O-TOLUIDINE

CAS number: 95-53-4

Synonyms: 1-Amino-2-methylbenzene, 2-aminotoluene, 2-methylaniline, 2-methylbenzenamine

Chemical formula: C_7H_9N

Workplace exposure standard (interim)

TWA: **2 ppm (8.8 mg/m^3)**

STEL: **—**

Peak limitation: **—**

Notations: **Carc. 1A, Sk.**

IDLH: **50 ppm**

Sampling and analysis: The recommended value is quantifiable through available sampling and analysis techniques.

Recommendation and basis for workplace exposure standard

A TWA of 2 ppm (8.8 mg/m^3) is recommended to minimise potential for methaemoglobinemia, bladder and kidney irritation and bladder cancer in exposed workers.

Given the limited data available from the primary sources, it is recommended that a review of additional sources be conducted at the next scheduled review.

Discussion and conclusions

o-Toluidine and its hydrochloride (HCl) salt are used in the manufacture of dyes, rubber, pesticides and pharmaceuticals.

Critical effects of exposure are methaemoglobinaemia, irritation of the bladder and kidneys and bladder cancer.

Chronic inhalational and dermal exposure is associated with methaemoglobinemia, anaemia, skin lesions and central nervous system (CNS) disturbances in workers. A threshold for these effects is not determined in the available database. However, poorly documented survey data indicate exposure above 5 ppm causes illness. Epidemiological data strongly associate occupational exposure with the development of bladder cancer and are supported by evidence for this endpoint in chronic feeding studies with animals. Results of the available *in vitro* and *in vivo* mutagenicity assays are equivocal and indicate mutagenic potential from genotoxic metabolites. SCOEL (2014) estimates a benchmark dose expected to result in 10 per cent increased tumorigenicity (BMD_{10}) at 210 mg/m^3 in humans from the dose-response for the incidence of bladder cancer in rats.

Numerical OEL equivalents are not recommended by DFG (2007) and SCOEL (2014) due to suspected genotoxic carcinogenicity. The ACGIH (2018) TLV-TWA recommendation is based on analogy to structurally and toxicologically related aniline, for which the critical endpoint is also methaemoglobin formation. In volunteers, a daily oral dose of 35 mg aniline is associated with blood methaemoglobin concentration of 3.7 per cent, which is below the maximally tolerated level of

approximately 5 per cent (SCOEL, 2010) and is the basis of the TWA for aniline and its homologues of 0.5 ppm.

In view of the uncertainty of a potential genotoxic mechanism of carcinogenicity and absence of a suitable inhalation carcinogenic unit risk factor, the TWA of 2 ppm recommended by ACGIH (2018) is retained in the interim. However, further assessment of the carcinogenic mechanism is recommended as a priority during subsequent reviews of the WES. Due to the toxicological similarities with aniline and the evaluation approach presented by ACGIH (2018), future reviews should also consider adoption of the TWA for aniline and its homologues for *o*-toluidine.

Recommendation for notations

Classified as a category 1A carcinogen according to the Globally Harmonized System of Classification and Labelling of Chemicals (GHS).

Not classified as a skin sensitiser or respiratory sensitiser according to the GHS.

A skin notation is recommended due to evidence of dermal absorption and contribution to adverse systemic effects.

APPENDIX

Primary sources with reports

Source	Year set	Standard
SWA	***1991***	***TWA: 2 ppm (8.8 mg/m^3)***
ACGIH	***2001***	***TLV–TWA: 2 ppm (8.8 mg/m^3)***

TLV-TWA intended to protect for methaemoglobinemia and irritation of the eyes, skin and bladder. Skin notation recommended based on reports of systemic toxicity following dermal absorption. Carcinogenicity in animals is unaffirmed in epidemiological data and relevance of carcinogenic potential to humans is uncertain (A3).

Summary of information:

TLV-TWA based on analogy to aniline and nitrobenzene, which are toxicologically and structurally related.

Human data:

- Signs of intoxication include methaemoglobinemia, haematuria, irritation of bladder and kidney and physiological and psychological disturbances (no further details provided)
- Severe intoxication at 40 ppm (1 h, no further details provided)
- Available epidemiological data confounded by mixed exposures to other aromatic amines
- Increased incidence of bladder cancer in workers co-exposed to aniline (no details on exposure provided)
- Calculated skin permeability coefficient: 0.0037 cm/h.

