GreenScreen® Chemical Assessment

[Tert-Butyl Alcohol (75-65-0)]

Method Version: GreenScreen® Version 1.4[1]

Assessment Details[2]:

Assessment Type:	Certified
Assessment Prepared By:	WAP Sustainability
Assessment Prepared For:	WA Ecology
Date Assessment Completed:	2/31/2023
Assessment Expiration Date:	2/31/2026
Assessor Type: (Licensed GreenScreen Profiler or equivalent, Authorized GreenScreen Practitioner or Unaccredited)	Licensed GreenScreen® Profiler

[1] Use GreenScreen® Chemical Hazard Assessment Guidance (Guidance) v1.4 in Section I
[2] **Assessment Type**: GreenScreen reports are either "UNACCREDITED" (by unaccredited person), "AUTHORIZED" (by Authorized GreenScreen Practitioner), or "CERTIFIED" (by Licensed GreenScreen Profiler or equivalent);
Assessment Prepared By: Licensed GreenScreen Profilers must provide name of organization; Authorized GreenScreen Practitioners must provide their name.
Assessment Prepared For: Optional for Licensed GreenScreen Profilers, mandatory for Authorized Practitioners.
Date Assessment Completed: Assessments by Licensed GreenScreen Profilers require quality control tracked via internal documentation.
Assessment Expiration Date: Assessments expire three years from the date of completion.

GREENSCREEN BENCHMARK™ SUMMARY:

This chemical assessment report includes a GreenScreen Benchmark™ score and results for Tert-Butyl Alcohol (75-65-0) only.

No marketing claims can be made without licensing through Clean Production Action.

GreenScreen Benchmark Score: 2
Tert-Butyl Alcohol (75-65-0) was assigned a GreenScreen® Benchmark Score of 2 ("Use but Search for Safer Substitutes") as it has Moderate toxicity for Group I endpoints (Carcinogenicity, and Developmental Toxicity) and a High for flammability. This corresponds to GreenScreen® Benchmark criteria 2e and 2g in CPA 2018. Data gaps (DG) exist for endocrine activity, repeat dose neurotoxicity and respiratory sensitization. As outlined in CPA (2015) Section 13.2 (Step 8 – Conduct a Data Gap Analysis to assign a final Benchmark score), Tert-Butyl Alcohol meets requirements for a GreenScreen® Benchmark score of 2 despite hazard data gaps. In a worst-case scenario, if Tert-Butyl Alcohol were assigned a High score for endocrine activity it would be assigned a score of Benchmark 1.

HAZARD CLASSIFICATION SUMMARY

Table 1. GreenScreen Hazard Summary Table:[3,4]

GreenScreen Hazard Summary Table for Tert-Butyl Alcohol (75-65-0)																			
Group I Human					**Group II and II* Human**									**Ecotox**		**Fate**		**Physical**	
Carcinogenicity	**Genotoxicity/Mutagenicity**	**Reproductive Toxicity**	**Developmental Toxicity**	**Endocrine Activity**	**Acute Toxicity**	**Systemic Toxicity**		**Neurotoxicity**		**Skin Sensitization***	**Respiratory Sensitization***	**Skin Irritation**	**Eye Irritation**	**Acute Aquatic Toxicity**	**Chronic Aquatic Toxicity**	**Persistence**	**Bioaccumulation**	**Reactivity**	**Flammability**
						single	repeat*	single	repeat*	*	*								
M	**L**	*L*	*M*	DG	*M*	**L**	*M*	**M**	DG	**L**	DG	**L**	**H**	**L**	**L**	**M**	**vL**	*L*	**H**

Note: Hazard levels (Very High (vH), High (H), Moderate (M), Low (L), Very Low (vL)) in *italics* reflect lower confidence in the hazard classification while hazard levels in **BOLD** font reflect higher confidence in the hazard classification. Group II Human Health endpoints differ from Group II* Human Health endpoints in that Group II Human Health endpoints have four hazard scores (i.e., vH, H, M and L) instead of three (i.e., H, M and L), and are based on single exposures instead of repeated exposures. Group II* Human Health endpoints are indicated by an * after the name of the hazard endpoint or after "repeat" for repeated exposure sub-endpoints.

[3] See Appendix A for a glossary of hazard endpoint acronyms.
[4] See Appendix B for alternative GreenScreen Hazard Summary Table (Classification presented by exposure route). If such summaries are presented, they must be included in addition to the Hazard Summary Table above and placed in an Appendix to the report.

SCOPE OF ASSESSMENT

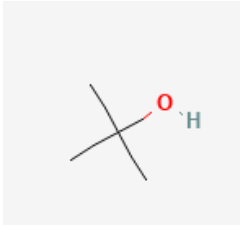
Chemical Name (CASRN): Tert-Butyl Alcohol (75-65-0):

Also Called (List Synonyms):

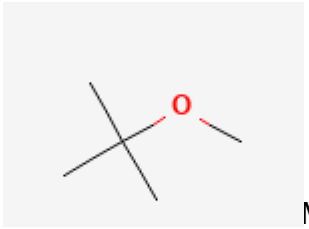
ECHA 2023
1,1-Dimethylethanol
2-Methyl-2-propanol
2-Propanol, 2-methyl- (9CI)
TBA
TBA azeotrop
TBA-84
TEBOL 88
TEBOL 90
Trimethylcarbinol
Trimethylmethanol
t-Butanol
tert-Butanol
tert-Butyl alcohol (8CI)

Chemical Structure:

PubChem 2023



Chemical Structure(s) of suitable analog(s) and/or moieties:



Methyl tertiary butyl ether (MTBE) (CAS# 1634-04-4)

Selected based on structural similarity and used as an additional line of evidence to support hazard conclusions for the following endpoints:

- Reproductive toxicity
- Endocrine activity
- Respiratory sensitization

For Inorganic Chemicals and relevant particulate organics (*if not relevant, list NA*) Define Properties:

ECHA 2023

1. Particle size (e.g., silica of respirable size): NA
2. Structure (e.g., amorphous vs. crystalline): liquid
3. Mobility (e.g., water solubility, volatility): tertiary butyl alcohol is considered to be miscible in water.
4. Bioavailability: NA

Identify potential applications/functional uses of the chemical:

PubChem 2023

1. Used for cleaning or safety in an occupational or industrial setting
2. Used in home air fresheners, including candles with a fragrance
3. In products used to control microbial pests on hard surfaces or laundry
4. Used in products for removing grease and other hydrophobic materials on hard surfaces
5. Used as part of a process, or in a piece of equipment (e.g., lubricants, adhesives, sealants, oils, paints, coatings)
6. Used as a fragrance component or ingredient
7. Used in body cleaners, washes, shower gels, facial cleaning products
8. Deodorants and antiperspirants
9. Hair care and styling products

Table 2. Environmental Transformation Products Summary

Life Cycle Stage	Transformation Pathway	Environmental Transformation Product	CAS #	Feasible (Yes or No)	Relevant (Yes or No)	GreenScreen List Translator Score or GreenScreen Benchmark Score
End of life	Biodegradation	Isobutyrate	79-31-2	Y	N	LT-UNK
End of life	Biodegradation	2-Propanol	67-63-0	Y	N	BM-2
End of life	Biodegradation	Lactate	79-33-4	Y	N	LT-UNK

Report rationale for each determination as to whether an identified environmental transformation product is feasible and relevant:

- The degradation pathway for tert-butyl alcohol was sourced from Zhong 2006 and Rosendahl 1998.
- Transformation products are determined as feasible as the transformations are likely to occur based on the functional use of the chemical across its life cycle.
- Transformation products are determined as not relevant as the transformations are likely to be transient and represent compounds that are commonly formed in the ambient environment.

HAZARD CLASSIFICATION SUMMARY[5]

GROUP I HUMAN HEALTH EFFECTS (GROUP I HUMAN)

Carcinogenicity (C): M

Tert-Butyl Alcohol was assigned a hazard classification level of Moderate for carcinogenicity based on tert-Butanol induced kidney tumors in male (but not female) rats and thyroid tumors (primarily benign) in male and female mice following long-term administration in drinking water. A moderate score is assigned as the animal data only provides suggestive evidence of cancer and does not provide unequivocal evidence of carcinogenicity in humans. The hazard conclusion is based on high quality study data specific to the compound of interest and is therefore reported with high confidence.

Data

- Lists
  - *Authoritative:*
    - *US EPA - Suggestive evidence of carcinogenic potential*
  - *Screening:*
    - *Québec CSST - WHMIS 1988 Class D2B - Toxic material causing other toxic effects.*

- Measured Data

ECHA 2023

- In a 2-year chronic toxicity study, tertiary butyl alcohol was administered to 60 F344/N male rats/dose ad libitum in drinking water at dose levels of 0, 1.25, 2.5 and 5 mg/mL for 103 weeks. Survival was decreased at the mid- and high-dose levels (24% and 20% in the control and low-dose group versus 8% and 4% in the mid- and high-dose groups). Final mean body weight was 24% less than controls in the high-dose group; low- and mid-dose groups were decreased 15% and 17%, respectively. There were no gross lesions. At the 15-month interim evaluation, incidences and severities of mineralization (type not specified) of the kidney were greater than controls in the mid- and high-dose groups, chronic progressive nephropathy was present in all males, and severity of nephropathy was slightly but not statistically significantly increased in all exposed groups. By study termination, severity of nephropathy was significantly increased in the high-dose group, incidences of linear papillary mineralization were significantly increased in all dose groups, and there was a dose-related increase in focal renal tubule hyperplasia in all dose groups. The standard microscopic evaluation showed a non-statistically significant increase in renal tubule tumors (adenomas and carcinomas combined). When the standard and extended evaluations were combined, there was an increased incidence of renal tubule tumors in all groups which reached statistical significance in the mid-dose

[5] Include computer-modeling outputs in Appendix C.

group but not the low- or high-dose groups. These results indicate that, in male rats, the kidney is the target organ for tertiary butyl alcohol repeated exposure toxicity.

- In a 2-year chronic toxicity study, tertiary butyl alcohol was administered to 60 F344/N female rats/dose ad libitum in drinking water at dose levels of 0, 2.5, 5 and 10 mg/mL for 103 weeks. Survival was decreased at the high-dose level (34% versus 58% in control group) and final mean body weight was 21% lower than the control. At the 15-month interim evaluation, necropsy body weight was significantly decreased in the 10 mg/mL group and there was a statistically significant dose-related increase in absolute and relative kidney weights at all dose levels. There were no gross lesions. Microscopic lesions included chronic progressive nephropathy in all groups at 15-months with a statistically significant increase in severity in all exposed groups by the end of the study. Kidney transitional cell hyperplasia was also significantly increased in the high-dose group. There was no increase in any tumor type at any dose level. These results indicate that, in female rats, the kidney is the target organ for tertiary butyl alcohol repeated exposure toxicity.

- In a 2-year chronic toxicity study, tertiary butyl alcohol was administered to 60 B6C3F1 male mice/dose ad libitum in drinking water at dose levels of 0, 5, 10 and 20 mg/mL for 103 weeks. Survival was decreased at the high-dose level (28% survived to study termination versus 45% in the control group). Mean body weight was decreased in the high-dose group and remained 5-10% lower than controls until the final 10 weeks of the study. There were no gross lesions. Incidences of thyroid follicular cell hyperplasia were statistically significantly increased in all groups and incidences of chronic inflammation and transitional epithelial cell hyperplasia of the bladder were increased in the high-dose group. There was no progression of the bladder hyperplasia to neoplasia. The incidence of follicular cell adenoma was marginally increased in the mid-dose group and a single follicular cell carcinoma was found in one male in the high-dose group. These results indicate that, in male mice, the thyroid gland and bladder are target organs for tertiary butyl alcohol repeated exposure toxicity.

