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Dichloroacetic acid and its salts

MAK Value Documentation, supplement – Translation of the German version from 2019

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Keywords

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Abstract

The German Commission for the Investigation of Health Hazards of Chemical Compounds in the Work Area has re-evaluated dichloroacetic acid [79-43-6] to derive a maximum concentration at the workplace (MAK value) and to review its carcinogenicity classification. The critical effects are irritation, carcinogenicity in the liver of rats and mice as well as neurotoxicity. After oral application, dichloroacetic acid or its sodium salt has tumour-promoting effects and is carcinogenic in the liver of rats and mice. The NOAEL for carcinogenic effects in rats is 3.6 mg/kg body weight. In mice, a NOAEL cannot be derived. Dichloroacetic acid is not mutagenic in most tests in vitro and in vivo. The mechanisms involved in the tumour development in the liver are most likely interference with energy metabolism, oxidative stress and inhibition of apoptosis. As the primary mode of action is non-genotoxic, dichloroacetic acid and its salts are classified in Carcinogen Category 4. Dichloroacetic acid is corrosive to the eye and the skin of rabbits. As no inhalation studies are available to evaluate possible irritating effects on the respiratory tract, the structurally similar trichloroacetic acid is used as a read-across. Therefore, a MAK value of 0.2 ml/m^3 corresponding to 1.1 mg/m^3, is derived for dichloroacetic acid. Accordingly, a MAK value of 1.1 mg/m^3 for the inhalable fraction, measured as the acid, is set for the salts. As the local effect is critical, the acid and its salts are assigned to Peak Limitation Category I with an excursion factor of 1. As skin contact to dichloroacetate may contribute significantly to systemic toxicity, the salts are designated with "H". Skin contact is not expected to contribute significantly to the systemic toxicity of dichloroacetic acid. Sensitization is not expected from the available data.

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MAK value (2018)	**dichloroacetic acid: 0.2 ml/m³ ≙ 1.1 mg/m³** **dichloroacetates: 1.1 mg/m³ I (inhalable fraction)** **as acid**
Peak limitation (2018)	**Category I, excursion factor 1**
Absorption through the skin (2018)	**dichloroacetic acid: –** **dichloroacetates: H**
Sensitization	–
Carcinogenicity (2018)	**Category 4**
Prenatal toxicity	–
Germ cell mutagenicity	–
BAT value	–
CAS number	79-43-6
pKa value, acid	1.48 (US EPA 2003)
Vapour pressure, acid	0.19 hPa (Greim 2010, available in German only)
1 ml/m³ (ppm) ≙ 5.26 mg/m³	**1 mg/m³ ≙ 0.19 ml/m³ (ppm)**

Note: Dichloroacetic acid can occur simultaneously as vapour and aerosol.

In 2010, dichloroacetic acid and its salts were classified in Carcinogen Category 3 A (Greim 2010). On the basis of more recent data, this supplement reviews the earlier classification.

The dichloroacetate anion is responsible for the systemic effects. Therefore, also studies with sodium dichloroacetate or neutralized dichloroacetic acid are included in this supplement. The evaluation applies to the acid and its salts.

Mechanism of Action

Dichloroacetic acid induces xenobiotic-metabolizing enzymes in the liver and has a tumour-promoting effect. The inhibition of apoptosis together with the selective advantage of spontaneously initiated cells has been suggested as the mechanism of action for the formation of liver tumours (Greim 2010). A disturbance in the fat and carbohydrate metabolism, mitochondrial dysfunction, increased oxidative phosphorylation and oxidative stress are further mechanisms considered to be responsible for the carcinogenicity in the liver (Wehmas et al. 2017; Wood et al. 2015). As a result of these effects, there is considerable interference with the energy metabolism, particularly in the liver cells, and probably also, due to their high energy requirement, in the nerve cells. Mutagenic and mitogenic effects, and persistent cytotoxicity do not play a role in tumour formation (Greim 2010).

More recent studies with gavage administration of dichloroacetic acid in mice confirm the occurrence of oxidative stress. Depending on exposure duration and dose, increased concentrations of superoxide anions, changes in the activities of superoxide dismutase, catalase and glutathione peroxidase and increased lipid peroxidation are observed (Table 1).

Tab. 1 Studies of the mode of action of dichloroacetic acid in the liver in vivo published since 2008

Species, strain, number per group	Exposure	Findings	References
mouse, B6C3F1, 8 ♂	**once**, 300 mg/kg body weight, gavage	**300 mg/kg body weight: 6 and 12 hours:** superoxide anions ↑, lipid peroxidation ↑, DNA single strand breaks ↑	Hassoun and Dey 2008
mouse, B6C3F1, 7 ♂	**4 and 13 weeks, 7 days/week**, 0, 7.7, 77, 154, 410 mg/kg body weight and day, gavage, neutralized	**4 weeks:** **7.7 mg/kg body weight and above:** superoxide dismutase ↓, superoxide anions ↑, lipid peroxidation ↑, DNA single strand breaks ↑ catalase, glutathione peroxidase: not affected **13 weeks:** **7.7 mg/kg body weight and above:** superoxide anions ↑, lipid peroxidation ↑, DNA single strand breaks ↑ **7.7 and 77 mg/kg body weight:** superoxide dismutase ↓, catalase: not affected, glutathione peroxidase ↓ **77 and 154 mg/kg body weight:** total glutathione ↓ **154 and 410 mg/kg body weight:** superoxide dismutase ↑, catalase ↑, glutathione peroxidase ↑ **410 mg/kg body weight:** absolute and relative liver weights ↑	Hassoun et al. 2010; Hassoun and Cearfoss 2011
mouse, B6C3F1, 6 ♂	**13 weeks, 7 days/week**, 0, 7.5, 15, 30 mg/kg body weight and day, gavage, neutralized	**7.5 mg/kg body weight and above:** superoxide anions ↑, lipid peroxidation ↑, DNA single strand breaks ↑	Hassoun et al. 2014

Inhibition of the pyruvate dehydrogenase kinase complex

Due to the inhibition of pyruvate dehydrogenase kinase (PDK), dichloroacetic acid interferes in a central metabolic step: the oxidative decarboxylation of pyruvate to acetyl coenzyme A. The kinetic parameters for various isoenzymes of PDK in rats are shown in Table 2 (Bowker-Kinley et al. 1998). In humans, three isoenzymes of PDK have been found to date, namely PDK1, PDK2 and PDK3; PDK1 and PDK2 are almost identical to PDK1 and PDK2 in rats. PDK1 and PDK2 are mainly expressed in the heart and in the skeletal muscle and to a lesser extent in the placenta, lungs, brain, kidneys, liver and pancreas. PDK3 is expressed only in the heart and in skeletal muscle (Gudi et al. 1995). Thiamine deficiency is assumed to be the cause of CNS damage; this itself is a sequel of the activation of the pyruvate dehydrogenase complex due to the inhibition of pyruvate dehydrogenase kinase.

