Propylene oxide (PO)

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Citation for most recent IARC review

IARC Monographs 60, 1994

Current evaluation:

Conclusion from the previous Monograph

Propylene oxide is *possibly carcinogenic to humans (Group 2B)*.

There is *inadequate* evidence in humans for the carcinogenicity of propylene oxide.
There is *sufficient* evidence in experimental animals for the carcinogenicity of propylene oxide.

Exposure and biomonitoring

Biomarkers of exposure or effect

There are three biomarkers of effect currently being applied to measure PO exposure in humans: DNA adducts, hemoglobin adducts, and sister chromatid exchanges (SCE). These effect biomarkers integrate exposure for different time intervals, and correlation between all three end points is not expected.

DNA adducts of propylene oxide are formed in various organs of mice, rats and dogs. PO binding in mouse liver DNA was about one-twentieth that of ethylene oxide (EO). The major adduct formed in DNA after *in vitro* reaction of PO is 7-HP-guanine, followed by the adducts 3-HP-, 1-HP-adenine, and 3-HP-cytosine. These adducts are chemically unstable: 7-HP-guanine and 3-HP-adenine will depurinate, forming apurinic sites, and 1-HP-adenine and 3-HPcytosine will spontaneously convert to *N*6-HP-adenine and 3-HP-uracil, respectively. The level of 1-HP-adenine corresponded to 2% of 7-HP-guanine. 1-HP-adenine is chemically more stable ($t_{1/2}$ = 9.2 days for rearrangement to an *N*6 adduct compared with $t_{1/2}$ = 5 days for depurination of 7-HP-guanine). Therefore, this minor adduct could be an alternative for monitoring PO exposures (Czene et al., 2002 and references therein). DNA adducts are subject to DNA repair and necrosis of the cell, which will alter the cumulative dose estimate depending if the exposure is intermittent (allowing for DNA repair time) or constant (no DNA repair time).

PO forms adducts with albumin and hemoglobin (Hb) in man, dog, rat and mouse. Adducts to Hb are especially suitable because small molecules such as PO form chemically stable adducts with the N-terminal valine of Hb, N-(3-hydroxypropyl)valine (HOPrVal). Concentration of HOPrVal (Hb) is linearly related to air concentrations of PO (see below Boogaard et al., 1999). These adducts do not appear to affect the average life span of the erythrocytes. The exposure dose as measured by determination of Hb-adduct concentrations is integrated over the average life span of human erythrocytes, which is approximately 120 days.

Hb-adducts are not subject to repairs and therefore a cumulative dose over the past 120 days is achieved.

PO may cause SCE, which is a nonmutational genotoxic event (do not alter genetic information); however, when they arise in vivo, they do indicate that a putative mutagen/carcinogen has reached the critical cellular target for direct genotoxicity. SCE is not specific to PO exposures, and may be caused by other chemicals.

For dose estimates, monitoring Hb adducts is preferred to DNA adducts for several reasons: availability of large amounts (erythrocytes in whole blood), availability of methods for chemical identification (specific and sensitive), and the well-defined life span due to absence of DNA repair (Ogawa et al., 2006). Also, the hemoglobin adduct is strongly correlated with measured concentrations of PO (8h TWA) (Boogaard et al., 1999). Limitation of Hb adducts is that they cannot be used for monitoring of acute or short term exposures.

Polymorphisms:
Hypoxanthine phosphoribosyl transferase (*HPRT*) gene has been used for studies of the mutagenic effect of EO but not for PO. High exposures to EO in workers showed a significant increase in *HPRT* mutant frequency compared to controls, while at low exposures no such association was found.

Occupational exposure

Occupational exposure occurs during the production of propylene oxide and its derivatives and during production of hydroxypropyl starch ethers. (IARC, 1994). Since the IARC review, three new occupational biomonitoring studies have been conducted: one in a chemical plant manufacturing glycols and glycol ethers from PO and two in PO producing plants. Included also is an exposure to polyethylene (PE) study because PE metabolizes to PO.

