Proposed RfD and RfC for Trifluoroacetic Acid (TFA): Report to the Hawaii State Department of Health

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Trifluoroacetic Acid

Trifluoroacetic acid, or TFA, is a strong organic acid and has the shortest chain (only two carbons) of all perfluorinated carboxylates (PFCA) (Solomon et al. 2016; Boutonnet et al. 1999). It has a pKa of 0.23, is miscible with water, and has a low octanol/water partition coefficient (log P_{OW}= -2.1) which shows no potential for bioaccumulation (Boutonnet et al. 1999). TFA is manufactured as an industrial chemical, exists naturally in the environment, and is also produced from the atmospheric degradation of fluorocarbons including hydrofluorocarbons (Garavagno et al. 2024; Solomon et al. 2016). TFA is used in the production of fluorinated compounds, as a laboratory chemical, and for the surface treatment of glass (UBA 2021). TFA is highly soluble in water and adsorbs poorly to soil, sediment and organic matter, and thus is introduced into the natural water cycle quickly from the atmosphere, soils, and through wastewater (UBA 2021). The ultimate sink of TFA is in surface waters (Solomon et al. 2016).

The sodium salt of TFA (sodium trifluoroacetate, NaTFA) is much less toxic than the free acid (Boutonnet et al. 1999). At the pH levels of environmental media (pH range approximately 4.0-8.5), TFA often presents as NaTFA.

TFA is a severe irritant to the skin and eyes (Boutonnet et al. 1999). The literature regarding toxicity of trifluoroacetate in stream mesocosms, algae, higher plants, fish, animals and humans, has shown overall low toxicity in these systems (Boutonnet et al. 1999).

Literature Search

Methods

A literature search was conducted to obtain both peer-reviewed literature and grey literature such as government reports. PubMed was utilized to identify the relevant studies in the peer-reviewed literature. Government documents were identified via Google and were also searched to identify additional peer-reviewed studies. However, some of the industry studies and older studies identified in both government documents and the peer-reviewed literature were not accessible.

Government documents

Various government and international documents describe points of departure[1] (PODs) for TFA including the European Chemicals Agency (ECHA) and European Food Safety Authority (EFSA), the German Environment Agency (UBA), and the United Nations Environment Programme (UNEP).

Peer-reviewed literature

Multiple reviews were identified in the peer-reviewed literature that described toxicology studies of TFA and NaTFA including Dekant & Dekant (2023), Ema et al. (2010), and Boutonnet et al. (1999). These studies, along with the results from PubMed searches, were used to identify the original literature.

Terminology

Several of the studies discussed in this report describe no-observed-adverse-effect levels (NOAELs) or no-observed-effect level (NOEL). The NOAEL is the "highest exposure level at which there are no biologically significant increases in the frequency or severity of adverse effect between the exposed population and its appropriate control; some effects may be produced at this level, but they are not considered adverse or precursors of adverse effects" (EPA 2024a). The NOEL is the "exposure level at which there are no statistically or biologically significant increases in the frequency or severity of adverse effect between the exposed population and its appropriate control; some effects may be produced at this level, but they are not considered adverse or precursors of adverse effects" (EPA 2024a).

Various studies describe LD_{50} ("lethal dose 50") and LC50 ("lethal concentration 50") values. The LD_{50} is the dose that is lethal to 50% of the animals (EPA 2024a). The LC_{50}, on the other hand, is the concentration that is lethal to 50% of the animals (EPA 2024a). Similarly, the LD_0 is the maximum tolerable dose or the highest non-toxic dose while LD_{100} is the lowest dose that kills 100% of the test animals.

ECHA (2024) describes Derived No Effect Levels (DNELs) for TFA. ECHA describes the DNEL as "the level of exposure above which humans should not be exposed" (ECHA 2012). The DNEL should reflect the likely route(s), duration, and frequency of exposure (ECHA 2012). DNELs should also consider the following: 1) the uncertainty caused by the experimental data and intra- and interspecies variation; 2) the nature and severity of

[1] The point of departure is the point on a dose-response curve established from experimental data, generally corresponding to an estimated low effect level (e.g., 1% to 10% incidence of an effect) (EPA 2024b).

effects; and 3) the sensitivity of the human population (ECHA 2012). A NOAEL or NOAEC is utilized to calculate the DNEL.[2]

The Germany Environmental Agency (Umweltbundesamt, UBA) calculated a tolerable daily intake (TDI) for TFA. TDIs indicate the tolerable daily intake of a non-intentionally added contaminant in food and drinking water (UBA Undated). The TDI is based on a NOAEL and takes various safety factors into account including extrapolation of test duration, toxicodynamic and toxicokinetic differences between and within species (UBA Undated). The acceptable daily intake (ADI) is similar to the TDI but is the amount of a chemical a person can be exposed to on a daily basis over an extended period of time (usually a lifetime) without suffering deleterious effects (EPA 2024a).

Results of the Literature Search

Acute toxicity

Dekant & Dekant (2023) discuss a study cited by ECHA (2022) in which the potential effects of TFA were examined after a single oral dose of 2000 mg/kg bw (body weight) in rats. The absence of mortality in this study showed that the acute toxicity of trifluoroacetate was very low.