Tab. 2 Kinetic parameters for different isoenzymes of pyruvate dehydrogenase kinase in the rat (Bowker-Kinley et al. 1998)

Parameter	Specific activity [nmol/min per mg protein]	Ki for ADP [µM]	Ki for DCA [mM]	High level of expression
PDK1	650 ± 80	370 ± 20	1.0 ± 0.2	heart
PDK2	50 ± 5	120 ± 20	0.2 ± 0.05	skeletal muscle, heart, testes, liver, brain, kidneys
PDK3	1250 ± 200	80 ± 10	8.0 ± 1	testes
PDK4	400 ± 60	100 ± 15	0.5 ± 0.2	skeletal muscle, heart

ADP: adenosine diphosphate; DCA: dichloroacetic acid, Ki: inhibitory constant

Epigenetic mechanisms

Also the results of a study in which the amount of the methylated base 5-methylcytosine was determined speak against a genotoxic mechanism. As demonstrated by the authors, 5-methylcytosine plays a role in gene expression; hypomethylation of the DNA due to a decrease in 5-methylcytosine levels is thought to be an epigenetic, non-genotoxic

mechanism of tumour promotion facilitating the expression of aberrant genes. Female, 15-day-old B6C3F1 mice were given single intraperitoneal injections of *N*-methyl-*N*-nitrosourea of 25 mg/kg body weight. At the age of 6 weeks, the mice received 25 mmol dichloroacetic acid/l (neutralized with NaOH to pH 6.5–7.5) with the drinking water for 43 or 44 weeks. Some animals were given this drinking water for 11 days without pretreatment with *N*-methyl-*N*-nitrosourea. The 5-methylcytosine level in the hepatic DNA was significantly decreased after 11 days of treatment. After administration for 44 weeks, the values corresponded to those of the controls treated with *N*-methyl-*N*-nitrosourea only. The amount of 5-methylcytosine in the DNA in liver adenomas of these animals was significantly reduced. In the adenomas of the animals that had been treated with dichloroacetic acid for 43 weeks after the administration of *N*-methyl-*N*-nitrosourea and then investigated one week later, the 5-methylcytosine levels were no longer reduced (Tao et al. 1998). This research group had earlier demonstrated that adenomas and hyperplastic nodules induced by dichloroacetic acid regress after termination of the exposure to the substance (Pereira and Phelps 1996).

In a follow-up study it was demonstrated that in female B6C3F1 mice given gavage doses of dichloroacetic acid (neutralized with NaOH) of 500 mg/kg body weight and day for 5 days, the promotor regions of the proto-oncogenes c-jun and c-myc were hypomethylated and the expression of their mRNA and the levels of c-jun and c-myc proteins were increased. When methionine was injected intraperitoneally 30 minutes after each dose of dichloroacetic acid, it inhibited in a dose-dependent manner the hypomethylation and as a result the expression of the corresponding mRNA and proteins at doses of 100 mg/kg body weight and above. The authors discussed whether dichloroacetic acid may cause the hypomethylation of proto-oncogenes c-jun and c-myc via the depletion of *S*-adenosylmethionine, thereby modifying the control of cell proliferation and apoptosis and, in the long run, acting as an epigenetic carcinogen (BG Chemie 2006; Tao et al. 2000).

Toxicokinetics

The toxicokinetic parameters and the metabolism of rats and humans are described in detail in the documentation of 2010 (Greim 2010). In the following, mainly the studies published since 2009 are discussed.

The zeta-1 family of glutathione transferase (GSTz1) is the only known enzyme that metabolizes dichloroacetic acid. Therefore, the activity of GSTz1 determines the toxicokinetics of dichloroacetic acid. GSTz1 is active also in the form of maleylacetoacetate isomerase (MAAI), which catalyzes the penultimate step of tyrosine metabolism. During the biotransformation of dichloroacetic acid, a part of the GSTz1 is inhibited in a dose-dependent manner by the formation of adducts. Besides the wild type (EGT haplotype), a further four polymorphisms of the GSTz1 gene are known. These are designated KGT, EGM, KGM and KRT haplotypes, and occur to different degrees in various ethnic groups. EGM, KRT and KGM haplotypes result in greatly reduced enzyme activity in some cases. Persons with haplotypes EGM, KRT and KGM eliminate dichloroacetic acid only very poorly from the blood, which, compared with persons with the wild-type haplotype, leads to an increased toxicity of dichloroacetic acid (Shroads et al. 2012).

Absorption, distribution, elimination

After oral intake dichloroacetic acid is absorbed rapidly and completely; it has a plasma half-life of approximately one hour and inhibits its own metabolism, which over time after repeated administration results in an increase in its level in plasma and its elimination half-life. Evidence is available that the clearance of dichloroacetic acid and possible neurotoxic degradation products decreases with age and depends on the individual's GSTz1 genotype (Michelakis et al. 2010).

From kinetic studies, it has been estimated that in humans the elimination of dichloroacetic acid is no longer linear at doses of more than 2 mg/kg body weight. Higher dichloroacetic acid doses therefore result in overproportional internal exposure to dichloroacetic acid (Schultz and Shangraw 2006).

After gavage administration of 1, 5, 20 or 100 mg/kg body weight to rats, the bioavailability of the substance could not be determined at the low dose of 1 mg/kg body, as dichloroacetic acid was not detectable in the plasma. Its bioavail-

ability after doses of 5 and 20 mg/kg body weight was 10% and 13%, respectively. At the high dose of 100 mg/kg body weight the bioavailability increased to 81%. In rats, after intravenous administration of 20 mg/kg body weight, the AUC (area under the curve) was 13.8 µg/ml × hour and thus not 20 times, but 100 times as high as that at 1 mg/kg body weight (0.15 µg/ml × hour). In GSTz1-depleted rats, the AUC at 20 mg/kg body weight was 10 times as high as that in naive rats (Saghir and Schultz 2002).

The oral doses administered twice daily in clinical studies were between 4 and 25 mg/kg body weight. At a dose of 6.25 mg/kg body weight given twice daily, the plasma levels necessary to inhibit pyruvate dehydrogenase kinase are attained (Michelakis et al. 2010).