An occupational exposure study was performed to establish a relationship between ambient exposure to EO and PO in air and the formation of their respective adducts to Hb (Boogaard et al., 1999*). The personal air monitoring (PAM) data were measured by gas-diffusion monitors in the breathing zone in three groups of workers: (1) In 1990: male operators (N=20) and controls (N=36 operators from a different division of the same plant with no exposure to EO or PO) in a chemical plant manufacturing glycols and glycol ethers from EO and PO; (2) In 1997 follow-up: male operators (N=18) from the same plant maintaining and inspecting a shutdown of the plant; (3) In 1997: male workers (N=28) maintaining a styrene-PO plant during shutdown. No correlations between random PAMs and Hb-adducts were found in the 1990 survey of male operators and controls, but correlations were found between PO air concentrations spanning four months (the life span of the human erythrocyte) and the Hb-adducts of male operators in the 1997 follow-up. However, in the follow-up study, 89 of 112 PAMs (79.5%) were below the limit of detection (0.2 mg m−3 = 0.08 ppm 8-h TWA), but did allow calculations of increments for the 4-mo interval from which daily (8-h) increments and steady-state concentrations were derived. Background levels of HOPrVal were found in some controls; however, the publication does not reveal how many nor what level of HOPrVal

levels were found in controls and exposed operators. In the 1997 study of maintenance workers the median HOPrVal concentrations increased 21.3 pmol/g globin, a 2-fold increase from pre-maintenance activities (24.4 pmol/g globin) to end of shutdown period (45.7 pmol/g globin). A PO air concentration of 10 mg m-3 is equal to 5.3 nmol/g globin, and the operators' blood levels were measured to 0.005 to 0.161 nmol/g globin in 1997.

Hemoglobin adducts were assessed as a measure of PO exposure in a chemical plant using PO, EO, acrylamide and acrylonitrile in the production of surfactants for the textile industry in exposed (N=62) and controls selected from the laboratory (N=10) (Schettgen 2002). In addition to PO globin adduct (HOPrVal), other globin adducts (N-2-carbamoylethylvaline, N-2-cyanoethylvaline, HOEtVal) were also measured for workers at this plant. HOPrVal were not found in either exposed workers or controls. The authors contribute this fact to either the limit of the analytical detection (LOD, 80 pmol/g globin), or due to the low adduct formation of PO, which is five times lower than for EO. This study does not report PO air concentrations or describe what type of jobs the workers were performing.

A small study in China was conducted to determine DNA adducts (1-hydroxypropyl-adenine or 1HPAdenine), hemoglobin adducts (HOPrVal), and SCE in blood of workers (N=8) occupationally exposed to PO at a PO-producing plant and control subjects (N=8) at an institute of occupational health (Czene et al., 2002). PO concentrations were determined based on area air samples collected in areas workers occupied the day before blood samples were drawn. PO concentrations ranged from 0.9 to 6.9 ppm, and workers were reported to be present in the areas of highest PO concentration for 1 to 1.5 h per day. HOPrVal adduct concentrations in the exposed workers ranged from 0.13 to 4.92 pmol mg−1 globin for a mean of 2.69 ± 1.52 pmol mg−1 globin, which is in the same range as found in the glycol and glycol ether from PO producing plants (Boogard et al., 1999). For comparison, values for controls ranged from 0.005 to 0.008 pmol mg−1 globin. The mean difference between exposed and controls was highly significant. In this study, Hb- and DNA-adducts were measured in the same worker (Czene et al., 2002). DNA adducts were detected in 7 of the 8 exposed workers with a mean value of 0.66 ± 0.34 mol per 109 mol normal nucleotides. DNA adducts were not detected in controls. The LOD was 0.1 mol per 109 mol normal nucleotides. 1HPAadenine rather than the N7HPG adducts were measured in this study because they are more stable. The group mean difference for DNA adduct concentrations between exposed workers and controls was highly significant, as was the correlation between the individual OHPrVal hemoglobin and 1HPAdenine DNA adduct concentrations when all subjects were included in the analysis (including the controls with no detectable adducts) ($r = 0.887$, $P < 0.0001$). However, limiting the correlation to only the exposed workers resulted in a correlation with marginal statistical significance ($r = 0.713$, $p = .047$). For the third biomarker, the difference in mean SCE frequency was significant ($p \geq 0.011$) with mean SCE frequency was 3.7 ± 2.11% for the exposed workers and 2.0 ±0.52% for the control group (Czene et al., 2002). Strong and significant correlations were found between levels of N7HPG and SCE frequencies ($r = 0.792$; $p = 0.00026$) and between OHPrVal and SCEs ($r = 0.766$; $p = 0.0014$). Excluding smokers gave significant associations between SCE frequency and 1HPAdenine adducts $r = 0.851$ ($p = .00044$), and between SCE and OHPrVal $r = 0.757$ ($p = 0.011$).