Muijser et al. (2010)[3] studied male Wistar rats that were exposed to TFA vapors (nose-only exposure) at a concentration of 0, 30, 100, or 300 mg/m^3 for four hours (ECHA 2024). The animals in the first dose group (30 mg/m^3) were sacrificed one day after the end of the exposure and necropsied (ECHA 2024). The rats in the second group were sacrificed after a 14-day observation period (ECHA 2024). Clinical signs were noted, body weight was measured at day 0, 1, 3, 7, and 14, and gross pathology and histopathology were examined (ECHA 2024). TFA did not induce mortality at any of the tested concentrations including 300 mg/m^3. However, at 300 mg/m^3, TFA induced local irritation of the nasal cavity epithelium in the rats. This effect was reversible as no irritation of the nasal cavity was noted in the animals necropsied at the end of the period of observation.

Boutonnet et al. (1999) describe the following publications regarding the acute toxicity of TFA: Patty (1963) and Kheilo and Kremneva (1966). Patty (1963) describes an LD_{50} for the oral route of administration of a 2-5% solution in the range of 200 to 400 mg/kg bw in rats and mice. Oral LD_{100}, LD_{10}, and LD_0 values were found to be 1000, 500, and 250 mg/kg bw, respectively (Kheilo and Kremneva 1966). Kheilo and Kremneva (1966) also describe

[2] The LOAEL or LOAEC should be used only when a NOAEL/NOAEC is not available (ECHA 2012).
[3] It was not possible to obtain a reference for this citation.

acute inhalation toxicity for trifluoroacetic acid. The LC_{50}, LC_{100}, and LC_{10} for a 2-hour exposure in mice were reported to be 13.5 mg/l, 20.4 mg/l, and 9.2 mg/l, respectively. For a 2-hour exposure in rats, the LC_{50}, LC_{100}, and LC_{10} were reported to be 10 mg/l, 11.5 mg/l, and 8.3 mg/l, respectively. Kheilo and Kremneva (1966) described threshold concentrations of 4 mg/l for rats based on body temperature and 1.5 mg/l based on "neuromuscular excitability" (Boutonnet et al. 1999)

Boutonnet et al. (1999) also describe several LD_{50} values for the sodium salt of TFA via the intraperitoneal route of administration: >4000 mg/kg (Rosenberg 1971); >2000 mg/kg (Airaksinen and Tammisto 1968; Airaksinen et al. 1970). An intravenous LD_{50} for sodium trifluoroacetate was reported to be 1200 mg/kg for mice (Airaksinen and Tammisto 1968). Intraperitoneal administration of sodium trifluoroacetate to mice was described resulting in a slight increase in glucose-6-phosphate dehydrogenase activity in livers and erythrocytes (Rosenberg 1971). There was also a transient decrease in glutathione and NADPH content in the liver 12 hours after treatment. This decrease in glutathione and NADPH content was also seen in erythrocytes at 24 hours (Rosenberg 1971). Boutonnet et al. (1999) also describe a study that identified an intravenous LD_{50} for sodium triflouroacetate of 1200 mg/kg in mice (Airaksinen and Tammisto 1968).

Boutonnet et al. (1999) also describe a 1971 study by Rosenberg and Wahlstrom. In this study Swiss male rats were treated with either a single intraperitoneal dose of 1000 mg/kg or 2000 mg/kg sodium trifluoroacetate. Hepatocytes were found to have a cloudy swelling with slight accumulation of fat and an increase in liver glycogen at the 1000 mg/kg dose. At the 2000 mg/kg dose, vacuolization was noted.

Boutonnet et al. (1999) identified a 1969 study by Blake and colleagues in which they administered trifluoroacetaldehyde, trifluoroacetic acid and sodium trifluoroacetate via intraperitoneal injection to five groups of five mice each. The mice were observed for 96 hours. Blake and colleagues found that trifluoroacetaldehyde and trifluoroacetic acid were of "a low order of toxicity when administered to mice." There were no deaths observed even when mice received up to 800 mg/kg of trifluoroacetaldehyde or up to 5000 mg/kg of sodium trifluoroacetate. Trifluoroacetic acid caused death in two of the five animals at the 150 mg/kg dose.

Finally, Boutonnet et al. (1999) described a 1988 study by Fraser and Kaminsky. This study administered a single intraperitoneal dose of 240 mg/kg of trifluoroacetic acid to male Wistar rats that did not produce any toxic effects.

Chronic/repeated dose toxicity

Dekant & Dekant (2023) describe a 28-day oral study by Bayer (2014) in which trifluoroacetate was administered in the diet at concentrations of 0, 600, 1800, 5400, and 16000 ppm to male and female Wistar rats (n=5 per group). There was no mortality, effects on body weight, or consumption observed. However, there was a small increase in the activity of alanine aminotransferase (ALT) at the highest dose levels in both male and female rats. There were small effects on serum cholesterol and glucose levels. Along with the minor changes in ALT, there were dose-dependent increases in absolute and relative liver weight and liver to body/brain weight ratios at trifluoroacetate concentrations >600 ppm and liver enlargement at 5400 and 16000 ppm. However, although there was liver enlargement there were no histopathological changes in the liver. The highest dose, 16000, which corresponded to 1315 mg/kg bw per day in males and 1344 mg/kg bw/day in females, was considered the NOAEL.