- In a 2-year chronic toxicity study, tertiary butyl alcohol was administered to 60 B6C3F1 female mice/dose ad libitum in drinking water at dose levels of 0, 5, 10 and 20 mg/mL for 103 weeks. There were no adverse effects on survival. Mean body weight was decreased 10-15% in the high-dose group, 6% in the mid-dose group, and slightly less in the low-dose group. There were no gross lesions. Incidences of thyroid follicular cell hyperplasia were statistically significantly increased in the mid- and high-dose groups and increased incidences of chronic inflammation and transitional epithelial cell hyperplasia of the bladder were seen in the high-dose group. There was no progression of the bladder hyperplasia to neoplasia. The incidence of follicular cell adenoma was significantly greater in the high-dose group. These results indicate that, in female mice, the thyroid gland and bladder are target organs for tertiary butyl alcohol repeated exposure toxicity.

NTP 1995

- Under the conditions of these 2-year drinking water studies, there was some evidence of carcinogenic activity of t-butyl alcohol in male M44/N rats based on increased incidences of renal tubule adenoma or carcinoma (combined). There was no evidence of carcinogenic activity in female F344/N rats receiving 2.5, 5, or 10 mg/mL t-butyl alcohol. There was equivocal evidence of carcinogenic activity of t-butyl alcohol in male B6C3Fl mice based on the marginally increased incidences of follicular cell adenoma or carcinoma (combined) of the thyroid gland. There was some evidence of carcinogenic activity of t-butyl alcohol in female B6C3Fl mice based on increased incidences of follicular cell adenoma of the thyroid gland.

- Exposure to t-butyl alcohol was associated with mineralization and renal tubule hyperplasia in male rats, transitional epithelial hyperplasia and increased severity of nephropathy of the kidney in male and female rats, follicular cell hyperplasia of the thyroid gland in male and female mice, and chronic inflammation and hyperplasia of the urinary bladder in male mice and to a lesser extent in female mice.

US EPA 2021

- Under EPA Cancer Guidelines, there is suggestive evidence of carcinogenic potential for tert-butanol. tert-Butanol induced kidney tumors in male (but not female) rats and thyroid tumors (primarily benign) in male and female mice following long-term administration in drinking water (NTP, 1995). The potential for carcinogenicity applies to all routes of human exposure.

- Estimated Data: None

Mutagenicity/Genotoxicity (M): L

Tert-Butyl Alcohol was assigned a hazard classification level of Low for Mutagenicity/Genotoxicity based on weight of evidence which report negative results in most in vitro assays and all in vivo assays using the chemical of interest. Although a positive response was reported in the Ames test for strain TA102, this result was not replicated in two tests conducted in two independent GLP-accredited laboratories and, therefore, tertiary butyl alcohol is not considered mutagenic in bacteria. The results of the other in vitro studies did not provide any convincing evidence that tertiary butyl alcohol was mutagenic. The results of an in vivo micronucleus study indicate tertiary butyl alcohol is not clastogenic or aneugenic. Overall, tertiary butyl alcohol is not considered genotoxic. The low hazard conclusion is based on high quality studies reported for the chemical of interest therefore reported with high confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening: None*

- Measured Data

ECHA 2023

- The in vitro genotoxicity of tertiary butyl alcohol has been investigated in four Ames tests, a mammalian cell gene mutation assay (mouse lymphoma assay), a chromosome aberration study, a sister chromatid exchange study and a comet assay. The majority of studies were non-guideline but were conducted using methods considered equivalent to guideline. The majority of the Ames tests were negative. A positive result was reported in strain TA 102 in the presence of metabolic activation. In this study, the number of revertants increased in a dose dependent manner up to approximately 2.75 mg/plate. At this dose level, the number of revertants had almost doubled. At higher doses the number of revertants decreased dose dependently. Although the authors concluded tertiary butyl alcohol was mutagenic, no criteria as to what constituted a positive result were included in the paper and no information on toxicity was provided, which is imperative given the very high dose level employed. In follow up, the mutagenic potential of tertiary butyl alcohol in strain TA102 was investigated in a study conducted at two independent laboratories. The first laboratory used a pre-incubation method to restrict evaporation, whereas the second used the plate incorporation procedure. Both studies were conducted in accordance with OECD 471 in GLP-accredited laboratories. No significant increase in the numbers of mutations per plate was observed in either laboratory. Given the increase reported was less than 2-fold and the results of the follow-up studies showed no significant increase in mutations, tertiary butyl alcohol is not considered mutagenic in bacteria. The mutagenic potential of tertiary butyl alcohol was investigated in mammalian cells in a mouse lymphoma assay. In this study, a small dose-related increase was observed in one study conducted in the absence of S9; however, the increase was only 1.6-fold at the top dose (5000 ug/plate) and, as such, is not considered to constitute a positive result. The results of both trials conducted in the presence of metabolic activation were clearly negative. Overall, tertiary butyl alcohol does not appear to be mutagenic in mammalian cells.

- The results of a chromosome aberration study (non-guideline) and a sister chromatid exchange study have been included in NTP summary of tertiary butyl alcohol. A weak positive result was observed in one test of the SCE assay; however, as this result was not reproducible, the overall result of the study is considered negative (in accordance with the criteria outlined in OECD guideline 476). There were a number of deficiencies in the in vitro chromosome aberration study; however, a statistically significant increase in the number of cells with aberrations was observed at the top dose in one trial of the chromosome aberration study (6% of cells had aberrations compared to none in the controls) in the presence of S9. The increase is small compared to the positive control, was not dose-related, and may have been exacerbated by the absence of aberrations in control cells. On this basis the biological significance of this increase is doubtful. This conclusion is supported by the negative response observed in the in vivo micronucleus study (see discussion below). Severe toxicity in the repeat trial meant that only 13 metaphases could be scored at the top dose.

- A dose-related positive response was reported in an in vitro comet assay following incubation of tertiary butyl alcohol with HL-60 cells for one hour. No OECD guideline is available for the in vitro comet assay and at the time the methodology is unlikely to

have been well developed. There are a number of issues with the reporting and interpretation of the data that reduce confidence in the result. Firstly, the results are presented as % DNA damage; however, it is not clear what this relates to. No information on the % DNA present in the tail or tail length or the number of cells undergoing apoptosis has been reported (a minimum requirement for acceptance of an in vivo comet assay by EFSA: http://www.efsa.europa.eu/en/efsajournal/doc/2977.pdf). Furthermore, the results of three separate experiments have been grouped together and no information on the variability of the data is available to determine whether this approach was justified. Finally, there are concerns with the assessment of cytotoxicity (estimated by measuring % LDH released). The authors concluded that tertiary butyl alcohol was not cytotoxic based on a similar % of LDH released in treated cells (3.62 - 4.49%) and controls (3.29%). However, similar levels of LDH release were also reported for other substances in the same paper, but in these cases (due to a comparison to the internal control) the extent (2.79 - 4.15%) was considered indicative of cytotoxicity, raising doubts over the use of LDH as a measure of cytotoxicity. Overall, the results of this study are not considered reliable.

- One study investigating the potential for tertiary butyl alcohol to cause cytogenetic damage to peripheral blood cells of mice following 13 week exposure is available. No increase in micronucleus formation was observed following oral administration of very high doses of tertiary butyl alcohol. No change in the P/N ratio was observed; however, detection of tertiary butyl alcohol in the blood suggests the bone marrow will have been exposed. In addition, deaths in the high dose group suggest the maximum tolerated dose was exceeded.

- There are a number of studies available investigating the mutagenic potential of tertiary butyl alcohol in vitro and in vivo. Although a positive response was reported in the Ames test for strain TA102, this result was not replicated in two tests conducted in two independent GLP-accredited laboratories and, therefore, tertiary butyl alcohol is not considered mutagenic in bacteria. The results of the other in vitro studies did not provide any convincing evidence that tertiary butyl alcohol was mutagenic. The results of an in vivo micronucleus study indicate tertiary butyl alcohol is not clastogenic or aneugenic. Overall, tertiary butyl alcohol is not considered genotoxic.

<u>NTP 1995</u>

- t-Butyl alcohol was tested for induction of genetic damage in vitro and in vivo, and all results were negative. In vitro, t-butyl alcohol was negative in Salmonella typhimurium and mouse lymphoma cell mutation tests, and it did not induce sister chromatid exchanges or chromosomal aberrations in cultured Chinese hamster ovary cells. These in vitro studies were conducted with and without metabolic activation (S9). In vivo, no increase in micronucleated erythrocytes was observed in peripheral blood samples from mice administered t-butyl alcohol in drinking water for 13 weeks.

- Estimated Data: None

Reproductive Toxicity (R): *L*

Tert-Butyl Alcohol was assigned a hazard classification level of Low for Reproductive Toxicity based on weight of evidence using the available study data. Specifically in males, the only observed effect was a slight decrease in sperm motility for F0 males in the highest dose group of rats treated with tert-butanol. This effect was not observed, however, in other studies with orally treated rats and mice or in rats exposed via inhalation. In female rats, an increased length of the estrous cycle was reported in the highest dose group of orally exposed mice. This effect was not observed in similarly treated rats or in mice and rats exposed via inhalation. In addition, there was limited evidence of increased numbers of animals with long, unclear, or absent cycles in exposed rats and mice. However, these effects were limited to the highest doses tested (some with accompanying body-weight loss or lethality) and were not consistent across species or route of exposure. Furthermore, no adverse effects were reported in one- and two-generation reproductive/developmental studies on the analog MTBE providing additional support for the lack of evidence supporting reproductive effects as possible human hazards following tert-butanol exposure. This hazard conclusion was based on guideline studies. However, the hazard conclusion is based on discounting of effects observed in some studies at otherwise toxic dose levels (e.g., gestation lengths) and is therefore reported as low confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening:*
    - *GHS - Japan H361 - Suspected of damaging fertility or the unborn child [Toxic to reproduction - Category 2]*
    - *MAK Pregnancy Risk Group C*

- Measured Data

ECHA 2023

- The reproductive toxicity of tertiary butyl alcohol was investigated in an enhanced guideline reproductive/developmental screening study. In this study, Sprague-Dawley rats (12/dose/sex) were administered, via oral gavage, 0, 64, 160, 400 or 1,000 mg/kg bw/day tertiary butyl alcohol for 4 weeks pre-mating and then until termination: week 9 (males) and PND 21 (females). Selected pups were then administered tertiary butyl alcohol directly for a period of seven days before termination on PND 27. At 1,000 mg/kg bw/day, F0 toxicity manifested itself as clinical signs (unresponsiveness/ lethargy, ataxia, increased vocalization and rapid breathing). Body weight was mildly affected in males (non-significant decreases in male body weight from week one) and during the later stages of gestation in females. The only effect on feed consumption was a 15 % reduction in females over the first two weeks of lactation. During lactation a large increase in female body weight gain was observed. The significance of this finding is unknown. Increases in both liver and kidney weight were observed in males. At 400 mg/kg bw/day, lower incidences of clinical signs (as compared to the high dose) were observed in females, although only transiently (weeks 2-4). Kidney weight was increased in males of this dose level and was the only effect observed at 160

and 64 mg/kg bw/day. This increase was not statistically significant at the lowest dose and, therefore, is not considered adverse. There was no effect on mating performance or number of pregnancies observed in any treatment group. A slight effect on gestation length was observed; with half the females from the top dose group and almost half the females in the mid dose with a gestation length greater than 22 (all but one was a shift to 23 days). The shift was reported to be within the normal range (21-23 days, although with a distinct node of 22) and is therefore considered a chance finding. In the F1 generation, there was no effect on the number of implantations per pregnancy; however, significant pup mortality was observed at 1,000 mg/kg bw/day. Six out of 153 pups were stillborn with a further 32 pups dying between days 1-4 (the majority were found dead on day 1). The deaths include one total litter loss. The incidence of still born deaths is likely to be within the normal variation for this type of effect and is therefore not considered treatment related. The deaths observed post-parturition were only observed in the presence of maternal toxicity (unresponsiveness/lethargy and ataxia) and are considered likely to be a secondary consequence of this toxicity and not a direct effect of tertiary butyl alcohol. The parental NOAEL was 64 mg/kg bw/day based on increased kidney weight in males. The NOAEL for this study for reproductive toxicity is 1,000 mg/kg bw/day, the highest dose tested.