Exposures to PO at three manufacturing sites in France and the Netherlands were assessed by determining Hb-adducts (n>800 samples) over a 2-year period from operators, maintenance fitters and office staff (Jones et al., 2005***). The geometric means range from the three plants was 2.9-12.6 pmol g-1 globin, which is four to 16 times lower than the other two plants mentioned previously (Boogard et al., 1999, Czene et al., 2002). To evaluate smoking as a confounder, workers at one of the plants were separated into smokers (N=51) and non-smokers (N=177) and their GMs were 3.8 and 3.0 pmol g-1 globin, respectively. Smoking does not seem to be a major confounder when measuring Hb-adduct in blood of very low exposed workers.

Polyethylene (PE) is metabolized to PO. PO concentrations in blood from healthy male non-smoking volunteers (N=4) resulting from inhalation of PE (mean concentrations of 9.82 and 23.4 ppm for 180 min) was measured to 0.44 and 0.92 nmol/l for the two PE air levels (Filser et al., 2008**). The authors found a distinct species difference between rats and human in the activities of PO metabolizing enzymes of liver and lung cell fractions. Human microsomal epoxide hydrolase activities toward PO 2-4x (liver) and 6-8x (lung) higher in humans than those of rats.

Environmental exposures
Household and industrial detergents, paints, adhesives, textiles, defoamers, oil field chemicals, cosmetics, functional fluids and lubricants, heat transfer fluids, and automotive brake fluids contain PO. PO is also an additive in food. Levels of PO exposures from using these articles have not been measured (or reported).

PO is also found in tobacco smoke and is an environmental pollutant.

In a 2-year cancer bio-assay study in rats, the authors compared the results obtained from rats being exposed to a constant PO concentration with an unexposed human. The unexposed human level of PO Hb-adduct level was 0.006 pmol/mg globin (compared to Boogaard et al., 1999 for occupational exposure of 5.32 pmol/mg globin) (Rios-Blanco et al., 2002*****).

Human carcinogenicity:
From the last Monograph (vol. 60, 1994): One case-control study provides information about cancer risk in relation to exposure to propylene oxide specifically but does not allow any firm conclusion regarding carcinogenicity. Since then only one epidemiological study has been conducted.

Mortality study of workers formerly employed in a ethylene chlorohydrin and propylene chlorohydrin process plant (Olsen et al., 1997****). The objective was to compare the SMRs at this plant with previous excess mortality from pancreatic cancer and lymphopoietic and hematopoietic cancer found among workers in another chlorohydrin unit. All male workers (N=1361) who had worked in the ethylene or propylene production area for a month and worked at either manufacturing site for a year were included in the study. These workers were identified using work histories. Vital status was determined from 1940 to 1992. SMR was non-significantly elevated for lymphopoietic and hematopoietic cancers (SMR=129; 95%CI 62-238, observed 10 cases, expected 7.7), and not elevated for malignant neoplasms