Dekant & Dekant (2023) describe another Bayer (2014) repeated dose oral study that was carried out to examine possible modes of action regarding changes in liver enzymes and mild liver hypertrophy. Male and female rats (n=5 per group) were given diets with sodium TFA at concentrations of 0, 600, 1200, and 2400 ppm for 14 days. Animals were sacrificed after 14 days. TFA did not affect body weight gain, food consumption, hematology, and clinical chemistry but did result in increased liver weights and hepatocellular hypertrophy. Dekant & Dekant (2023) state that based on the findings of this study, trifluoroacetate "has to be considered a weak peroxisome proliferator."

An ECHA (2024) dossier[4] describes Bayer (2007)[5] as a 90-day oral study where Wistar rats were administered sodium trifluoroacetate orally via diet. There were 10 male and 10 female rats per dose group: 160, 1600, and 16000 ppm. A similarly constituted group of 10 males and 10 females were administered a normal diet and served as controls. No toxicologically significant changes were noted regarding clinical appearance, functional observations, food consumption, and ophthalmoscopy (ECHA 2024). Dekant & Dekant (2023) describe a Bayer (2014)[6] study in which sodium trifluoroacetate was administered to male and female Wistar rats (10 of each per dose group). Reduced body weight gain was observed in males in the 16000 ppm dose group. Treatment-related effects on clinical chemistry including bilirubin, glucose and liver enzyme levels and urine composition (higher ketone bodies) were seen at concentrations greater than 160 ppm. Both mean absolute and relative liver weights were also increased at feed concentrations above 160

[4] https://echa.europa.eu/de/registration-dossier/-/registered-dossier/5203/7/6/1#
[5] It was not possible to obtain a reference for this citation.
[6] It is possible that Bayer (2007) described by ECHA (2024) and Bayer (2014) described by Dekant & Dekant (2023) are the same study.

ppm. Minimal to moderate diffuse centrilobular hepatocellular hypertrophy and an increased incidence of hepatocellular necrotic foci were observed in males in the highest dose group. From this study, a NOAEL of 8.4 (males) and 10.1 mg/kg bw (females) TFA was determined.

Just et al. (1989) was also identified in EFSA's 2014 document and in Boutonnet et al. (1999). In this study, male Wistar rats were fed 500 mg/kg TFA for 5-14 days (EFSA 2014; Boutonnet et al. 1999). The authors did not see an effect on body weight but did see a slight increase in liver weight (EFSA 2014; Boutonnet et al. 1999).

The EFSA (2014) document also describes a UK assessment in which repeated dose studies were carried out between 75 and 750 mg/kg bw. Effects including changes in liver weight, liver serum enzymes and liver histopathology were observed.

The German Environment Agency describes a study in which rats were administered TFA in drinking water at concentrations of 0, 30, 120, and 600 ppm for one year (UBA Undated; Dekant & Dekant 2023). The study was conducted by manufacturer Solvay Hanover according to OECD guidelines 417 and 452 for the assessment of toxicokinetic and chronic effects (UBA Undated). Significant differences (p=0.005 and 0.009, respectively) were found between the ALT concentrations in the control group (0 ppm) and the groups that received 120 ppm and 600 ppm TFA at day 370 (UBA Undated). The differences were no longer significant (p=0.55) after recovery[7] at day 412 (UBA Undated). A no observed effect level (NOEL) of 30 ppm (corresponding to 1.8 mg/kg bw per day) was observed.

Boutonnet et al. (1999) identified a 1969 study by Blake et al. in which trifluoroacetic acid or sodium trifluoroacetate was administered via drinking water at a concentration of 114 g/l to male Sprague-Dawley rats. The rats rejected the solution which resulted in dehydration and weight loss. Within 10 days, an increase (<30%) was seen in the liver-to-body weight ratio.

Boutonnet et al. (1999) identified a study by Stier et al. (1972) in which sodium trifluoroacetate was administered to male Wistar rats in drinking water at approximately 130 µmol per 100 g body weight per 24 hours (~150 mg/kg bw per day) for five to six days. After five days of treatment, the relative liver weights were increased by approximately 43% and the glycogen content of the liver was decreased.

According to Boutonnet et al. (1999), Kheilo and Kremneva (1966) exposed rats via inhalation to trifluoroacetic acid for four hours daily six times a week at concentrations of

[7] The recovery period between days 370 and 412 was only for the control group and the group with the highest dose of TFA (600 ppm) (UBA Undated).

0.4 to 0.7 mg/l (86-150 ppm) for five months. Irritation of mucous membranes, increased proteinuria, and "altered neuromuscular excitability" were observed. The authors found that chronic exposure to approximately 5 to 11 ppm caused only minor symptoms in rats and that these are considered close to the threshold concentrations for chronic exposures (Boutonnet et al. 1999).

A study of male rats and male guinea pigs by Warheit (1993) was also identified by Boutonnet and colleagues. The rats and guinea pigs were fed diets containing 7500 ppm (750 mg/kg per day in rats) for 25 days. Among the rats, decreased body weight was observed (without changes in food consumption). Decreases in serum cholesterol, triglycerides, glucose, and insulin levels were also found in rats, and to a lesser extent, in guinea pigs. Increased liver weights and diffuse liver hypertrophy were seen in treated rats. However, the liver weight in guinea pigs was reduced.