Animal data:

- LD_{50} of 3,250 mg/kg (rabbits, dermal)
- Mean maximal methaemoglobinemia of 70.1% following IV injection of 27 mg/kg (cats); effects similar to those reported for aniline
- Methaemoglobinemia and increased markers for anaemia at 35 mg/kg/d in subchronic gavage study (rats, 2.5 mo)
- Bladder epithelial keratosis, metaplasia and low incidence of papillomas (no further details) in subchronic feeding study at 5,714 mg/kg/d as 7.5% solution in peanut oil (rats, 64 d, then 27 d at 50% dose, 91 d total). No further information provided
- Dose-dependent decreased body weight gain up to 50,000 ppm of diet (mice, rats) and renal hepatic and splenic pigmentation at 12,500 ppm (rats) reported in subchronic feeding studies (mice, rats, 7 wk)
- Significant dose-dependent increased incidence of vascular tumours in abdomen and bladder at 16,000 (3 mo) and 8,000 (15 mo) ppm of diet, or 32,000 (3 mo) and 16,000 ppm of diet reported in chronic feeding study (mice, 18 mo)
- Significant increased incidence of vascular tumours (males) and liver tumours (females) at 3,000 ppm of diet, but not at 1,000 ppm in chronic feeding study (mice, 2 yr)
- Significant increased incidence of subcutaneous tumours and non-significant increase in bladder tumours at 8,000 (3 mo) and 4,000 (15 mo) ppm of diet, or 16,000 (3 mo) and 8,000 ppm of diet (rats, 18 mo)
- Equivocal results for mutagenicity *in vitro* in bacteria, yeast and mammalian cells using pure substance in the presence or absence of metabolic activation, or as concentrated urine from exposed rats

Source	Year set	Standard
• Single-strand DNA breaks in liver and kidney cells at IP dose of 100 mg/kg (mice), inhibition of DNA synthesis in testis at gavage dose of 200 mg/kg (mice) and inhibition of renal DNA synthesis in pups (mice) reported *in vivo.* Insufficient data to recommend a TLV-STEL or sensitiser notation.		
DFG	***2007***	***Not assigned***
Summary of additional information: MAK not assigned due to suspected genotoxic carcinogenicity. Available human exposure data are suggestive of carcinogenic potential but are confounded by mixed exposures. DFG classifies substance as category 1 carcinogen based on weight of evidence from workplace studies with mixed exposure to aniline. Aniline concentrations reported in one study were however below those expected to elicit the observed carcinogenic effects, which is considered substantial evidence for carcinogenicity in humans by the DFG. No safe level of exposure can be assessed, skin notation therefore assigned despite low calculated dermal penetration rate. Human data: • Increased standardised mortality ratio (SMR) for deaths from bladder tumours in rubber production workers (n=2,160, >6 mo) exposed to mixtures containing substance (components not specified), regression analysis of these data showed dose-response relationship for substance, but not for exposure to other chemicals: o relative risk increased from 6.7 for 4-yr exposure to 7.6 for 5-yr exposure o results potentially confounded by exposure to *N*-phenyl-2-napthylamine • Significant increased standardised incidence ratio (SIR) for bladder cancer in rubber production workers (n=1,749) exposed to mixtures containing substance, aniline and potentially 4-aminobiphenyl not attributable to aniline; bladder carcinogenicity of 4-aminobiphenyl in humans noted in cited article • Exposure and biomonitoring data reviewed in follow-up study to the above SIR study: o average air concentrations estimated at 0.412±0.366 mg/m^3 and 0.187±0.181 mg/m^3 o urinary excretion 3 times higher than that of aniline; 4-aminobiphenyl was the same between controls and exposed workers o cited authors conclude highly likely the causal carcinogenic agent, prior exposure to 4-aminobiphenyl could however not be ruled out o agency argues exposure to aniline at these concentrations unlikely to cause the observed carcinogenic effect regarding MAK of 7.7 mg/m^3 for aniline • Dermal penetration rate *in vitro*: 0.37±0.12 µg/cm^2/h: o skin barrier creams may increase dermal penetration by a factor of 10. Animal data: • Substance forms genotoxic *N*-hydroxyl metabolites, analogous to other aromatic amines • Positive and negative *in vitro* mutagenicity results (also reported in ACGIH, 2018) were independent of metabolic activation • Evidence for mutagenicity *in vivo* included covalent binding to DNA, RNA and proteins in liver *in vivo* (rats), induction of DNA repair in bladder (rats) and DNA strand breaks in liver and kidneys (mice); chromosomal aberration and micronucleus assays were negative (mice) • Overall, considered results of *in vitro* and *in vivo* mutagenicity assays consistent with those of other genotoxic aromatic amines • Limited evidence for bladder tumorigenicity at 125 mg/kg/d after 11 yr in chronic feeding/gavage study (dogs, n=5, 5 d/wk, 9 yr); all dogs died of different causes, 1 dog, in		

