Phosphoric acid, trimethyl ester: Human health tier II assessment

08 March 2019

CAS Number: 512-56-1

Preface

This assessment was carried out by staff of the National Industrial Chemicals Notification and Assessment Scheme (NICNAS) using the Inventory Multi-tiered Assessment and Prioritisation (IMAP) framework.

The IMAP framework addresses the human health and environmental impacts of previously unassessed industrial chemicals listed on the Australian Inventory of Chemical Substances (the Inventory).

The framework was developed with significant input from stakeholders and provides a more rapid, flexible and transparent approach for the assessment of chemicals listed on the Inventory.

Stage One of the implementation of this framework, which lasted four years from 1 July 2012, examined 3000 chemicals meeting characteristics identified by stakeholders as needing priority assessment. This included chemicals for which NICNAS already held exposure information, chemicals identified as a concern or for which regulatory action had been taken overseas, and chemicals detected in international studies analysing chemicals present in babies' umbilical cord blood.

Stage Two of IMAP began in July 2016. We are continuing to assess chemicals on the Inventory, including chemicals identified as a concern for which action has been taken overseas and chemicals that can be rapidly identified and assessed by using Stage One information. We are also continuing to publish information for chemicals on the Inventory that pose a low risk to human health or the environment or both. This work provides efficiencies and enables us to identify higher risk chemicals requiring assessment.

The IMAP framework is a science and risk-based model designed to align the assessment effort with the human health and environmental impacts of chemicals. It has three tiers of assessment, with the assessment effort increasing with each tier. The Tier I assessment is a high throughput approach using tabulated electronic data. The Tier II assessment is an evaluation of risk on a substance-by-substance or chemical category-by-category basis. Tier III assessments are conducted to address specific concerns that could not be resolved during the Tier II assessment.

These assessments are carried out by staff employed by the Australian Government Department of Health and the Australian Government Department of the Environment and Energy. The human health and environment risk assessments are conducted

and published separately, using information available at the time, and may be undertaken at different tiers.

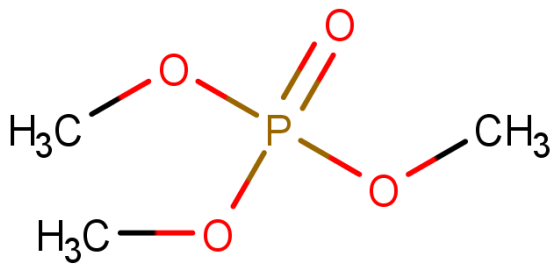
This chemical or group of chemicals are being assessed at Tier II because the Tier I assessment indicated that it needed further investigation.

For more detail on this program please visit:www.nicnas.gov.au

Disclaimer

Acronyms & Abbreviations

Chemical Identity

Synonyms	trimethyl phosphate TMP/TMeP tris(methyl) phosphate trimethyl orthophosphate O,O,O-trimethyl phosphate
Structural Formula	
Molecular Formula	C3H9O4P
Molecular Weight (g/mol)	140.07
Appearance and Odour (where available)	Clear colourless to slightly yellow liquid. Pleasant sweet odour.
SMILES	COP(=O)(OC)OC

Import, Manufacture and Use

Australian

No data is available for the chemical.

International

The following international uses have been identified by the United States (US) National Library of Medicine's Hazardous Substances Data Bank (HSDB); the US National Toxicology Program (NTP); European Union (EU) Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH) dossier (REACHa); Perkins & Will White Paper, 2014; the United Kindom Environment Agency (UK Environment Agency, 2003); the International Labour Organization (ILO) Encyclopaedia of Occupational Health & Safety, the Organisation for Economic Co-operation and Development (OECD) Screening information data set Initial Assessment Report (OECD SIAR, 1996); US Environment Protection Agency (US EPA) ChemView; the US EPA Chemical and Product Categories (CPCat); and the National Cancer Institute Technical Report (NCI, 1978).

The chemical has reported commercial uses as a:

- lubricant additive (<1 %);
- gasoline additive (<0.01 %);
- antifoulant for spark plugs; and
- flame retardant in paints, polymers, textile fabrics and building materials.

Whilst some of these uses could have application in the domestic setting, the chemical is not listed in the US Department of Health & Human Services Household Products Database (US HPD).

The chemical has reported site-limited uses as:

- a methylating agent;
- a catalyst in polymers and resin production; and
- an intermediate in the production of polymethyl phosphates.

The chemical has reported non-industrial uses as a raw material for making pesticides.

Restrictions

Australian

No known restrictions have been identified.

International

No known restrictions have been identified.

Existing Work Health and Safety Controls

Hazard Classification

The chemical is not listed on the Hazardous Chemical Information System (HCIS) (Safe Work Australia).

Exposure Standards

Australian

No specific exposure standards are available.

International

No specific exposure standards are available.

Health Hazard Information

Toxicokinetics

Absorption

Trimethyl phosphate (TMP) is absorbed rapidly and completely by inhalation of its vapour, through the skin and by ingestion (WHO).

Metabolism

In mice and rats, the chemical is metabolised relatively quickly after oral administration, and less quickly after intraperitoneal administration. Almost 90 % of the chemical is metabolised in 16 hours and in 96 hours most of the chemical is metabolised (US EPA, 1985a; Jackson & Jones, 1968; REACHa).

The chemical is degraded to dimethyl phosphate (DMP), but not to monoalkyl phosphate or free phosphoric acid. Metabolism to DMP was reported in rats and in mice treated intraperitoneally (i.p.) and orally with ^{32}P-labelled TMP. Following i.p. adminstration metabolism in the rat was slower with unmetabolised chemical detected in the urine. Other metabolites detected in urine included S-methyl cysteine, S-methyl cysteine N-acetate, and small amounts of S-methyl glutathione. Metabolism was again slower in the rat compared to the mouse. Based on an in vitro study, the chemical is metabolised in rat liver and intestinal tissue, but not in kidney tissue (US EPA, 1985a; Jackson & Jones, 1968; REACHa).