Reproductive and developmental toxicity

Dekant & Dekant (2023) cites ECHA (2022) and describes a one-generation reproductive toxicity study in rats. Sodium TFA was given to Wistar rats in their diet. In the F0 generation, male and female rats (25 males and females per dose group) were fed diets with 0, 120, 600, or 3000 ppm TFA for ten weeks during pre-mating, gestation, and lactation. The received doses were approximately 10, 50, and 250 mg/kg bw/day. During lactation, TFA was reduced to 60, 300, and 1500 ppm in females. The two cohorts of F1 animals (20 males and females per group in each cohort) were not given TFA during lactation and received TFA-containing diets from postnatal day 21 to postnatal day 35 (doses of 60, 300, and 1500 ppm) and from postnatal day 35 to sacrifice (doses of 10, 600, and 3000 ppm). The study examined estrous cyclicity, thyroid hormones, and sperm parameters. Treatment-related adverse effects were not seen in the parental animals. The F1 generation (offspring) showed treatment-related changes such as changes in relative liver weights, some clinical chemistry parameters, and reductions in serum T4. However, the other thyroid parameters (T3, TSH, weight, histopathology) and reproductive parameters were not affected. Thus, it was found that the top dose level in this study (3000/1500 ppm) with intakes between 242 and 265 mg of the sodium salt of TFA/kg bw/day was considered the NOAEL for reproductive performance, offspring development and general toxicity.

Ema et al. (2010), an ECHA dossier[8], and Boutonnet et al. (1999) identified a 1988 study by Lloyd and colleagues. According to Ema et al. (2010), Lloyd et al. dosed Alpk/AP male rats one time via oral gavage with either 10 or 20 mg/kg bw; no adverse testicular

[8] https://echa.europa.eu/de/registration-dossier/-/registered-dossier/10848/7/9/1

effects were seen. According to the ECHA dossier, Lloyd et al. (1988) administered a single oral dose of 25 mg/kg bw and saw no effect on the male reproductive system. Boutonnet et al. (1999) also describes Lloyd et al. (1988) but indicates that single oral doses of 10 and 25 mg/kg bw were administered; sodium TFA did not cause any observable testicular effects on weight or morphology. Boutonnet et al. (1999) indicates that another study by Lloyd and colleagues (1986) administered a single intraperitoneal dose of TFA to 10-week-old male AlpK/AP strain rats. Like Lloyd et al. (1988), TFA did not cause any observable effects on weight or morphology in the testes.

Dekant & Dekant (2023) cite ECHA (2022) regarding a developmental toxicity study of TFA. TFA was administered to groups of pregnant female rats (n=22 per group) from gestational day (GD) 6 to 19. Doses of 0, 37.5, 75, and 150 mg/kg bw per day were administered via gavage in water. Fetuses were birthed via C-section on GD 20. In maternal rats, the only effect of TFA was a small increase in liver and kidney weight. Fetal effects were not observed. Because there was a lack of maternal and embryo/fetal effects, the highest dose administered was selected as the NOAEL (150 mg/kg bw per day); the small increases in maternal liver and kidney weight were not considered adverse.

Ema et al. (2010) identified a toxicity study by Saillenfait and colleagues. This study is also identified in an ECHA dossier.[9] Saillenfait et al. (1996) administered halothane or TFA to Sprague-Dawley rats on GDs 10 to 20. TFA was administered by gavage at concentrations of 75 or 150 mg/kg bw per day. Mothers and pups were examined for body weight, liver weight, and kidney weight. Liver weight among the mothers was significantly increased in both dose groups. Body weight change was significantly reduced at the 150 mg/kg bw for GD 10 to15. Absolute and relative liver weights were significantly increased in both dose groups. Exposed offspring were examined on postnatal days 3, 12, and 49 for hepatic and renal biochemistry and/or function through measurements of several serum and urinary metrics. No adverse outcomes were found regarding length of gestation, litter size and offspring survival in the first three days. There were slight but non-significant decreases in pup weights in both TFA dose groups on postnatal days 1 and 3. Prenatal exposure to TFA induced only slight and transient alterations in the neonatal rat liver but was not predictive of developmental effects.

Discussion

The major target organ for TFA appears to be the liver (Boutonnet et al. 1999; Dekant & Dekant 2023). However, the effects of TFA on the liver appear to be mediated by

[9] https://echa.europa.eu/de/registration-dossier/-/registered-dossier/10848/7/9/1

peroxisome proliferation, and this mode of action is not considered relevant to humans (Dekant & Dekant 2023).

Summary of acute studies

Several of the studies identified and described above also reported LD and LC values. Although these values would not be considered for selection or development of an RfD, these values are summarized in Table 1, below, for completeness.