- Data from a two-generation reproductive toxicity study with methyl tertiary butyl ether was also used to evaluate the potential for tertiary butyl alcohol to cause reproductive toxicity. Methyl tertiary butyl ether is rapidly and irreversibly metabolized to tertiary butyl alcohol and significant blood levels of tertiary butyl alcohol can be found following methyl tertiary butyl ether exposures. Methyl tertiary butyl ether and tertiary butyl alcohol have similar properties of toxicity including central nervous system effects and decreases in body weight or body weight gain at high exposure concentrations. In terms of reproductive toxicity, neither chemical caused any effect on measures of fertility or reproductive success, in either male or female rats.

US EPA 2020

- Several studies evaluated reproductive effects [a one-generation, oral reproductive study and sub-chronic effects in rats and mice following oral and inhalation exposure (NTP, 1997, 1995)] in animals exposed to tert-butanol via oral gavage, drinking water, or inhalation for ≥63 days. The collection of studies evaluating reproductive effects of tert-butanol is limited by the absence of two-generation reproductive oral or inhalation studies and by the lack of human studies on reproduction. The design, conduct, and reporting of each study were reviewed, and each study was considered adequate to provide information pertinent to this assessment. Reproductive endpoints, such as reproductive organ weights, estrous cycle length, and sperm effects were examined following either oral or inhalation exposure. In males, the only significant effect observed was a slight decrease in sperm motility for F0 males treated with 1,000 mg/kg-day tert-butanol. No significant changes in sperm motility were reported following oral exposure in other rat studies or via inhalation exposure in mice or rats. In addition, the reduced motility in treated animals falls within the range of historical control data, and therefore, its biological significance is uncertain. In female B6C3F1 mice, estrous cycle length was increased 28% following oral exposure to 11,620 mg/kg-day (NTP, 1995). No significant changes in estrous cycle length were

observed following oral exposure in rats or inhalation exposure in mice or rats. However, there was some evidence of increased numbers of animals with long, unclear, or absent cycles in tert-butanol-exposed mice (oral/inhalation) and rats. It is noteworthy that these effects were limited to the highest doses tested with some doses accompanied by body-weight loss or lethality.

NTP 1995

- In vitro studies showed that ethanol reduced the fertilizing capacity of mouse spermatozoa at concentrations commonly observed after ethanol ingestion by man and experimental animals (100 to 44 mg%). At similar concentrations, t-butyl alcohol had no effect on fertilization.

- Estimated Data: None

Developmental Toxicity incl. Developmental Neurotoxicity (D): *M*

Tert-Butyl Alcohol was assigned a hazard classification level of Moderate for developmental toxicity based on reduction in mean litter size, a decrease in the number of live born per pregnancy, an increase in the number of stillborn pups, increased pup mortality up to PND 4, a decrease in mean pup body weight at birth which continued to weaning, decreased fetal weights and increased skeletal variations and developmental delay in postnatal physiological and psychomotor performance. A moderate score is assigned as the observed effects do not sufficiently provide convincing evidence to place the substance as a GHS Category 1 for developmental toxicity. Specifically, the conclusion of developmental toxicity is mixed amongst the available studies and numerous studies report developmental effects occurring alongside with maternal toxicity This hazard conclusion was based on quality studies for the compound of interest; however, the developmental effects occurred at doses that were also associated with maternal toxicity. Therefore, the hazard score is reported as low confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening:*
    - *GHS - Japan H361 - Suspected of damaging fertility or the unborn child [Toxic to reproduction - Category 2]*
    - *MAK Pregnancy Risk Group C*

- Measured Data

ECHA 2023

- In a reproduction/developmental toxicity screening study, tertiary butyl alcohol was administered to 12 Sprague-Dawley rats/sex/group by oral gavage at dose levels of 0, 64, 160, 400, and 1,000 mg/kg bw/day for up to 63 days in males and from 4 weeks prior to mating through PND 21 in females. For dams receiving 1,000 mg/kg bw/day tertiary butyl alcohol through gestation and lactation, there was a significant reduction in mean litter size, a decrease in the number of live born per pregnancy, an increase in the number of stillborn pups, increased pup mortality up to PND 4,

and a decrease in mean pup body weight at birth which continued to weaning. At this dose level, mild to moderate transient systemic toxicity was observed in both sexes in the parental generation including reversible CNS effects such as lethargy and ataxia, and reduced feed consumption and weight gain. No significant toxicity was observed at any other dose level. The NOAEL for developmental/reproductive effects was 400 mg/kg bw/day. The NOAEL for overall toxicity was 160 mg/kg bw/day.

- In a teratological evaluation, groups of 15-20 pregnant Sprague-Dawley rats were exposed to tertiary butyl alcohol by whole body inhalation at concentrations of 0, 2000, 3,500 or 5,000 ppm for 7 hr/day on gestation days 1-19. Dams were sacrificed on Gestation Day 20 and fetuses were individually weighed and examined for external, skeletal and visceral malformations and variations. All concentrations were maternally toxic, as manifested by observations of an unsteady gate at the end of each exposure period. In addition, exposure at 5,000 ppm caused a statistically significant decrease in food consumption and maternal body weight. There was a dose-related increase in developmental toxicity, in the form of decreased fetal weights and increased skeletal variations, but no evidence of teratogenicity, even at maternally toxic exposures. Under conditions of the study, the NOAEC was 5,000 ppm for teratogenicity and the LOAEC was 2,000 ppm for developmental toxicity and maternal toxicity.

- Although this study used a limited number of animals at a single dose level and was not strictly conducted according to current regulatory guidelines, it did show that tertiary butyl alcohol does not cause teratogenicity, even at a dose level causing feto lethality. Groups of gravid CBA/J and C57BL/6J mice receiving 10.5 mmoles/kg of tertiary butyl alcohol twice daily by oral intubation on days 6 through 18 of gestation had a statistically significant increase in the number of resorptions and a significant decrease in live fetuses per litter. No external malformations were observed while skeletal variations consisted of a non-statistical increase in the number of minor variations (misaligned or under ossified sternebrae and under ossified supraoccipital bones) in tertiary butyl alcohol-treated animals. No data were presented on maternal toxicity in the present study. Other authors have reported that when the CBA/J strain of mice was treated with ethanol, the presence of fetal resorptions was always associated with abnormalities or fetal abnormalities occurred in the absence of significant fetal mortality. When a group of non-pregnant C57BL/6J mice was treated with 10.5 mmoles/kg of tertiary butyl alcohol twice daily for 3 days, there was no evidence that repeatedly treated mice eliminated tertiary butyl alcohol more rapidly than control animals receiving a single dose. Elimination was complete after 12 hours suggesting that accumulation did not occur at that dose-level. The blood concentration time profile in non-pregnant mice indicated that peak concentrations of about 13 mM and average concentrations of 8 mM were attained. Under these test conditions, teratogenicity did not occur at a dose level that caused significant fetotoxicity.

- In a postnatal toxicity study, tertiary butyl alcohol was administered to groups of 15 pregnant Swiss Webster mice/group at concentrations of 0, 0.5, 0.75, and 1.0% (w/v) in a modified liquid diet from gestation day 6-20. Fecundity was significantly

affected in the 0.75% and 1.0% groups. There was a decrease in total number of litters, neonates per litter and the total number of stillborn pups. Administration of the test material also caused a dose-related effect on developmental parameters in the pups in this study in terms of fetal weight, open field behavior, cliff avoidance, and roto-rod performance. With the administration of 1% tertiary butyl alcohol, there was an increase in time of eye opening of 2-4 days compared to the other groups. Also, control pups raised by their own dams took over a second longer to right themselves (statistically significant) than pups fostered with dams given standard rat chow during pregnancy. In this study, administration of high concentrations of tertiary butyl alcohol to dams during gestation caused behavioral deficits in the pups at exposure concentrations that were not reported to cause gross structural anomalies.

NTP 1995

- (Same study reported under ECHA 2023) The effects of prenatal administration of t-butyl alcohol and ethanol on postnatal development were compared in Swiss-Webster mice. A liquid diet containing either 0.5%, 0.75%, or 1.0% t-butyl alcohol or 3.6% ethanol was fed to pregnant mice from day 6 to day 20 of gestation. The t-butyl alcohol diet resulted in a dose-related reduction in number of litters, litter size, and birth weights, and an increase in the number of stillborn. Testing of pups indicated that t-butyl alcohol was approximately five times more potent than ethanol in producing a developmental delay in postnatal physiological and psychomotor performance.

- t-Butyl alcohol and ethanol were compared for their ability to induce microcephaly in the neonatal rat. Neonatal Long-Evans rats were reared using an artificial feeding technique (cannulation) from postnatal day 4 through day 18. On postnatal days 4 through 7, t-butyl alcohol or ethanol was administered in the milk formula. Mean daily doses of t-butyl alcohol were 0.60, 1.44, 2.16, and 2.69 g/kg body weight. The doses were calculated to be of equal anesthetic value to the ethanol doses by using membrane-to-buffer partition coefficients. Following the 4-day alcohol exposure, all animals were given a plain milk formula until day 18. A similar degree of microcephaly were present in both alcohol-treated groups but not in controls. The general impairment of brain growth could be due to the membrane solubilizing properties of the alcohols.

- (Same study reported under ECHA 2023) In a teratology assessment of t-butyl alcohol, 1-butanol, and 2-butanol, pregnant Sprague-Dawley rats were exposed by inhalation to 0, 2,000, 3,500, or 5,000 ppm t-butyl alcohol for 7 hours per day on gestation days 1 through 19. Dams were sacrificed on day 20, and fetuses were examined for skeletal abnormalities or visceral defects. Dose-related reductions in fetal weight were observed for each of the butanol isomers; however, concentrations 50 times the current permissible exposure limit (100 ppm) did not produce teratogenicity. In further investigations, pregnant Sprague-Dawley rats were exposed to 2,000 or 4,000 ppm t-butyl alcohol by inhalation for 7 hours per day on gestation days 1 through 19; males were similarly exposed for 6 weeks and mated to unexposed females. The high concentration of t-butyl alcohol was maternally toxic, reducing maternal feed intake and weight gain. However, the few behavioral or neurochemical effects noted in the offspring on tests conducted through 90 days of age were not considered biologically significant.

US EPA 2020

- Developmental effects of tert-butanol observed after oral exposure (liquid diets or gavage) in several mouse strains and one rat strain include measures of embryo-fetal loss or viability (e.g., increased number of resorptions, decreased numbers of neonates per litter) and decreased fetal body weight observed decreases in pup body-weight gain during postnatal days (PNDs) 2 - 10; however, the data suggest that this effect might be due to altered maternal behavior or nutritional status. In addition, a single-dose study reported a small increase in the incidence of variations of the skull or sternebrae in two mouse strains. Although variations in skeletal development were noted in the study, no malformations were reported. Similar developmental effects were observed after whole-body inhalation exposure in Sprague-Dawley rats for 7 hours/day on gestation days (GDs) 1−19. Fetal effects included dose-related reductions in body weight in male and female fetuses and a higher incidence of skeletal variations when analyzed based on individual fetuses (but not on a per litter basis).