Excretion

The metabolites are primarily excreted in the urine (REACHa). The metabolites of trialkyl phosphates in general may also be excreted in exhaled air and in the faeces (US EPA, 1985a; US EPA, 1985b).

Acute Toxicity

Oral

Based on the reported median lethal doses (LD50) in experimental animals, the chemical has moderate oral toxicity, warranting hazard classification (see ***Recommendation*** section).

In an acute oral toxicity study in rats conducted according to the Organisation for Economic Cooperation and Development (OECD) Test Guideline (TG) 401 (Acute oral toxicity), the reported median lethal dose (LD50) value was 840 mg/kg bw (OECD SIAR, 1996; CCOHS RTECS; REACHa).

In other acute oral toxicity studies conducted in rats, mice, rabbits and guinea pigs, the reported LD50 values ranged between 1110–3390, 1470–3610, 940–1271 and 1050–1275 mg/kg bw, respectively (US EPA, 1985a; US EPA, 1985b; CCOHS RTECS).

Clinical signs of toxicity in rabbits, guinea pigs and rats included hyperexcitability, fine tremors incoordination, muscular weakness, paralysis and respiratory failure (see ***Neurotoxicity*** section).

Dermal

The chemical has low acute toxicity based on limited data from an animal test following dermal exposure.

In an acute dermal toxicity study, TMP was applied in liquid form on the skin of 4 male New Zealand White (NZW) rabbits for 24 hours (occlusive), with observation for 14 days. No other study details were reported. The LD50 value was 2.83 mL/kg bw (calculated as 3388 mg/kg bw based on density of 1.197g/mL) (US EPA, 1985a; REACHa).

Inhalation

No data are available for this chemical. However, the low vapour pressure (0.74 Pa at 25 °C) of the chemical indicate that inhalation exposure would not be significant (unless aerosolised) under normal conditions (OECD SIAR, 1996).

Corrosion / Irritation

Skin Irritation

Limited data are available. While the chemical is reported to be a strong irritant to skin (HSDB), the data available do not support this. The chemical is not corrosive based on an in vitro OECD TG study and available animal data indicate only slight irritant effects.

In an in vitro study conducted according to OECD TG 431 (In vitro skin corrosion: Reconstructed human epidermis (RHE) test method), the chemical (97 % purity; 50 µL) was applied to human non-transformed keratinocytes for 4 hours. The mean tissue viability following TMP treatment was 95 % demonstrating that the chemical is not expected to be corrosive to skin (REACHa).

The chemical was rated 4 on the scale of 1-10 (10 most severe) for skin irritation in rabbits. No other study details are available (US EPA, 1985a; US EPA, 1985b).

In a non-guideline study, 6 albino rabbits were treated with 2 mL/kg bw/day (2394 mg/kg bw/day) TMP for 2 hours (see ***Repeated dose toxicity: Dermal*** section). Treatment was repeated for 20 days over a total of 28 days. No signs of dermal irritation were reported (Deichmann & Witherup, 1946).

The related chemical, triethylphosphate (CAS No. 78-40-0), was non-irritating in skin irritation study in rabbits conducted similarly to the OECD TG 404 (REACHb).

Eye Irritation

Limited data are available. While the chemical is reported to be a strong irritant to eyes (HSDB), the data available do not support this claim. Based on the OECD TG study, the chemical is not expected to cause severe eye irritation.

In an in vitro study conducted according to OECD TG 438 (Isolated chicken eye test method), the chemical (97 % purity; 30 µL) was applied onto the centre of the cornea of chickens for 10 seconds. The chemical did not cause ocular corrosion or severe irritation in the enucleated chicken eyes (REACHa).

The chemical was rated 2 on the scale of 1-10 (10 most severe) for eye irritation in rabbits. No other study details are available (US EPA, 1985a; US EPA, 1985b).

The related chemical, triethylphosphate (CAS No. 78-40-0), was slightly to moderately irritating in an eye irritation study in rabbits conducted similarly to the OECD TG 405 (REACHb). Whilst effects in the cornea (score 1.7) and conjunctivae (score 2) were seen in all three animals, these were reversed in two animals by 7 days and one animal by 14 days.

Observation in humans

The alkyl phosphate esters in general are suggested to cause skin, eye and respiratory irritation in exposed individuals (US EPA, 1985b). However, no chemical specific information is available.

Sensitisation

Skin Sensitisation

Limited data are available. Based on weight of evidence, the chemical is not expected to be a skin sensitiser. In chemico and in vitro guideline studies covering the first two key events of the adverse outcome pathway (AOP) for skin sensitisation were negative. A structurally similar chemical, triethyl phosphate (CAS No. 78-40-0) is not a skin sensitiser based on a guideline local lymph node assay (LLNA).

In a study conducted according to OECD TG 442C (In chemico skin sensitisation: Direct peptide reactivity assay (DPRA)), TMP (100 mM) and positive control (100 mM) were incubated in excess with cysteine and lysine peptides at 1:10 and 1:50 ratios, respectively. The positive control results were a depletion of 51.55 % and 75.11 % for lysine and cysteine, respectively. The chemical showed a mean depletion of 0% for lysine and 2.87 % for cysteine, indicating no or minimal activity and; therefore, a negative prediction of DPRA (REACHa).

In a study conducted according to OECD TG 442D (In vitro skin sensitisation: antioxidant/electrophile response element (ARE)-Nrf2 luciferase test method), luciferase activity was measured in keratinocyte cells (KeratinoSensTM) treated with the chemical (up to 200 mM), as an indicator of activation of endogenous Nrf2 dependent genes. A negative result is indicated by an Imax (maximal average fold induction of luciferase activity observed at any concentration of the tested chemical and positive control) value that is less than 1.5. Two out of three repetitions were negative (Imax values of 0.87, 1.27 and 2.12) and; therefore, it was concluded that the chemical did not induce the keratinocyte Nrf2 pathway linked to skin sensitisation (REACHa).