Table 1. Summary of identified lethal doses (LDs) and lethal concentrations (LCs)

Type of study	Potential POD	Exposure route	Species	Duration of exposure (hours)	Endpoint(s) evaluated	Study
Acute toxicity	LD_{50}: >2000 mg/kg	Oral	Rats	NR[b]	Death	ECHA 2022
Acute toxicity (2-5% solution)	LD_{50}: 200-400 mg/kg	Oral	Rats and mice	NR[b]	Death	Patty 1963; Ema et al. 2010
Acute toxicity	LD_{50}: >4000 mg/kg	Intra-peritoneal	U[a]	NR[b]	Death	Rosenberg 1971; Boutonnet et al. 1999
Acute toxicity	LD_{50}: >2000 mg/kg	Intra-peritoneal	U[a]	NR[b]	Death	Airaksinen and Tammisto 1968; Boutonnet et al. 1999
Acute toxicity	LD_{50}: >1200 mg/kg	Intravenous	Mice	NR[b]	Death	Airaksinen and Tammisto 1968; Boutonnet et al. 1999
Acute toxicity	LD_{50}: >2000 mg/kg	Intra-peritoneal	Mice	NR[b]	Death	Airaksinen et al. 1970; Boutonnet et al. 1999
Repeated dose	LD_{100}: 1000 mg/kg bw	Oral	Rats	NR[b]	Death	Kheilo and Kremneva 1966; Boutonnet et al. 1999
Repeated dose	LD_{10}: 500 mg/kg bw	Oral	Rats	NR[b]	Death	Kheilo and Kremneva 1966; Boutonnet et al. 1999
Repeated dose	LD_{0}: 250 mg/kg bw	Oral	Rats	NR[b]	Death	Kheilo and Kremneva 1966; Boutonnet et al. 1999
Acute toxicity	LC_{100}: 11.5 mg/l	Inhalation	Mice	2	Death	Kheilo and Kremneva 1966;

						Boutonnet et al. 1999
Acute toxicity	LC_{50}:10 mg/l	Inhalation	Mice	2	Death	Kheilo and Kremneva 1966; Boutonnet et al. 1999
Acute toxicity	LC_0: 8.3 mg/l	Inhalation	Mice	2	Death	Kheilo and Kremneva 1966; Boutonnet et al. 1999

[a] U=unknown
[b]NR=not reported

Summary of chronic/repeated dose and reproductive and developmental toxicity studies

Several sources identified POD values for TFA. These studies are described in detail in the Discussion section and summarized in Table 1, below. A UNEP document on TFA and its salts describes an acute oral no-observed-effect-concentration (NOEC) for TFA in rats of 250 mg a.e.[10]/kg bw based on Boutonnet et al. (1999). The acute NOEC for the sodium salt of TFA in rats is ≥5,000 mg/kg bw. The UNEP document describes an "8-day chronic" oral NOEL for the sodium salt of TFA in rats of 114 mg/kg bw/day. These values are not included in Table 2 as the basis of these values were unclear.

Several studies were identified that examined reproductive and developmental toxicity of TFA (ECHA 2022; Lloyd et al. 1988; Lloyd et al. 1986; Sallenfait et al. 1996). The review of data for setting chronic reference doses includes evaluation of reproductive and developmental toxicity data (NRC 2001). Two prenatal developmental toxicity studies in two species and a two-generation reproduction study are considered a portion of the minimum database necessary for setting a chronic RfD (NRC 2001).

Table 2. Point of departure values identified for TFA

Type of study	Potential POD	Exposure route	Species	Duration of exposure (days)	Endpoint(s) evaluated	Study
Repeated dose/ "mechanistic"	NOAEL: M: 43 mg/kg bw per day F: 190 mg/kg bw per day	Oral	Rats	14	Body weight gain, food consumption, hematology, and clinical chemistry, liver weight, hepatocellular hypertrophy	EFSA 2014; Dekant & Dekant 2023
Repeated dose	NOAEL:	Oral	Rats	28	Mortality, body weight, food consumption, liver effects	Bayer 2014; Dekant &

[10] a.e.=acid equivalent (UNEP 2016).

	M: 1315 mg/kg bw per day F: 1344 mg/kg bw per day					Dekant 2023
Repeated dose	NOAEL: M: 8.4 mg/kg bw per day F: 10.1 mg/kg per day	Oral	Rats	90	Body weight, hematology, clinical chemistry and urine composition, liver weight, hepatocellular hypertrophy, hepatocellular necrotic foci	Bayer 2007; Bayer 2014; ECHA 2024; Dekant & Dekant 2023
Repeated dose	NOAEL: M: 10 mg/kg bw per day F: 12 mg/kg bw per day	Oral	Rats	90	Hematology parameters, urine composition, body weight gain	Bayer 2014
Drinking water	NOEL: 1.8 mg/kg bw per day	Oral	Rats	365	Changes in liver enzymes & liver effects	UBA 2021; Dekant & Dekant 2023
Repeated dose	NOAEL: 242-265 mg/kg bw per day	Oral	Rats	During pre-mating, gestation, and lactation	Reproductive performance, offspring development and general toxicity.	Dekant & Dekant 2023; ECHA 2022
Repeated dose	NOAEL: 150 mg/kg bw per day	Oral	Rats	GD	Developmental toxicity	ECHA 2022

Proposed toxicological values

EFSA, ECHA, and the German Environment Agency describe proposed toxicological values for TFA including an ADI, DNELs, and a TDI. The derivation of these values are described below and are summarized in Table 3.