- In these studies, fetal effects were generally observed at high doses that cause toxicity in the dams as measured by clinical signs (e.g., decreased [~7−36%] body weight gain and food consumption and reported ataxia and lethargy). As stated in the Guidelines for Developmental Toxicity Risk Assessment (US EPA, 1991), “an integrated evaluation must be performed considering all maternal and developmental endpoints.…[W]hen adverse developmental effects are produced only at doses that cause minimal maternal toxicity; in these cases, the developmental effects are still considered to represent developmental toxicity and should not be discounted.” Although, at doses of “excessive maternal toxicity…information on developmental effects may be difficult to interpret and of limited value.” In considering the observed fetal and maternal toxicity data following tert-butanol exposure and the severity of the maternal effects, the role of maternal toxicity in the developmental effects observed at the doses used remains unclear. Specifically, discerning from the available data whether the fetal effects are directly related to tert-butanol treatment or are secondary to maternal toxicity is not possible.

- Estimated Data: None

Endocrine Activity (E): DG

Tert-Butyl Alcohol was assigned a hazard classification level of data gap for Endocrine Activity based on lack of adequate studies. While one study reporting negative findings for potential interactions with the steroidogenic pathway was located, this single study does not provide sufficient evidence to suggest the absence of endocrine activity is associated with the chemical. No additional data was located for any of the compound of interest of analogs of the compound.

Data

- Lists
    - *Authoritative: None*
    - *Screening: None*

- Measured Data

Peyster 2014

- The analog MTBE is a solvent and fuel additive included in reformulated gasoline to increase combustion efficiency. While widespread use in motor fuels in the U.S. was discontinued after MTBE was detected in surface and ground waters due to concerns about environmental persistence and water quality, it is still manufactured in the U.S. for export. Questions concerning the etiology of rat Leydig cell and mouse liver tumors identified in extremely high dose cancer studies have led to an interest in evaluating potential hormonal imbalances and endocrine system involvement. To address the possibility that MTBE or its metabolite, tert-butanol (TBA), are interacting with components of the endocrine system that are involved in steroidogenesis a number of targeted experiments were performed focusing mostly on the primary gonadal steroids, estradiol and testosterone. The goal of the experiments was to gain a better understanding of potential interactions with the steroidogenic pathway, including effects specifically on aromatase, the P450 enzyme that converts testosterone to estradiol. In three GLP-compliant in vitro guideline studies, MTBE and TBA were classified as non-binders to the androgen receptor, were classified negative for effects on testosterone and estradiol in the steroidogenesis assay and were classified as non-inhibitors of aromatase activity. In three 14-day in vivo experiments involving gavaging of male Sprague-Dawley rats with doses of MTBE ranging from 400 to 1,500 mg/kg bw/day, the lack of definitive and consistent supporting statistically significant findings in steroid hormone measurements and aromatase activity and mRNA measured in liver and testis microsomes further suggested that it is unlikely that MTBE is interacting with the endocrine system directly. Evidence of other underlying systemic effects were also seen, including reduced body weight gain, increased adrenal weights, and elevated corticosterone suggestive of a more general stress response. Taken together, the results from these studies suggest that MTBE and TBA do not directly impact the steroidogenic pathways involved in estrogen and androgen production.

- Estimated Data: None

GROUP II AND II* HUMAN HEALTH EFFECTS (GROUP II AND II* HUMAN)

Note: Group II and Group II endpoints are distinguished in the v1.4 Benchmark system (the asterisk indicates repeated exposure). For Systemic Toxicity and Neurotoxicity, Group II and II* are considered sub-endpoints. See GreenScreen Guidance v1.4, Annex 2 for more details.*

Acute Mammalian Toxicity (AT): *M*

Tert-Butyl Alcohol was assigned a hazard classification level of Moderate for acute mammalian toxicity based the authoritative listing as a GHS H332 Category 4 acute toxicant. It should be noted that the available LD50 values >2000 mg/kg bw and LC50

values >20 mg/L do not support a moderate classification. The hazard score is based on authoritative a listing however the available data suggests a low score. Therefore, the hazard score is reported with low confidence.

Data

- Lists
  - *Authoritative:*
    - *GHS Harmonized H332 – Harmful if inhaled [Acute toxicity (inhalation) – Category 4]*
  - *Screening:*
    - *GHS - Australia H332 - Harmful if inhaled [Acute toxicity (inhalation) - Category 4]*
    - *GHS - Malaysia H332 - Harmful if inhaled [Acute toxicity (inhalation) - Category 4]*

- Measured Data

ECHA 2023

- The acute oral toxicity of tertiary butyl alcohol is well understood. In acute oral toxicity studies conducted according to US EPA OPPTS 870.1100, tertiary butyl alcohol had an acute oral LD50 value of 3384 mg/kg bw in male rats, 2743 mg/kg bw in female rats and 3046 mg/kg bw (combined).

- Under the conditions of this study, the oral LD50 of tertiary butyl alcohol in an unspecified strain of rabbit was 3558 mg/kg bw. .

- Under the conditions of this study, the oral LD50 of tertiary butyl alcohol in an unspecified strain of rat was 3500 mg/kg bw. .

- The acute dermal toxicity of tertiary butyl alcohol is well understood. In an acute dermal toxicity study conducted according to US EPA Guidelines (1978), tertiary butyl alcohol had an acute dermal LD50 value of >2,000 mg/kg bw in male and female rabbits. Based on these data, tertiary butyl alcohol is demonstrated to be a low order of acute toxicity following skin contact.

- In an acute inhalation study, a single group of 5 male and 5 female rats was exposed whole-body for 4 hours to a vapor concentration of 10,000 ppmV tertiary butyl alcohol (single dose level; approximately 30 mg/L)and observed for 14 days. One female was found dead on study Day 3. The LC50 is reported as >10,000 ppm.

- Estimated Data: None

Systemic Toxicity/Organ Effects incl. Immunotoxicity (ST-single): L

Tert-Butyl Alcohol was assigned a hazard classification level of Low for single dose systemic toxicity/organ effects. The hazard score is based on no indication of specific target organ toxicity reported following oral, inhalation or dermal oral exposures. While some test-material related hemorrhage and congestion in various visceral organs were

observed in a few animals following oral exposures this effect is determined to be not of toxicological significance and was only observed to occur at very high concentrations (i.e., >2,000 mg/kg bw). The low hazard conclusion is based on study data specific to the chemical and therefore is reported with high confidence.

Data

- Lists
  - *Authoritative:*
    - *H335 - May cause respiratory irritation [Specific target organ toxicity - single exposure; Respiratory tract irritation - Category 3]*
  - *Screening:*
    - *GHS - Japan H335 or H336 [Specific target organs/systemic toxicity following single exposure - Category 3]*

- Measured Data

ECHA 2023

- In an acute oral toxicity test, the oral LD50 was 3384 mg/kg bw in male rats, 2743 mg/kg bw in female rats, and 3046 mg/kg bw for both sexes combined when administered a single dose of tertiary butyl alcohol. No compound-related macroscopic lesions were observed in the low-dose group. Test-material related hemorrhage and congestion in various visceral organs were observed in a few animals during postmortem examination of animals dying on study or sacrificed at the termination of the study. This effect on blood vessel integrity was found generally among males and females in the two highest dose groups.
- In an acute dermal toxicity test, no deaths were reported when a limit dose of 2000 mg/kg bw of undiluted tertiary butyl alcohol was applied to the abraded skin of male and female New Zealand rabbits for twenty-four hours. Microscopically, dermal inflammatory cell infiltrate occurred in both treated and untreated skin of rabbits. However, in general, the severity and extent of these changes were comparatively less in the untreated skins. Trace or mild acanthosis and hyperkeratosis were seen in the treated skin of five rabbits (one male and four females), but not in any of the untreated skins examined. Mild dermal fibroplasia was observed in the treated skin of four rabbits (one male and three females) but was absent in any of the untreated skin examined.
- Under the conditions of this study, the 4-hour inhalation LC50 of tertiary butyl alcohol in male and female Sprague-Dawley rats was greater than 10000 ppm (actual concentration) or 36 mg/L (nominal concentration). Exposure to high concentrations of tertiary butyl alcohol may cause reversible CNS effects and ocular irritation. Under the conditions of this study, the 4-hour inhalation LC50 of tertiary butyl alcohol in male and female Sprague-Dawley rats was greater than 10000 ppm (actual concentration) or 36 mg/L (nominal concentration). Exposure to high concentrations of tertiary butyl alcohol may cause reversible CNS effects and ocular irritation. Both male and female groups appeared to gain weight normally during the two week post-exposure observation period. At necropsy, the most notable observation was that four of the ten animals (3 males, 1 female) in the study were observed to have focal areas of redness on the lungs. The only female rat that exhibited macroscopic abnormalities at necropsy was the animal which died on the third day of the post-

exposure period.

Systemic Toxicity/Organ Effects incl. Immunotoxicity (ST-repeat) (Group II*): *M*

Tert-Butyl Alcohol was assigned a hazard classification level of Moderate for repeated dose systemic toxicity/organ effects based on a 2-year rat oral LOAEL of 1.25 mg/mL (90 mg/kg bw/day) derived for general toxicity (decreased body weight) and a 90-day rat inhalation LOAEL of 135 ppm (0.4 mg/L; equivalent to 406 mg/m^3) based on mild nephropathy observed in kidneys of animals from both sexes compared to minimal effects in the control. Both the oral and inhalation LOAELs exist within the Moderate classification break. In both cases the LOAEL is the lowest concentration tested and a NOAEL could not be determined. Based on the absence of a NOAEL value the hazard score is reported with low confidence .

Data

- Lists
  - *Authoritative: None*
  - *Screening: None*

- Measured Data

ECHA 2023

- In a 90-day NTP study, groups of 10 male and 10 females F344/N rats were administered 0, 2.5, 5, 10, 20 and 40 mg/mL TBA (equivalent to 0, 230, 490, 840, 1,520 or 3,610 mg/kg bw/day in males and 290, 590, 850, 1,560, 3,620 mg/kg bw/day in females) via the drinking water. All males and six of the females in the top dose died during the study. Bodyweight was significantly lower in top dose females (21%), and in males receiving 20 mg/mL (17%) and 10 mg/mL (12%). Water consumption in both sexes dosed ≥10 mg/mL was less than control animals, correlating with the decrease in urine volume. A dose-related increase in clinical signs (ataxia, hypoactivity (males), hyperactivity (females and emaciation) were reported (specific details of the effects at each dose were not provided). The kidney, bladder and liver were identified as target organs. An increase in absolute and relative kidney weight was observed in both sexes at all doses (>12% absolute weight at 2.5 mg/mL). A dose related increase in the severity of nephropathy (also referred to as chronic progressive nephropathy; CPN) was observed in males from 2.5 mg/mL (see table below – note at the top dose level all males died). This lesion was described in the NTP report as spontaneous background lesion in F344 rats, which was exacerbated by treatment. Exacerbation of this lesion is regarded as relevant to human health (see section 7.9.6.1 for a more detailed discussion). A statistically significant increase in the incidence of mineralization was observed in males ≥10 mg/mL. In females, a dose related increase in the incidence of nephropathy but not of severity was observed from 10 mg/m^2 . Staining of kidneys (Mallory Heidenhaim and Lee's methylene blue basic fuschin stains) revealed an increase in the presence of hyaline droplets and crystalline structures associated with the hyaline droplets within the renal tubule epithelium and tubule lumina of male kidneys (but not females) of all dose groups. The presence of the hyaline droplets

and mineralization suggests TBA induces α2u-globulin nephropathy in male kidneys; a rat-specific effect not considered relevant to humans. Both types of nephropathy are known to increase kidney weights. As exacerbation of CPN is considered relevant to human health and it is not known whether the increased kidney weights were caused by CPN or α2u-globulin nephropathy, the kidney weight findings cannot be dismissed and are considered relevant for human health. Absolute and relative liver weights of females at all dose levels and relative liver weights of males at ≥ 5 mg/mL were increased. Apart from increased alanine transaminase activity in top dose females, these increases were not accompanied by any other effects. In the bladder, grossly visible calculi, microscopic inflammation3 of the lamina propria and hyperplasia of the transitional epithelium was observed in males in the 20 mg/mL group. Inflammation and hyperplasia were also observed in females dosed 40 mg/mL. Due to the effects in the kidney in males, a LOAEL of 2.5 mg/mL (equivalent to 230 mg/kg bw) is derived from this study.