In a phase 1 study with brain tumour patients, it was confirmed in one person with the GSTz1 haplotype EGM/EGM that a dose of 16 mg/kg body weight and day produces a considerably higher level of dichloroacetic acid in the blood (280 mg/l, 2.2 mM) than in persons with the wild-type haplotype (2–55 mg/l) (Dunbar et al. 2014).

In a phase 1 study with 24 patients, after twice daily administration of 6.25 mg dichloroacetic acid/kg body weight, the median dichloroacetate level in the plasma was 121.7 µM (59.8–733.1 µM) after 15 days, and after twice daily administration of 12.5 mg/kg body weight it was 436 µM (210.6–1583.5 µM). After administering 6.25 mg dichloroacetic acid/kg body weight twice daily for 28 days, a value of almost 1 mM was attained (Chu et al. 2015).

In a chamber diffusion study, the penetration through human skin (n = 3) of neutralized dichloroacetic acid at 40 °C was investigated. The exposure lasted between 24 and 48 hours, the lag time was 5 hours. The receptor phase consisted of phosphate-buffered physiological saline solution. At a concentration of 1000 mg neutralized dichloroacetic acid/l, a permeability coefficient of 1.9×10^{-3} cm/hour was found. In addition, the study revealed that the permeability of the simultaneously investigated haloketones was three times as high at 40 °C than at 20 °C (Xu et al. 2002). From the permeability coefficient for dichloroacetic acid, a flux of 1.9 µg/cm² and hour at 40 °C can be calculated. A threefold increase in permeability at 40 °C compared with at 20 °C can be expected also for dichloroacetic acid, so that a flux of 0.63 µg/cm² and hour is assumed for a 0.1% solution at usual workplace temperatures. With a pKa value of 1.48, dichloroacetic acid is present in a practically completely ionized form on the skin, which has a pH of 5.5, so that adjustment to a pH of 7 did not change the dissociation equilibrium to any relevant extent. The flux therefore applies to both dichloroacetic acid and dichloroacetate. According to ECHA (2018), dichloroacetic acid is corrosive to the skin. For such substances, skin irritation at concentrations of 1% and above is to be assumed according to the CLP regulation. After linear extrapolation of the flux of 0.63 µg/cm² to a non-irritating 0.5% solution, the amount absorbed after the exposure of a 2000 cm² skin surface for one hour would be 6.3 mg.

Sodium dichloroacetate and other dichloroacetates have not been classified as regards skin irritation. In analogy to sodium monochloroacetate, classification as a skin irritant is assumed. For such substances, skin irritation at concentrations at and above 10% is to be assumed according to the CLP regulation. After linear extrapolation of the flux of 0.63 µg/cm² to a non-irritating 5% solution, the amount absorbed after the exposure of a 2000 cm² skin surface for one hour would be 63 mg.

Effects in Humans

Sodium dichloroacetate is used as a therapeutic agent for various metabolic conditions such as lactate acidosis, hyperglycaemia or hypercholesterolaemia. No relevant side-effects have been found to date in volunteer studies following short-term treatment with sodium dichloroacetate. After long-term therapy with sodium dichloroacetate for chronic diseases, care must be taken that an appropriate low dose is selected due to the severe neurological side-effects. As dichloroacetic acid or sodium dichloroacetate have been used therapeutically for a considerable time, extensive toxicokinetic and toxicodynamic data are available (Stacpoole et al. 1998; see also Greim 2010).

In about 50% of all patients given sodium dichloroacetate in doses of 25 to 50 mg/kg body weight and day, anxiolytic and sedative effects occurred. Peripheral neuropathy was reported in three persons. The symptoms were no longer present in all persons six months after discontinuing the medication. The administration of further doses of 10 to

25 mg/kg body weight and day to these persons over a period of two years no longer resulted in neurological effects (Stacpoole et al. 1998).

Use in tumour therapy

A possible anti-tumour effect of dichloroacetic acid has been under discussion since 2007. Tumour cells have an increased energy requirement, which is provided also by the degradation of glucose to lactate (Warburg effect). As dichloroacetic acid inhibits the oxidative decarboxylation of pyruvate to acetyl coenzyme A via the inhibition of pyruvate dehydrogenase kinase, there is a reduction in the available energy (Anti-Warburg effect). Attempts have been made to make use of this effect in tumour therapy, although the success of this is, to date, unclear.

No controlled clinical studies of the use of dichloroacetic acid in tumour patients are available.

In a phase 1 study with 5 patients, the effect of dichloroacetic acid on glioblastomas was investigated. Three patients received dichloroacetic acid twice daily in increasing oral doses of between 6.25 mg/kg body weight and 25 mg/kg body weight as monotherapy, and two were given dichloroacetic acid in addition to radiation therapy and the cytostatic drug temozolomide. The authors reported a partial remission in one of the three patients who had received dichloroacetic acid alone. On the basis of the results of this study, the evidence for the efficacy of dichloroacetic acid against glioblastomas is very weak. At a dose below 6.25 mg/kg body weight given twice daily (12.5 mg/kg body weight and day), none of the patients had peripheral neuropathy. No dichloroacetic acid was detectable in the plasma during the first two to three months. After three months, the plasma levels were 0.44 ± 0.16 mM in 4 patients, which is within the range of the inhibition concentration of 0.2 mM for PDK2 (Michelakis et al. 2010).

In another phase 1 study, the safety of dichloroacetic acid therapy and the response in 13 patients with high-degree gliomas and 2 patients with brain metastases from adenocarcinomas of the uterus or the lungs were investigated. The patients were given oral doses of dichloroacetic acid of 8 mg/kg body weight twice daily for 4 weeks. The dichloroacetic acid dose was adapted according to GSTz1 genotype. The best objective result after 4-week treatment was stabilization of the disease in 8 patients. Based on these results, the hypothesis was put forward that dichloroacetic acid has a limited effect on glioblastomas or brain metastases. In 2 of the 8 patients, sensory peripheral neuropathy (grade 1) in the fingertips and toes occurred without muscle weakness and ataxia. In one "slowly metabolizing" patient, the symptoms occurred in the second cycle after four weeks and in one "rapidly metabolizing" patient in the sixth cycle after 24 weeks of treatment. Gait abnormalities occurred in three other patients. One patient with the GSTz1 haplotype EGM/EGM, who had received doses of 16 mg/kg body weight and day, reacted after 26 days with neuropathy and increased transaminase activity in the blood. The authors recommend a dose of 10 mg/kg body weight and day for persons who do not have the wild-type allele of the GSTz1 gene, to avoid side-effects such as neuropathy as far as possible (Dunbar et al. 2014).