EFSA derived an ADI of 0.05 mg/kg body weight per day (EFSA 2014). To derive this value, EFSA examined information provided by Bayer CropScience in 2013 and by the applicant, BASF.[11] EFSA (2014) agreed with Bayer CropScience's 2013 position paper that the NOAEL (8.4 mg/kg bw per day) from a 90-day oral rat study and the application of an extra uncertainty factor of 2 (applied to the standard 100 to account for the potential chronic toxicity; 2 x 100 = 200) was appropriate.

[11] EFSA cited BASF (2011).

ECHA (2024) established a derived no effect level (DNEL) of 3.37 ppm for acute inhalation toxicity based on Muijser et al. (2010). According to Dekant & Dekant (2023), the animals in this study were exposed to a mixture of vapors and aerosols for 4 hours per day, six days per week for 4 months (guinea pigs) or 5 months (rats). Severe irritation of the respiratory pathway and the eyes in both rats and guinea pigs was observed (Dekant & Dekant 2023). Liver and kidney dystrophy and lack of body weight gain were also found (Dekant & Dekant 2023). The no observed adverse effect concentration (NOAEC) for local effects based on the nasal cavity epithelium irritation is 300 mg/m^3. Factoring in modification for respiratory volume (0.210/0.315 = the respiratory rate difference under standard conditions and under conditions of light activity for 15 minutes), the relevant dose descriptor is 200 mg/m^3 (ECHA 2024). Assessment factors were also applied including 2.5 for remaining differences in toxicokinetics and toxicodynamics, 5 for intraspecies differences (worker), 1 for issues related to dose-response, and 1 for quality of the whole database. This results in an overall assessment factor of 12.5 (2.5 x 5 x 1 x 1). Dividing 200 mg/m^3 by the assessment factor of 12.5 results in a DNEL of 16 mg/m^3 or 3.37 ppm.

ECHA produced a DNEL of 0.042 mg/kg bw/day for long-term exposure. The key study was Bayer (2007). The Bayer study identified a NOAEL of 8.4 mg/kg bw in rats. The most sensitive endpoint was repeated dose toxicity dosed orally. An assessment factor of 2 was applied for extrapolation from subchronic to chronic exposure. An assessment factor of 4 was utilized for interspecies differences between rats and humans. An assessment factor of 2.5 was applied for remaining interspecies differences (default factor for remaining differences). Finally, an assessment factor of 10 was applied as the default for the general population (default for the general population). Thus, an overall assessment factor of 200 was used (2 x 4 x 2.5 x 10). Dividing 8.4 mg/kg bw by the overall assessment factor of 200 results in a DNEL of 0.042 mg/kg bw/day.

The German Environment Agency utilized the one-year drinking water study previously described and the corresponding NOEL to calculate their “health guide value” for TFA. Dividing 1.8 mg/kg by the safety factor of 100 (10 for possible differences in sensitivity between rats and humans and 10 for sensitivity within human species), a tolerable daily intake (TDI) of 0.018 mg/kg was calculated (UBA 2021). Using the standard assumptions of 70 kg body weight and 2 liters of drinking water consumption per day and an allocation of 10% of the TDI for drinking water itself, the “health guide value” was calculated:

$$\text{Health Guide Value (TFA)} = \frac{0.018 \text{ mg/kg} \times 70 \text{ kg} \times 0.1}{2 \text{ l}} = 0.063 \text{ mg/l} \approx 0.060 \text{ mg/l}$$

Table 3. Overview of proposed toxicological values

Reference value	Value	Endpoint	Study	UF	Source
ADI	0.05 mg/kg bw per day	Body weight, hematology, clinical chemistry and urine composition, liver weight, hepatocellular hypertrophy, hepatocellular necrotic foci	90-day oral rat study	200	EFSA 2014
DNEL for acute toxicity (inhalation)	3.37 ppm	Nasal cavity epithelium irritation	Acute toxicity in rats	12.5	ECHA 2024
DNEL for long-term exposure (oral)	0.042 mg/kg bw per day	Body weight, hematology, clinical chemistry and urine composition, liver weight, hepatocellular hypertrophy, hepatocellular necrotic foci	90-day oral rat study	200	ECHA 2024
TDI	0.018 mg/kg bw	Changes in liver enzymes & liver effects	1-year rat drinking water study	100	UBA 2021

Selection of an RfD

The TDI (0.018 mg/kg bw per day) developed by the German Environment Agency is the best substitute for an RfD. Gibb & O'Leary recommend the TDI because it is based on a one-year study. The endpoint is changes in liver enzymes and liver effects; the liver is thought to be the target organ of TFA. As described above, uncertainty factors of 10 for possible differences in sensitivity between rats and humans and 10 for sensitivity within human species were used resulting in an overall uncertainty factor of 100 (10 x 10=100). The German Environment Agency's "health guide value" for drinking water of 0.060 mg/L is based on the TDI of 0.018 mg/kg bw, a 70 kg body weight for humans, 2 liters of drinking water consumption per day, and an allocation of 10% of the TDI for drinking water (UBA Undated).[12] The uncertainty factors used by the German Environment Agency (UBA Undated) to estimate the TDI and the "health guide value" are reasonable. Furthermore, the NOAELs for reproductive and developmental toxicity are well above the NOEL of 1.8 mg/kg bw per day that was used to set the TDI.