In a study conducted according to OECD TG 429 (Skin sensitisation: LLNA), CBA/Ca mice (5/group) were treated with 50 µL of the undiluted triethyl phosphate (CAS No 78-40-0) (at concentrations of 25 % or 50 % v/v in dimethyl formamide). All concentrations produced negative results, with stimulation index (S.I.) values of 2.53, 1.97 and 2.47 (REACHb).

Repeated Dose Toxicity

Oral

Based on the available information, the chemical may cause adverse effects on the nervous system (see ***Neurotoxicity*** section) following repeated exposure, and is recommended for classification (see ***Recommendation*** section).

In a study similar to OECD TG 422 (Combined repeat dose and reproductive/developmental screening toxicity test), TMP was administered daily by oral gavage to Crj:CD(SD) rats (13/sex/dose) at doses of 0, 40, 100 or 250 mg/kg bw/day. Males were exposed for 42 days, including 14 days before mating. Females were exposed from 14 days before mating to day 3 of lactation. The highest dose was lethal to 12 males and 1 female. Clinical signs of toxicity at the highest dose included progressive paralytic gait and decreased motor activity that became evident after week 2 (see ***Neurotoxicity*** section). None of the females at highest dose and only two females at the 100 mg/kg bw/day fell pregnant; however, the two females at 100 mg/kg bw/day did not deliver any pups (see ***Reproductive toxicity*** section). Therefore, only females receiving 40 mg/kg bw/day were followed until the end of the study. Body weights were significantly reduced at the highest dose when compared to controls, with significantly reduced food intake in males. In addition, pregnant females given 40 or 100 mg/kg bw/day had a significant decrease in body weight gain during mid and late pregnancy. Changes in haematology and clinical chemistry were reported in males at 100 mg/kg bw/day or more. The absolute and relative kidney weights were significantly increased in males receiving 40 mg/kg bw/day or more. The absolute and relative thymus weights were significantly increased in females at 40 mg/kg bw/day and in males at 100 mg/kg bw/day. Renal lesions were observed in males at all doses, but a dose related increase was not observed. Nephropathy was characterised by tubular and papillary alteration (increased eosinophilic droplets in tubular

epithelium, and increased regeneration of tubules and papillary necrosis). Degeneration of nerve fibres was observed at doses of 100 mg/kg bw/day or more (see ***Neurotoxicity*** section). Other histopathological findings included atrophy of the liver hepatocytes, thymus (cells not specified), adrenal (cortical cells), spleen follicles and testes were reported in high dose males rats. Other than effects in nerve fibres, no histopathological findings were reported in females. A no observed adverse effect level (NOAEL) was not determined based on increased absolute and relative kidney weights accompanied by histological changes in males at the lowest tested dose at 40 mg/kg bw/day (US EPA, 2010; OECD SIAR, 1996; HSDB).

In a 30-month well described toxicity study, TMP was administered to Wistar rats (50/sex/dose) in drinking water at doses of 0, 1, 10 or 100 mg/kg bw/day. The highest dose was not tolerable and was reduced to 50 mg/kg bw/day in week 54. Additional groups of animals (10/sex/dose) were treated for 12 months only. Mortality was increased at the highest dose despite the dose reduction. Clinical signs of toxicity at the highest dose included weakness of hind limbs, increased incidence of sunken flanks (especially in males), distended abdomen (especially in females), and poor general condition. Bodyweight gain was significantly decreased in males at the two highest doses and in females at the highest dose. The increased relative organ weights in the high-dose group were attributed to decreased body weight. The chemical had minor effects on the haematopoietic system. Neurotoxic effects including histological damage to the nervous system were observed in the high-dose group (see ***Neurotoxicity*** section) in both the 12 month and full study groups. Necropsy of animals that died during the study showed higher incidences of changes in the lungs (mottled, reddish, pale) and heart (thick, hard, abnormal colour), and scarring of the kidneys. Scarring of kidneys and small testes were noted in animals at 24 months but no changes attributable to treatment were noted at 30 months. The NOAEL, based on suppression of body weight gain was 1 mg/kg bw/day in males and 10 mg/kg bw/day in females (Bomhard et al., 1997; REACHa).

In a 9-week study, male Wistar rats were fed the chemical at concentrations of 0 or 0.5 % in diet (0 or 461 mg/kg bw/day). Body weights in treated rats were significantly lower than the control. Differences between the treated and control rats were observed in organ weights (elevated absolute and relative kidney weights and reduced testes weight), haematology and clinical chemistry including serum enzyme activities (US EPA, 2010; OECD SIAR, 1996; Oishi et al., 1982).

In a dose finding study for chronic carcinogenicity studies (see ***Carcinogenicity*** section), rats and mice (5/sex/dose) were orally (gavage) treated with the chemical at doses up to 1470 or 2150 mg/kg bw/day, respectively for 3 days a week for seven weeks. Reduced bodyweights and mortalities were reported with effects more pronounced in rats. The body weight gain in rats was reduced to approximately 80 % of control following treatment at 316 mg/kg bw or above (NTP, 1978).

Dermal

Limited information is available for the chemical.

In a non-guideline study, 6 NZW rabbits were treated with 2 mL/kg bw/day (2394 mg/kg bw/day) TMP for 2 hours. Treatment was repeated for 20 days over a total of 28 days. Half of the animals showed reduced body weight. Another 3 albino rabbits were treated as above but the chemical was left on the skin for 3 hours instead of 2. The chemical caused significant body weight loss and was lethal to 2 out of 3 rabbits. All rabbits dermally treated with the chemical showed fine tremors, unsteadiness and weakness, and one rabbit developed flaccid paralysis (see ***Neurotoxicity*** section; Deichmann and Witherup, 1946).