Another phase 1 study was carried out to assess the safety of dichloroacetic acid treatment and to document a possible effect on the tumour in 24 patients with therapy-resistant, advanced solid malignancies. 16 patients were given oral dichloroacetic acid doses of 6.25 mg/kg body weight twice daily. The dichloroacetic acid dose was administered twice daily on 28-day cycles and was increased to 12.5 mg/kg body weight in 7 patients. The treatment was continued until progression of the disease, occurrence of unacceptable toxicity or consent withdrawal. Objectively, likewise in this study, the best result was disease stabilization in 8 patients. On the basis of this phase 1 study, the efficacy of dichloroacetic acid in the treatment of advanced and therapy-resistant solid tumours can neither be confirmed nor refuted (Chu et al. 2015).

Animal Studies and in vitro Studies

Subacute, subchronic and chronic toxicity

Inhalation

No information is available.

Oral administration

The NOAELs (no observed adverse effect levels) and LOAELs (lowest observed adverse effect levels) derived from the various studies after oral administration are shown in Table 3. The individual studies are described in detail in the documentation of 2010 (Greim 2010). For the end points liver adenomas, liver carcinomas and hepatocyte proliferation in male rats, a systemic NOAEL of 3.6 mg/kg body weight and day can be derived from the 100-week drinking water study. Female mice reacted to dichloroacetic acid less sensitively than the males. Increased relative liver weights and vacuolization of the hepatocytes were found at 52 mg/kg body weight and day. At this dose, however, no liver carcinomas occurred. A NOAEL cannot be derived for male mice. Increased incidences of liver carcinomas were found even at the lowest dose tested of 8 mg/kg body weight and day. Other effects were not described.

It is also not possible to provide a NOAEL for male and female dogs, as effects on the central nervous system and histopathological changes in the testes occurred even at the lowest dose tested of 12.5 mg/kg body weight and day.

Tab. 3 NOAELs and LOAELs after repeated oral administration of dichloroacetic acid or sodium dichloroacetate

Species	Dose [mg DCA/SDCA/kg body weight and day] duration (administration)	Effects	References
rat			
F344 ♂	3.6 DCA 100 weeks (drinking water)	NOAEL for peroxisome proliferation, hepatocyte proliferation, liver carcinomas	DeAngelo et al. 1996
Sprague Dawley ♂	3.9 DCA 13 weeks (drinking water)	NOAEL (no histopathological changes in the brain)	Mather et al. 1990
F344/Long Evans ♂	16 DCA 13 weeks (drinking water)	LOAEL locomotor disturbances (see Table 4)	Moser et al. 1999
mouse			
B6C3F1 ♂	8 DCA 100 weeks (drinking water)	LOAEL liver carcinoma incidences significantly increased	DeAngelo et al. 1999; DeAngelo and Daniel 1990
B6C3F1 ♀	40 DCA 576 days (drinking water)	NOAEL for liver adenomas and carcinomas, LOAEL liver effects	Pereira 1996
B6C3F1 ♀	94 DCA 104 weeks (drinking water)	NOAEL for liver carcinomas	Schroeder et al. 1997
dog			
beagle ♂ and ♀	12.5 SDCA 13 weeks (gelatine capsules)	LOAEL histopathological changes: central nervous system, testes, gall bladder, liver	Cicmanec et al. 1991

DCA: dichloroacetic acid; SDCA: sodium dichloroacetate

For adult Long Evans rats, a NOAEL of 23 mg/kg body weight and day can be derived for the end point neurotoxicity (Table 4) from the 13-week drinking water study by Moser et al. (1999). At the lowest dose (16 to 18 mg/kg body weight and day), deficits in gait occurred in adult and young F344 rats; these were evaluated as the earliest indicators of neurotoxicity. A BMDL (benchmark dose lower confidence limit) cannot be calculated as no incidences are given. A dose of 16 mg/kg body weight would correspond to exposure at the workplace to 10 mg/m^3 (16 mg/kg body weight / 4 × 70 kg/10 m^3 × 7/5 / 2 / 2).

Tab. 4 Neurotoxic effects of dichloroacetic acid after administration with the drinking water to young and adult Long Evans and F344 rats (Moser et al. 1999)

Strain	Age (days)	Duration (weeks)	Dose: effects
10 LE per group	29–30	13	17 mg/kg body weight: deficits in gait 88 mg/kg body weight: decrease in grip strength of fore and hind limbs (no dose–response relationship) 192 mg/kg body weight: tremor, hypotension, inhibition of pupil reflex
10 F344 per group	29–30	13	≥ 16 mg/kg body weight: deficits in gait 173 mg/kg body weight: decrease in grip strength of fore and hind limbs, tremor, hypotension, inhibition of pupil reflex
10 LE per group	68–69	8	23 mg/kg body weight: NOAEL ≥ 122 mg/kg body weight: decrease in grip strength of fore and hind limbs, no effect on foot splay, righting reflex deficits in 33%
10 F344 per group	68–69	8	18 mg/kg body weight: deficits in gait ≥ 91 mg/kg body weight: decrease in grip strength of fore and hind limbs, increase in landing foot splay, righting reflex deficits in 40%, unusual chest-clasping response on being lifted by the tail

LE: Long Evans

In another study of the neurotoxicity of dichloroacetate (no other details), young and adult female Sprague Dawley rats were given gavage doses for 16 weeks. In the first experiment, 0 or 50 mg/kg body weight and day were given to young rats aged 4 to 6 weeks. In the second experiment, 0, 100, 200 or 500 mg/kg body weight and day were administered to the same rats and 0 or 500 mg/kg body weight and day to adult rats (aged 12 to 14 weeks). In the first experiment, the motor and sensory nerve conduction velocity (SNCV) were significantly reduced and the tactile response threshold significantly increased at 50 mg/kg body weight. In the second experiment, the most sensitive end point was the SNCV, which was slowed in a dose-dependent manner after 16 weeks. The motor fibres were not affected to the same extent by this change. The reaction to thermal pain was unchanged (Calcutt et al. 2009). The fact that the changes in the 100 mg/kg group were less pronounced than in the first experiment after administration of 50 mg/kg body weight and day cannot be explained. No statistically significant results were obtained in any of the post-hoc tests. The initial weights of the animals varied somewhat, but there were no differences in the other dependent variables between the two control groups. The absence of significant effects in the post-hoc test on the SNCV may be a result of the higher variance in the control group. The absolute values for these parameters are very similar in the 50 and 100 mg/kg body weight dose groups of the two experiments. The reason for this effect is the reduction in the diameter of the non-myelinated nerve fibres in the treated animals. Here too, nominal differences between the control group and the 100 mg/kg group are evident; they are, however, not significant. This effect is not found in the first experiment, and the reason for the slowing of the SNCV in this experiment is unclear. The tactile response threshold was not determined in the second experiment, and the pain thresholds are not as sensitive as this end point. The fact that this information is lacking makes it difficult to compare the two experiments. The other biochemical and neuropathological findings obtained in the older animals clearly indicate that the mechanism of action for the effects of dichloroacetic acid in the peripheral nervous system is a reduction of the fibre diameter of non-myelinated nerves mediated by oxidative stress. As the results of the two experiments with young rats are not consistent, a NOAEL cannot be derived from this study.