Source	Year set	Standard
		which bladder hyperplasia and papillomas were detected, died after 11 yr from thyroid tumour.
SCOEL	***2017***	***Not assigned***
Summary of additional data: No OEL recommended due to suspected genotoxic carcinogenicity. Skin notation recommended based on reports of systemic availability following dermal exposure in workers. Incidence of bladder carcinomas in female rats in chronic feeding study used in Benchmark dose (BMD) analysis: • best fit for estimating dose expected to cause tumorigenicity at 10% above the background level (BMD_{10}) achieved with Quantal-linear model; BMD_{10} estimated at 42.2 mg/kg/d in rats • equivalent inhalational dose extrapolated as 840 mg/m^3 assuming a respiratory volume of 10 m^3 during an 8-h shift for a 70-kg worker working for 5 d/wk, 48 wk/yr for 40/75 yr • a factor of 4 is applied to account for allometric scaling to derive the human BMD_{10} of 210 mg/m^3 • tumour incidence at 8.8 mg/m^3 was therefore estimated to be 1:239. Human data: • Dermal absorption evidenced in several workplace studies, in which biomonitoring showed systemically available concentrations greater than those expected from inhalational exposure alone • Exposure at 40 ppm (1 h) of toluidine (isomer not specified) caused severe intoxication, 10 ppm caused illness and unsatisfactory conditions >5 ppm (no further details provided) reported in survey article. Human data for *aniline* (reported in SCOEL, 2010): • Blood methaemoglobin concentrations levels of ≈5% considered tolerable by analogy to carboxyhaemoglobin, for which 4% is tolerable • Methaemoglobin formation reported in volunteer repeat oral dose study with aniline dose groups 0, 5, 15, 25, 35, 45, 55 and 65 mg/person/d (n=5/group, 3 d): o increase in methaemoglobin formation non-significant up to 15 mg (1.2–1.8%) o doses of 35, 45, 55 and 65 mg aniline caused methaemoglobin levels of 3.7, 7.1, 5.2% and 16%, respectively o daily dose of 35 mg aniline, corresponding to 3.7% increase in blood methaemoglobin concentrations, used as basis for TWA of aniline (0.5 ppm ≡1.94 mg/m^3). Animal data: • Increased tumour incidence in bladder (females), spleen (females, males), abdominal cavity and scrotum (males), subcutaneous tissue (males) and mammary gland (females) reported in chronic feeding study with dose groups 0, 3,000 and 6,000 ppm of diet (rats, mice, 2 yr, also reported in ACGIH, 2018): o observations for mice not reported o dose-response relationship for bladder carcinomas in females used to calculate BMD; incidence of carcinomas reported as 0, 20 and 47% for dose groups 0, 3,000 and 6,000 ppm of diet ≡0, 150, 300 mg/kg/d of the *o*-toluidine hydrochloride salt.		
OARS/AIHA	***NA***	***NA***
No report.		

Source	Year set	Standard
HCOTN	***NA***	***NA***
No report.		

Secondary source reports relied upon

Source		Year	Additional information
NICNAS	✓	2014	• Anaemia, weight loss, anorexia, cyanosis, skin lesions, methaemoglobinemia and disturbance in the CNS such as headache, dizziness and confusion reported in chronically exposed workers (no further details provided) • Haemoglobin adducts found in the urine of exposed workers: o higher levels identified in those with existing impaired skin condition • NICNAS considers substance mutagenic based on the weight of evidence from available, well-documented *in vitro* and *in vivo* studies.
IARC	✓	2012	• Metabolism of substance not yet fully characterised: o available data indicate preferential arene oxidation or *N*-acetylation rather than *N*-oxidation; the latter would generate *N*-hydroxyl metabolites • Cancers of the urinary bladder associated with occupational exposure may result from oxidation in bladder epithelium • Haemoglobin adduct levels increased in patients treated with prilocaine and rubber manufacturing workers • Classified as Group 1; carcinogenic to humans: o sufficient evidence for carcinogenicity in humans; causes bladder cancer o sufficient evidence for carcinogenicity in animals o moderate mechanistic evidence indicating carcinogenicity involves metabolic activation, formation of DNA adducts and induction of DNA-damaging effects.
ECHA	✓	2020	• High hazard due to carcinogenicity (no threshold derived).
US NIOSH	✓	1994	• IDLH based on acute inhalation toxicity data in humans.