- In a 2-year NTP carcinogenicity study, groups of 60 male and 60 females F344/N rats were administered 0, 1.25 (males only), 2.5, 5 or 10 (females only) mg/mL TBA (equivalent to 0, 90, 200 and 400 mg/kg bw/day in males and 0, 180, 330 and 650 mg/kg bw/day in females) via the drinking water. After 15 months, 10 animals/sex/group were sacrificed and urinalysis, hematological and organ weight (brain, kidney and liver) investigations conducted. At 15 months, females in the top two doses had reduced urine specific gravities and volume consistent with their decreased water intake. Terminal survival rates were reduced in a dose dependent manner in males (12, 10, 4 and 2 animals from the control to high dose group), whereas survival in females was lower in top dose females only (29, 28, 26 and 17 animals from the control to the high dose group). In males, terminal bodyweight was adversely reduced from the lowest dose (15%), whereas female bodyweight was only reduced at the top dose (20%). There was a dose related increase in water consumption in males and a dose-related decrease in females. In this study, the kidney was identified as the target organ. At the 15 month interim, relative kidney weights of 2.5 and 5 mg/mL treated males and absolute and relative kidney weights of all treated females were statistically significantly increased compared to controls. Nephropathy was present in all animals, with the severity slightly increased in all exposed groups of males (non-statistically significantly) (see table below). Increased incidence and severity of mineralization of the kidney was observed in 2.5 and 5 mg/mL treated males, although this increase was only statistically significant at 5 mg/mL. At termination, a dose related increase in incidence and severity of foci of linear mineralization was observed in males. This type of mineralization has been specifically associated with accumulation of α2u-globulin in male rats4 . The severity of the kidney nephropathy was increased in all female dose groups and in 5 mg/mL treated males when compared to controls. Signs associated with nephropathy (inflammation, mineralization and transitional epithelial hyperplasia) were also increased from 2.5 mg/mL in males and 5 mg/mL in females. There was no progression of transitional cell hyperplasia to neoplasms. The incidence of focal renal tubule hyperplasia was increased in all treated male groups (not dose dependently). Renal tubule hyperplasia was also observed in one 10 mg/mL female. Additional male rats with hyperplasia (11, 13, 11 and 19 animals in control through to high dose groups) were identified following examination of further sections of the

kidney in male rats. The extent of the increased hyperplasia was statistically significant in males at 5 mg/mL. A LOAEL for toxicity of 1.25 mg/L (equivalent to 90 mg/kg bw/day) TBA was derived, since adverse effects were observed at all doses; in the low-dose group, these comprised effects on bodyweight in males.

- In a 13-week NTP study, groups of 10 male and 10 female B6C3F1 mice were administered TBA via the drinking water at doses of 0, 2.5, 5, 10, 20 and 40 mg/mL (equivalent to 0, 350, 640, 1,590, 3,940 or 8,210 mg/kg bw/day in males and 500, 820, 1,660, 6,430, 11,620 mg/kg bw/day in females). At 40 mg/mL, 2 males and 1 female died. At this dose, terminal bodyweight was lower in both males (25%) and females (15%) and was also lower in males of the 20 mg/mL dose group (14%). Clinical findings included emaciation, ataxia and hypoactivity in males and emaciation in females. The increased estrous cycle length observed in females of this dose was considered a consequence of the toxicity observed and not a specific effect of TBA. Water consumption was reduced in the top dose, although not throughout the study in males. Slight increases in hemoglobin and hematocrit values at the top two doses are consistent with slight dehydration. The kidney and bladder were identified as target organs. Absolute (12%) and relative (35%) kidney weights of 40 mg/L females were increased compared to controls. No nephropathy was noted. In the bladder, hyperplasia of the transitional epithelium was present in all males and three females dosed with 40 mg/mL and in six male mice dosed with 20 mg/mL7. Chronic inflammation of the bladder (primarily macrophages, lymphocytes and plasma cells) was observed in six males and six females in the 40 mg/mL and six males in the 20 mg/mL group. A NOAEL of 10 mg/mL (equivalent to 1590 mg/kg bw/day) can be derived from this study based on the effects on bodyweight and bladder at 20 mg/mL.

- In a 2-year NTP carcinogenicity study, groups of 60 male and 60 female B6C3F1 mice were administered 0, 5, 10 or 20 mg/mL TBA (0, 540, 1,040 and 2,070 mg/kg bw/day in males and 0, 510, 1,020 and 2,110 mg/kg bw/day in females) via the drinking water. Survival was reduced in high dose males (27, 36, 34, 17 survivors from control to high dose) and as a consequence the interim kill was not conducted in either sex. Survival was similar in females from all treated groups (36, 35, 41, 42 from control to high dose). In the 20 mg/mL group, female bodyweight was 10-15 % lower than control from week 13 and was 12% lower than controls at the end of the study. Male bodyweight gain was 5-10% lower at various stages during the treatment period. There was no difference at termination. Mean bodyweight of 10 mg/mL treated females was about 6% lower than controls throughout the study and bodyweight was slightly lower than controls in 5 mg/mL females as well. Water consumption in both sexes was similar to the controls. In this study the thyroid, urinary bladder and liver were identified as target organs. In the thyroid a dose-related increase in the incidence of follicular cell hyperplasia was observed in both sexes. The extent of this increase reached statistical significance in all dose groups in males and in the top two dose groups in females. In the urinary bladder, the incidence of chronic inflammation and transitional cell epithelium hyperplasia was increased at the top dose in both sexes. Similarly, an increase in the number of males with fatty liver was also observed at the top dose. A LOAEL of 5 mg/mL (510 mg/kg bw/day) was derived for toxicity, since adverse effects were reported at all

doses. At the LOAEL, a statistically significant increase in the incidence of follicular cell hyperplasia in both sexes occurred.

- In a 90-day NTP study, groups of 10 male and 10 female F344/N rats were exposed to TBA by inhalation at concentrations of 0, 135, 270, 540, 1,080 and 2,100 ppm (equivalent to 0, 406, 825, 1,643, 3,274 and 6,369 mg/m^3) for 6 hours a day, 5 days per week. At 2100 ppm, emaciation and hypoactivity were observed at one observation time point only. No effects on bodyweight were observed. The minor changes in clinical chemistry and hematology were not sufficiently severe to be considered adverse. At the top concentration, absolute kidney weight was increased in males (10%) and was accompanied by signs of mild chronic nephropathy in all males. In top concentration females, relative liver weight was increased (9%). At the next concentration (1,080 ppm), similar effects were noted in the male kidney (11% ↑ weight and nephropathy) and female liver (9% ↑ relative weight). At the lower concentration levels, nephropathy was less severe than at the higher concentrations, but still more severe than in the control (nephropathy considered mild in controls). Sections of kidney from male rats in the 0, 1,080 ppm and 2,100 ppm concentration group were stained by Mallory Heidernhain method for the presence of tubular hyaline droplets. There were no differences between the controls and treated groups in the number, shape or size of the droplets. In this study no NOAEC could be identified due to the increase in severity of the chronic nephropathy observed in males of all concentration groups. The LOAEC is the lowest concentration tested 135 ppm (406 mg/m^3)

- In a 90-day NTP study, groups of 10 male and 10 female B6C3F1 mice were exposed to TBA by inhalation to 0, 135, 270, 540, 1,080 and 2,100 ppm (equivalent to 0, 406, 825, 1643, 3274 and 6369 mg/m^3) for 6 hours a day, 5 days per week. In the top concentration group, 1 male died in week. Five males also died in the 1,080 ppm concentration group; however, these deaths were attributed to problems with the water/food and not treatment with TBA. Bodyweight (↓ 19%) and bodyweight gain (↓ 24%) were adversely affected in top concentration females, whereas only bodyweight gain (↓ 19%) was affected in the 1,080 ppm concentration group. Relative female liver weights were also increased in these two groups (9 and 20% in females of the 1,080 and 2,100 ppm groups, respectively); however, as there were no histopathological changes observed, these increases may be secondary to the reduced bodyweight, particularly at the top concentration. No adverse treatment related effects were noted at ≤540 ppm. The NOAEC for this study is 540 ppm (1643 mg/m^3); at the next concentration level (1080 ppm) there were effects on female bodyweight gain.

ECHA 2023 Summary

The study NOAELs and LOAELs are summarized in the following table:

Route	Duration	Species	LOAEL/C	NOAEL/C	Reference
Oral, drinking water	90-day	Rat	230 mg/kg bw/day	Not derived	NTP (1995)
Oral, Drinking water	2-year	Rat	90 mg/kg/bw/day	Not derived	NTP (1995)
Oral, Drinking	90-day	Mice	3940 mg/kg	1590 mg/kg	NTP (1995)

water			bw/day	bw/day	
Oral, Drinking water	2-year	Mice	510 mg/kg/bw/day	Not derived	NTP (1995)
Inhalation	90-day	Rat	406 mg/m^3	1,385 mg/m3	NTP (1997)
Inhalation	90-day	Mice	3274 mg/m^3	1643 mg/m^3	NTP (1997)

- In rats, for the oral route, sub-chronic and carcinogenicity studies consistently showed the kidney to be the principal target organ for tertiary butyl alcohol toxicity. The effects in the kidney ranged from increased weight, increased incidence/severity of CPN and the presence of hyaline droplet (males only) indicative of α2u-nephropathy. Other effects, seen at higher exposures (40 mg/mL) in the sub-chronic studies included death and body weight effects. In the carcinogenicity studies, bodyweight effects occurred in males of all dose levels (≥90 mg/kg bw/day) and may be a result of the increased kidney toxicity in these animals. A dose related increase in clinical signs (ataxia, hypoactivity, hyperactivity and emaciation) was observed in the oral sub-chronic study and similar effects observed in the top dose of the chronic study (10 mg/mL). LOAELs (but not NOAELs) of 230 mg/kg bw/day for sub-chronic and 90 mg/kg bw/day for chronic exposure were derived via the oral route.

- In the sub-chronic inhalation study similar effects were observed as via the oral route, with the kidney as the target organ. In the sub-acute study, the driving effect was clinical signs (hypo/hyperactivity and ataxia) observed at all doses ≥2,759 mg/m^3. At higher doses (≥10,680 mg/m^3) other effects included deaths and body weight effects. A NOAEC of 1,385 mg/m3 was derived for sub-acute exposure and a LOAEC of 406 mg/m^3 was derived for sub-chronic exposure.