Local effects on skin and mucous membranes

Dichloroacetic acid is corrosive to the skin and eyes of rabbits (Greim 2010). No information is available for sodium dichloroacetate.

Genotoxicity

The genotoxicity studies are described in detail in the documentation of 2010 (Greim 2010). In in vitro studies, weak mutagenic effects have been found only in the Salmonella typhimurium strains TA100 and TA1535. The clastogenic effects observed in vitro can be explained by the extreme acidity, as the medium was not neutralized after the addition of dichloroacetic acid.

Positive and negative results were obtained in tests for DNA crosslinks and DNA strand breaks in vivo. The discrepancies cannot be explained by the methodological details reported. The positive results for the induction of micronuclei are not considered meaningful due to the great variation in control values. These are in contrast to a number of methodologically sound studies with negative results, so that the suspected clastogenic effects of dichloroacetic acid could not be confirmed in vivo. Weak gene mutation activity was found in the Big Blue mouse at doses far above those causing carcinogenic effects in the mouse; mutations are therefore not the primary cause of carcinogenicity (Greim 2010).

In vitro studies published since 2009 are described below.

A test for the induction of the SOS response in the Salmonella typhimurium strain TA1535/pSK1002 (SOS/umu test) yielded positive results at and above dichloroacetic acid concentrations of 15 511 µM. The cytotoxicity, investigated in parallel in a microplate cytotoxicity test, was determined in a concentration range from 200 to 77 600 µM. The lowest cytotoxic concentration was 7760 µM. In the authors' opinion, the cytotoxicity correlated to a great extent with the genotoxic effect (Zhang et al. 2016). No information is given regarding neutralization or the pH.

In a comet assay with CHO (Chinese hamster ovary) cells (AS52 cell line) exposed for 4 hours to dichloroacetic acid concentrations of between 1 and 25 mM, no increase in DNA strand breaks was observed (Plewa et al. 2002). The concentration producing a decrease in cell growth to 50% of the control value was 7.3 (Plewa et al. 2010) or 11.46 mM (Plewa et al. 2002). Even below cytotoxic conditions, no DNA damage could be detected. No information is given regarding neutralization or the pH.

In another comet assay with HepG2 cells exposed to dichloroacetic acid concentrations of 0 to 10 000 µM for 4 hours, DNA damage was recorded at concentrations of 10 µM and above. At these concentrations, cytotoxicity was below 25% (Zhang et al. 2012). No information is given regarding neutralization or the pH.

In a micronucleus test with human lymphocytes, dichloroacetic acid was tested at concentrations of 0, 25, 50 or 100 µg/ml (0, 194, 388, 776 µM). Treatment of the cells lasted 48 hours. At the middle and high concentrations, the increase in micronuclei was statistically significant, but not concentration-dependent. The survival of the lymphocytes compared with the control value was 62%, 41% and 24%, respectively. Compared with the control group, significantly more cells were in the S phase, and the rate of apoptosis was increased, although not in a concentration-dependent manner. An increase in micronuclei thus occurs only at cytotoxic concentrations and when the rate of apoptosis is increased (Varshney et al. 2013). Also this publication does not mention whether neutralization was carried out after the addition of the test substance, or whether the pH was determined.

In an HPRT (hypoxanthine guanine phosphoribosyl transferase) gene mutation test with CHO cells (0–10 000 µM) without the addition of metabolic activation (the test was not carried out with metabolic activation), an increase in mutation frequency was demonstrated at 1000 µM and above in two test series carried out consecutively (surviving cells in the second test series at 1000, 5000 and 10 000 µM: 75.5%, 53.1% and 42.5%, respectively, compared with the control value). No information is given regarding neutralization or the pH (Zhang et al. 2010).

Summary: The in vitro tests published since the documentation of 2010 yielded positive results in the SOS/umu test with Salmonella typhimurium, in the comet assay with HepG2 cells, in the micronucleus test with human lymphocytes and in the HPRT gene mutation test. Details regarding neutralization after the addition of the test substance or

a pH control are not given in any of the publications, so that the positive results can presumably be attributed to the acidity.

Carcinogenicity

No studies with inhalation exposure are available. In all drinking water studies, dichloroacetic acid was adjusted to a pH of about 7, which means that the carcinogenic effects are independent of the acidity and are the result of the exposure to dichloroacetate. The carcinogenicity studies with the lowest dose levels after administration with the drinking water in rats and mice are presented in Table 5.

Tab. 5 Studies of the carcinogenicity of dichloroacetic acid in rats and mice

Author:	DeAngelo et al. 1996; DeAngelo and Daniel 1992; Richmond et al. 1995				
Substance:	dichloroacetic acid (purity > 99%), neutralized				
Species:	**rat**, F344/N, 50–78 ♂ per group				
Administration route:	drinking water				
Concentration:	1st experiment: 0 (2.0 g NaCl/l); 0.05, 0.5, 5.0 g/l (after 9 weeks reduced from 5.0 g/l to 2.5 g/l; after 23 weeks to 2.0 g/l; after 52 weeks to 1.0 g/l and finally withdrawn from the study) (0, 3.6, 40.2 mg/kg body weight and day) 2nd experiment: 0 (water), 2.5 g/l (after 8 weeks reduced to 1.5 g/l; after 26 weeks to 1.0 g/l: mean 1.6 g/l, corresponding to 139.1 mg/kg body weight and day)				
Duration:	1st experiment: 100 weeks 2nd experiment: 103 weeks				
Toxicity:	1st experiment: high dose group: irreversible peripheral neuropathy of the hind legs, therefore sacrificed after 60 weeks 2nd experiment: body weights significantly ↓				
	Dose [mg/kg body weight and day]				
	0	**3.6**	**40.2**	**0**	**139**
survivors after 100 and 103 weeks, respectively	23/50	21/60	23/60	33/78	28/78
Tumours and preneoplasms[a]					
Liver:					
hyperplastic nodules	1/23 (4.4%)	0/26	3/29 (10%)	1/33 (3.0%)	1/28 (3.6%)
adenomas[b]	1/23 (4.4%)	0/26	6/29 (21%)*	0/33	3/28 (10.7%)
carcinomas[c]	0/23	0/26	3/29 (10%)	1/33 (3.0%)	6/28 (21.4%)*
adenomas and carcinomas[d]	1/23 (4.4%)	0/26	9/29 (31%)**	1/33 (3.0%)	8/28 (28.6%)**
hyperplastic nodules, adenomas and carcinomas	2/23 (8.7%)	0/26	12/29 (41%)**	2/33 (6.1%)	9/28 (32.1%)**