(SMR=94) and pancreatic cancer (SMR=25),. Including a latency of 25 years gave increased the SMR to 144 for lymphopoietic and hematopoietic cancers, but did not reach statistical significance (95%CI 52-312). Comparing the SMRs across plants gave similar results except for lymphopoietic and hematopoietic cancer deaths at one plant SMR=181 (95%CI 66-393, observed 6 cases and expected 3.3). Comparing SMRs across process gave similar non-significant results; however, including a latency of 25 years gave a SMR= 194 (95%CI 71-423, 6 observed, 3.1 expected,) for lymphopoietic and hematopoietic cancer among those employees with exposure only to the ethylene chlorohydrin process. At 10-20 years of employment in the chlorohydrin plants, there was a significant Mantel-Haenszel RR (RR 3.56, 95% CI 1.23-10.29) for lymphopoietic and hematopoietic cancer based on three observed deaths. This effect disappeared with different categories were used in a Poisson regression. Limitation of this study is that following exposures are not considered: cigarette smoking, the ratio of exposure to chlorohydrin versus oxide, exposure in enclosed versus outdoor production facility, earlier versus later exposure periods (usually industrial hygiene improves over the years). The mean follow-up time was 25 years and additional follow-up time might change the outcome.

Animal cancer data (excluding mechanisms):

From the last Monograph (vol. 60, 1994): Propylene oxide was tested by oral gavage in one study in rats, by inhalation in one study in mice and in three adequate studies in rats and by subcutaneous administration in one study in mice and in one study in rats.

Propylene oxide administered by oral gavage to rats produced tumours of the forestomach, which were mainly squamous-cell carcinomas. In mice exposed by inhalation, propylene oxide produced hemangiomas and hemangiosarcomas of the nasal cavity and a few malignant nasal epithelial tumours. In a study in rats of each sex exposed by inhalation, papillary adenomas of the nasal cavity were observed in males and females and thyroid adenomas and carcinomas were found in females; in the second study, in males, papillary adenomas of the nasal cavity and an increased incidence of adrenal phaeochromocytomas were observed; in the third study, in females, increased incidences of mammary fibroadenomas and adenocarcinomas were observed. Subcutaneous administration of propylene oxide to mice produced local sarcomas; the study in rats was inadequate for evaluation.

Male Fischer 344/N rats in closed exposure chambers were exposed to constant PE concentrations, between 20.1 and 3000 ppm (7 h at least) and the PO concentrations were measured in the blood of these rats (Filser et al., 2008**). The PO blood concentrations ranged from 53 nmol/l at 20.1 ppm PE to 1750 nmol/l at 3000 ppm PE. The PO blood concentrations measured in this study are too low for resulting in PO treatment–related tumors seen in a long-term study (Renne et al., 1986; U.S. National Toxicology Program, 1985b).

Mechanisms of carcinogenicity:

From the last Monograph (vol 60, 1994): In rats exposed by inhalation, there is strong uptake of propylene oxide, which is then metabolized extensively and eliminated rapidly. Metabolism occurs predominantly by conjugation with glutathione. Propylene oxide can also

be hydrolyzed by epoxide hydrolase to 1,2-propanediol, which is subsequently metabolized to lactic and pyruvic acids.

Dominant lethal mutations were not induced in rats or mice, and sperm abnormalities were not observed in mice exposed to propylene oxide in vivo. Micronuclei and, in single studies, chromosomal aberrations and sister chromatid exchange were induced in mouse bone marrow after intraperitoneal injection of propylene oxide. Neither sister chromatid exchange nor chromosomal aberrations were induced in monkeys exposed by inhalation to 300 ppm. Propylene oxide induced chromosomal aberrations and sister chromatid exchange in human lymphocytes and DNA damage, gene mutation, chromosomal aberrations and sister chromatid exchange in mammalian cells in vitro. It caused dominant lethal mutation in Drosophila and was mutagenic to yeast, fungi and bacteria.