[12] Note that UBA (Undated) indicates that the drinking water concentration of TFA should be kept as low as possible and that 0.010 mg/L is recommended.

Route-Extrapolated RfC

Because of the volatile nature of TFA (Garavagno et al. 2024), a route-to-route extrapolation was performed to produce a reference concentration (RfC) for inhalation exposure. EPA (2009) provides guidance on the validity of route-to-route extrapolation. If there is no quantitative toxicity information for the inhalation route, a qualitative evaluation of the exposure route should be conducted to ensure that the hazard will not be misrepresented when data from one route are substituted for another without consideration of pharmacokinetic differences (EPA 2009). A recent article examined whether route-to-route extrapolation to derive guidelines for inhalation exposure was appropriate for PFAS[13] compounds (Monnot et al. 2023). The authors noted that "the current knowledge of PFAS toxicokinetics indicates that there is nothing that would preclude employing route-to-route extrapolation in the derivation of provisional inhalation toxicological screening levels for individual PFAS when acceptable health-based ingestion standards are available" (Monnot et al. 2023).

EPA provides the following method for extrapolating from an RfD to an RfC (EPA 1994; Monnot et al. 2023):

Extrapolated RfC (mg/m^3) = RfD (mg/kg-day) x (70 kg/20 m^3/day)

where 70 kg is the default body weight and 20 m^3/day is the default inhalation rate (EPA 1994; Monnot et al. 2023). Using this equation, and applying it to the TDI above, yields the following RfC:

(0.018 mg/kg-day) x (70 kg/20 m^3/day) = <u>0.063 mg/m^3</u>

[13] According to the EPA, trifluoroacetic acid is a member of the broad class of compounds known as PFAS even though it is not specifically listed or targeted for EPA action (EPA 2022).

References

Airaksinen MM, Tammisto T. 1968. Toxic actions of the metabolites of halothane: LD 50 and some metabolic effects of trifluoroethanol and trifluoroacetic acid in mice and guinea pigs. Ann Med Exp Biol Fenn 46(2):242-248.

Airaksinen MM, Rosenberg PH, Tammisto T. 1970. A possible mechanism of toxicity of trifluoroethanol and other halothane metabolites. Acta Pharmacol Toxicol (Copenh) 28(4):299-304. doi:10.1111/j.1600-0773.1970.tb00556.x

BASF. 2011. Trifluoroacetic Acid (TFA): Environmental and Consumer Safety Considerations Arising from Saflufenacil (BAS 800 H) Agricultural Uses BASF RegDoc ID 2011/7000035, 14 January 2014, 1-30.

Bayer C. 2014. Summary of toxicological and metabolism studies for flurtamone. https:// www.bayer.com/ sites/default/files/M- 482307-01-5. PDF

Blake DA, Cascorbi HF, Rozman RS, Meyer FJ. 1969. Animal toxicity of 2,2,2-trifluoroethanol. Toxicol Appl Pharmacol 15(1):83-91. doi:10.1016/0041-008x(69)90135-5

Boutonnet JC, Bingham P, Calamari, et al. 1999. Environmental Risk Assessment of Trifluoroacetic Acid. Human and Ecological Risk Assessment: An International Journal *5*(1):59-124. https://doi.org/10.1080/10807039991289644

Dekant W, Dekant R. 2023. Mammalian toxicity of trifluoroacetate and assessment of human health risks due to environmental exposures. Arch Toxicol 97(4):1069-1077. doi:10.1007/s00204-023-03454-y

ECHA. 2012. How to prepare toxicological summaries in IUCLID and how to derive DNELs: Practice Guide 14, European Chemicals Agency (ECHA). July 2012.

ECHA. 2024. Registration Dossier of Trifluoroacetic acid, European Chemical Agency (ECHA). Last updated on March 7, 2024. Accessed on September 27, 2024. Available at: https://chem.echa.europa.eu/100.000.846/dossier-list/reach/dossiers/active?searchText=Trifluoroacetic%20acid

EFSA. 2014. Reasoned opinion on the setting of MRLs for saflufenacil in various crops, considering the risk related to the metabolite trifluoroaceitc acid (TFA). EFSA Journal 12(2):3585.

Ema M, Naya M, Yoshida K, Nagaosa R. 2010. Reproductive and developmental toxicity of degradation products of refrigerants in experimental animals. Reprod Toxicol 29(1):1-9. doi:10.1016/j.reprotox.2009.09.005

EPA. 1994. Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry (EPA/600/8-90/066F), U.S. Environmental Protection Agency (EPA). October 1994. Available at: https://www.epa.gov/sites/default/files/2014-11/documents/rfc_methodology.pdf

EPA. 2009. Risk Assessment Guidance for Superfund, Volume I: Human Health Evaluation Manual (Part F, Supplemental Guidance for Inhalation Risk Assessment; EPA-540-R-070-002), Environmental Protection Agency (EPA). January 2009. Available at: https://nepis.epa.gov/Exe/ZyPDF.cgi/P1002UOM.PDF?Dockey=P1002UOM.PDF

EPA. 2022. Fact Sheet: Proposed Revision to the Regulatory Definition of Volatile Organic Compounds – Exclusion of trans--1,1,1,4,4,4-hexafluorobut-2-ene, and HFO-1336mzz(E); (also known as HFO-1336mzz-E), Environmental Protection Agency (EPA). April 2022. Available at: https://www.epa.gov/system/files/documents/2022-04/hfo-1336mmz-e-fact-sheet.pdf