Carcinogenicity — non-threshold based genotoxic carcinogens

Is the chemical mutagenic?	Yes
Is the chemical carcinogenic with a mutagenic mechanism of action?	Insufficient data

Insufficient data are available to determine if the chemical is a non-threshold based genotoxic carcinogen.

Notations

Source	Notations
SWA	Carc. 2, Skin
HCIS	Carcinogenicity – category 1A
NICNAS	Carc. Cat. 1
EU Annex	Carcinogenicity – category 1B
ECHA	Carcinogenicity – category 1B
ACGIH	Carcinogenicity – A3
DFG	Carcinogenicity – 1, H (skin)
SCOEL	Carcinogenicity – A, Skin
HCOTN	NA
IARC	Carcinogenicity – Group 1
US NIOSH	NA

NA = not applicable (a recommendation has not been made by this Agency); — = the Agency has assessed available data for this chemical but has not recommended any notations

Skin notation assessment

Calculation		
Adverse effects in human case study:	yes	
Dermal LD_{50} ≤1000 mg/kg:	no	
Dermal repeat-dose NOAEL ≤200 mg/kg:		
Dermal LD_{50}/Inhalation LD_{50} <10:		
In vivo dermal absorption rate >10%:	yes	
Estimated dermal exposure at WES >10%:	yes	
		a skin notation is warranted

IDLH

Is there a suitable IDLH value available?	Yes

Additional information

Molecular weight:	107.15
Conversion factors at 25°C and 101.3 kPa:	1 ppm = 4.38 mg/m^3; 1 mg/m^3 = 0.229 ppm
This chemical is used as a pesticide:	☐
This chemical is a biological product:	☐
This chemical is a by-product of a process:	☐
A biological exposure index has been recommended by these agencies:	✓ ACGIH ✓ DFG ✓ SCOEL

Workplace exposure standard history

Year	Standard
Click here to enter year	

References

American Conference of Industrial Hygienists (ACGIH®) (2018) TLVs® and BEIs® with 7th Edition Documentation, CD-ROM, Single User Version. Copyright 2018. Reprinted with permission. See the *TLVs® and BEIs® Guidelines section* on the ACGIH website.

Deutsche Forschungsgemeinschaft (DFG) (2007) o-Toluidine – MAK value documentation.

EU Scientific Committee on Occupational Exposure Limits (SCOEL) (2017) Recommendation from the Scientific Committee on Occupational Exposure Limits for o-Toluidine, 2-methylaniline. SCOEL/REC/301.

EU Scientific Committee on Occupational Exposure Limits (SCOEL) (2010) Recommendation from the Scientific Committee on Occupational Exposure Limits for aniline. SCOEL/SUM/153.

European Chemicals Agency Regulation (ECHA) No 1907/2006 of the European Parliament and of the Council of 18 December 2006 concerning the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH).

European Chemicals Agency (ECHA) (2020) o-toluidine – REACH assessment.

International Agency for Research on Cancer (IARC) (2012) Volume 100 F, Chemical agents and related occupations. IARC Monographs on the evaluation of the carcinogenic risk to humans.

National Industrial Chemicals Notification and Assessment Scheme (NICNAS) (2014) Benzenamine, 2-methyl: Human health tier II assessment – IMAP report.

Tenth Adaptation to Technical Progress Commission Regulation (EU) No 2017/776 amending, for the purposes of its adaptation to technical and scientific progress, Regulation (EC) No 1272/2008 of the European Parliament and of the Council on classification, labelling and packaging of substances and mixtures (the CLP Regulation).

US National Institute for Occupational Safety and Health (NIOSH) (1994) Immediately dangerous to life or health concentrations – o-Toluidine.