Inhalation

No data are available for this chemical.

Observation in humans

No medically significant depression of cholinesterase were reported in an occupational exposure study among a group of 175 factory workers exposed via inhalation to a mixture of chemicals, including trimethyl phosphate, during the manufacture or formulation of pesticide products. The levels of trimethyl phosphate at the plant (based on six personal air samples) were below the detection limit (US EPA, 2010).

Genotoxicity

The chemical was consistently positive in in vivo genotoxicity assays. Mixed or equivocal findings were reported in genotoxicity assays in vitro. Due to consistently positive in vivo genotoxicity assay findings, including heritable germ cell tests and effects in sperm, classification is warranted (see ***Recommendation*** section).

In vitro tests

The chemical was positive in:

- multiple reverse mutation assays with *Salmonella typhimurium* TA 100 and *Escherichia coli* (strains WP2, WP2 uvrA) in the presence and absence of activation. Other strains were positive in a single study or produced mixed results.
- DNA repair assay in *E. coli*;
- DNA damage assay in rat hepatocytes without activation;
- micronucleus assay in Chinese hamster lung cells and
- chromosomal aberration assay in human lymphocytes (REACHa, Connor, 1979; OECD SIAR, 1996; US EPA 1985a, US EPA 1985b; US EPA, 2010).

The chemical was negative in:

- bacterial mutation assays with *S. typhimurium* (strains TA97, TA98, TA100, TA102, TA1530, TA1535, TA1531, TA1532, TA1534) and *E. coli* (strains CM561, CM571, P3110 (polA+), P3478 (polA-));
- induction of DNA double-strand breaks in rat hepatocytes; and
- induction of chromosomal aberrations in cultured Chinese Hamster lung cells (OECD TG 473) (REACHa, Connor, 1979; OECD SIAR, 1996; US EPA 1985a, US EPA 1985b; US EPA, 2010).

In vivo tests

In multiple studies, the chemical induced (following gavage or intraperitoneal injection):

- chromosome aberrations in bone marrow cells of rats and mice;
- chromosome aberrations in spermatocytes in hamsters and mice;
- micronuclei in mouse bone marrow cells; and
- heritable translocations in mice (REACHa, Connor, 1979; OECD SIAR, 1996; US EPA 1985a,b; US EPA, 2010).

The chemical produced dominant lethal effects in several strains of mice and recessive lethal mutations in male *Drosophila melanogaster* fruit fly. The chemical induced somatic mutations and was positive for eye mosaic assay in *D. melanogaster* (Connor, 1979; US EPA, 2010).

The chemical was reported negative in one mouse micronuclei test (no details available) (US EPA, 2010).

Carcinogenicity

Based on limited evidence in experimental animals, the chemical has carcinogenic potential and warrants classification (see ***Recommendation*** section). The chemical produced neoplasms in female mice and benign fibromas in male rats.

In a long-term carcinogenicity study, B6C3F1 mice (50/sex/dose) and F344 rats (50/sex/dose) were treated, by gavage, with the chemical 3 times per week, at doses of 250 or 500 mg/kg bw/day in mice, and 50 or 100 mg/kg bw/day in rats. Mice and rats were treated for 103 and 104 weeks, respectively. The chemical caused significantly higher (high dose compared to control) dose-related incidence of adenocarcinomas of the uterus/endometrium in female B6C3F1 mice. This type of tumour was not observed in historical control rats. The chemical induced a significantly higher (high dose compared to control) dose-related incidence of benign fibromas of the subcutaneous tissue in male Fischer 344 rats. There was no evidence of carcinogenicity in female rats or male mice (OECD SIAR, 1996; NCI, 1978; HSDB; REACHa).

The chemical was not carcinogenic in a 30-month study. The chemical was administered to Wistar rats (50/sex/dose) in drinking water at doses of 0, 1, 10 or 50/100 mg/kg bw/day (see ***Repeated dose toxicity*** section) (Bomhard et al., 1997; REACHa).

Reproductive and Developmental Toxicity

The chemical caused adverse effects on the reproductive system of male rats and associated reproductive impairment in females. High doses (≥100 mg/kg bw/day in rats and at 1000 mg/kg bw/day in mice; US EPA, 2010) induced reversible male sterility in various animal models. Due to the findings hazard classification is warranted (see ***Recommendation*** section). The chemical does not cause specific developmental toxicity based on available data.

Fertility and reproductive system

In a study conducted according to OECD TG 422, Crj:CD(SD) rats (13/sex/dose) were exposed to the chemical (0, 40, 100 or 250 mg/kg bw/day) via oral gavage, from 14 days before mating for a total of 42 days (males) or from 14 days before mating to day 3 of lactation (females) (see ***Repeated dose toxicity*** section). Only two females receiving the highest dose copulated, and only two females that received 100 mg/kg bw/day fell pregnant (out of 13 that copulated). The fertility index was 100, 92.3, 15.4 and 0 % for groups receiving 0, 40, 100 or 250 mg/kg bw/day. All control females and 12/13 females treated with 40 mg/kg bw/day successfully delivered litters. The two pregnant females receiving 100 mg/kg bw/day did not deliver live pups. In the 40 mg/kg group, the fertility index and number of implantation sites were non-significantly reduced. Intrauterine mortality of embryos was significantly increased, with significantly less live pups born for females treated with the chemical. Three females treated with the chemical lost their litters by day 4 of lactation. At day 4 of lactation, the pups in the 40 mg/kg group had significantly higher body weights than those in the control group. This is assumed to be due to the significantly smaller litter size. At terminal necropsy, the males receiving 100 mg/kg bw/day and above (only 1 surviving male at the highest dose) had significantly reduced absolute and relative epididymal weights. Histopathological examination showed atrophy of the testes and increased atretic follicles in the ovary at the highest dose (interim necropsies of the deceased animals). A NOAEL for reproductive toxicity could not be determined based on increased intrauterine mortality at the lowest dose of 40 mg/kg bw/day (OECD SIAR, 1996). A NOAEL for parental toxicity (see ***Repeated dose toxicity*** section) could not be determined based on kidney effects in males at the lowest dose of 40 mg/kg bw/day (OECD SIAR, 1996).