*p ≤ 0.05; **p ≤ 0.01
[a] only animals were examined which survived more than 78 weeks
[b] historical controls: incidence 2.3%; range 0%–10% (Haseman et al. 1998)
[c] historical controls: incidence 0.7%; range 0%–6% (Haseman et al. 1998)
[d] historical controls: incidence 2.8%; range 0%–10% (Haseman et al. 1998)

Author:	DeAngelo et al. 1999
Substance:	dichloroacetic acid (purity > 99%), neutralized
Species:	**mouse**, B6C3F1, 35–88 ♂ per group
Administration route:	drinking water
Concentration:	0, 0.05, 0.5, 1, 2, 3.5 g/l (0, 8, 84, 168, 315, 429 mg/kg body weight and day)
Duration:	100 weeks, interim sections 10 animals/group after 26, 52, 78 weeks

Tab. 5 *(continued)*

Toxicity:	at 315 mg/kg body weight and above: absolute and relative liver weights ↑, body weights ↓					
	Dose [mg/kg body weight and day]					
	0 (water)	**8**	**84**	**168**	**315**	**429**
animals at start of study	88	35	55	71	55	46
interim sections	35	–	30	30	30	30
number of animals that died	3	2	1	9	11	8
survivors after 100 weeks	50	33	24	32	14	8
histopathological examination	85	33	55	65	51	41
Tumours						
Liver:						
adenomas[a)]	5/50 (10%)	1/33 (3%)[c)]	5/25 (20%)	18/35 (51%)*	9/21 (43%)*	5/11 (45%)*
carcinomas[b)]	13/50 (26%)	11/33 (33%)*	12/25 (48%)*	25/35 (71%)*	20/21 (95%)*	11/11 (100%)*

*p ≤ 0.05; **p ≤ 0.01
[a)] historical controls: incidence 29.4%; range 4%–60% (Haseman et al. 1998)
[b)] historical controls: incidence 17.9%; range 6%–29% (Haseman et al. 1998)
[c)] according to US EPA (2003)

Author:	Pereira 1996
Substance:	dichloroacetic acid, neutralized
Species:	**mouse**, B6C3F1, 38–134 ♀ per group
Administration route:	drinking water
Concentration:	1st experiment: 0 (NaCl), 2, 6.67, 20 mmol/l (258, 860, 2580 mg/l, about 40, 133, 400 mg/kg body weight and day, estimated from the data of DeAngelo et al. (1999); pH value 6.5–7.5) 2nd experiment: 20 mmol/l intermittent (24 days treatment, 48-day interval, corresponding to on average 6.67 mmol/l (133 mg/kg body weight and day))
Duration:	360, 576 days
Toxicity:	body weights at 400 mg/kg body weight ↓, dose-dependent increase in relative liver weights, vacuolization of hepatocytes, no data for survivors

	Dose [mg/kg body weight and day]				
	0	**40**	**133**	**400**	**133 (intermittent)**
Tumours and preneoplasms (only liver investigated):					
360 days					
foci of cellular alteration	0/40	0/40	1/20 (5%)	8/20 (40%)*	0/15
adenomas	1/40 (2.5%)	0/40	3/20 (15%)	7/20 (35%)	0/15
carcinomas	0/40	0/40	0/20	1/20 (5%)	0/15
576 days					
foci of cellular alteration	10/90 (11%)	7/50 (14%)	11/28 (39%)*	17/19 (89%)*	14/34 (41%)*
adenomas[a)]	2/90 (2.2%)	3/50 (6.0%)	7/28 (25%)*	16/19 (84%)*	3/34 (8.8%)
carcinomas[b)]	2/90 (2.2%)	0/50	1/28 (3.6%)	5/19 (26%)*	1/34 (2.9%)

*p ≤ 0.05
[a)] historical controls: incidence 17.3%, range 2%–50% (Haseman et al. 1998)
[b)] historical controls: incidence 8.4%, range 0%–20% (Haseman et al. 1998)

After the administration of dichloroacetic acid with the drinking water at a dose level of 40 mg/kg body weight and day, the incidence of liver tumours was significantly increased in male rats. No liver tumours occurred at the lowest dose tested of 3.6 mg/kg body weight and day. In male B6C3F1 mice, the number of animals with liver carcinomas was significantly increased at the lowest dose tested of 8 mg/kg body weight and day and above. In female mice, a NOAEL of 52 mg/kg body weight was derived for carcinogenicity. Liver tumours occur very frequently in B6C3F1 mice. The

number of liver tumours was, however, dose-dependently increased in male and in female mice, and the incidences were markedly above the historical control data given by the NTP at that time. Dichloroacetic acid was thus found to be carcinogenic in the liver of rats and mice. Furthermore, a tumour-promoting effect of dichloroacetic acid was observed in all initiation–promotion studies. A significantly increased incidence of proliferation was observed after 5 days exposure in the liver of mice but not, however, after exposures lasting from 12 days to 100 weeks (Greim 2010).

A new drinking water study in mice is available which has confirmed the carcinogenic effects in males and females. The animals were exposed to neutralized dichloroacetic acid for 10 weeks, and the treatment was then followed by a recovery period lasting 84 weeks or further exposure to phenobarbital. The incidence of liver carcinomas in the male mice was not increased up to 232 mg/kg body weight. In the females, the incidence of liver carcinomas was increased, though not in a statistically significant manner (Table 6). The relative mRNA expression of *CAT* (catalase), *Nqo1* (NAD(P)H dehydrogenase (quinone 1)), *Hmox1* (haemoxigenase 1), *Gclm* (glutamate cysteine ligase), *Cyp2b10* (cytochrome P450 2b10), *Cyp4a10* (cytochrome P450 4a10), *Bcl2* (B cell lymphoma 2), *Mki67* (protein phosphatase 1 regulatory subunit) was increased after single doses of phenobarbital. The genes are markers for oxidative stress, mitogenesis, apoptosis or cell proliferation. After exposure to dichloroacetic acid alone, no significant changes in these mRNA profiles and no changes in DNA methylation or the distribution of the single nucleotide variants were observed (Wood et al. 2015).