- Repeated oral exposure of mice showed them to be less sensitive to tertiary butyl alcohol than rats. In the sub-chronic study effects were only observed at doses ≥20 mg/mL. At this dose level, effects on body weight and in the bladder (transitional cell hyperplasia and chronic inflammation) were observed. Deaths, clinical signs, kidney and liver effects were observed at the highest dose (40 mg/mL). A NOAEL was derived for sub-chronic exposure of 10 mg/mL (1590mg/kg bw/day). In the carcinogenicity study, effects on the bladder and body weight were again observed at 20 mg/mL (highest dose tested). The target organ in this study was the thyroid with a dose-related increase in the incidence of thyroid follicular cell hyperplasia observed from the lowest dose. A LOAEL of 510 mg/kg bw/day was derived from this study.

- Via the inhalation route in mice, effects were more varied. In the subacute study, clinical signs (hypoactivity, hyperactivity and urogenital wetness) were observed at concentrations of ≥1,750 ppm (5305 mg/m^3). Other effects were observed at the highest dose only (7,000 ppm or 21,194 mg/m^3) and included death, liver weight increase and decreased thymus weight. In the sub-chronic study, no clinical signs were observed, although it is noted that the top concentration in this study (2,100 ppm) is relatively close to the LOAEC for these effects in the 18-day study and may reflect experimental variation. In the sub-chronic study, reduced body weight was the lead effect and was reduced from 1,050 ppm (3,274 mg/m^3). A NOAEC of 900 ppm (2,759 mg/m^3) was derived for sub-acute exposure and 540 ppm (1,643 mg/m^3) for sub chronic exposure.

- Estimated Data: None

Neurotoxicity (N-single): M

Tert-Butyl Alcohol was assigned a hazard classification level of Moderate for single dose neurotoxicity based on reversible neurological effects reported in numerous acute mammalian toxicity studies. Based on the reported reversible nature of these effects a GHS Category 3 for single exposure is assigned to the compound. The hazard score is based on high quality study data for the compound of interest and therefore is reported with high confidence.

Data

- Lists
    - *Authoritative: None*
    - *Screening:*
        - *H336 - May cause drowsiness or dizziness (unverified) [Specific target organ toxicity - single exposure; Narcotic effects - Category 3]*

- Measured Data

ECHA 2023

- In an acute oral toxicity test, the oral LD50 was 3,384 mg/kg bw in male rats, 2,743 mg/kg bw in female rats, and 3,046 mg/kg bw for both sexes combined when administered a single dose of tertiary butyl alcohol. Clinical signs indicating reversible effects on the central nervous system included piloerection, ataxia, decreased limb tone, low carriage, prostration, impaired righting reflex, bradypnea, hypoactivity and lacrimation.

- In an acute oral toxicity test, the oral LD50 was 3,558 mg/kg bw for an unspecified strain and sex of rabbit receiving a single dose of tertiary butyl alcohol. Clinical signs were indicative of reversible central nervous system effects.

- In an acute inhalation study, a single group of 5 male and 5 female rats was exposed whole-body for 4 hours to a vapor concentration of 10,000 ppmV tertiary butyl alcohol and observed for 14 days. One female was found dead on study Day 3. The principal signs observed during exposure were indicative of central nervous system depression and included dyspnea and prostration in all animals. These same signs, along with generalized weakness, appeared in several animals of each sex during the post-exposure period but were fully reversible by study termination. Gross pathological effects observed at necropsy were limited to focal areas of redness on the lungs.

- In an acute dermal toxicity test, no deaths were reported when a limit dose of 2,000 mg/kg bw of undiluted tertiary butyl alcohol was applied to the abraded skin of male and female New Zealand rabbits for twenty-four hours. The only effects related to test substance administration were reversible central nervous system effects in all animals and injected iris in all males and four of five females.

- Estimated Data: None

Neurotoxicity (N-repeated) (Group II*): DG

Tert-Butyl Alcohol was assigned a hazard classification level of data gap for repeated dose neurotoxicity based on absence of data specific to this endpoint. While numerous repeat dose studies indicate that repeat exposures to tert-Butyl Alcohol result in CNS depression (see above), this effect appears to be reversible. Therefore, there the available studies do not provide sufficient evidence that the CNS effects represent organ dysfunction or a significant function change in the central or peripheral nervous system. None of the available repeat dose studies include behavioral observations or function observation battery endpoints. Repeat dose neurotoxicity was not located for analogs of the substance.

Data

- Lists
  - *Authoritative: None*
  - *Screening: None*

- Measured Data: None Found

- Estimated Data: None

Skin Sensitization (SnS) (Group II*) L

Tert-Butyl Alcohol was assigned a hazard classification level of Low for skin sensitization based on the absence of a sensitization reaction report in a guideline study. The low hazard score is based on study data specific to the compound of interest and is therefore reported with high confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening: None*

- Measured Data

ECHA 2023

- In a Guinea Pig Maximization Test for skin sensitization, a group of 20 female guinea pigs was exposed to undiluted tertiary butyl alcohol. Under the conditions of this study, a concentration of 100% tertiary butyl alcohol at challenge did not cause a sensitization reaction after prior intradermal exposures of 1% and topical exposures of 100% tertiary butyl alcohol. Based on an absence of positive effects in this study, tertiary butyl alcohol is not classifiable for skin sensitization according to GHS

- Estimated Data: None

Respiratory Sensitization (SnR) (Group II*): DG

Tert-Butyl Alcohol was assigned a hazard classification level of data gap for respiratory sensitization based on lack of adequate studies. While ethanol is known to be an immunomodulatory agent and can cause some forms of respiratory hyperresponsiveness no data was available to suggest this same effect was caused by tert-Butyl Alcohol. In addition, there is no evidence of respiratory symptoms in volunteers following acute exposure to low levels of MTBE.

Data

- Lists
  - *Authoritative: None*
  - *Screening: None*

- Measured Data

Oldenburg 2011

- There is very limited knowledge about the effects of alcohol on airway hyperresponsiveness and inflammation in asthma. Historical accounts of alcohol administration to patients with breathing problems suggest that alcohol may have bronchodilation properties. We hypothesized that alcohol exposure will alter airway hyperresponsiveness (AHR) and pulmonary inflammation in a mouse model of allergic asthma. To test this hypothesis, BALB/c mice were fed either 18% alcohol or water and then sensitized and challenged with ovalbumin (OVA). AHR was assessed by means of ventilation or barometric plethysmography and reported as either total lung resistance or enhanced pause, respectively. Airway inflammation was assessed by total and differential cell counts in bronchoalveolar lavage fluid (BALF), cytokine levels in BALF, lung histology, and serum immunoglobulin E (IgE) levels. Alcohol feeding significantly blocked methacholine-induced increases in AHR compared with water-fed controls. Alcohol feeding significantly reduced total cell numbers (64%) as well as the number of eosinophils (84%) recruited to the lungs of these mice. Modest changes in lung pathology were also observed. Alcohol exposure led to a reduction of IgE in the serum of the EtOH OVA mice. These data demonstrate that alcohol exposure blunts AHR and dampens allergic airway inflammation indices in allergic mice and suggest that there may be an important role for alcohol in the modulation of asthma. These data provide an in vivo basis for previous clinical observations in humans substantiating the bronchodilator properties of alcohol and for the first time demonstrates an alcohol-induced reduction of allergic inflammatory cells in a mouse model of allergic asthma.

ATSDR 2022

- In controlled exposure human studies, there is no evidence of respiratory symptoms in volunteers following acute exposure to low levels of MTBE.

- Estimated Data: None

Skin Irritation/Corrosivity (IrS): L

Tert-Butyl Alcohol was assigned a hazard classification level of Low for skin irritation/corrosivity based on study data that reported minimal erythema at 24 hours in some rabbits, but no signs of erythema or edema were present at the 72 hour observation period. This observed effect does not result in a classification for skin irritation. The low hazard score is based on study data specific to the compound of interest and is therefore reported with high confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening: None*

- Measured Data

ECHA 2023

- In a primary dermal irritation study, young adult New Zealand white rabbits (3 males, 3 females) were dermally exposed to 2 mL of tertiary butyl alcohol for 24 hours. Test material (0.5 mL) was applied to each of 2 intact and 2 abraded sites for each rabbit. Animals were then observed for 72 hours. Irritation was scored by the method of Draize. No edema was noted. The test material caused minimal erythema at 24 hours in some rabbits, but no signs of erythema or edema were present at the 72 hour observation period. Based on a total absence of edema, a mean erythema score of 0.7 at 24 hours, and full reversibility of erythema by 72 hours following application of tertiary butyl alcohol to intact and abraded rabbit skin for 24 hours, tertiary butyl alcohol is not classifiable for skin irritation/corrosion according to GHS.

- Estimated Data: None

Eye Irritation/Corrosivity (IrE): H

Tert-Butyl Alcohol was assigned a hazard classification level of High for eye irritation/corrosivity based on a GHS harmonized H319 classification. This hazard conclusion is supported by available study data. The hazard score is based on an authoritative a-list and study data and is therefore reported with high confidence.

Data

- Lists
  - *Authoritative:*
    - *GHS Harmonized H319 - Causes serious eye irritation [Serious eye damage/eye irritation - Category 2A]*
  - *Screening:*
    - *EU - GHS (H-Statements) Annex 6 Table 3-1 H319 - Causes serious eye irritation [Serious eye damage/eye irritation - Category 2A]*
    - *GHS - Australia H318 - Causes serious eye damage [Serious eye damage/eye irritation - Category 1]*

  - *GHS - Japan H319 - Causes serious eye irritation [Serious eye damage / eye irritation - Category 2A]*
  - *GHS - New Zealand Eye irritation category 2*
  - *EU - Manufacturer REACH hazard submissions H319 - Causes serious eye irritation (unverified) [Serious eye damage/eye irritation - Category 2A]*

- Measured Data

ECHA 2023

- In a primary eye irritation study, 0.1 mL of Arconol (a product containing predominantly tertiary butyl alcohol [91% minimum] and low levels of acetone, water, other organics, and butanes) was instilled into the eyes of young adult New Zealand Albino rabbits (4 males, 5 females). Of these, 3 had their eyes washed after treatment and 6 were unwashed. Animals were observed for 16 days (unwashed eyes) or 34 days (washed eyes). Irritation was scored by the method of Draize. For all six animals in the unwashed group, a positive response of corneal opacity ≥1 and conjunctival redness ≥2 calculated as the mean scores following grading at 24, 48 and 72 hours after instillation of the test material was observed. All effects in unwashed eyes were fully reversible in 16 days. For all three animals in the washed group, a positive response of corneal opacity ≥1 was observed. All animals were normal on day 22. Corneal effects were again observed in a single animal in the washed group on day 25 and these effects continued until study termination on day 34. Since the score for corneal effects (6.3) on day 25 was higher than the scores on day 13 (5.0), day 16 (5.0), day 19 (2.5) and day 22 (0), no conclusion can be drawn about the significance of the effects observed in this single animal. In this study, Arconol containing ≥91% tertiary butyl alcohol was an eye irritant.

- Estimated Data: None

ECOTOXICITY (ECOTOX)

Acute Aquatic Toxicity (AA): L

Tert-Butyl Alcohol was assigned a hazard classification level of Low for acute aquatic toxicity based on LC50 and EC50 values >100 mg/L for all three trophic levels (i.e., fish, invertebrates, and algae). The hazard conclusion is based on study data and is therefore reported with high confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening:*
    - *German FEA - Substances Hazardous to Waters Class 1 - Low Hazard to Waters*

- Measured Data

ECHA 2023

- The 96-hour LC50 for fathead minnow exposed to tertiary butyl alcohol is >961 mg a.i./l. The 96 h NOEC was 961 mg a.i./l based on the lack of mortality and sublethal effects at this and all lower concentrations.
- The 96-hour LC50 for fish exposed to tertiary butyl alcohol was >856 mg/L.
- The 48-hour EC50 for Daphnia magna was measured as 933 mg/L for tertiary butyl alcohol.
- Tertiary butyl alcohol is not toxic to aquatic invertebrates.
- In this study tertiary butyl alcohol is not toxic to algae. The 96-hour EC50 based on biomass and growth was >976 mg/L (this level was also a NOEC).
- The 72-hour EC50 of algal growth in the presence of tertiary butyl alcohol >1,000 mg/L (this level was also a NOEC).