Following oral (gavage) treatment of male Wistar rats at 100 or 250 mg/kg bw/day of TMP for 5 days, dose-dependent reduction was reported in a number of offspring 2-5 weeks after treatment. The fertility was restored to normal 6 weeks after the end of treatment. Limited data and details are available (US EPA, 2010; Jackson & Jones, 1968).

In a non-guideline study, male SD rats (number not specified) were orally (gavage) treated at 0 or 250 mg/kg bw/day of TMP 5 days a week, for 30 days, or 6 days/week for 60 days. Following treatment, the males were mated with untreated females. No vaginal plugs were reported in females suggesting a TMP effect on mating behaviour of males (US EPA, 2010; Hanna & Kerr, 1981; HSDB).

In another non-guideline study, fertility was evaluated in male SD rats, male albino Swiss mice and NZW rabbits. Animals were treated with the chemical either i.p. or orally (gavage) (the actual route used or number of animals in each study was not specified). Mating behaviour was not affected and the frequency of vaginal plugs and mountings were similar between TMP treated and in control groups. Significantly reduced fecundity was reported in each species following exposure to various doses of TMP. In rats and mice, based on reduced fecundity and persistent sterility, the LOAELs were 100 and 1500 mg/kg bw/day of TMP (5 days a week for 1 month), respectively. Effects on sterility were reversible. In rats treated with the chemical 5 days a day for 90 days, the LOAEL was 200 mg/kg bw/day based on reduced fecundity. The administration method used in different experiments was not clearly reported (US EPA, 2010; Harbison et al, 1976).

Following treatment of Wistar male rats (5 rats for TMP and 7 for vehicle control) orally with 100 mg/kg bw/day for 5 consecutive days, the prostate weights relative to body weight were significantly reduced in TMP treated males. Weights of the seminal vesicles, testes or pituitary weights were not significantly affected. Based on histology, spermatogenesis appeared normal and motile sperm were present in epididymis and vas deferens. The number of immature looking Leydig cells was considered to be increased in the testicular interstitial tissue (Carstensen, 1971).

In a 30-month repeated dose toxicity study in Wistar rats (50/sex/dose), TMP was administered in drinking water at doses of 0, 1, 10 or 100 mg/kg bw/day (see ***Repeated Dose Toxicity*** section). The highest dose was not tolerable and was reduced to 50 mg/kg bw/day in week 54. There was an increased incidence of changes in the testes (small size, contents fluid) in the high-dose group. Small seminal vesicles were also observed at necropsy of males receiving the highest dose (Bomhard et al., 1997; REACH).

Spermatogenesis and sperm abnormalities

The chemical was shown to impair spermatogenesis, induce sperm structural abnormalities and impair epididymal sperm motility.

In a non-guideline study, male SD rats (number not specified) were orally (gavage) treated with 0 or 250 mg/kg bw/day of TMP, 5 days a week, for 30 days, or 6 days/week for 60 days. The epididymal sperm from TMP treated males had structural abnormalities. Histological examination of testes revealed impaired spermatogenesis due to defects in final stage of spermatogenesis leading to depletion of mature spermatids. Following 60 days of treatment, germ cells were absent and only Sertoli cells were present in the seminiferous tubules. A NOAEL was not established in this study (US EPA, 2010; Hanna & Kerr, 1981; HSDB).

In a non-guideline study, male SD rats (20/dose) were orally (gavage) treated with 0 to 1500 mg/kg bw/day, 5 days a week up to 5 weeks. Surviving rats (4/dose) were sacrificed weekly. None of the rats receiving 750 mg/kg bw/day or more survived until the end of the experiment, and 90% mortality was reported for the group receiving 500 mg/kg bw/day. Histological examination of testes revealed effects on spermatogenesis with aggregation of multinucleated giant cells and maturation arrest at the spermatid stage (US EPA, 2010; Cho & Park, 1994).

Following oral (gavage) administration of 0 or 100 mg/kg bw/day of TMP to SD rats (10/dose) for 28 days, cauda epididymal sperm motility was reduced while no effects were reported on sperm numbers or viability. Presence of degenerated sperm was reported in the epididymis of 3 out of 10 rats. No significant histological changes were reported in the testes, seminal vesicles or prostate. No effects were reported on food intake or body weights (US EPA, 2010; Takizawa et al., 1998).

Following oral treatment of male Long-Evans hooded rats with 0, 100, 250, or 600 mg/kg bw/day of TMP in water for 5 days, body weight gain was significantly reduced (all doses). The absolute weights of testes and epididymides were not affected by the chemical. The epdididymal sperm count and the number of motile sperm were significantly reduced at the highest dose (≤50 % of the control) (Toth et al., 1992).

Male Crj-CD(SD) IGS strain rats were orally treated with a single dose of the chemical at 600 mg/kg bw, sperm motility was significantly impaired at 1, 2 and 3 weeks after the treatment. Testes weights and sperm numbers were not affected (Fukunishi et al., 2000).

Concentration dependent inhibition of sperm motility was reported following treatment of epididymal sperm suspension with the chemical in vitro. The inhibition of motility may be due to the inhibition of sperm choline acetyltransferance by the chemical (Harbison et al., 1976).

