

Please contact the IRIS Hotline for all questions concerning this assessment or IRIS, in general, at (202)566-1676 (phone), (202)566-1749 (FAX) or hotline.iris@epa.gov (internet address).

III. [reserved]

IV. [reserved]

V. [reserved]

VI. Bibliography

Substance Name — Azobenzene

CASRN — 103-33-3

VI.A. Oral RfD References

None

VI.B. Inhalation RfD References

None

VI.C. Carcinogenicity Assessment References

Gaudin, D., R.S. Gregg and K.L. Yielding. 1971. DNA repair inhibition: A possible mechanism of action of co-carcinogens. *Biochem. Biophys. Res. Commun.* 45(3): 630-636.

Innes, J.R.M. 1968. Evaluation of carcinogenic, teratogenic and mutagenic activities of selected pesticides and industrial chemicals. Vol. 1, Carcinogenic study. Bionetics Research Labs, Inc., Bethesda Natl. Tech. Info. Service, U.S. Department of Commerce.

Innes, J.R.M., B.M. Ulland, M.G. Valerio, et al. 1969. Bioassay of pesticides and industrial chemicals for tumorigenicity in mice: A preliminary note. *J. Natl. Cancer Inst.* 43: 1101-1114.

NCI (National Cancer institute). 1979. Bioassay of azobenzene for possible carcinogenicity. NCI Carcinogenesis Technical Report Series. No. 154. p. 112.

Sina, J.F., C.L. Bean, G.R. Dysart, V.I. Taylor and M.O. Bradley. 1983. Evaluation of the alkaline elution/rat hepatocyte assay as a predictor of carcinogenic/mutagenic potential. *Mutat. Res.* 113(5): 357-391.

U.S. EPA. 1985. Health and Environmental Effects Profile for Azobenzene, Final Draft. Prepared by the Office of Health and Environmental Health and the Environmental Criteria and Assessment Office, Cincinnati, OH for the Office of Solid Waste and Emergency Response, Washington, DC. EPA/600/X- 86/082.

VII. Revision History

Substance Name — Azobenzene
CASRN — 103-33-3

Date	Section	Description
09/07/1988	II.	Carcinogen summary on-line
12/03/2002	II.D.2.	Screening-Level Literature Review Findings message has been added.

VIII. Synonyms

Substance Name — Azobenzene
CASRN — 103-33-3
Last Revised — 09/07/1988

- 103-33-3
- AZOBENZEEN
- Azobenzene
- AZOBENZIDE
- AZOBENZOL
- AZOBISBENZENE
- AZODIBENZENE
- AZODIBENZENEAZOFUME
- AZOFUME

- BENZENEAZOBENZENE
- BENZENE, AZODI
- BENZOFUME
- DIAZOBENZENE
- DIPHENYLDIAZENE
- 1,2-DIPHENYLDIAZENE
- DIPHENYLDIIMIDE
- ENT 14,611
- NCI-C02926
- USAF EK-704