- Estimated Data: None

Chronic Aquatic Toxicity (CA): L

Tert-Butyl Alcohol was assigned a hazard classification level of Low for chronic aquatic toxicity based on measured data indication NOEC values >10 mg/L and modeled data for neutral organics indicating ChV values of >10 mg/L. The conclusion is based on study data and modeled estimates and is therefore reported with high confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening:*
    - *German FEA - Substances Hazardous to Waters Class 1 - Low Hazard to Waters*

- Measured Data

ECHA 2023

- A concentration dependent increase in egg mortality was observed ($p < 0.001$), with 10% non-viable eggs in the control group increasing to 89% in the top (1,371 mg/L) group. Larvae mortality (up to 96 hours post-hatch) showed little change (<10% mortality) except for the top dose group which had 16% mortality. The number of least severe (category 1) deformations increased (not significantly) with tert-butanol concentration (11% control, 19% top dose group), but there were no significant changes in category 2 or 3 deformations. A non-significant decrease in the number of healthy larvae with increasing tert-butanol concentration was noted (74% control, 61% top-dose group). The NOEC for fish (based on egg mortality) was determined to be 332 mg/L.

- In a 21 Day reproductive toxicity assay with Daphnia magna, exposure to a limit concentration of tertiary butyl alcohol resulted in no effects to immobilization, reproductive parameters or body length. As such the relevant 21 Day EC50 values

for immobilization and reproductive parameters are all >100 mg/L with the NOEC determined as 100 mg/L.

- Estimated Data:

ECOSAR

ECOSAR Class	Organism	End Pt	Predicted mg/L (ppm)
Neutral Organics	Fish	ChV	71.657
Neutral Organics	Daphnid	ChV	29.913
Neutral Organics	Green Algae	ChV	39.328
Neutral Organics	Fish (SW)	ChV	52.566
Neutral Organics	Mysid (SW)	ChV	232.733

ENVIRONMENTAL FATE (FATE)

Persistence (P): M

Tert-Butyl Alcohol was assigned a hazard classification level of Moderate for persistence based on analytical testing which shows a half-life in inoculum of <60 days and modeled data using EpiSuite estimating a half-life of 720 hours in soil (30-days). The hazard conclusion is based on study data and modeled estimates and is therefore reported with high confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening:*
    - *EC - CEPA DSL Persistent*

- Measured Data

ECHA 2023

- The % ThCO2 produced by the test substance, tertiary butyl alcohol was 2.6 and 5.1% ThCO2 by day 29 of the study. Therefore, tertiary butyl alcohol cannot be classified as readily biodegradable under the conditions of this test. The %ThCO2 produced by the reference substance, sodium benzoate, was 70.6% ThCO2 by day 5 and 87.1% ThCO by day 29, proving that the inoculum was viable.

- Tertiary butyl alcohol was inherently and ultimately biodegradable under the conditions of the test. 66% degradation was observed after 56 days using adapted inoculum. Although the substance achieved 99% degradation over 28 days, it did not

pass the 10-day window criterion and is therefore not considered to be readily biodegradable.

EU RAR 2006

- TBA is inherently biodegradable (although variable results were reported, depending on test system and inoculum used: adapted versus non-adapted micro-organisms). TBA is poorly degradable because tertiary compounds are not common in the environment. TBA is degraded by adapted micro-organisms when the microorganisms have no access to compounds that are more easily degradable. Several tests conducted for inherent biodegradability (Zahn-Wellens tests, see OECD 302B; *discussed above*) with adapted and un-adapted activated sludge and found that TBA (analyzed in the test medium) was fully degraded within about 2 weeks. In unadapted sludge there was a lag time of about 1 week before the micro-organisms were adapted and started to degrade TBA.

HSDB 2014

- If released to air, a vapor pressure of 40.7 mm Hg at 25°C indicates t-butyl alcohol will exist solely as a vapor in the atmosphere. Vapor-phase t-butyl alcohol will be degraded in the atmosphere by reaction with photochemically-produced hydroxyl radicals; the half-life for this reaction in air is estimated to be 14 days. t-Butyl alcohol does not contain chromophores that absorb at wavelengths >290 nm and, therefore, is not expected to be susceptible to direct photolysis by sunlight. If released to soil, t-butyl alcohol is expected to have very high mobility based upon a reported Koc of 37. Volatilization from moist soil surfaces is expected to be an important fate process based upon a Henry's Law constant of 9.05X10-6 atm-cu m/mole. t-Butyl alcohol may volatilize from dry soil surfaces based upon its vapor pressure. The half-life of t-butyl alcohol under anoxic conditions in a non-amended soil was about 200 days, but the half-lives in the same soil amended with nitrate and sulfate nutrients were 100 and 50 days, respectively. Biodegradation of t-butyl alcohol in unamended soils collected at different depths had rates of <0.01 to 0.15 mg/L/day/gram dry soil. If released into water, t-butyl alcohol is not expected to adsorb to suspended solids and sediment based upon the Koc. The biodegradation half-life of t-butyl alcohol was reported to range from about 28 to 180 days in aerobic water and 100 to 500 days in anaerobic water. Volatilization from water surfaces is expected to be an important fate process based upon this compound's Henry's Law constant. Estimated volatilization half-lives for a model river and model lake are 3.6 and 29 days, respectively.

- Estimated Data:

EpiSuite

Level III	Fugacity Model:		
	Mass Amount (percent)	Half-Life (hr)	Emissions (kg/hr)
Air	7.78	229	1000
Water	43.3	360	1000
Soil	48.9	720	1000

Sediment	0.083	3.24E+03	0
Persistence Time: 364 hr			

Bioaccumulation (B): vL

Tert-Butyl Alcohol was assigned a hazard classification level of very Low for bioaccumulation based on a report logKOW of 0.317. This is a measured value reported for the chemical of interest and therefore is reported as high confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening: None*

- Measured Data

  - ECHA 2023: The partition coefficient using the shake-flask method has been determined as 2.08 (Log Pow = 0.317) at 22.5 C
  - HSDB 2014: A reported BCF of <5 in carp suggests bioconcentration in aquatic organisms is low.
  - EU RAR 2006: Very low bioaccumulation potential (Log Kow: 0.35), thus food chain effects (secondary poisoning) not expected.

- Estimated Data: None

PHYSICAL HAZARDS (PHYSICAL)

Reactivity (Rx): *L*

Tert-Butyl Alcohol was assigned a hazard classification level of Low for reactivity based on the absence of chemical groups that are associated with explosivity or oxidizing properties. The hazard score is based on the chemical structure and professional judgement and therefore is reported as low confidence.

Data

- Lists
  - *Authoritative: None*
  - *Screening: None*

- Measured Data

  ECHA 2023
  - The molecule has no chemical groups that are associated with explosive properties.
  - On the basis of structure, the substance is expected to be incapable of reacting exothermically with combustible materials.

  PubChem 2023

- Instability: 0. 0 = This degree includes materials that are normally stable, even under fire exposure conditions, and that do not react with water.

- Estimated Data: None

Flammability (F): H

Tert-Butyl Alcohol was assigned a hazard classification level of High for flammability based on a harmonized GHS classification of H225. This classification is based on available study data and is therefore reported with high confidence.

Data

- Lists
  - *Authoritative:*
    - *GHS Harmonized H225*
  - *Screening:*
    - *EU - GHS (H-Statements) Annex 6 Table 3-1 H225 - Highly flammable liquid and vapour [Flammable liquids - Category 2]*
    - *GHS - Australia H225 - Highly flammable liquid and vapour [Flammable liquids - Category 2]*
    - *GHS - Japan H225 - Highly flammable liquid and vapour [Flammable solids - Category 1]*
    - *GHS - Malaysia H225 - Highly flammable liquid and vapour [Flammable liquids - Category 2]*
    - *GHS - New Zealand Flammable liquids category 2*
    - *Québec CSST - WHMIS 1988 Class B2 - Flammable liquids*
    - *EU - Manufacturer REACH hazard submissions H225 - Highly flammable liquid and vapour (unverified) [Flammable liquids - Category 2]*

- Measured Data

ECHA 2023

- Auto-ignition temperature for tertiary butyl alcohol is 470 degrees C.
- The flash point of tertiary butyl alcohol is 15 degrees C. The substance is therefore classified as a Flammable Liquid category 2 under GHS.

- Estimated Data: None

REFERENCES

ATSDR 2022. Toxicological Profile for Methyl tert-Butyl Ether (MTBE) Available at https://www.atsdr.cdc.gov/ToxProfiles/tp91.pdf.

EU RAR 2006. European Union Risk Assessment Report Tertiary Butyl Hydroperoxide (TBHP) CAS No: 75-91-2 EINECS No: 200-915-7 RISK ASSESSMENT. Available at https://echa.europa.eu/documents/10162/dcfcec71-84a5-4f5c-a438-4aa8fa5ddbb9.

GreenScreen Guidance 2018. Available at https://www.greenscreenchemicals.org/learn/guidance-and-method-documents-downloads.

HSDB 2014 DATA SOURCES Hazardous Substances Data Bank (HSDB) Annotation Record T-Butyl Alcohol. Available at https://pubchem.ncbi.nlm.nih.gov/source/hsdb/50.

NTP (National Toxicology Program) 1995. Toxicology and carcinogenesis studies of t-butyl alcohol (CAS no 75-65-0) in F344/N rats and B6C3F1 mice (Drinking water studies). (NTPTR436). Research Triangle Park, NC: U.S. Department of Health and Human Services, Public Health Service, National Institutes of Health. https://ntp.niehs.nih.gov/ntp/htdocs/lt_rpts/tr436.pdf

NTP (National Toxicology Program). 1997. NTP technical report on toxicity studies of t-butyl alcohol (CAS no 75-65-0) administered by inhalation to F344/N rats and B6C3F1 mice. (NIH Pub 97-3942). Research Triangle Park, NC: U.S. Department of Health and Human Services, Public Health Service, National Institutes of Health. http://ntp.niehs.nih.gov/ntp/htdocs/ST_rpts/tox053.pdf

Oldenburg 2011. Oldenburg PJ, Poole JA, Sisson JH. Alcohol reduces airway hyperresponsiveness (AHR) and allergic airway inflammation in mice. Am J Physiol Lung Cell Mol Physiol. 2012 Feb 1;302(3):L308-15

Peyester 2014. Peyster A, Mihaich E, Kim DH, Elyea WA, Nemec MJ, Hirakawa BP, Leggieri SE. Responses of the steroidogenic pathway from exposure to methyl-tert-butyl ether and tert-butanol. Toxicology. 2014 May 7;319:23-37

PubChem 2023. National Center for Biotechnology Information (2023). PubChem Compound Summary for CID 6386, tert-Butanol. Available at https://pubchem.ncbi.nlm.nih.gov/compound/tert-Butanol

Rosendahl 1998. Methyl tert-butyl ether Pathway Map. Available at http://eawag-bbd.ethz.ch/mtb/mtb_map.html

US EPA 2020. Toxicological Review of tert-Butyl Alcohol (tert-Butanol). IRIS Database. Available at https://cfpub.epa.gov/ncea/iris/iris_documents/documents/toxreviews/1036tr.pdf

US EPA 2021. Toxicological Review of tert-Butyl Alcohol (tert-Butanol) [CASRN 75-65-0] . Available at https://cfpub.epa.gov/ncea/iris/iris_documents/documents/toxreviews/1036tr.pdf

Zhong WH, Lu Z, Chen JM, Chen X, Sun KD. [Conditions and intermediate metabolites of MTBE degradation by beta-Proteobacteria]. Huan Jing Ke Xue. 2006 Dec;27(12):2536-41.