EPA. 2024a. IRIS Glossary, U.S. Environmental Protection Agency (EPA). Last updated on May 23, 2024. Available at: https://www.epa.gov/iris/iris-glossary

EPA. 2024b. Benchmark Dose Software (BMDS) Glossary of Terms, Environmental Protection Agency (EPA). Last updated on January 17, 2024. Available at: https://sor.epa.gov/sor_internet/registry/termreg/searchandretrieve/glossariesandkeywordlists/search.do?details=&vocabName=BMDS%20Glossary&uid=1821254&taxonomyName=BMDS%20Glossary

Fraser JM, Kaminsky LS. 1988. 2,2,2-Trifluoroethanol intestinal and bone marrow toxicity: the role of its metabolism to 2,2,2-trifluoroacetaldehyde and trifluoroacetic acid. Toxicol Appl Pharmacol 94(1):84-92. doi:10.1016/0041-008x(88)90339-0

Garavagno MdlA, Holland R, Khan MAH, Orr-Ewing AJ, Shallcross DE. 2024. Trifluoroacetic Acid: Toxicity, Sources, Sinks and Future Prospects. Sustainability 16:2382. https://doi.org/10.3390/su16062382

Just WW, Gorgas K, Hartl FU, Heinemann P, Salzer M, Schimassek H.1989. Biochemical effects and zonal heterogeneity of peroxisome proliferation induced by perfluorocarboxylic acids in rat liver. Hepatology 9(4):570-581. doi:10.1002/hep.1840090411

Kheilo GI, Kremneva SN. 1966. [Comparative assessment of trifluoro-acetic and pentafluoropropionic acid toxicity]. Gig Tr Prof Zabol 10(3):13-17.

Lloyd SC, Blackburn DM, Foster PM. 1988. Trifluoroethanol and its oxidative metabolites: comparison of in vivo and in vitro effects in rat testis. Toxicol Appl Pharmacol 92(3):390-401. doi:10.1016/0041-008x(88)90179-2

Monnot AD, Massarsky A, Garnick L, Bandara SB, Unice KM. 2023. Can oral toxicity data for PFAS inform on toxicity via inhalation? Risk Anal 43(8):1533-1538. doi:10.1111/risa.14039

National Research Council (NRC) Subcommittee on Reproductive and Developmental Toxicity. 2001. Evaluating Chemical and Other Agent Exposures for Reproductive and Developmental Toxicity. Washington (DC): National Academies Press (US); 2001. Appendix B, Ascertaining Information on the Reproductive and Developmental Toxicity of Agent Exposures. Available from: https://www.ncbi.nlm.nih.gov/books/NBK222212/

Patty FA 1963. Industrial Hygiene and Toxicology, Vol. II, 2nd revised edit, pp. 1801–1803.

Rosenberg PH. 1971. Decrease in reduced glutathione and NADPH and inhibition of

glucose-6-phosphate dehydrogenase activity caused by metabolites of fluoroxene and halothane. Ann Med Exp Biol Fenn 49(2): 84-88.

Rosenberg PH and Wahlstrom T. 1971. Hepatotoxicity of halothane metabolites in vivo and inhibition of fibroblast growth in vitro. Acta Pharmacol Toxicol 29(1):9-19.

Saillenfait AM, Roure MB, Ban M, et al. Postnatal hepatic and renal consequences of in utero exposure to halothane or its oxidative metabolite trifluoroacetic acid in the rat. J Appl Toxicol. 1997;17(1):1-8. doi:10.1002/(sici)1099-1263(199701)17:1<1::aid-jat386>3.0.co;2-z

Solomon KR, Velders GJ, Wilson SR, et al. 2016. Sources, fates, toxicity, and risks of trifluoroacetic acid and its salts: Relevance to substances regulated under the Montreal and Kyoto Protocols. J Toxicol Environ Health B Crit Rev 19(7):289-304. doi:10.1080/10937404.2016.1175981

Stier A, Kunz HW, Walli AK, Schimassek H. 1972. Effects on growth and metabolism of rat liver by halothane and its metabolite trifluoroacetate. Biochem Pharmacol 21(16):2181-2192. doi:10.1016/0006-2952(72)90033-0

UBA. 2021. Reducing the input of chemicals into waters: trifluoroacetate (TFA) as a persistent and mobile substance with many sources, Umwelt Bundesamt (UBA). November 2021.

UBA. Undated. Derivation of a health guide value for trifluoroacetic acid (TFA), Umwelt Bundesamt (UBA); originally published in German, translated using Google Translate. November 2021. Available at: https://www.umweltbundesamt.de/sites/default/files/medien/479/publikationen/hgp_reducing_the_input_of_chemicals_into_waters.pdf

UNEP. 2016. Sources, fates, toxicity, and risks of trifluoroacetic acid and its salts: relevance to substances regulated under the Montreal and Kyoto protocols. United Nations Environment Programme. February 2016.

Warheit DB. 1993. Mechanistic studies with HCFC-123. Dupont Company, Haskell Laboratory Report HLR 828–92.