Other Health Effects

Neurotoxicity

The chemical had no anticholinesterase activity normally associated with organophosphorous compounds, nor did it cause related organophosphate induced delayed neuropathy (OPIDN) (Connor et al., 1979; Jackson & Jones, 1968; NICNAS). However, based on the available information, the chemical can cause neurotoxicity following repeated exposure and warrants hazard classification (see ***Recommendation*** section). The chemical may also cause acute neurotoxicity at high doses.

In a neurotoxicity study, 5 beagle dogs were fed daily with gelatine capsules containing 1 mL of the chemical for 1–4 months. The approximate daily doses were 88 and 121 mg/kg bw/day for males exposed for 29 or 50 days, and 105, 89, and 106 mg/kg-day for females exposed for 71, 101, or 121 days, respectively. One female dog received 2 mL of the chemical in gelatine capsules, 5 days a week for 150 days (appr. 181 mg/kg bw/day). The effects were compared to previous studies on untreated control dogs. After 4 weeks, signs of neurotoxicity (impairment of hopping, tactile placing and tactic gait) were reported. After week 9, electrophysiological tests showed prolonged latency of neuromuscular impulse transmission followed by a decrease in maximum conduction velocity of sensory fibres. Neuropathology examination revealed degenerative changes in nerve fibers and demyelination of axons among the female dogs treated for the longest periods (101 and 121 days). Significant body weight loss and inactivity after day 88 were reported in the dog treated at 181 mg/kg bw/day of the chemical. Neuropathology showed advanced distal degeneration of the long spinal tracts and peripheral nerve fibers, and demyelination of nerve fibers (US EPA, 2010; HSDB).

The chemical did not cause delayed neurotoxicity (evaluated as ataxia) in adult white leghorn hens treated with daily intraperitoneal injections of 50 mg/kg bw/day of TMP for 10 days (Hollingshaus et al., 1981).

In an OECD TG 422 combined repeated dose toxicity study with the reproduction/developmental toxicity screening test conducted in Crj:CD(SD) rats (13/sex/dose), animals were administered the chemical (0, 40, 100 or 250 mg/kg bw/day) via oral gavage (see ***Repeated dose toxicity*** section). Rats given 100 mg/kg bw/day or more showed degeneration of nerve fibres in the spinal cord and peripheral nerves (e.g. sciatic nerve) (US EPA, 2010; OECD SIAR, 1996).

In a 30-month study, Wistar rats (50/sex/dose) received the chemical in drinking water at doses of 0, 1, 10 or 100 mg/kg bw/day (see ***Repeated dose toxicity*** section). The highest dose was not tolerable and was reduced to 50 mg/kg bw/day in week 54. Additional groups of animals (10/sex/dose) were treated for 12 months only. Histopathologically, most animals in the high-dose group (100/50 mg/kg bw/day), in both the 12 month and full study groups, showed peripheral nerve degeneration and spinal cord degeneration. Myopathy of skeletal muscles was also observed in several animals. The incidence and degree of the myopathy of skeletal muscles increased in a dose-dependent manner. At the 12-month interim necropsy, some rats of the highest dose group showed signs of hind limb skeletal muscle wasting. Three high-dose males showed signs of emaciation (Bomhard et al., 1997; REACHa).

In an acute toxicity study, albino rabbits and rats (n=6–10/dose) or guinea pigs (n=2/dose) were orally administered a single dose up to 4.7 mL/kg bw of the chemical. Another group of 9 albino rabbits received cutaneous application of 2.0 mL/kg bw/day of TMP for 2–3 hours, 5 days a week for 4 weeks. Acutely lethal doses of the chemical induced a gradually decreasing rate and amplitude of respiratory movements, general weakness, mild hyperirritability, and fine tremors, followed by dyspnoea, collapse and respiratory failure. These findings indicate significant neuromuscular disturbance at high doses. The repeated dermal application in rabbits caused similar signs of poisoning, frequent body weight loss, unsteadiness, general weakness, incoordination of lower extremities and flaccid and spastic paralysis of the extremities (Deichmann & Witherup, 1946).

Endocrine Disruption

Based on the limited available information, the chemical can modify testicular testosterone synthesis.

Following treatment of Wistar male rats (n=4–6) orally at 100 mg/kg bw/day of the chemical for 5 consecutive days, the plasma concentration as well as testicular content and concentration of testosterone were reduced by 59, 66 and 77 % when compared to the control. The chemical altered the histochemical localisation of 3-beta-hydroxysteroid dehydrogenases within the testes (Carstensen, 1971).

Risk Characterisation

Critical Health Effects

The critical health effects for risk characterisation include systemic long-term effects including reproductive toxicity and carcinogenicity as well as potentially heritable genotoxicity. The chemical causes adverse effects in the nervous system.

Public Risk Characterisation

The uses of the chemical in Australia are unknown. The chemical is used overseas as a gasoline additive and in paints and polymers as a flame retardant. However, the chemical is not expected to have frequent uses in consumer products that could expose the public directly to the chemical. The chemical is not listed in the US Household Products Database and in Europe the chemical is registered for use as an intermediate and processing aid (REACHa).

Although it is expected that the chemical will be bound within articles or coated surfaces, consumers may be directly exposed to the chemical that is released from articles through, for example, abrasion or dissolution. However, the presence of the chemical in household air or dust was negligible (Araki at al., 2013; Kanazawa et al., 2010). While many phosphate triester flame retardants were commonly detected in the household dust, TMP was not above the detection limit in any dust samples (n=40) (Kanazawa et al., 2010). Hence, the public risk from this chemical is not considered to be unreasonable.

Occupational Risk Characterisation

During product formulation, dermal and inhalation exposure might occur, particularly where manual or open processes are used. These could include transfer and blending activities, quality control analysis, and cleaning and maintaining equipment. Worker exposure to the chemical at lower concentrations could also occur while using formulated products containing the chemical. The level and route of exposure will vary depending on the method of application and work practices employed.