APPENDIX A: HAZARD CLASSIFICATION ACRONYMS

(Alphabetical order)

(AA) Acute Aquatic Toxicity

(AT) Acute Mammalian Toxicity

(B) Bioaccumulation

(C) Carcinogenicity

(CA) Chronic Aquatic Toxicity

(D) Developmental Toxicity

(E) Endocrine Activity

(F) Flammability

(IrE) Eye Irritation/Corrosivity

(IrS) Skin Irritation/Corrosivity

(M) Mutagenicity and Genotoxicity

(N) Neurotoxicity

(P) Persistence

(R) Reproductive Toxicity

(Rx) Reactivity

(SnS) Sensitization- Skin

(SnR) Sensitization- Respiratory

(ST) Systemic/Organ Toxicity

APPENDIX B – OPTIONAL HAZARD SUMMARY TABLE

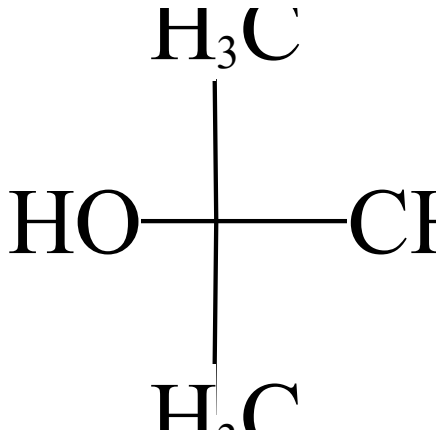
Exposure Route	GreenScreen Hazard Ratings: [*Chemical Name*]																	
	Group I Human					Group II and II* Human									Ecotox		Fate		Physical	
	C	M	R	D	E	AT	ST		N		SnS*	SnR*	IrS	IrE	AA	CA	P	B	Rx	F
							single	repeated*	single	repeated*										
oral																				
dermal																				
inhalation																				

APPENDIX C – MODELING RESULTS

Attach:

- **EPISuite Results for TERT-BUTYL ALCOHOL (CAS# 75-65-0)**
-

EPI Suite Results for CAS 000075-65-0



```
SMILES : OC(C)(C)C
CHEM   : 2-Propanol, 2-methyl-
MOL FOR: C4 H10 O1
MOL WT : 74.12
------------------------------ EPI SUMMARY (v4.11) --------------------------
Physical Property Inputs:
Log Kow (octanol-water):   ------
Boiling Point (deg C)  :   ------
Melting Point (deg C)  :   ------
Vapor Pressure (mm Hg) :   ------
Water Solubility (mg/L):   ------
Henry LC (atm-m3/mole) :   ------

Log Octanol-Water Partition Coef (SRC):
Log Kow (KOWWIN v1.68 estimate) =  0.73
Log Kow (Exper. database match) =  0.35
Exper. Ref:  HANSCH,C ET AL. (1995)

Boiling Pt, Melting Pt, Vapor Pressure Estimations (MPBPVP v1.43):
Boiling Pt (deg C):  70.42  (Adapted Stein & Brown method)
Melting Pt (deg C):  -73.81  (Mean or Weighted MP)
VP(mm Hg,25 deg C):  46.9  (Modified Grain method)
VP (Pa, 25 deg C) :  6.25E+003  (Modified Grain method)
```

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MP  (exp database):  25.4 deg C
BP  (exp database):  82.4 deg C
VP  (exp database):  4.07E+01 mm Hg (5.43E+003 Pa) at 25 deg C
Subcooled liquid VP: 41.1 mm Hg (25 deg C, exp database VP )
: 5.48E+003 Pa  (25 deg C, exp database VP )

Water Solubility Estimate from Log Kow (WSKOW v1.42):
Water Solubility at 25 deg C (mg/L):  2.175e+005
log Kow used: 0.35 (expkow database)
no-melting pt equation used
Water Sol (Exper. database match) =  1e+006 mg/L (25 deg C)
Exper. Ref:  RIDDICK,JA ET AL. (1986)

Water Sol Estimate from Fragments:
Wat Sol (v1.01 est) =  1.511e+005 mg/L

ECOSAR Class Program (ECOSAR v1.11):
Class(es) found:
Neutral Organics

Henrys Law Constant (25 deg C) [HENRYWIN v3.20]:
Bond Method :   9.99E-006  atm-m3/mole  (1.01E+000 Pa-m3/mole)
Group Method:   1.04E-005  atm-m3/mole  (1.06E+000 Pa-m3/mole)
Exper Database: 9.05E-06  atm-m3/mole  (9.17E-001 Pa-m3/mole)
For Henry LC Comparison Purposes:
User-Entered Henry LC:  not entered
Henrys LC [via VP/WSol estimate using User-Entered or Estimated values]:
HLC:  2.103E-005 atm-m3/mole  (2.131E+000 Pa-m3/mole)
VP:   46.9 mm Hg (source: MPBPVP)
WS:   2.18E+005 mg/L (source: WSKOWWIN)

Log Octanol-Air Partition Coefficient (25 deg C) [KOAWIN v1.10]:
Log Kow used:  0.35  (exp database)
Log Kaw used:  -3.432  (exp database)
Log Koa (KOAWIN v1.10 estimate):  3.782
Log Koa (experimental database):  3.500

Probability of Rapid Biodegradation (BIOWIN v4.10):
Biowin1 (Linear Model)         :   0.5283
Biowin2 (Non-Linear Model)     :   0.5580
Expert Survey Biodegradation Results:
Biowin3 (Ultimate Survey Model):   2.8232  (weeks       )
Biowin4 (Primary Survey Model) :   3.5874  (days-weeks  )
MITI Biodegradation Probability:
Biowin5 (MITI Linear Model)    :   0.5605
Biowin6 (MITI Non-Linear Model):   0.6991
Anaerobic Biodegradation Probability:
Biowin7 (Anaerobic Linear Model):  0.2631
Ready Biodegradability Prediction:   YES

Hydrocarbon Biodegradation (BioHCwin v1.01):
Structure incompatible with current estimation method!

Sorption to aerosols (25 Dec C)[AEROWIN v1.00]:
Vapor pressure (liquid/subcooled):  5.48E+003 Pa (41.1 mm Hg)
Log Koa (Exp database): 3.500
Kp (particle/gas partition coef. (m3/ug)):
```

```
Mackay model           :  5.47E-010
Octanol/air (Koa) model:  7.76E-010
Fraction sorbed to airborne particulates (phi):
Junge-Pankow model     :  1.98E-008
Mackay model           :  4.38E-008
Octanol/air (Koa) model:  6.21E-008

Atmospheric Oxidation (25 deg C) [AopWin v1.92]:
Hydroxyl Radicals Reaction:
OVERALL OH Rate Constant =   1.6904 E-12 cm3/molecule-sec
Half-Life =     6.327 Days (12-hr day; 1.5E6 OH/cm3)
Half-Life =    75.930 Hrs
Ozone Reaction:
No Ozone Reaction Estimation
Fraction sorbed to airborne particulates (phi):
3.18E-008 (Junge-Pankow, Mackay avg)
6.21E-008 (Koa method)
Note: the sorbed fraction may be resistant to atmospheric oxidation

Soil Adsorption Coefficient (KOCWIN v2.00):
Koc    :  2.111  L/kg (MCI method)
Log Koc:  0.324       (MCI method)
Koc    :  5.096  L/kg (Kow method)
Log Koc:  0.707       (Kow method)

Aqueous Base/Acid-Catalyzed Hydrolysis (25 deg C) [HYDROWIN v2.00]:
Rate constants can NOT be estimated for this structure!

Bioaccumulation Estimates (BCFBAF v3.01):
Log BCF from regression-based method = 0.500 (BCF = 3.162 L/kg wet-wt)
Log Biotransformation Half-life (HL) = -1.1827 days (HL = 0.06566 days)
Log BCF Arnot-Gobas method (upper trophic) = 0.030 (BCF = 1.071)
Log BAF Arnot-Gobas method (upper trophic) = 0.030 (BAF = 1.071)
log Kow used: 0.35 (expkow database)

Volatilization from Water:
Henry LC:  9.05E-006 atm-m3/mole  (Henry experimental database)
Half-Life from Model River:      56.58  hours   (2.357 days)
Half-Life from Model Lake :      689.4  hours   (28.72 days)

Removal In Wastewater Treatment:
Total removal:               2.35  percent
Total biodegradation:        0.09  percent
Total sludge adsorption:     1.76  percent
Total to Air:                0.51  percent
(using 10000 hr Bio P,A,S)

Level III Fugacity Model:
Mass Amount    Half-Life    Emissions
(percent)        (hr)       (kg/hr)
Air       7.78            229          1000
Water     43.3            360          1000
Soil      48.9            720          1000
Sediment  0.083           3.24e+003    0
Persistence Time: 364 hr
```

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- **ECOSAR Results for TERT-BUTYL ALCOHOL (CAS# 75-65-0)**

ECOSAR Version 1.11 Results Page

SMILES : OC(C)(C)C
CHEM : 2-Propanol, 2-methyl-
CAS Num: 000075-65-0
ChemID1:
MOL FOR: C4 H10 O1
MOL WT : 74.12
Log Kow: 0.730 (EPISuite Kowwin v1.68 Estimate)
Log Kow: (User Entered)
Log Kow: 0.35 (PhysProp DB exp value - for comparison only)
Melt Pt: (User Entered for Wat Sol estimate)
Melt Pt: 25.40 (deg C, PhysProp DB exp value for Wat Sol est)
Wat Sol: 2.597E+005 (mg/L, EPISuite WSKowwin v1.43 Estimate)
Wat Sol: (User Entered)
Wat Sol: 1E+006 (mg/L, PhysProp DB exp value)

Values used to Generate ECOSAR Profile

Log Kow: 0.730 (EPISuite Kowwin v1.68 Estimate)
Wat Sol: 1E+006 (mg/L, PhysProp DB exp value)

ECOSAR v1.11 Class-specific Estimations

Neutral Organics

ECOSAR Class	Organism	Duration	End Pt	Predicted mg/L (ppm)
Neutral Organics	: Fish	96-hr	LC50	841.850
Neutral Organics	: Daphnid	48-hr	LC50	425.112
Neutral Organics	: Green Algae	96-hr	EC50	194.994
Neutral Organics	: Fish		ChV	71.657
Neutral Organics	: Daphnid		ChV	29.913
Neutral Organics	: Green Algae		ChV	39.328
Neutral Organics	: Fish (SW)	96-hr	LC50	1051.798
Neutral Organics	: Mysid	96-hr	LC50	1847.973

Neutral Organics	: Fish (SW)		ChV	52.566
Neutral Organics	: Mysid (SW)		ChV	232.733
Neutral Organics	: Earthworm	14-day	LC50	174.529

Note: * = asterisk designates: Chemical may not be soluble enough to measure this predicted effect. If the effect level exceeds the water solubility by 10X, typically no effects at saturation (NES) are reported.

Class Specific LogKow Cut-Offs

If the log Kow of the chemical is greater than the endpoint specific cut-offs presented below, then no effects at saturation are expected for those endpoints.

Neutral Organics:

Maximum LogKow: 5.0 (Fish 96-hr LC50; Daphnid LC50, Mysid LC50)
Maximum LogKow: 6.0 (Earthworm LC50)
Maximum LogKow: 6.4 (Green Algae EC50)
Maximum LogKow: 8.0 (ChV)

- **Other**