Tab. 6 Drinking water study of the carcinogenic mechanism of action after administration of dichloroacetic acid and phenobarbital

Author:	Wood et al. 2015
Substance:	DCA (purity > 99%), neutralized
Species:	**mouse**, B6C3F1, 48 ♂, 48 ♀ per group
Administration route:	drinking water
Concentration:	0 (deionized water); ♂: 1, 2, 3.5 g/l (136, 232, 297 mg/kg body weight and day); ♀: 1, 2 g/l (142, 253 mg/kg body weight and day)
Study design:	10 weeks: ♂: 0, 1, 2, 3.5 g DCA/l, ♀: 0, 1, 2 g DCA/l thereafter: 84 weeks deionized water or 0.06% phenobarbital
Toxicity:	body weights and food consumption without effects, no increased mortality, reduced water consumption, at and above 2 g DCA/l: liver weights ↑

		Dose [g/l]						
		0	**PB**	**1 DCA**	**1 DCA + PB**	**2 DCA**	**2 DCA + PB**	**3.5 DCA**
Liver:								
hepatoblastomas	♂	0/27	5/20	1/27	7/19*	0/27	8/19**	0/26
	♀	0/27	0/20	0/26	0/19	0/28	0/20	–
adenomas	♂	5/27	16/20**	13/27*a)	14/19**	11/27	16/19**	15/26*
	♀	0/27	16/20**	9/26**	17/19**	6/28	19/20**	–
carcinomas	♂	8/27	13/20	8/27	11/19	6/27	13/19*a)	19/26*
	♀	0/27	4/20	2/26	4/19	3/28	8/20**	–
sum	♂	12/27	20/20**	15/27	16/19	14/27	19/19**	24/26**
	♀	0/27	17/20**	10/26**	18/19**	9/28**	19/20**	–

DCA: dichloroacetic acid; PB: phenobarbital
*$p \leq 0.05$; **$p \leq 0.01$
a) subsequently calculated with one-sided Fisher's exact test

In another study, the progression of cell proliferation and metabolism during the formation of liver cell carcinomas was investigated in male mice given dichloroacetic acid with the drinking water either for 93 weeks or for only 4 to 52 weeks and then just water up to week 93. In addition, a short-term study (6, 15 and 30 days) with 8 animals per dose group (0, 0.5, 1, 2 or 3.5 g/l) was carried out. Cell proliferation was slightly increased during the 10th week only.

After administration of dichloroacetic acid for 4 weeks, the number of animals with carcinomas was significantly increased only during the 86th week of the recovery period. The first adenomas and carcinomas occurred after exposure for 52 weeks and were significantly increased (Table 7). The gene expression was changed in genes responsible for mitochondrial dysfunction, oxidative phosphorylation, the NRF2-mediated response to oxidative stress and activation of Farnesoid X receptor/Retinoid X receptor-alpha (FXR/RXR α), LXR (liver X receptor)/RXR and peroxisome proliferator-activated receptor alpha (PPAR α)/RXR α. An increase in DNA sequence variants, irrespective of age and exposure to dichloroacetic acid, could not be detected. There was no effect on apoptosis either in the short-term study or in the chronic study after 57 weeks (the only determination). The authors mention that cytotoxicity was not observed at any time; the results are, however, not described. The authors attribute the carcinogenic effect to mitochondrial dysfunction, changed oxidative phosphorylation, the response to oxidative stress and an influence on the fat and carbohydrate metabolism in the cell. They more or less exclude the involvement of mitogenesis, mutagenesis or persistent cytotoxicity as mechanisms (Wehmas et al. 2017).

Tab. 7 Long-term administration of dichloroacetic acid with the drinking water to investigate cell proliferation and metabolism

Author:	Wehmas et al. 2017					
Substance:	DCA (purity > 99%), neutralized					
Species:	mouse, B6C3F1, in total 404 ♂, (see Table for distribution in groups)					
Administration route:	drinking water					
Concentration:	0 (deionized water) or 3.5 g/l (about 429 mg/kg body weight and day)					
Study design:	0, 3.5 g/l, 93 weeks continuously, or 3.5 g/l for 4, 10, 26, 52 weeks, then untreated up to week 93					
Toxicity:	body weights 20% ↓, no increased mortality, reduced water consumption, relative liver weights up to week 57 ↓, thereafter ↑, reversible liver cell hypertrophy and liver cell necrosis					
	DCA exposure / water (in weeks)					
	0/93	**4/89**	**10/83**	**26/67**	**52/41**	**93/0**
Liver:						
adenomas	12/52 (23%)	7/28 (25%)	18/55 (33%)	22/54 (41%)*[a)]	30/54 (56%)*	26/44 (59%)*
carcinomas	9/52 (17%)	23/28 (82%)*	27/55 (49%)*	32/54 (59%)*	35/54 (65%)*	41/44 (93%)*
adenomas and carcinomas	19/52 (37%)	24/28 (86%)*	34/55 (62%)*	39/54 (72%)*	49/54 (91%)*	44/44 (100%)*

DCA: dichloroacetic acid
*$p < 0.05$
[a)] subsequently calculated with one-sided Fisher's exact test

Manifesto (MAK value classification)

The most sensitive end points are irritation, the hepatocarcinogenicity in rats and mice, and the demonstrated neurotoxicity in humans.

MAK value. No data are available for repeated inhalation exposure of humans or of animals. However, as with monochloroacetic acid and trichloroacetic acid, in addition to the systemic effects, due to the acidic properties of the substance, effects on the respiratory tract are to be expected. The pKa value of dichloroacetic acid is lower than that of phosphoric acid, for which reason the MAK value has been established at 0.2 ml/m^3 (1.1 mg/m^3) in analogy to that for the likewise stronger acid trichloroacetic acid as related to the molar masses. At a MAK value of 1.1 mg/m^3 for dichloroacetic acid, 11 mg or 0.16 mg/kg body weight and day would be absorbed at a respiratory volume of 10 m^3 and a body weight of 70 kg. This dose is markedly below the dose used therapeutically of 12.5 mg/kg body weight and day.

At 2 mg/kg body weight and above, the elimination of dichloroacetic acid is no longer linear in humans (Schultz and Shangraw 2006). The internal exposure to dichloroacetic acid at 12.5 mg/kg body weight can be expected to be overproportional compared with that at the dose of 0.16 mg/kg body weight.