Given the critical systemic long-term health effects, the chemical could pose an unreasonable risk to workers unless adequate control measures to minimise dermal and inhalation exposure are implemented. Good hygiene practices to minimise oral exposure are expected to be in place. The chemical should be appropriately classified and labelled to ensure that a person conducting a business or undertaking (PCBU) at a workplace (such as an employer) has adequate information to determine the appropriate controls.

The data available support an amendment to the hazard classification in the Hazardous Chemical Information System (HCIS) (Safe Work Australia) (see ***Recommendation*** section).

NICNAS Recommendation

Assessment of the chemical is considered to be sufficient, provided that the recommended amendment to the classification is adopted, and labelling and all other requirements are met under workplace health and safety and poisons legislation as adopted by the relevant state or territory.

Regulatory Control

Work Health and Safety

The chemical is recommended for classification and labelling aligned with the Globally Harmonized System of Classification and Labelling of Chemicals (GHS) as below. This does not consider classification of physical hazards and environmental hazards.

From 1 January 2017, under the model Work Health and Safety Regulations, chemicals are no longer to be classified under the Approved Criteria for Classifying Hazardous Substances system.

Hazard	Approved Criteria (HSIS)[a]	GHS Classification (HCIS)[b]
Acute Toxicity	Not Applicable	Harmful if swallowed - Cat. 4 (H302)
Repeat Dose Toxicity	Not Applicable	May cause damage to nervous system through prolonged or repeated exposure - Cat. 2 (H373)
Genotoxicity	Not Applicable	May cause genetic defects - Cat. 1B (H340)
Carcinogenicity	Not Applicable	Suspected of causing cancer - Cat. 2 (H351)
Reproductive and Developmental Toxicity	Not Applicable	May damage fertility - Cat. 1B (H360F)

[a] Approved Criteria for Classifying Hazardous Substances [NOHSC:1008(2004)].

[b] Globally Harmonized System of Classification and Labelling of Chemicals (GHS) United Nations, 2009. Third Edition.

[*] Existing Hazard Classification. No change recommended to this classification

Advice for consumers

Products containing the chemical should be used according to the instructions on the label.

Advice for industry

Control measures

Control measures to minimise the risk from oral and dermal exposure to the chemical should be implemented in accordance with the hierarchy of controls. Approaches to minimise risk include substitution, isolation and engineering controls. Measures required to eliminate, or minimise risk arising from storing, handling and using a hazardous chemical depend on the physical form and the manner in which the chemical is used. Examples of control measures that could minimise the risk include, but are not limited to:

- using closed systems or isolating operations;
- health monitoring for any worker who is at risk of exposure to the chemical, if valid techniques are available to monitor the effect on the worker's health;
- minimising manual processes and work tasks through automating processes;
- work procedures that minimise splashes and spills;
- regularly cleaning equipment and work areas; and
- using protective equipment that is designed, constructed, and operated to ensure that the worker does not come into contact with the chemical.

Guidance on managing risks from hazardous chemicals are provided in the *Managing risks of hazardous chemicals in the workplace—Code of practice* available on the Safe Work Australia website.

Personal protective equipment should not solely be relied upon to control risk and should only be used when all other reasonably practicable control measures do not eliminate or sufficiently minimise risk. Guidance in selecting personal protective equipment can be obtained from Australian, Australian/New Zealand or other approved standards.

Obligations under workplace health and safety legislation

Information in this report should be taken into account to help meet obligations under workplace health and safety legislation as adopted by the relevant state or territory. This includes, but is not limited to:

- ensuring that hazardous chemicals are correctly classified and labelled;
- ensuring that (material) safety data sheets ((M)SDS) containing accurate information about the hazards (relating to both health hazards and physicochemical (physical) hazards) of the chemical are prepared; and
- managing risks arising from storing, handling and using a hazardous chemical.

Your work health and safety regulator should be contacted for information on the work health and safety laws in your jurisdiction.

Information on how to prepare an (M)SDS and how to label containers of hazardous chemicals are provided in relevant codes of practice such as the *Preparation of safety data sheets for hazardous chemicals—Code of practice* and *Labelling of workplace hazardous chemicals—Code of practice*, respectively. These codes of practice are available from the Safe Work Australia website.

A review of the physical hazards of the chemical has not been undertaken as part of this assessment.

References

Araki A, Saito I, Kanazawa A, Morimoto K, Nakayama K, Shibata E, Tanaka M, Takigawa T, Yoshimura T, Chikara H, Saijo Y, Kishi R (2014). Phosphorus flame retardants in indoor dust and their relation to asthma and allergies of inhabitants. Indoor Air 24(1):3-15. doi: 10.1111/ina.12054.

Bomhard EM, Krinke GJ, Rossberg WM, Skripsky TH (1997). Trimethylphosphate: A 30-month chronic toxicity/carcinogenicity study in Wistar rats with administration in drinking water. Fundamental and applied toxicology 40: 75–89

Canadian Centre for Occupational Health and Safety Registry of Toxic Effects of Chemical Substances (CCOHS RTECS). Accessed September 2018 at http://ccinfoweb2.ccohs.ca/rtecs/records/TC8225000.html

Carstensen H (1971). Inhibiting effect of a methylating agent, trimethylphosphate, on testosterone synthesis in the male rat. Life Sci I 10(16):917-25.

Carstensen H (1971). Inhibiting effect of a methylating agent, trimethylphosphate, on testosterone synthesis in the male rat. Life Sciences 10: 917-925

Cho NH and Park C (1994). Effects of dimethylmethylphosphonate (DMMP) and trimethylphosphate (TMP) on spermatogenesis of rat testis. Yonsei Medical journal 35 (2), 198-208.

Connor TH (1979). The mutagenicity of trimethylphosphate. Mutat Res 65(2):121-31.