PO binds covalently to the cyclic ring nitrogens in DNA producing hydroxypropyl (HP) adducts. HP adducts can be non-promutagenic lesions being eliminated spontaneously, leaving behind the depurinated sites that can result in mutations only if not repaired before DNA replication occurs, or the adduct may be at the coding site resulting in potentially promutagenic lesions (Albertini et al., 2007). These adducts may cause mutation by transversions. PO can also react with the phosphate groups in the DNA backbone.

PO is a less potent mutagen than the week EO mutagen.

In a 2-year cancer bioassay with rats exposed to constant PO concentration the pattern for DNA and Hb-adduct accumulation did not correlate with the incidence curve for nasal tumors (Rios-Blanco et al., 2002*****). Tumor formation was only seen at >300 ppm exposure (highest levels tested 300 and 500 ppm). Neither adduct accumulation in tissues can explain the threshold in nasal tumor formation. The authors suggest an increased cell proliferation in the nose occurs at high PO concentration to be a critical factor for tumorigenesis in this tissue.

Research needs and recommendations:

The animal data consist of oral, inhalation and subcutaneous studies in three strains of rats and two strains of mice. Propylene oxide caused tumors at or near the site of administration in rodents, causing nasal tumors after inhalation exposure (NTP, 1985).

One of the major limitations in cancer epidemiological studies is that records of exposure are often incomplete or lacking (Kolman et al., 2002). Measuring hemoglobin adducts, an extremely sensitive effect biomarker, which does not undergo repair as DNA-adducts, may overcome this problem. In addition, low background level of OHPrVal shows that contributions from non-occupational sources are minor. The levels of OHPrVal of the smokers were similar to those of the nonsmokers, indicating that tobacco smoking is not the major contributing factor to the found background levels. Therefore, a future prospective study exploring cytogenetic effect would be feasible even though PO exposures are low because:

1) the great difference in means between exposed and controls seen in the pilot study will require few participants in the two groups (exposed and unexposed workers) and

2) the OHPrVal is a sensitive and specific biomarker to PO exposures (even detected in controls with unknown exposures).

The OHPrVal adduct represents cumulative exposures over a 3-month time period. The number of workers recruited to such a study would only be about 11 assuming equal variances in the two groups; however, other possible confounders should be included in the study such as gender, age, ethnicity, years of education, smoking, and possibly polymorphisms in the GST metabolizing pathway of PO. In addition, loss of follow-up and errors in power calculations would benefit from a larger study (~ 100 exposed and 100 controls in a balanced design). Various exposure-selective cross sectional epidemiological studies that look at OHPrVal adducts and cytogenetic effects would be useful.

Possible sources of future cohorts for such a study might be the already established cohorts in PO manufacturing workers (USA) (Olsen et al., 1997), which did not show an increased mortality rate due to cancer by duration with or without latency or cancer risk by process (PO versus EO); or the manufacturing cohorts in France and the Netherlands (Jones et al., 2005), and China (Czene et al., 2008). PO production is being expanded, and workers at these new PO manufacturing sites should be recruited for a future study. Other possible cohorts for propylene oxide biomarker epidemiological studies are: processing workers where PO is used as a starting material in polyurethane polyols (NTP 11th RoC), surfactants for textiles (Schettgen et al., 2002) and glycol/glycol ether manufacturing (Boogaard et al., 1999), and manufacturing of polyethylene (PE), which metabolizes to PO. Women should be included in the study as PO might be mammary carcinogen (Rudel et al., 2007). Workers' exposures to PO in paint and automotive fluids have not been adequately characterized. If these workers have exposures to PO then these could potentially be included in a future biomarker epidemiological study. Therefore recruiting sufficient workers to a future prospective study should be feasible.

Selected relevant publications since IARC review:

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