Tab. 8 Survey of the NOAELs and LOAELs and the resultant workplace concentrations

Species	End point	NOAEL [mg/kg body weight and day]	LOAEL [mg/kg body weight and day]	Extrapolated workplace concentration [mg/m³]
rat F344	liver	3.6	40	4
mouse B6C3F1	liver	40 ♀	8 ♂	3.7
rat F344 (young)	neurotoxicity	–	16	10
rat, Long Evans	neurotoxicity	23	122	28
humans	neurotoxicity	< 10	> 10	70
humans	irritation analogy to TCA	absorption 1.1 mg/m³ × 10 m³ = 11 mg/day 0.16 mg/kg body weight up to 2 mg/kg body weight: linear elimination		0.2 ml/m³ ≙ 1.1 mg/m³

TCA: trichloroacetic acid

From a chronic drinking water study in rats, a NOAEL of 3.6 mg/kg body weight and day can be derived for the effects on the liver. From this NOAEL of 3.6 mg/kg body weight for liver effects in F344 rats, a concentration of 4 mg/m³ (3.6 mg/kg body weight / 4 × 70 kg/10 m³ × 7/5 / 2 = 4 mg/m³) would result. Neuropathy was not observed at this level, however it was observed in the high dose group given about 160 mg/kg and day. Effects on the brain did not occur (Table 8).

According to the PBPK (physiologically based pharmacokinetic) model of Li et al. (2008), 2.1 mg/kg body weight and day in the mouse corresponds to a dose of 0.1 mg/kg body weight in humans, based on the daily AUC of dichloroacetic acid in the liver. The LOAEL (lowest observed adverse effect level) of 8 mg/kg body weight and day from the carcinogenicity study with male mice therefore corresponds to a dose of 0.53 mg/kg body weight and day on 5 days per week for humans. For a person of 70 kg body weight and a respiratory volume of 10 m³, assuming absorption of 100%, a concentration of 3.7 mg/m³ is obtained. The NOAEL (no observed adverse effect level) was 40 mg/kg body weight and day in female mice. The 10-fold spontaneous incidence of liver carcinomas in male B6C3F1 mice (26%; DeAngelo et al. 1999) compared with that in the females (2.2%; Pereira 1996) indicates a considerable sex-specific difference in the effects, making the extrapolation of the dose obtained in the male animals to that for humans a worst-case scenario.

In the 13-week drinking water study by Moser et al. (1999) neuropathy occurred at dose levels of 16 mg/kg body weight and day and above in F344 rats. By comparison, Long Evans rats were somewhat less sensitive. A BMDL cannot be calculated as no incidences are given. A dose of 16 mg/kg body weight would correspond to 10 mg/m³ (16 mg/kg body weight / 4 × 70 kg/10 m³ × 7/5 / 2 / 2) for exposure at the workplace. This concentration is 9 times as high as the MAK value of 1.1 mg/m³.

All in all, it is difficult to establish a NOAEL for systemic effects, as the male B6C3F1 mouse is not very suitable for a quantitative comparison with humans due to the high spontaneous incidence of liver tumours. In addition, it is not clear whether the rat can be quantitatively compared with humans as regards neuropathy. As the elimination of the substance is no longer linear at about 2 mg/kg body weight and above, and as twice daily doses of 6.25 mg/kg body weight and day led to a dichloroacetic acid level of about 1 mM in patients with slow metabolism after two weeks of treatment (Chu et al. 2015)—where the half maximum inhibitory concentration for pyruvate dehydrogenase kinase is about 0.2 mM—neurotoxicity is not to be expected even in the least favourable case at a dose of 1 mg/kg body weight. In addition, the patients were treated on 7 days per week, whereas exposure at the workplace is on 5 days per week only.

For the salts, a MAK value of 1.1 mg/m³ I (inhalable fraction), as acid, has been established.

Peak limitation. Acid: Irritation is the main effect, so that dichloroacetic acid has been classified in Peak Limitation Category I. In analogy to trichloroacetic acid, an excursion factor of 1 has been set.

Salts: Data for irritation are not available. In analogy to sodium monochloroacetate and sodium trichloroacetate, an irritant effect is assumed for the salts so that the salts of dichloroacetic acid are assigned to Peak Limitation Category I with excursion factor 1.

Carcinogenicity. Dichloroacetic acid was found to be carcinogenic in the liver of rats and mice. Furthermore, in all initiation–promotion studies, tumour-promoting effects of dichloroacetic acid were observed. Quantitative evaluation of the results of the in vivo genotoxicity tests indicates that genotoxic mechanisms are not the primary mechanisms of liver tumour formation. Also, more recent studies with mice indicate that mutagenic and mitogenic effects as well as persistent cytotoxicity are not responsible for tumour formation; the decisive factors are extensive interference with energy metabolism, oxidative stress and the inhibition of apoptosis.

As genotoxicity is not a major factor in the mechanism of carcinogenesis, the prerequisites for the classification of dichloroacetic acid and its salts in Carcinogen Category 4 are fulfilled.

Germ cell mutagenicity. Also on the basis of the data for genotoxicity published since the documentation of 2010, germ cell mutagenicity is not to be suspected. The substance is therefore not classified in one of the categories for germ cell mutagens.

Absorption through the skin. In humans, the dermal absorption of 6.3 mg dichloroacetate can be estimated from an in vitro study (Xu et al. 2002) after exposure to a non-irritating 0.5% solution of dichloroacetic acid, assuming one-hour exposure of a 2000 cm^2 skin surface. On the basis of the information given above, for a body weight of 70 kg, an amount of 70 mg is estimated from the systemically tolerable dose of 1 mg/kg body weight and day. Therefore, absorption through the skin makes up less than 25% of the systemically tolerable amount. Dichloroacetic acid is therefore not designated with an "H" (for substances which can be absorbed through the skin in toxicologically relevant amounts).

Taking, as calculated above, an absorbed amount of 6.3 mg for 0.5% dichloroacetic acid as a basis, the absorption of 63 mg would be expected after linear extrapolation for a non-irritating 5% dichloroacetate solution. For dichloroacetate, the same tolerable amount of 70 mg applies, as derived above. Therefore, absorption through the skin makes up more than 25% of the systemically tolerable amount, and dichloroacetates are designated with an "H".

Sensitization. As there are still no data available for sensitization, dichloroacetic acid and its salts are not designated with "Sh" (for substances which cause sensitization of the skin) or "Sa" (for substances which cause sensitization of the airways).

Notes

Competing interests

The established rules and measures of the Commission to avoid conflicts of interest (https://www.dfg.de/en/dfg_profile/statutory_bodies/senate/health_hazards/conflicts_interest/index.html) ensure that the content and conclusions of the publication are strictly science-based.

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