Deichmann WB, Witherup S (1946). Observations on the effects of trimethyl phosphate upon experimental animals. J Pharmacol Exp Ther 88(4):338-42.

Fukunishi K, Teraoka M, Awata N (2000). Effect of a single treatment of trimethyl phosphate (TMP) on sperm motility and other characteristic parameters measured by the CASA-system. Teratology 62(3):46A.

Globally Harmonised System of Classification and Labelling of Chemicals (GHS) United Nations, 2009. Third edition. Accessed at http://www.unece.org/trans/danger/publi/ghs/ghs_rev03/03files_e.html

Hanna PJ, Kerr JB (1981). Antifertility activity of trimethylphosphate in male rats. Experientia 37(9):999-1001.

Harbison RD, Dwivedi C, Evans MA (1976). A proposed mechanism for trimethylphosphate-induced sterility. Toxicol Appl Pharmacol 35(3):481-90.

Jackson H, Jones AR (1968). Antifertility action and metabolism of trimethylphosphate in rodents.Nature 9;220(5167), 591-592

Kanazawa A, Saito I, Araki A, Takeda M, Ma M, Saijo Y, Kishi R (2010). Association between indoor exposure to semi-volatile organic compounds and building-related symptoms among the occupants of residential dwellings. Indoor Air. 2010 Feb;20(1):72-84. doi: 10.1111/j.1600-0668.2009.00629.x.

National Cancer Institute (NCI) Technical Report No. 81 (1978). Bioassay of trimethylphosphate for possible carcinogenicity. Accessed September 2018 at https://ntp.niehs.nih.gov/ntp/htdocs/lt_rpts/tr081.pdf

National Industrial chemicals Notification and Assessment Scheme (NICNAS).). Human health Tier II assessment for Phosphoric acid, tris(methylphenyl) ester (CAS RN 1330-78-5). Assessed at https://www.nicnas.gov.au/

National Toxicology Program (NTP). Testing status of trimethylphosphate C03781. Accessed September 2018 at https://ntp.niehs.nih.gov/testing/status/agents/ts-c03781.html

Oishi H, Oishi S; Hiraga K (1982). Toxicity of several phosphoric acid esters in rats. Toxicology Letters 13: 29–34

Organisation for Economic Co-operation and Development Screening information data set Initial Assessment Report (OECD SIAR) (1996). Trimethyl phosphate (512-56-1). Accessed September 2018 at https://hpvchemicals.oecd.org/UI/handler.axd?id=76217775-c234-44ef-95a6-df6bb5e46f5a

Perkins& Will White Paper (2014). Healthy Environments: Strategies for Avoiding Flame Retardants in the Built Environment. Sparking a conversation about opportunities to design healthier building environments. Accessed November 2018 at https://perkinswill.com/sites/default/files/PerkinsWill_FlameRetardantAlternatives.pdf

Registration, Evaluation, Authorisation and Restriction of Chemicals (REACHa). Registration dossier for Trimethyl Phosphate (CAS No. 512-56-1). Accessed September 2018 at https://echa.europa.eu/registration-dossier/-/registered-dossier/22505

Registration, Evaluation, Authorisation and Restriction of Chemicals (REACHb). Registration dossier for Triethyl Phosphate (CAS No. 78-40-0). Accessed September 2018 at https://echa.europa.eu/registration-dossier/-/registered-dossier/14738

Safe Work Australia (SWA). Hazardous Chemicals Information System (HCIS). Accessed November 2018 at http://hcis.safeworkaustralia.gov.au/HazardousChemical

Takizawa S, Katoh C, Inomata A, Horii I (1998). Flow cytometric analysis for sperm viability and counts in rats treated with trimethylphosphate or pyridoxine. J Toxicol Sci 23(1):15-23.

Toth GP, Wang SR, McCarthy H, Tocco DR, Smith MK (2000). Effects of three male reproductive toxicants on rat cauda epididymal sperm motion. Reprod Toxicol 6(6):507-515.

United Kindom (UK) Environment Agency (2003). Prioritisation of flame retardants for environmental risk assessment. Fisk PR, Girling AE and Wildey RJ. Accessed November 2018 at https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/291681/scho1008bote-e-e.pdf

United States Environmental Protection Agency (US EPA) (1985a). Health and Environmental Effects Profile for Trimethyl Phosphate. Technical Report Data. Accessed November 2018 at https://cfpub.epa.gov/ncea/risk/recordisplay.cfm?deid=33917

United States Environmental Protection Agency (US EPA) (1985a). National Service for Environmental Publications (NSCEP). Chemical Hazard Information Profile. Draft report for tri (alkyl/alkoxy) phosphates. Accessed October 2018 at www.nepis.epa.gov

United States Environmental Protection Agency (US EPA) (2010). Provisional Peer-Reviewed Toxicity Values for Trimethyl Phosphate (CASRN 512-56-1). Accessed November 2018 at https://cfpub.epa.gov/ncea/pprtv/documents/TrimethylPhosphate.pdf

United States Environmental Protection Agency (US EPA) Chemical and Product Categories (CPCat). Accessed September 2018 at https://actor.epa.gov/cpcat/faces/chemicalUse.xhtml?casrn=512-56-1

United States Environmental Protection Agency (US EPA) ChemView. Accessed September 2018 at https://chemview.epa.gov/chemview

United States National Library of Medicine's Hazardous Substances Database (HSDB). National Library of Medicine. Trimethyl Phosphate. Accessed September 2018 at https://toxnet.nlm.nih.gov/cgi-bin/sis/search2/f?./temp/~IYTPWz:3

World Health Organisation (WHO). International Chemical Safety Cards (ICSC) 0686. Accessed October 2018 at http://www.ilo.org/dyn/icsc/showcard.display?p_card_id=0686&p_version=2&p_lang=en

Last update 08 March 2019

Share this page